Abstracts for

Ontario Biology Day



Department of Biology | McMaster University | March 16-17, 2013

McMaster University

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Welcome to Ontario Biology Day 2013!

On behalf of the Department of Biology at McMaster University, we welcome you to the 26th Ontario Biology Day (OBD) conference. OBD has become an annual event where the focus is a celebration of senior undergraduate research project accomplishments. This year we are hosting about 200 students from 10 universities across Ontario. Given the broad array of different topics covered in over 170 oral and poster presentations, your engagement as undergraduates in biological research has been remarkable over the past year.

We hope that you will take advantage of all of the scientific and social activities planned for OBD 2013. You will have a chance to hear from several distinguished invited speakers including an opening address from Dr. Patrick Deane, President of McMaster University. For many of you this will be your first opportunity to present your research to an audience of peers but hopefully this will be only one of many more occasions in your career. With career decisions ahead, keep in mind that OBD is a great networking event so take this opportunity to learn more about research programs underway here at McMaster and across the province Also, several of our industrial sponsors will be in attendance and in addition to thanking them for their support, use this event to find out more about careers in industry.

The OBD 2013 Organizing Committee would like to acknowledge the support of many people who have helped make this event possible. Our appreciation extends to the many McMaster graduate students, staff and faculty members who have volunteered their time to this event. We thank our many sponsors from McMaster University and industry who have made it possible for us to keep registration fees modest without compromising the scientific or social activities that OBD represents. Finally, we thank the students and supervisors who have come to share their hard work and enthusiasm for research. Without you, this conference would never happen.

Best wishes for a great conference,

The Ontario Biology Day Organizing Committee

André Bedard, Professor
George Bijelic, Manager of Instructional Programs
Amanda File, Graduate Student
Jonathan Stone, Associate Professor
Elizabeth Weretilnyk, Professor
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Saturday March 16th, Session 1, Room 1 – MDCL 1309, 1:15 – 2:45 pm
Genetics -

111

The Expression of the GABABR1 subunit in the Rat's Cochlear Nucleus

Mehwish Kamal, Department of Biology, University of Windsor

Keywords: Hearing, auditory brainstem, cochlear nucleus, GABAB receptor, immunohistochemistry

The type B γ-aminobutyric acid receptor (GABAB receptor) is a major inhibitory neurotransmitter receptor in the central nervous system. A functional GABAB receptor consists of two subunits: GABABR1 and GABABR2. We used immunohistochemical techniques to examine the level and localization of the GABABR1 and GABABR2 subunits in three brainstem auditory structures: the cochlear nucleus (CN), superior olivary complex, and nucleus of lateral lemniscus, with this presentation focused on results related to the GABABR1 in the CN. Our results indicate that the level of the receptor is higher in the molecular layer of the dorsal CN and granule cell region of the CN and lower in the ventral CN and deep layers of the CN. The level in cell body is similar across different regions in the CN. The regional distribution of immunoreactivity in the neuropil was parallel to that of the combined immunoreactivity in the neuropil and cell bodies. Our results suggest that GABAB receptors make different contributions in different regions of the CN.

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Identifying Gene Families Affected By Copy Number Variation In Mice Using The Mouse Diversity Genotyping Array

A.E. Marshall, University of Western Ontario

Gene expression; selective advantage; genome; structural variation; transcriptome

Copy number variation (CNV), the deletion or duplication of genetic material, is hypothesized to be another means by which gene expression can be altered. Depending on the location and the extent of this duplication or deletion, the expression of genes within affected regions can be greatly altered. It is not known, though, the extent to which CNVs to alter gene expression. Through the use of the Mouse Diversity Genotyping Array, genome-wide CNV studies can be performed with high-resolution for mice. By processing publically available data from this array, CNVs have been discovered within 336 mouse samples from diverse genetic backgrounds. Regions within some of the 336 mice were seen to be completely deleted, and these are referred to as having a copy number state of zero. The genes within these regions were identified as

potentially knocked out. Genes within CNVs with a copy number state of zero were categorized according to their protein products' properties. Within these deleted genes, the most enriched clusters were related to sensory perception and olfaction. Moreover, the three pathways that were most affected by these deleted genes were olfactory transduction, retinol metabolism and graft-versus-host disease. A similar result has been seen in humans, where genes involved with immunity and perception are overrepresented within CNVs. This led to the hypothesis that genes related to environmental response are most variable in copy number within mice as well. To further test this, the genes affected by CNVs in 19 wild-caught mice subject to natural selection were compared to those in 116 classical inbred mouse strains subject to artificial selection through strict breeding practices. CNVs in wild and inbred mice have both unique and shared functional enrichments. Moreover, within the genes affected by a copy number state of zero, the only pathway significantly affected is olfactory transduction in wild-caught mice. In inbred strains, the three most significantly affected pathways are graft-versus-host disease, allograft rejection, and type 1 diabetes mellitus. Differences between wild-caught and inbred mice are hypothesized to reflect different natural versus artificial selective pressures. Inbred mice have been bred in conditions of artificial selection and they also live in an environment where there are controlled conditions. Wild-caught mice, in contrast, are acted upon by natural selection and thus the genes affected by CNVs within these samples may be fine-tuned for optimal survival in the wild. In conclusion, there are some functions that are significantly overrepresented in the genes that are affected by CNVs within mice. This study indicates that regions of the genome related to environmental response in mice are very dynamic due to different selective pressures and, as a result, have variable transcriptomes.

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Effect of Environmental Toxins on Mammalian Cell Cycle Gene Expression

K. Mayrand, University of Windsor; A. Hubberstey, University of Windsor

Keywords: Natural selection, evolution, environmental toxins, aryl hydrocarbon pathway, gene expression

Natural selection puts pressure on a population to adapt to environmental changes and stressors such as toxins. I have investigated the effects of toxin Benzo(a)pyrene (BaP), on protein expression in human HepG2 cells. One way cells respond to toxic stressors is through use of tumor suppressors like p53. A second way cells may respond to toxins is through the aryl hydrocarbon (ArH) pathway. BaP activates the ArH pathway which expresses CYP1A1, an enzyme responsible for metabolizing the toxin. This results in the production of toxic metabolites. When the cells were exposed to BaP, expression of p53 increased and CYP1A1 levels decreased. p53 should be up regulated to protect the cell from toxins like BaP and CYP1A1 should decrease to prevent the buildup of toxic metabolites. This study examines potential novel interactions between cellular signaling pathways that may shed light on how cells respond to toxin exposure in their environment.

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Mechanisms Inferred For Copy Number Variants In C57BL/6J Inbred Mice and 19 Wild Caught Mice

J.A. Morris, University of Western Ontario

Mouse; DNA; Genome; Copy Number Variants; Genotyping Microarrays

Genome plasticity can range from changes at the level of single nucleotides to changes in copy number up to Megabase pairs long. Changes in copy number can be neutral or can influence gene dosage with deleterious or beneficial effects. Copy number variants (CNVs) are structural alterations of genomic DNA, such as duplications, deletions, or insertions, detectable from 500 bp to 1 Mbp with current microarray technology. CNVs are an important source of genetic variation, making it important to understand the mechanisms underlying their occurrence throughout genomes. Sequence contexts such as short interspersed elements (SINEs), long interspersed elements (LINEs), long-tandem repeats, or high GC-content can contribute to CNV formation and still exist after the formation event. An examination of sequence contexts surrounding CNV start and end points can lead to inferring mechanisms of occurrence for the event. CNVs were identified from a 336 mouse sample set using the Mouse Diversity Genotyping Array (MDGA) and a Hidden Markov Model-based approach for CNV detection to search for sequence contexts. The mouse sample set contains a wide variety of mouse strains, but only eight inbred C57BL/6J mice and 19 wild-caught mice were selected for further analysis to capture extreme genetic diversity in a smaller sample size. CNVs were analyzed from the 27 mice using the UCSC Genome Browser, to obtain 10 kilobase pairs of DNA sequence surrounding CNV start-end points, and RepeatMasker, to determine contexts within the selected sequence. Chromosome 19 was first analyzed for mechanisms of CNV occurrence. A prevalence for Alu/B1, B2-B4, and LINE1 elements surrounding CNV breakpoints was found, with the former two being SINEs and the latter a LINE. 20 random locations within the mouse genome were inspected for DNA contexts to see if Alu/B1, B2-B4, or LINE1 elements occurred more often than not at CNV breakpoints. GC-content was unchanged between DNA surrounding CNVs and randomly selected sequences. It was determined that only Alu/B1 and B2-B4 elements occur preferentially around CNV breakpoints, with implications for CNV occurrence as due to retroviral transposition. This work will contribute to understanding the CNVs within the 336 mouse sample set, facilitating its use as an effective reference when genotyping mice using the MDGA.

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The Effects of Benzo[a]Pyrene on the Developing Mammary Gland

E. Okurapa, University of Windsor; A. Issa, University of Windsor; B. Fifield, University of Windsor; R. Ferraiuolo , University of Windsor; L.A. Porter, University of Windsor. Biological Sciences, University of Windsor ON

Benzo[a]pyrene; development; mammary gland

Benzo[a]pyrene, also known as B[a]P, is a well studied polycyclic aromatic hydrocarbon that has been known to suppress the immune system eventually leading to an increase in the susceptibility to disease. At high doses this carcinogen has also been known to cause tumors in several different tissues of the body. The purpose of this study was to determine the effects of low dose exposures of B[a]P on the developing mammary gland over an extended period of time. Tissue was collected from FVB mice over different time points and the morphology of the cells was determined using hematoxylin and eosin stain. The tissue collected was also stained to detect the presence of different cell markers using immunohistochemistry. To date we have not found any statistically significant difference in the development of the mammary gland between control and

B[a]P treated virgin mice that were treated during puberty. We do however see increase in the incidence of mammary tumors in mice that were treated during puberty. Collectively, our data indicated that exposure to B[a]P over a window covering does not alter overt development of the gland. Future work is being conducted to determine if there are cellular changes not indicated in our studies. It is possible that the susceptibility to tumorigenesis is not linked to any change in early development, more comprehensive statistical analysis and fine tuning of timing of B[a]P delivery is required to make this conclusion.

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Copy Number Variants Identified By The Mouse Diversity Genotyping Array For The Purpose Of Inferring Phylogenetic Relationships Among *Mus musculus* Strains

D.C. Wallace, University of Western Ontario

Copy Number Variation; Mouse Diversity Genotyping Array; Mus musculus; Phylogeny; Bioinformatics

Copy number variants (CNVs) are a heritable form of genomic structural variation characterized by large gains or losses of genetic material (>500 bp). CNVs have been associated with disease susceptibility or resistance, and as heritable markers they have potential applications in fields such as paternity testing, forensics, and evolutionary biology. Like other genetic markers, it was hypothesized that CNVs will be more commonly shared within than between mouse strains. It was also expected that strains more highly related to one another would share a greater number of CNV events than those more distantly related. Using the Mouse Diversity Genotyping Array (MDGA), as well as CNV calling software, CNV events were identified in a large collection of *Mus musculus* strains. Hotspot Detection software identified recurrent CNVs across mice. These data were arranged into distance matrices, and software was used to infer the phylogenetic relationships among the strains from the pair-wise comparisons of CNV events. A total of 17,069 CNV events were identified using the Mouse Diversity Genotyping Array and other software. Samples of the same strain shared a greater number of CNVs than the average, and usually paired together in the inferred phylogenetic trees. The inferred phylogenetic trees closely resemble the known genealogies of these *Mus musculus* strains. These results suggest that CNV events can be used to infer phylogenetic relationships among *Mus musculus* strains. Therefore CNVs, as heritable forms of genetic variation, can be used as genetic markers. As a form of genomic variation that has been linked with disease susceptibility, studying CNVs is imperative to understanding exactly what role they play in human health. This is a pioneering effort to infer phylogenetic trees from distance matrices created using pair-wise comparisons of shared CNV events. .

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The Effects of Water Quality on Dark Septate Endophyte and Arbuscular Mycorrhiza Colonization in the Emergent Wetland Plant *Phalaris arundinacea*

B. R. Boutette, Wilfrid Laurier University, K. J. Stevens, Wilfrid Laurier University

Arbuscular mycorrhiza; Dark septate endophyte; Wetland; Phalaris arundinacea; Water quality

Symbiotic fungal associations are integral components of terrestrial ecosystems effecting ecosystem processes including ecosystem cycling, soil stability and plant community structure. Mycorrhizal associations are associations that form within plant host root systems and improve host nutrient status while the plant provides photosynthates to the fungi. Although long though absent from wetlands, mycorrhizal associations have been noted in most wetland types. Wastewater treatment plants in urban centers discharge numerous compounds derived from pharmaceutical and personal care products (PPCP) that may effect mycorrhizal associations. In laboratory studies, triclosan a common contaminant in WTTP effluent has been shown to effect arbuscular mycorrhiza (AM) colonization, spore germination and hyphal growth. Field studies to confirming these findings are lacking. The goal of this study is to determine if there are effects of water quality on AM and dark septate endophyte (DSE) colonization of the wetland plant *Phalaris arundinacea* in an urbanized watershed. Twenty-seven sites were chosen along the upper Grand River watershed to represent areas of contrasting water quality. Water quality data from the Provincial Monitoring Quality Network (PMQN) were used to categorize sites into poor, intermediate or high water quality. Root samples were obtained from 5 replicate plants per site. Roots were processed and levels of AM and DSE colonization were quantified. Preliminary data suggests a reduced colonization in poor quality sites. Total colonization (AM +DSE) was significantly lower in poor quality sites (48.3±8.89%) compared to good quality sites (65±6.01%). Intracellular structures (arbuscules, arbusculate coils and coils) tended to be higher in good quality sites (30.94±3.77%) than poor quality sites (23.6±6.43%). Further analyses are being conducted to explore relationships between colonization and water quality.

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The Effects of Reduced Diets on Tardigrade Resistance to Cyclical Freezing

A.L. Duncan, McMaster University; J.R. Stone, McMaster University; C.D Rollo, McMaster University

Tardigrade; Diet; Stress Tolerance; Growth Rate; Cryptobiosis

Tardigrades are capable of undergoing cryobiosis- entering a dormant state from which they can later be revived. This, among many other features contributes to their reputations as some of the most resilient eukaryotes in existence. An antagonistic relationship been shown in other organisms, including *mus musculus* (Rollo 2002) between protein-expression pathways that promote rapid growth (favoured when food is abundant) and pathways that promote stress tolerance (favoured when food is scarce). However, while there have been some studies into how tardigrades respond to starvation it has not yet been well documented how this particular relationship in expression affects various species already renowned for their stress tolerance.

Populations of *Hypsibius dujardini* were raised under various dietary conditions: abundant food (control), above-minimum diet, minimum diet (based on the protocols established by Goldstein et. al), and below-minimum diet. There was a single set of duplicates, for a total of eight populations. At two week intervals, representatives from each population were subjected to 1, 3, or 5 cycles of cooling to -40º C, lasting a total of 9 hours. After a recovery period of several hours, the number of individuals who successfully revived were recorded.

While data is still sparse, the first couple of trials suggest a higher rate of survivorship within some of the reduced diet groups as compared to the control group, which would suggest that a lean diet prepares a tardigrade for cryobiosis better than does a generous diet.

I anticipate this experiment opening the door to further studies identifying the precise molecular mechanisms responsible for this effect, generalizing it to other forms of stress (desiccation, radiation, etc.), generalizing it to to other organisms, or further investigating the effects of cryobiosis.

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Effects of Landscape Disturbance and Wetlands Characteristics on the Quality of Terrestrial Organic Matter in Aquatic Systems

F.B. Lester, Wilfrid Laurier University; J.C. McGeer, Wilfrid Laurier University.

Dissolved organic matter; toxicology; Hyalella azteca; excitation emission matrix spectroscopy; landscape disturbance

My research is directed to understanding variations in the quality of dissolved organic matter (DOM) across different ecosystems. Terrestrially derived DOM is an important component of aquatic systems, and landscape characteristics, such as vegetation type, could influence its quality, particularly in relation to recovery from ecosystem disturbance. The project is investigating these terrestrial-aquatic linkages by testing how forest fires, logging practices, and wetland vegetation influence DOM quality, as measured by its optical properties and ability to mitigate acute Cu toxicity. DOM mitigates against waterborne dissolved Cu toxicity by complexing Cu^{2+} ions, the most bioavailable and toxic form. Previous studies have shown that the binding capacity of DOM for Cu2+ varies depending on its source. The potential influence of ecosystem level disturbance on DOM characteristics related to functional quality has not been studied previously. DOM was collected and concentrated from 9 different streams representing burned (2), logged (2), wetland (3) and undisturbed (2) locations. Acute toxicity tests, 96 h in duration and following standard Environment Canada methods (EPS1/RM/33) assessed the capacity to bind Cu2+ and render it unavailable. All testing was done with water at a temperature of 22-22 degrees Celsius and pH of 7.3+/-0.1. The artificial soft water used in testing consisted of the following salts: 0.1 mM $CaCl_2$, 0.1 mM Na HCO_3, 0.025 mM $MgSO_4$, 0.005 mM KCl, and 0.001 mM NaBr. All sources reduced toxicity. Optical characterizations included absorbance at 340nm

and excitation emission matrix spectroscopy (EEMS) combined with parallel factor (PARAFAC) analysis. As well, through other studies (data from the Canadian Forest Service), vegetation and physical characteristics were available for sites. Landscape, optical and toxicity mitigation properties were assessed to determine how quality measures were correlated with sources characteristics.

This research will provide increased understanding of the effects of terrestrial landscape characteristics on DOM and how landscapes are altered by ecosystem disruptions (fire, logging and smelter emission). This will then be linked to DOM quality in aquatic systems. This research is supported by NSERC with contributions from Vale, Xstrata, Canadian Forest Service and the City of Sudbury.

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Using Optical Signatures of the Toxic Alga *Heterosigma akashiwo* to Delineate Cellular Physiological States

J.S.P. McCain, Western University; C.G. Trick, Western University

Harmful algal blooms; bio-optics; Heterosigma akashiwo; fluorescence excitation-emission matrix; oceanography

Harmful algal blooms (HABs) represent a significant threat to ocean ecosystems, and due to their unpredictable distribution, toxicity and duration, characterizing the magnitude of HAB events remains a challenge. Even when blooms are readily accessible, determining the community composition and toxin levels requires extensive and tedious sampling and microscopic enumeration techniques, even before understanding which of the myriad of toxins should be studied.

In this study we examined the ichthyotoxic microalga *Heterosigma akashiwo*. HAB progression of this alga involves physiologically distinct growth phases, with only the stationary phase considered to be toxic.Therefore accurate temporal characterization is critical to understanding bloom physiology. The current method for characterizing growth phase involves inefficient microscopic techniques, and the use of bio-optics in this context is relatively unexplored. We have used bio-optical characteristics to delineate physiological states, specifically absorbance and fluorescence over different growth conditions. By comparing two *Heterosigma akashiwo* isolates (one toxic isolate, one non-toxic isolate), at three distinct points in their growth, we can assess if there is an optical signal associated with the onset of toxicity.

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Aquatic Invertebrate-Macrophyte Relationships In Boreal Wetlands: Do Macrophyte Structures Or Oil Sand Process Material Constructed Wetlands Exert A Greater Effect?

J. Mockler, University of Windsor; J. Ciborowski, University of Windsor

Wetlands; Invertebrate; Macrophyte; Oil Sands

Northern Alberta is the location of the Athabasca Oil Sands. This area has been greatly impacted by human disturbance through surface mining. The oil sands companies are required to reclaim mined land to the original levels of productivity, diversity, and habitat abundance. Wetlands are an extremely important component of the boreal ecosystem, covering approximately half of the area's natural landscape. Wetlands are a crucial indicator of environmental reclamation success since they support a significant amount of biodiversity. Wetland aquatic invertebrates are an especially important indicator since they are a food source for many larger species. Macrophyte structure and water chemistry (salinity, conductivity, pH, and dissolved oxygen content) both affect the invertebrate community, but which of these factors exerts a greater effect? My research assesses the impacts of both macrophyte structure and oil sands process material on the invertebrate-macrophyte relationships of constructed wetlands.

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The Effect Of Variation In Habitat Complexity On Species Richness Using Aquatic Macrophytes As An Example.

J.I. St.Pierre, University of Windsor; K.E. Kovalenko, University of Windsor; A.K. Pollock, University of Windsor; J.J.H. Ciborowski, University of Windsor

Complexity; Macroinvertebrates; Richness; Aquatic; Macrophytes

Many studies have reported a positive relationship between habitat complexity and species diversity. However, the mechanisms behind this relationship are still unclear. Theoretical work suggests that both variation in complexity and complexity itself may mediate functional relationships among organisms, which may be the mechanism behind increase in richness. We tested the postulates that variation in complexity is more important than overall complexity in explaining macroinvertebrate richness. We sampled invertebrates associated with several species of macrophytes covering a wide complexity gradient from two wetlands in Lake Huron. Plant complexity was quantified by vertical and horizontal interstitial distances. Regression analysis was used to relate genus richness to plant species complexity, and variance of interstitial distance. Results of this experiment will bring us closer to the mechanisms by which habitat complexity affects richness in aquatic ecosystems. This has implications for the selection of protected areas and habitat restoration by the enhancement of habitat structures.

Saturday March 16th, Session 1, Room 3 – MDCL 1105, 1:15 – 2:45 pm
Molecular Biology -

311

Evidence Of Non-Clonal Multiple Triazole Resistant Populations Of Aspergillus *Fumigatus* in India

Howard Chang, McMaster University; Jianping Xu, McMaster University

Aspergillus; fumigatus; triazole; resistance; India

Aspergillus fumigatus is a widespread fungal opportunistic pathogen responsible for millions of hospitalizations worldwide that result in an alarmingly high mortality rate. The incidence of infection and mortality rate are increasing due to social and demographic changes and this is further compounded by the recent emergence of multiple triazole resistant strains which frustrate traditional therapies. *A. fumigatus* nosocomial infections are also of great concern, and more research is needed in order to understand pathogen dispersal, transmission, and evolution, thereby minimizing its burden on patients and society. It is unknown how *A. fumigatus* strains from India acquired the L98H and TR34 mutations on the Cyp51A gene and thus developed resistance to azole drugs. It has previously been suggested that the multiple triazole resistant strains found in India are of clonal origin. This study constructs the genealogy of triazole resistant strains found in India by collecting MLST data using seven loci. The results successfully rejects the hypothesis that multiple triazole resistant strains of A. *fumigatus* are of clonal origin.

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Immunity and Transcription Regulation of Lysosomal Genes

R. M. Dayam, Ryerson University; C. H. Choy, Ryerson University; and R. J. Botelho, Ryerson University

Macrophages; Lysosomes; TFEB; phagocytosis; Endocytosis

Control and prevention of infection remains a critical problem because many infectious agents have evolved strategies that circumvent the immune system. The immune system is composed of different types of cells, which interact with each other and create a network that defends our body. Immune cells such as neutrophils, lymphocytes, and macrophages are responsible for defending against pathogens as well as clearance of dying and damaged host cells. For example, macrophages are involved in phagocytosis, a process in which solid particles are ingested to form phagosomes. The newly formed phagosome then fuses with early endosomes and finally with lysosomes to form the phagolysosome. During phagosome maturation, phagosomes fuse with lysosomes which contain lysosomal hydrolases and involve in the degradation of the macromolecules. Since lysosomes play an important role in different physiological processes inside the cell, it is important to

understand what controls expression of lysosomal genes. Transcriptional factor EB (TFEB) is known to regulate lysosomal biogenesis. Under normal conditions, TFEB is phosphorylated and remains cytosolic; however, stress such as starvation causes dephosphorylation of TFEB, which leads to its nuclear translocation and an increase in the expression of lysosomal genes. We hypothesized that Fc receptor phagocytosis as well as endocytosis will activate TFEB and increase lysosome gene expression. Our preliminary data supports our hypothesis; therefore, we would like to further investigate, whether up-regulation of lysosomal genes lead to an increase in the number of lysosomes or the hydrolytic activity of the lysosomes.

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Antibiotic Resistance Profiles Of Environmentally Isolated Pathogens In Response to Stress

R.M. Guay, Wilfrid Laurier University; R.M. Slawson, Wilfrid Laurier University

Antibiotic Resistance; Low Oxygen Stress; Salmonella; Yersinia

Antibiotic resistance in the natural environment is becoming increasingly prevalent. There are many concerns associated with antibiotics such as the number of infections that are resistant to antibiotics in humans and agriculturally used animals increasing. Antibiotic resistance of *Salmonella* spp., *Enterococcus* spp., *Yersinia* spp., and *Aeromonas* spp. was tested in two different ways. First, Campy packs were used to simulate low oxygen and no oxygen environments and resistance was tested following this exposure. Second, growth curves were determined for *Salmonella* and *Yersinia* using Optical Density measurements. Antibiotic resistance was tested between log phase and stationary phase under different temperatures and oxygen availability. In the first test, no significant change in antibiotic resistance profiles was found between optimal conditions and stress conditions. In the second test, it was found that the duration of the lag phase increased and some changes in antibiotic resistance profiles were seen.

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The Effect Of Tubulation On Lysosomal Physiology

H. Gueorguieva, Ryerson University; A. Mrakovic, Ryerson University; R. Botelho, Ryerson University

lysosome; macrophages; tubulation; LPS; traffecking

Lysosomes are acidic organelles dedicated to the digestion of molecules internalized by endocytosis and phagocytosis. In most cells, lysosomes are often small and spherical, appearing as puncta (dots) when visualized by fluorescence microscopy. Indeed, this is a typical lysosomal morphology in macrophages, which are immune cells that internalize and kill bacteria and other pathogens by phagocytosis. Strikingly, a large subset of lysosomes become tubular in macrophages exposed to lipopolysaccharide (LPS), a bacterial

molecule. We do not yet understand exactly how tubulation occurs, how tubular lysosomes differ from "punctate" lysosomes and what tubular lysosomes do in immune cells. Through the use of fluorescent microscopy and fluid phase markers, we have begun to study the physiology of tubular lysososomes relative to punctate lysosomes. Previous work indicated that tubular lysosomes are extremely dynamic compared to the punctate lysosomes. Therefore, we hypothesized that endocytic cargo would arrive at tubular lysosomes faster than punctate lysosomes. To do this, we pre-labelled all lysosomes with "green" fluid-phase marker and then followed with a pulse-chase experiment with a "red" fluid-phase marker and tracked the percent localization of the two markers overtime using imaging analysis software. Our preliminary evidence suggests that trafficking reaches tubular and punctate lysosomes at similar rates. However, we observed that LPS-stimulation increased the level of endocytic uptake and labeling of all lysosomes relative to non-LPS exposed cells. We are now in the process of examining additional lysosome traits like pH and proteolytic activity.

Regulatory Role of Non-Coding RNAs in *Streptomyces*

R.A. Young, McMaster University; M.J. Moody, McMaster University; M.A. Elliot, McMaster University

Microbial genetics; RNA regulation; non-coding RNAs

Streptomyces represent a unique group of soil dwelling bacteria, known for their complex developmental cycle and ability to produce a wide array of antibiotics. Due to this morphological and metabolic complexity, a large proportion of the streptomycete genome is dedicated to regulatory functions. These regulatory networks include not only regulatory proteins, but also non-coding RNAs (ncRNAs), which regulate a diverse range of cellular processes in all organisms. Recent transcriptome analyses of three model streptomycetes (*Streptomyces coelicolor*, *Streptomyces venezuelae* and *Streptomyces avermitilis*) identified numerous conserved ncRNAs; however, the regulatory roles of each of these RNAs remain poorly understood. ncRNAs typically act by base pairing with a target mRNA and influencing its transcription, stability or translation. In bacteria, ncRNAs can be classified into three broad categories: antisense RNAs (asRNAs), cutoRNAs (convergent untranslated overlapping RNAs), and intergenic small RNAs (sRNAs). Due to their genomic context, targets for asRNAs and cutoRNAs are easily identifiable. asRNAs are transcribed from the opposite strand of a protein coding gene, and share complete complementarity with their target mRNA. cutoRNAs are a product of inefficient transcription termination at the 3′ end of a gene, and may target a mRNA transcribed from the convergent downstream gene. In contrast, identifying sRNA targets is much more difficult, as sRNAs frequently interact with multiple mRNAs, without complete complementarity. In this study, representatives from these three different classes of ncRNAs were selected for characterization in *S. coelicolor*. In particular, the timing of expression of three different asRNAs relative to their sense gene was examined using RT-PCR; results show a similar expression profile for each asRNA and its respective sense transcript. Similarly, the expression and stability of cutoRNA-associated sequences was followed using RT-PCR, which showed simultaneous expression of the 3′ UTRs of the convergent genes. Co-expression of these ncRNAs and their respective targets presents an opportunity for RNA base pairing; however, further experiments are required to validate these interactions and determine their regulatory implications. Finally, to characterize the most highly expressed sRNA in *S. coelicolor*, a deletion strain was constructed, and its phenotype was then compared to the wild-type strain. The sRNA deletion strain did not display any obvious phenotypic abnormalities, suggesting that the target may not be involved in *Streptomyces* development or secondary metabolism. Overall, these studies lay a foundation for understanding the activity of a broad range of ncRNAs in these complex and medically important bacteria.

Saturday March 16th, Session 1, Room 4 – MDCL 1102, 1:15 – 2:45 pm
Physiology (in Fish) -

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The Effect of Acute Hypoxia Exposure on the Corticotropin Releasing Factor System in Zebrafish (*Danio rerio*) Hearts

J.C. Bergstrome, University of Guelph; N.J. Bernier, University of Guelph

Corticotropin releasing factor, hypoxia, zebrafish, heart

The fish corticotropin-releasing factor (CRF) system consists of CRF, urotensin 1 (U1), urocortin 3 (UCN3), two receptors, as well as a binding protein. Of these components, only urocortin 1 (homologous to U1), UCN3, and the second receptor are present in mammalian cardiac tissue. This system is conserved throughout vertebrates and is involved in stress regulation via the hypothalamus-pituitary-interrenal axis as well as protecting organs during ischemia. Of particular interest is the protection of oxygen-dependent organs like the heart. Studies have investigated urocortin effects on ischemic hearts, and the effects of each CRF paralogue in reperfusion experiments. Although zebrafish (*Danio rerio*) have renowned abilities to endure short-term severe hypoxic conditions with minimal cardiac effects, their CRF system expression during hypoxia has not been analyzed. It was hypothesized that if CRF system components are involved in cardiac protection during ischemia in *D.rerio* hearts, then U1, UCN3 and the second CRF receptor expression will increase during hypoxia. In this project 64 adult zebrafish were divided into 4 experimental conditions: an undisturbed control group, a 40 minute hypoxic group (5% DO), a 70 minute normoxic group, and a 70 minute reoxygenation group where air was infused for 30 minutes after a 40 minute hypoxia exposure. Gene expression was quantified with q PCR. Determining the expression of all CRF components in the heart of a teleost species will make it possible to identify components that have a predominant physiological role, as well as provide insight into the CRF system's involvement in cardioprotection.

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The Impacts Of Constant And Intermittent Hypoxia Acclimation On Oxygen Transport In Killifish (*Fundulus Heteroclitus*)

B.G. Borowiec, McMaster University; K.L. Darcy, McMaster University; G.R. Scott, McMaster University

Hypoxia, intermittent, constant, gill morphology, hematology

Hypoxia is encountered worldwide both as a consequence of ecosystem dynamics and anthropogenic influences. Estuarine fish like *Fundulus heteroclitus* encounter intermittent nocturnal hypoxia in the wild, but

most research focuses on constant hypoxia. Under low-oxygen conditions, fish should attempt to improve oxygen supply for metabolism to peripheral tissues by both increasing oxygen uptake and transport efficiency. Adult, wild-caught *F. heteroclitus* were held in constant (25% or 10% relative air saturation) or nocturnal intermittent hypoxia (25% or 10% relative air saturation overnight; and 100% relative air saturation during day) for one or four weeks. Oxygen uptake ability was quantified via changes in gill morphology, specifically total filament length and total number of filaments over all four arches and the average lamellar density of the first gill arch. Oxygen transport efficiency was quantified via hematocrit and whole-blood hemoglobin concentration. Here we show paradoxical declines in oxygen uptake ability but large improvements in oxygen transport efficiency. Fish acclimated to one week of 10% and four weeks of 25% constant hypoxia saw a lower total filament length than the normoxic controls. There were no significant differences from normoxia-acclimated animals in the total number of filaments or average lamellar density across exposure lengths. In terms of hematology, fish acclimated to one and four weeks of 25% as well as one week of 10% constant hypoxia had an average hematocrit and whole-blood hemoglobin concentration that was significantly higher than animals in normoxic conditions. Examination of both respiratory and circulatory parameters in *F. heteroclitus* can provide clues as to the mechanisms used by aquatic estuarine organisms to increase oxygen supply and transport ability under different degrees and schedules of hypoxic stress as well as the sensitivity of these mechanisms.

Species Comparison of 17α-ethynylestradiol Uptake in Teleost Fish

T.L. Chow, McMaster University; T.A. Blewett, McMaster University; D.L. MacLatchy, Wilfrid Laurier University; C.M. Wood, McMaster University

17α-ethynylestradiol (EE2); Teleost; Oxygen Consumption; Species

17α-ethynylestradiol (EE2), a synthetic estrogen used in birth control and hormone replacement therapies, is a potent endocrine disruptor found in aquatic environments. The present study used radiolabelled (^{3}H) EE2 to facilitate a species comparison of EE2 uptake in four teleost fish species: japanese medaka (*Oryzias latipes*), goldfish (*Carassius auratus*), zebrafish (*Danio rerio*) and fathead minnow (*Pimephales promelas*). Fish were dosed at 100 ng/L of EE2 for 2 h at 18°C. Absolute EE2 uptake rates, EE2 tissue distribution patterns and oxygen consumption rates (MO_2) were determined. Zebrafish had the greatest absolute EE2 uptake, followed by japanese medaka, goldfish and lastly fathead minnow. Meanwhile, MO_2 rates did not significantly differ across species. Each species had a unique tissue distribution profile. In goldfish, zebrafish and fathead minnow, the greatest EE2 accumulation occurred in the carcass (over 60%), followed by the liver and gall bladder (average of 14%). However, in japanese medaka, 50% of the EE2 was metabolized in the liver and gall bladder, followed by 30% in the carcass. A linear relationship between EE2 uptake rate and MO_2 was observed only in japanese medaka (R^2=0.81, $p<0.05$) and zebrafish (R^2=0.98, $p<0.05$), while no relationship was established for the other two species. This suggests a similar pathway for EE2 and oxygen across the gills in japanese medaka and zebrafish, and an alternate uptake mechanism for goldfish and fathead minnow. This study demonstrated that total EE2 accumulation rates and tissue-specific pattern of EE2 uptake vary amongst species. These findings have austere implications for the development of predictive models for the effects of EE2 in teleost fish.

Effect Of Embryonic Anoxia Exposure On The Endocrine Stress Response Of Adult Zebrafish (*Danio rerio*).

C.M. Ivy, University of Guelph; N.J. Bernier, University of Guelph

Cortisol; Hypoxia; Social Subordination; Stress Axis Programming; Zebrafish

The ability of prenatal stress to alter the endocrine and behavioural responses to stressors in adults has been demonstrated in several mammalian species but is poorly understood in fish. In zebrafish (*Danio rerio*), while many studies have shown that stressful conditions during development can affect the physiology of adults, whether these stressors also affect the developmental programming of the hypothalamus-pituitary-interrenal (HPI) stress axis is not know. Therefore, this study assessed whether anoxia exposure (< 0.5% dissolved oxygen) during zebrafish embryonic development would impact the development of the HPI axis and result in altered stress sensitivity and cortisol response. Embryos at 36 hours post-fertilization (hpf), a sensitive developmental stage characterized by decreasing anoxia tolerance and HPI axis formation, were exposed to anoxia or maintained in normoxia conditions for 4 hours and then raised to adults. Adults from each treatment were then exposed to two secondary stressors: hypoxia (10 minute exposure to 5% dissolved oxygen) or social subordination (1 hour of dyadic interaction between sized-matched fish from each treatment). Survival from embryos to adults did not differ between treatments. Adults derived from both embryonic treatments were characterized by a similar 3-fold increase in whole body cortisol in response to the hypoxia secondary stressor, suggesting that prenatal anoxia exposure does not affect the HPI axis response of adults to hypoxic conditions. In contrast, during dyadic social interactions, adults derived from anoxia-exposed embryos were significantly more likely to become dominant (89% of the time) and had lower whole body cortisol levels than adults derived from normoxia-exposed embryos. These findings suggest that prenatal anoxia exposure may differentially impact the various stress-sensitive pathways that regulate the HPI axis in zebrafish, resulting altered stressor-specific endocrine and behavioural responses.

Effects Of Copper And Hypoxia On Killifish

N. Jabari, McMaster University; G.B. McClelland, McMaster University

Hypoxia, Copper, Environmental Stress, Expression

Aquatic wildlife is continuously exposed to environmental stress, both natural and anthropogenic, and so has evolved a multitude of physiological and biochemical mechanisms aimed at maximizing metabolic efficacy to tolerate a multitude of pressures. This study attempts to determine the combined effects of copper and hypoxia exposure in the common Killifish, *Fundulus heteroclitus*. One of the most well studied response pathways associated with hypoxia is the stabilization of a hypoxia inducible factor (HIF). Acting as a master regulator, HIF1 induces an array of cellular processes to meet reduced oxygen availability. Water contamination with metallic compounds is one other major stress factor affecting fish and their ecosystems. The effects of

elevated copper exposure have been determined to mimic those induce by hypoxia, stabilizing HIF and stimulating expression of similar response pathways, although the exact mechanism that it does so is not known. The effects of both stresses will be analyzed by quantifying changes in gene and protein expression in key factors affecting HIF stabilization. HIF1a, factor inhibiting HIF (FIH), prolyl-2-hydroxylase (PHD2), as well as one of HIF's target genes involved with iron metabolism, Ceruloplasmin (CP) will be analyzed for their gene expression in Killifish gill, liver and muscle tissue. Considering that HIF1a regulation and modification occurs at the post-translational level, analyzing its protein expression through Western Blot analysis will provide better insight as to how each tissue responds to each stress. Together, the molecular analysis of how HIF1a and its associated proteins are regulated under multiple stressors will help to not only better understand how species may tolerate a changing environment, but will hopefully strengthen the protection of the freshwater systems we depend on.

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Prolonged Exposure to Environmentally Relevant Concentrations of Venlafaxine Reduces Exploration in Round Goby (*Neogobius melanostomus*)

M.J. Lee, McMaster University

Pharmaceutical Contamination; Fish; Behaviour

There is growing public concern over pharmaceutical contamination of water bodies. Captured invasive round goby fish (*Neogobius melanostomus*) were exposed to a commonly prescribed antidepressant, Venlafaxine. We investigated this drugs effect on movement in an anxiety-inducing environment focussing on two doses (low 1µg/L and high, 10µg/L) and at two time points (14 days, 28 days post exposure). We found that neither treatment dose had an effect on rates of movement, startle responses, and exploration bouts. However, exploration rates decreased over time, suggesting that fish subjected to prolonged exposure to environmentally relevant concentrations of Venlafaxine, may undergo anxiety response modification. Additionally we found that females were more likely to explore than were males but only in the second testing period. These results suggest that the duration of contamination exposure may influence fish responses to anxiety-inducing environments, and may also differentially affect sexes. High mortality rates resulted in small sample sizes in our experiment necessitating further study on the influence of environmental Venlafaxine to substantiate the potential risks of prolonged contaminant exposure.

Saturday March 16th, Session 1, Room 5 – MDCL 1009, 1:15 – 2:45 pm
Genetics (and Evolution) -

511

Phylogenetic Coordinates in 2D Space: Visualization of Genetic Distance Between Species Based on Genomic Signatures Portrayed By Chaos Game Representation

K.A. Davis, The University of Western Ontario

Genomic Signatures; Taxonomy; Phylogeny; DNA Barcoding; Mitochondrial Genome

While evolutionary relationships have long been considered to be key aspects of biological knowledge, no universal method exists for the determination or study of these relationships. Genomic signatures, species-specific compositional biases, provide a way to compare genomes without the need to align genomes, a time consuming process that is not possible for all organisms. Genomic signatures of closely related species are more similar than those of distantly related species, which provides insight into evolutionary relationships and could form the basis for a novel method of visualization for these relationships. A Chaos Game Representation (CGR) was made for each of the 3176 complete mitochondrial genomes larger than 3kb available on NCBI. A CGR is a two-dimensional scatterplot, where each point represents one base pair in the genome sequence and the position of each point is determined by all bases previously plotted, which allows for visualization of genomic signatures. Structural SIMilarity Index (SSIM) and MultiDimensional Scaling (MDS) were used to compare the structure of CGRs, representing genomic signatures, and ultimately create a CGR Phylogenetic Plot where each mitochondrial genome is assigned an (x,y) coordinate in Euclidean Space based on the genomic signature seen in the CGR. The distribution of points in a plot is influenced by the length of the genomes being plotted as well as how thoroughly the group of species being plotted has been sampled. Closely-related species plotted near each other on these plots, indicating that they do have more similar genomic signatures, and this similarity is being recognized when plotted. Species that have diverged more recently tended to be placed towards the outer edges of a plot, while species that diverged earlier from the others tended to be placed near the centre of the plot. The placement of basal species groups, those that were the first to diverge from a common ancestor, with few representative species is not always consistent with the evolutionary literature and could be a potential limitation of this methodology. A main advantage of these plots is that any sequence can be used, as long as it is of sufficient length, and no sequence alignment is required. This means that it is possible to study genomes that share little homology, unlike other methods such as DNA barcoding and whole genome sequences are not required. Overall these plots are an effective way to quickly visualize evolutionary relationships and could aid in the placement of any newly discovered species or help solve taxonomic controversies.

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Genetic Hitchhiking

S.J. Finlayson, University of Windsor; S. Injic, University of Windsor; M.E. Cristescu, University of Windsor

Selective sweep, genetic hitchhiking, Daphnia

One of the major roadblocks in understanding ecological adaptation is examining the underlying genetic basis. In particular, local adaptation - the evolution of an organism becoming better suited to its specific environment over another – has important ecological and conservation implications. However, concealed genetic processes responsible for clearly observed ecological adaptation continue to be elusive. Looking at patterns of genetic diversity can be used to detect loci under selection. We uncovered genetic signatures of local adaptation by quantifying allelic variation by a genome wide multi-locus screen using known microsatellite markers. Identification of ecologically responsive and important loci will help better understand the mechanisms of selection as well as environmental and anthropogenic impacts on all ecosystems

Evolutionary History Of The Bacterial Cellulose Synthesis Cluster

E.A. Kell, Wilfrid Laurier University; G. Moreno-Hagelsieb, Wilfrid Laurier University

Evolution; Cellulose; Prokaryotes; Bioinformatics; Phylogenetic

This study investigated the evolutionary history of the Bacterial Cellulose Synthesis cluster of genes. In *Escherichia coli*, this cluster contains 10 genes that code for a multi-protein complex that produces and excretes cellulose as a component of bacterial biofilms. Biofilms are produced by many bacteria, this extracellular matrix aids in their attachment and survival on surfaces, both artificial and biological. Cellulose is a main component of this matrix. Understanding the evolution, and therefore the conservation, of this cluster may help to guide future studies on biofilm formation or on these genes as drug targets.

To identify homologs of the cluster genes, we selected genes whose encoded proteins matched those coded by genes in the cluster using BLAST. We used an E-value cutoff of $1x10^{-6}$ and required a minimum coverage of 60% of the sequences in the alignments. From the NCBI Refseq database genomes that were chosen for investigation were those which contained homologs to specific genes in the cluster; the pair *bcsA*, and *bcsB*, the trio *bcsE*, *bcsF*, and *bcsG*, and those which contained both. Narrowing the approximately 2100 genomes from the database down to 247. Redundant, i.e. highly similar, genomes were grouped together with a random representative genome used for further analysis, making the total number of genomes explored in this study 94. The three different subsets of genomes, those with *bcsA/bcsB*, with *bcsE/bcsF/bcsG* and those with both, were subsequently used as independent groups to produce phylogenetic and similarity trees.

Phylogenetic trees, based on the alignment of the 16S ribosomal RNA gene using SSU-align software, were produced and used as a basis of evolutionary distances between genomes. The amino-acid sequences of the main genes (*bcsA*, *bcsB*, *bcsE*, *bcsF*, and *bcsG*) were aligned, concatenated, and used to build trees based on the differences found within the Bcs cluster. The protein alignments were performed using MUSCLE and the phylogenetic trees were built using evolutionary models through the statistical program R. These trees were compared for differences in topology and congruence between distance matrices. Similarity trees, based on the genome similarity score (GSS), were also used as a measure of evolutionary distance between entire genomes. GSSs compare BLAST bit scores of every gene in common between two genomes, giving each pair

of genomes a single score of similarity; the highest score occurs when a genome is compared to itself, this score would be 1. Cluster similarity scores were calculated using the same method but only comparing the genes of interest from the Bcs cluster. Cluster similarity trees were produced using these scores and compared to the genome similarity trees, once again revealing differences in the branching and the evolution of the cluster in comparison to the rest of the genome. Congruence between the distance matrices of these trees were also compared using R. Whether the Bcs cluster evolved at the same rate as the rest of the genome, was horizontally transferred, or conserved among prokaryotes was inferred by this study.

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Stressing For Fuel: A Series Stress Protocol To Screen Wild Samples Of Microalgae For Biofuel Production

S.J. Marcotte, Laurentian University

Biofuel, Chlorophyceae, Stress, Density, Heterogeneous

The most popular system for producing biofuel from algae is to cultivate a monoculture that produces high stores of fatty acids and triacylglycerides (TAGs). This study was designed to evaluate the difference between monocultures and mixed cultures.

The second objective of the study was to ascertain whether initial culture density influenced the growth rate and production of biomass in response to exposure to environmental stressors. The proposed production site for the biofuel production is situated on a mining smelter site, where contamination is a qualified issue.

Four wild samples consisting of natural communities adapted to extreme environmental conditions were compared to laboratory reared strains, *C. vulgaris, B. braunii, S. dimorphus,* and a mixture of these strains. The experiment compares lipid production after exposure to pH stresses (pH3 and pH10) and temperature stresses (30°C, 10°C, and freezing at below -20°C), in sequence, over a period of 12 weeks.

The diversity pre- and post- exposure to stressors was based on morphological characteristics such as size, shape and where possible visible internal organelle structure and placement. Measurements of biomass accumulation was taken as a function of optical density and lipid production was measured in situ only.

The results indicated that a mixed culture would survive a series of environmental stresses and continue to produce biomass and lipids and that monocultures can be severely impacted by stress in series. Secondly dilution effects were only significant in combination with the exposure to stress in series.

Saturday March 16th, Session 1, Room 6 – MDCL 1010, 1:15 – 2:45 pm
Health (Human) -

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Stimulation Of Kupffer Cells and Liver Metastasis in Mice

S.R. Asmail, Western University; E.MK. Lui, Western University

Panax quinquefolius; liver cancer and metastasis; mouse in vivo metastatic model

Liver resident macrophages, commonly known as Kupffer cells secrete pro-inflammatory cytokines responsible for mounting effective immune responses against pathogenic attacks and metastasizing cells. In this study, the pro-inflammatory effect of the aqueous extract of *Panax quinquefolius* polysaccharides on liver cells' stimulation was examined. This was tested in two *in vivo* experiments involving 11 C57Bl/6 mice involving the administration of ginseng polysaccharides (250 mg/kg) in water *ad libitum* for 7 days, and the pretreatment of another set of 12 C57Bl/6 mice with ginseng polysaccharides (250mg/kg) in water 3 days prior to the tail vein injection of Bl6F10 melanoma cells, and throughout the 14 days following the injection. The levels of three plasma and liver homogenate cytokines (IL-1β, IL-6, and TNF) were analyzed by ELISA. Future experimental approaches will include the exclusive isolation of Kupffer cells to confirm such results *in vitro*.

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The Role of WDR1 and Isoform WDRΔ35 in Mammalian Cell Migration

N.D. Cheikh, University of Windsor; N.M. Harabor, University of Windsor; and A. Hubberstey; University of Windsor.

Actin Cytoskeleton; Cell Migration; Mutagenesis; AIP1; Cofilin

Actin cytoskeletal maintenance is imperative for cell viability due to its role in activities like cell mobility. These structures are formed by the association of globular (G) actin monomers into filamentous (F) forms which can then be recycled. Actin turnover is controlled by regulatory proteins such as Cofilin which severs actin filaments. WD Repeat Protein-1 (WDR1),mammalian homolog of Actin Interacting Protein 1 (AIP1), acts to enhance Cofilin activity and thus depolymerization of actin filaments. Overexpression of WDR1 and isoform WDRΔ35 has shown to increase cell mobility. To understand the functional role that WDR1 and WDRΔ35 play in actin turnover and cellular movement mutants were created via a PCR driven mutagenesis system and expressed in HEK293 cells. Live cell fluorescent microscopy was employed to determine migration rate.These values have yet to be calculated but may provide invaluable implications that can be applied to cancer cells which exhibit abnormal motility.

Effects Of Select Cues On Salivary pH Through Food Imagery

S. Easwaran, McMaster University

Vegetative State; Salivary pH; Conscious Awareness

Vegetative state patients are individuals that show no evidence of awareness of their environment or self, despite signs of wakefulness. Positive diagnosis of the vegetative state is a complex condition that is difficult to assess. Recent findings suggest that patients originally thought to have no consciousness may in fact have some conscious ability formerly neglected by standard clinical methods. A novel method to identify awareness is through the use of physiological measures to assess behavioural responses. A previous study demonstrates a patient's capability to communicate by manipulating their salivary pH through food imagery. Given that changes in salivary pH are a good determinant of communication response, the objective of this study is to specify cues that are best suited to elicit a stronger response. I propose that select food imagery cues can be used to enhance communication by means of producing a greater change in pH from a baseline. These changes will also reflect gender and ethnicity biases to specific cues. Test subjects are normal individuals, not of the vegetative state. Favourable flavours (i.e. Sweet) will elicit a greater change in pH than unfavourable flavours (i.e. Bitter). In addition, individuals of an Asian descent will display a greater salivary pH change from a spicy flavour cue than other ethnic groups. Results of the experiment will evaluate optimal food imagery cues and classify unique cues with respect to gender and ethnic background. This information will be useful in selecting the most effective cue for salivary pH testing in patients of a vegetative state. This research contributes to the scientific community significantly by providing improvements to a novel solution of identifying conscious awareness in vegetative state patients. This can potentially serve as a vital early-marker in altering the diagnosis of a patient from a vegetative state, where no awareness or communication is possible, to a minimally conscious state, where some awareness or communication exists. If future research and progress in communication by the means of physiological response succeed, there lies the implication of potentially providing a patient with the decision-making power of maintaining life-sustaining intervention (nutrition and hydration) and the opportunity of ending possible suffering. A tremendous impact and countless possibilities await this emerging research.

The Effects of ZNF131 Misexpression on the Cellular Proliferation of HCT116 Colon Tumour Cells

E. J. Neely, McMaster University; S. Robinson, McMaster University; J. M. Daniel, McMaster University

Colorectal Cancer; Kaiso; ZNF131; c-Myc

In 2008, colorectal cancer accounted for 608,000 deaths worldwide (WHO, 2012). Researchers around the

world have focused on understanding the progression of this devastating disease in hopes of identifying novel diagnostic markers for its prognosis and targets for its future therapeutic treatment. The transcription factor Kaiso is implicated in colorectal cancer development due to its regulation of the proto-oncogenes *cyclin D1, c-Myc* and *ID2*. Recently, it was discovered that ZNF131, a family member and heterodimerization partner of Kaiso, also interacts with these three target genes. In an effort to understand role of ZNF131 in colorectal cancer development, experiments examining the effects of ZNF131 misexpression on the protein and mRNA levels of *cyclin D1, c-Myc* and *ID2* were performed in human colon HCT116 carcinoma cells. Western blotting and RT-PCR revealed that the transient overexpression of ZNF131 could potentially have an effect on the protein and mRNA levels of *c-Myc* but little effect on the expression of *ID2* and *cyclin D1*. This preliminary result raises the possibility that a relationship could exist between *c-Myc* and ZNF131 expression and suggests that ZNF131 could play a role in promoting the cell proliferation of HCT116 colon tumour cells.

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The Effects of Cytokines From A2780 Ovarian Cancer Cell Lines On HUVEC Angiogenesis

D. Sriram, Laurentian University; A.T. Kovala, Northern Ontario School of Medicine

Cytokines, Angiogenesis, Ovarian cancer, Malignancy, HUVEC

The development of tumours is a complex process involving the interaction of the cancer cells themselves, the microenvironment in which they develop, and the initiation of processes such as angiogenesis. The stimulation of pathological angiogenesis in tumours involves tumour cell expression of angiogenic factors such as cytokines. My work focused on investigating which cytokines expressed in the A2780 ovarian cancer cell lines are most crucial for angiogenesis. When A2780 cells are treated with the chemotherapeutic agent docetaxel, the expression of a number of proangiogenic and proinflammatory cytokines are induced. Supernatants from these cells were tested for angiogenic activity using Matrigel tube formation assays. The cytokines present in the supernatants were identified using BioRad BioPlex analysis. Antagonists for the cytokines CCL2 (MCP-1), TNF-α, IL-8, and CCL5 (RANTES) were tested for inhibition of angiogenesis. Inhibition with each of the drugs resulted in a significant reduction in total node formation. This suggests that all four cytokines may play a crucial role in the regulation of pathological angiogenesis, and that they may act synergistically. Future studies will focus on confirming the roles of these cytokines in tumour angiogenesis induced by ovarian cancer cells. These studies highlight the role of cytokines in both pathological angiogenesis and the tumour microenvironment in ovarian cancer.

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Gender Differences in Neurovascular Responses to Isometric Handgrip Exercise

S. M. Thompson, University of Windsor; K. Martin, University of Windsor; J. Seifarth, University of Windsor; Dr. C. McGowan, University of Windsor

Acute exercise; isometric handgrip; resistance vessel function; blood pressure

Isometric Handgrip (IHG) training is one alternative for lowering arterial blood pressure (BP), yet the mechanisms that mediate these adaptations are unknown. Also unknown is why older normotensive women respond to IHG training with greater post-training BP reductions than age-matched males. To elucidate mechanisms underlying this variation, this study will assess neurovascular responses to an IHG bout, and determine if sex-based differences exist. Thirty participants over 60 years of age (n=15 males, 15 females) will be recruited from Essex County. Resting BP (brachial artery oscillometry) and heart rate (electrocardiography) will be measured continuously during the bout, while resistance vessel function (RVF; strain-gauge plethysmography) will be assessed pre- and post-bout. It is hypothesized that, overall, women will have greater neurovascular responses to a bout of IHG. Quantifying the IHG stimulus would provide insight into the mechanisms underlying BP adaptions to IHG exercise, which could improve the treatment and prevention of hypertension.

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Investigating The Potential Neuroprotective Effects Of The Cytokines Erythropoietin And Granulocyte Macrophage Colony-Stimulating Factor of Dopamine Neurons In The 6-Hydroxydopamine Rat Model Of Parkinson's Disease

K. Farmer, Carleton University; C.A. Rudyk, Carleton University; S. Hayley, Carleton University

Parkinson's disease; 6-OHDA; GM-CSF; EPO; Neuroprotection

Parkinson's Disease (PD) is a progressive degenerative disease that targets the dopamine neurons within the nigrostriatal pathway. Although the exact mechanistic cause of the disease is still to be eluded, there is plenty of evidence that implicates oxidative damage as the primary initiator of the disease. We induced a progressive model of hemi-Parkinsonism in rats by administering 20 µg of 6-hydroxydopamine (6-OHDA), into the anterior-dorsal striatum. The compound 6-OHDA enters the dopamine neuron via the DAT transporter protein and begins to generate reactive oxygen species (ROS); these ROS's cause physical damage to the neuron, and has been shown to induce neuronal apoptosis. At 13 and 28 days post 6-OHDA infusion, a dose of either saline, erythropoietin (EPO) (50 µg/Kg), or granulocyte macrophage-colony stimulating factor (GM-CSF) (10 µg/Kg) was given via intraperitoneal injection (IP). In our lab we have demonstrated that GM-CSF has a neuroprotective effect in other models of PD, and literature sources suggest that EPO may have a neuroprotective effect as well. A stereological count of TH positive neurons within the substantia nigra pars compacta found no significant difference in treatment groups. We did find a significant lesion within the ipsilateral striatum of the saline treatment group, which was not present in the EPO or the GM-CSF treatment groups. There was no visible lesion in the contralateral striatum of any of the animals. These findings imply that EPO and GM-CSF have a neuroprotective and regenerative effect on the nigrostriatal dopaminergic neuron dendritic projections, and may play an important role in the treatment of PD in humans.

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Effect of Withdrawal of Ubisol-QE as a Therapeutic Treatment on Paraquat Induced Neurodegeneration in Rat Model of Parkinson's Disease

S.E. Leahy, University of Windsor; S. Pandey, University of Windsor

Parkinson's disease; Ubisol-QE; Water Soluble CoQ10; Paraquat; Dopaminergic Neurons

The standard pathophysiology in neurodegenerative diseases, such as Parkinson's disease (PD), is most often caused by oxidative stress which leads to neurodegeneration. It has been shown that exposure to environmental toxins such as paraquat, a commonly used herbicide, can lead to an increase in the incidence of PD. So far, there is no effective therapeutic treatment which can halt the progression of the neurodegeneration. We use a water soluble formulation of CoQ10 (Ubisol-QE) and are testing its ability to protect neurons in vivo. It has previously been shown that Ubisol-QE prevents progression of neurodegeneration in rat models of PD post injury. The primary experiment was performed to see the effects of withdrawn treatment; therefore, paraquat exposed rats were treated (4 weeks) with Ubisol-QE, subsequently half of them had their treatment withdrawn. We evaluated the neuroprotective effects in terms of the number of tyrosine hydroxylase positive neurons in the substantia nigra region of the brain, both after treatment and after treatment withdrawal. The results suggest that Ubisol-QE has a significant effect after 8 weeks of treatment and can be used as a treatment for PD.

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Is Serotonin An Upper Or A Downer? An Adaptationist Perspective On The Neurobiology Of Rumination Prompts Reconsidering Of The Relationship Between Serotonin And Depression

P.W. Andrews, McMaster University; R.K. Lee, McMaster University;

Depression; serotonin; high serotonin hypothesis; low monoamine hypothesis; ventrolateral prefrontal cortex; rumination hypothesis

Depression is the most common psychiatric condition for which people seek help, and understanding its etiology is of wide scientific interest. The attempt to develop a unifying hypothesis for antidepressant drug action led to the hypothesis that the proximate neurochemical cause of depression is low forebrain levels of monoamines—particularly serotonin and norepinephrine. However, serotonin has never been measured directly in the brains of living human beings, so all evidence supporting the hypothesis is circumstantial. This article reviews evidence that the low monoamine hypothesis may simply have the direction of association reversed. Specifically, depression is hypothesized to be a state of high forebrain serotonin, with the caveat that this is a necessary but not a sufficient condition for depression. A new hypothesis for antidepressant drug action is proposed that is based on evidence that antidepressants of all major classes suppress forebrain serotonin synthesis, which depletes forebrain serotonin levels with extended use. This hypothesis is supported by a large animal literature on stress and depression, and by literature of depression in humans, including metabolite measurement studies, genetics, diet change, and conditions comorbid with depression. It also provides explanation for puzzling neuroimaging studies that the low monoamine hypothesis could not explain. Overall, this hypothesis is relevant to the role of depression as an evolved adaptation to promote uninterrupted analysis.

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Detailed Study of Acetylcholine Deficient Mouse Retinas by Electroretinogram Measured Oscillatory Potentials

I. S. McDonald, University of Western Ontario

Mouse, vesicular acetylcholine transport protein knockout, electroretinogram, retinal development, Starburst Amacrine Cell

Understanding the role of vesicular acetylcholine transport (VAChT) protein during development in mouse retinas is important in understanding diseases in the eye as a consequence of cholinergic (acetylcholine) deficiency. 31 mice were generated as a model organism to study VAChT-related retinal degeneration. A VAChTSix3-Cre-flox/flox genotype was used to create a VAChT retinal knockout using Cre-Lox recombination with Cre driven by Six3 in early retinal development. Structure and function of the mouse retinas were examined in to observe the response to the VAChT deficiency. The ability of light to reach the retinas was assessed using anterior segment ocular coherence tomography. The thickness of the inner plexiform layer, a cell layer in the retina thought to be affected by the VAChT deficiency, was measured using hematoxylin and eosin staining and imaging. The oscillatory potential component of the electroretinogram (ERG) was used to examine the function of starburst amacrine cells, which are the only cells in the retina that produce acetylcholine. Mice with the VAChT-deficient phenotype were predicted to have degenerated retinal function as indicated by altered oscillatory potentials. Using Excel, MATLAB and SPSS statistical software to transform and analyze data, it was determined that VAChT deficient mice had significantly lowered amplitude of the oscillating electrical potential. There were no structural abnormalities indicated by the thickness of the inner plexiform layer thickness of ocular coherence tomography. The decline in function did not increase with age, indicating that VAChT has a significant role in the development of mouse eyes.

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Epigenetic And Gene Expression Changes In The Hippocampus Of A Mouse Model Of Fetal Alcohol Spectrum Disorder

R.L. Stringer, The University of Western Ontario; E.J. Diehl, The University of Western Ontario; S.M. Singh, The University of Western Ontario

Epigenetics, DNA Methylation, Neurodevelopment, Hippocampus, Gene Expression

Fetal Alcohol Spectrum Disorder (FASD) is an umbrella term encompassing the possible outcomes of prenatal alcohol exposure (PAE). Long-term gene expression changes resulting from epigenetic modifications may be one way in which changes due to PAE are induced and maintained throughout life. The hippocampus in particular is dramatically affected by PAE and is an excellent target for specific investigation. Mice were

treated in a third trimester binge-exposure model on postnatal days 4 and 7 and hippocampal tissue was isolated following sacrifice at P70. RNA and DNA isolated from ethanol-treated and control mouse hippocampus was utilized in microarray expression and MeDIP methylation analysis, respectively. Differential expression of *Tcf7l2*, *Adamts3*, and *Sgpl1* identified through microarray technology could not be confirmed using reverse transcription qPCR. Ongoing investigation is focused on confirming differential methylation seen through MeDIP analysis. The candidate gene *Cxxc1* is currently being analyzed for differential methylation of a site containing CpG dinucleotides as well as a CTCF binding site through the use of sodium bisulfite sequencing.

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Blood-Brain Barrier Breakdown As An Early Mechanism For Neurodegeneration In A *harlequin* Mouse

J. Wagner, Western University; K. Hill, Western University

Evans Blue; Spectrophotometry; Absorbance; Cerebellum; Inflammation

The prevalence of neurodegenerative disease is increasing in Canada as the average age of the population increases, but the mechanisms for neurodegeneration are not properly understood. These diseases, such as Alzheimer's disease, Parkinson's disease and ALS, not only decrease the quality of life of those with the disease, but also cause an economic strain on the current health care system. Characterized by cerebellar and retinal degeneration, the *harlequin* (*hq*) mouse is a model of mitochondrial dysfunction with granule neuron degeneration and loss, and can be used to determine the mechanisms of neurodegenerative disease. The *hq* mouse, Alzheimer's disease, Parkinson's disease and ALS all share common features of cerebellar degneration such as disturbances in movement, increased oxidative stress, mitochondrial dysfunction, inflammation and a progressive loss of neurons. The *hq* mouse contains a downregulating mutation on the X-chromosome resulting in microglial cell activation. Microglia are macrophages of the central nervous system and are activated by apoptotic cells or foreign material. Activation leads to an inflammatory response causing further neurodegeneration, which is characterized in the *hq* mouse at 4 months of age. It is hypothesized that foreign material, entering the cerebellum, through a break in the blood-brain barrier, causes this activation of microglial cells. The permeablility of the blood brain barrier in wild type (WT) and *hq* mice was tested by measuring the amount of experimentally injected Evans blue (EB) dye, into the tail vein, of young (under four months of age) and adult mice (over seven months of age). EB binds to serum albumin in blood, which is unable to cross the blood-brain barrier unless the barrier is compromised. Spectrophotometry was used to measure the amount of EB in cerebella tissue, as the dye absorbs light at 620 nm. As a positive control, the amount of dye was also measured in the livers of all mice, as there is no barrier separating blood flow from the liver. A difference in absorbance values between the liver and cerebellum confirmed there is a blood-brain barrier present (p-value < 0.001). A difference, however, in absorbance values was observed in the cerebellum between the two age groups of *hq* mice (p-value$= 0.006$) but a difference was also found in WT mice (p-value- 0.029) so it is concluded that these are age, not genotype, related differences. There is no observed evidence for a compromised blood-brain barrier, in *hq* mice, at all age groups tested. The neurodegeneration observed in *hq* mice may be due to an internal tissue malfunction.

Saturday March 16th, Session 2, Room 2 – MDCL 1110, 3:00 – 4:30 pm

Ecology -

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Cold Tolerance in the Purple Sea Urchin, *Strongylocentrotus purpuratus*

N. Azizi, McMaster University; D. Rollo, McMaster University; J. R. Stone, McMaster University

Purple Sea Urchin; Temperature Tolerance; Environmental Biology;

Purple sea urchins (*Strongylocentrotus purpuratus*) live in the intertidal zones, and their ecological range expands from Cedros Island in Baja California to Alaska. During daily and seasonal low tide, they experience ambient temperatures (down to -20° C in winter) that they would not experience when submerged in sea water (lowest, -2° C in winter). Results from a previous experimental study showed a decrease in survivorship as water temperature approached the limits of the natural range (maximum: 23.5° C and minimum: 5° C). I hypothesized that a similar decrease in survivorship of purple sea urchins would be observed when they are exposed to 0°C and -2° C (temperatures beyond the lower natural limit). Three sea urchins were housed in each of three separate tanks maintained at 5 °C (control), 0 °C and -2 °C. After exposing each tank to its designated temperature, survivorship was observed. Presence of survivors after exposure to 0 °C and -2 °C would be a possible indication of an innate thermal (cold) tolerance ability of sea urchins due to supercooling or freeze tolerance (osmoregulation). Study of tolerance to extreme conditions, such as temperature, in other animals could be used as a starting point for finding solutions to physiological and medical complications that humans face, such as short shelf lives of donor organs and blood components.

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Étude de la morphologie comparée chez les Mustélidés du nord de l'Ontario

A.M. Belanger, Université Laurentienne; J.-F. Robitaille, Université Laurentienne

Morphologie; Mustélidés; Planimétrie; Dimorphisme sexuel

L'étude de la morphologie des animaux en utilisant des techniques d'allométrie est importante pour nous permettre de voir la variation corporelle intra- ou inter-spécifique. Des études dans ce cadre peuvent aider à percevoir si un animal possède ou non une spécialisation pour sa niche, et étudier le degré potentiel de recouvrement des niches écologiques en mesurant le degré de similarité de forme entre les espèces sympatriques d'une même guilde. L'objectif de cette étude pilote était de comparer la forme corporelle selon

l'habitat chez les Mustélidés du nord de l'Ontario à l'aide de mesures morphologiques chez les mâles et les femelles de de l'hermine, *Mustela erminea*, de la belette à longue queue, *Mustela frenata*, du vison, *Neovison vison*, de la martre, *Martes americana*, du pékan, *Martes pennanti*, et de la loutre de rivière, *Lontra canadensis*. Cette étude est basée sur l'étude des carcasses données par les trappeurs de fourrures commerciaux du nord de l'Ontario et des techniques de planimétrie pour comparer les masses corporelles, les dimensions linéaires, les rapports surface/volume et les réserves énergétiques entre les espèces. Une analyse de la présence de corrélation en utilisant la donnée R^2 démontre les tendances et aspects morphologiques qui devraient être étudiés plus profondément. Les résultats démontrent la présence du dimorphisme sexuel dans la martre, le pékan et la loutre de rivière. De plus, les résultats suggèrent qu'il existe une contrainte ou spécialisation de la forme corporelle chez l'hermine, la belette à longue queue et le vison. Il semble aussi avoir une quantité moindre de 10 % de la masse corporelle de matière grasse dans la martre et le pékan qui démontre une faible réserve énergétique. Cependant, des recherches avec un plus grand nombre de représentants par âge, sexe et espèces devraient être effectuées pour vérifier la validité des tendances de cette étude. La présente étude était limitée par la disponibilité des carcasses, la durée de ma thèse de 4^{e} année et le travail intensif requis pour étudier en détail la morphologie en rapport au budget énergétique. Le projet, sans l'aide de bases de données existantes et le travail soigné de plusieurs individus, dépasserait largement le cadre d'un projet sous-gradué.

Social Interactions Effect Competitive Feeding Behaviours in the Snail Species *Pomacea canaliculata*

K. Feather-Adams, McMaster University; S. Balshine, McMaster University; J.R. Stone, McMaster University

Competitive Behaviour; Invasive Species; Pomacea canaliculata;

The native habitat of the snail species *Pomacea canaliculata* spans from Colombia to Argentina, in the La Plata River Basin. Populations now are found predominantly Southeast Asia, in shallow, calm, turbid bodies of water, such as those occurring between the rows in rice fields and puddles on roadsides. *P. canaliculata* is characterized by a generalist diet, consisting of aquatic vegetation, and is highly tolerant to changes in environmental temperatures and water pollution. The species was largely ignored by the research community until the late 1990s, when the species became a top priority for Southeast Asian farmers, who were affected by expanding pest populations that invaded rice fields and destroyed crops. *P. canaliculata* now sits on the list of the top 100 most-invasive species in the world, and research on the species has drastically increased in the last decade. While there has been a fair amount of research conducted on the anatomy, physiology, ecology, morphology and genealogy of *P. canaliculata*, very little research has been performed on behaviour, specifically relating to feeding or competition. The macrophytophagous generalist feeding habits of *P. canaliculata* observed outside laboratory settings constitute a major component of the detrimental effect on crops and aquatic vegetation. I hypothesized that individuals surrounded by cohorts would demonstrate a higher frequency and longer duration of competitive behaviours than would individuals who were fed in isolation. Current data show that individuals in the isolation spent significantly more time eating (p-value = 0.009, 2-tailed t-test) and less time performing aggressive behaviours (p-value = 0.106, 2-tailed t-test) than did individuals in groups. It was also observed that individuals in groups performed more acts of aggression (p-value = 0.040, 2-tailed t-test) and interrupted their feeding more frequently (p-value = 0.023, 2-tailed t-test) than did individuals in isolation. Findings from this study will enable experts to explore novel options to control the invasive spread of *P. canaliculata*.

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Behavioural Analyses Of Copper And Temperature Effects On Host-Ranavirus Systems

J. Kent, Laurentian University; D. Lesbarreres, Laurentian University; J. Leduc, Laurentian University

Host-Pathogen; Ranavirus; Multiple-Stressor; Behaviour; Leopard Frog

In recent decades the incidence of amphibian declines have been on the rise, causing great concern within the scientific community. The causes of these declines have been attributed to be due to such things as emerging infectious diseases, disturbed ecosystems and increasing temperatures, however little concern has traditionally been given to the effects of these sources combined. This study was undertaken in order to explore the potential utility of behavioural analysis in order to examine multiple stressor effects on a host-ranavirus system in place of traditional methods requiring animal sacrifice. In order to analyze behavioural effects, Northern Leopard Frog tadpoles (*Lithobates [Rana] pipiens*) were filmed and analyzed for several behavioural traits such as lethargy, balance, etc., that were deemed to be indicative of metal, temperature and pathogen stress by previous research. It was discovered that these stressors did indeed have a significant impact on tadpole behaviour, therefore verifying the relevance of the use of behaviour in order to assess stress impact on declining amphibian populations without sacrifice.

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The Effects of Population Density on Juvenile Growth Rates of Painted Turtles, *Chrysemys picta*

S.L. Sanders, Laurentian University; J.D. Litzgus, Laurentian University

Chrysemys picta; Density-dependence; Juvenile growth rate; Habitat productivity

Freshwater turtles often grow to large body sizes and occur in high density populations due to the lack of predation they experience at large sizes. This suggests that other factors may limit population density to levels that permit turtle growth and development. Previous studies have revealed that turtle abundance is often driven by primary productivity of the habitat. Open water nitrogen, phosphorous and euphotic depth, were measured at five lakes in Algonquin Provincial Park, Ontario, Canada, in order to determine the effect of habitat productivity on the population densities of the resident Painted Turtles (*Chrysemys picta*), and to in turn examine the effects of population density on juvenile growth rates. Primary productivity may be responsible for variations in turtle abundance because more productive environments have been shown to support higher densities of individuals. Studies have also shown that high density populations may decrease individual growth rates due to competition for limited resources. Thus, we predicted that as habitat productivity increased we

would observe greater Painted Turtle population densities and decreased individual growth rates. Nitrogen had no affect on density; however, greater phosphorous levels were found at sites with high turtle densities. Secchi disk readings revealed that lakes with thin, but dense, layers of primary producing organisms at their surfaces may support higher densities. Density appeared to have no affect on juvenile growth rate except at extremely high densities at which growth rates decreased, possibly as a result of density-dependent effects such as competition for limited food. The high density site was also identified as one of the most productive habitats, suggesting that despite elevated productivity and therefore presumably abundant food resources, the stress of high population density had a negative effect on growth rate in this northern population of Painted Turtles.

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Response of Nearshore Benthic Macroinvertebrate Communities to Artificial Deepening of a Lake Thermocline

K.B. Watkins, Laurentian University; M.S. Gillespie, Laurentian University; J.M. Gunn, Laurentian University

Climate change; lake thermocline; benthic macroinvertebrate community; biomonitoring

Climate change is expected to increase air temperature and surface wind speeds in Boreal regions, changes that will likely deepen the thermoclines of dimictic lakes and greatly affect the nearshore habitat of benthic macroinvertebrates (Forsius et al. 2010). The purpose of this study is to determine how increased lake water temperatures will affect the biodiversity of nearshore invertebrate communities. In order to achieve this, Lac Croche, in Quebec, Canada, was experimentally mixed in 2012 in one of three basins using a Solarbee® lake mixer; in comparison to 2011, a reference year without artificial mixing. Modified Hester-Dendy (HD) traps, a type of artificial substrate, were used to sample the benthic macroinvertebrates. Three shallow sites (60-80 cm) were used in each basin, with 8 HD traps at each site. HD traps were set out in July and recovered in September. Order/family level community composition as well as total abundance and biomass were measured to calculate various biological indices for each site, basin and year. No statistical differences among sites, basins or years (experimental vs. reference) were found in the benthic invertebrate communities. However, community changes associated with the thermocline deepening were seen with other organisms in the lake (zooplankton, fish) which suggests that the nearshore community is more tolerant of whole lake changes in thermal structure. This study suggests that benthic invertebrate communities may be quite resistant to changes brought on by increased surface wind speeds that are predicted to accompany climate change in some regions.

Saturday March 16th, Session 2, Room 3 – MDCL 1105, 3:00 – 4:30 pm
Molecular Biology -

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Molecular And Genetic Analysis Of *sen1* and *sen2* Genes In The Model Eukaryote, *Schizosaccharomyces pombe*

K. V. Carias, University of Western Ontario

Schizosaccharomyces pombe; sen1; sen2; senataxin; DNA damage response

Senataxin (*SETX*) loss of function mutations are thought to contribute to neurological diseases such as ataxia with oculomotor apraxia type 2 and juvenile amyotrophic lateral sclerosis. These abnormal phenotypes may be related to the DNA repair functions of *SETX*. Interestingly, *SETX* orthologs (*sen1* and *sen2*) are present in the model eukaryote, *Schizosaccharomyces pombe*, which provides a useful model system to study their molecular function(s). Senataxins in *S. pombe* were investigated by obtaining *sen1* and *sen2* deletion mutants and testing for their hyper-sensitivity to DNA damage through treatment with hydrogen peroxide and thiabendazole. Due to the possibility of functional redundancy, *sen1 sen2* double mutants were also created through tetrad analysis and assayed for their sensitivity to DNA damaging agents. Interestingly, s*en1 sen2* double mutants, but not either single mutant, were found to be sensitive to hydrogen peroxide and thiabendazole when compared to wild type. *sen1* and *sen2* genes are important players in the DNA damage response of *S. pombe.* Future direction would be to perform a recombinase mediated exchange to express human *SETX* gene in *S. pombe* in order to determine if human *SETX* can complement the fission yeast senataxin double mutants. If complementation can be established then fission yeast may represent a useful *in vivo* model for the study of the molecular function of human *SETX.*

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Evaluating the Ability of 2,3-dimercapto-1-propanol (DMP) to Activate Nrf2 in a Huntington's Disease Cell Model

L.M. Dow, Department of Biology, Western University; R.C. Cumming (Supervisor), Department of Biology, Western University

Nrf2; Keap1; Huntington's Disease; Non-canonical; Inclusion bodies

Nuclear factor erythroid-2-related factor 2 (Nrf2) is a transcription factor that regulates the expression of antioxidant enzyme genes containing promoters with antioxidant response elements (AREs). Nrf2 is negatively regulated by Keap1 (Kelch-like-ECH-associated protein), which promotes the polyubiquitination and

degradation of Nrf2 within the cytoplasm. Upon exposure to electrophilic or oxidative stress Nrf2 is released from Keap1 and translocates to the nucleus where it stimulates the enhanced expression of antioxidant enzyme genes. Normal function of this Nrf2-Keap1-ARE system is altered in the pathological states of many neurodegenerative disorders including Huntington's Disease (HD). Previous research by the Cumming lab has shown that in rat pheochromocytoma (PC12-103Q) cells, expressing a mutant form of the protein Huntingtin (Htt), Nrf2 fails to translocate to the nucleus resulting in a significant decrease in the expression of antioxidant enzyme genes such as Peroxiredoxin-1 (Prx-1). Interestingly, 2,3-dimercapto-1-propanol or DMP is able to attenuate mutant Htt induced toxicity and restore normal expression of Prx-1. The purpose of this study was to evaluate the mechanism by which DMP activates Nrf2 via subcellular and co-localization experiments. Here we demonstrate that in mouse hippocampal (HT22) cells transfected with Nrf2-GFP, DMP treatment is associated with the aggregation of Nrf2 into nuclear granules or puncta, which decrease in size and re-locate to the cytoplasm over time. Co-transfection of HT22 cells with Nrf2-GFP and Keap1-RFP results in the co-localization of Nrf2 and Keap1 regardless of DMP treatment. However, time-lapse fluorescent microscopy revealed co-localization of Nrf2 and Keap1 in large cytoplasmic inclusions which were associated with increased cell death. In contrast, DMP treated cells exhibited smaller Nrf2/Keap1 inclusions and decreased toxicity. Future studies should explore the potential role of DMP in promoting sequestration of Keap1 or Nrf2 into autophagic vesicles or lysosomes. Arsenic, which has recently been found to activate Nrf2 via a non-canonical pathway involving the p62/SQSTM1 (sequestosome 1) protein, and forms adducts with the same cysteine residues of Keap1 as DMP. Therefore, it is possible that DMP also activates Nrf2 in a non-canonical fashion involving p62, which has known roles in both autophagy and inclusion body formation.

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Characterization of Independent Parallel *In Vitro* Selection of Catalytic DNA

R.E. Gysbers, McMaster University; K. Tram, McMaster University; Y. Li, McMaster University

DNAzymes; Deoxyribozymes; In Vitro Selection

Historically, nucleic acids have been thought to encompass an exclusive function: the storage and transfer of genetic information. The discovery and subsequent characterization of natural RNA catalysts, called ribozymes, altered this view. In addition to the discovery of natural ribozymes, several artificial ribozymes and deoxyribozymes (DNAzymes) capable of competently catalyzing a wide variety of chemical reactions were isolated *via* a method termed *in vitro* selection. Selection of DNA performed *in vitro* consists of isolating a catalytically active DNA molecule from a large random library pool through selective pressures and enrichment. Within the data encompassing deoxyribozymes, several motifs have emerged; the most characterized DNAzyme motifs capable of RNA cleavage are 8-17 and 10-23. 8-17 is known as the simplest RNA-cleaving motif and multiple independent selection procedures have led to the isolation of an assortment of 8-17 variants. The objective of this project is to select two RNA-cleaving catalysts from two pools of DNA sequences flanked by primer-binding sites. This aspiration will be achieved by artificially selecting DNA sequences which exclusively cleave a single RNA site embedded in an all-DNA substrate sequence. Characterization of the products of this experiment will lead to the advancement of the study of DNAzymes, and could impact many disciplines of science from medical therapeutics to analytical biosensors.

Search for Conserved Regulatory Motifs in the FoxP2 Promoter Using PoRC Hunter, a Newly Developed Tool

M. Pesevski, McMaster University; J.R. Stone, McMaster University

FoxP2; PoRC Hunter; Conserved Regulatory Motifs; Conserved Promoter Region

The *FoxP2* (fork-head box P2) gene codes for a transcription factor that belongs to the fork-head box family and is primarily involved in central nervous system development in vertebrates(Simon E Fisher & Scharff, 2009; Scharff & Petri, 2011). This gene was strongly linked with development of human speech and language in the 1990s through the study of the KE family who displayed symptoms of developmental verbal dyspraxia inherited in an autosomal dominant fashion (S E Fisher, Vargha-Khadem, Watkins, Monaco, & Pembrey, 1998; Hurst, Baraitser, Auger, Graham, & Norell, 1990). Since the association of *FoxP2* with development of human language, most of this research has been focused on the structure and function of the FoxP2 transcription factor and its action on downstream targets(Vernes et al., 2007, 2011). The structure of the FoxP2 transcription factor is strikingly conserved across most vertebrate species and therefore it is still unknown how and why this gene is so intimately linked with the development of language, a human-specific trait (Enard et al., 2002) Shifting the focus towards the highly understudied regulation of the *FoxP2* gene may in fact solve this mystery. In this study, we attempt to develop a better understanding of the regulation of *FoxP2* by analyzing the promoter region of this gene. The objective of this study is to develop a new computational tool called PoRC Hunter (Promoter Regulatory Conserved Hunter) that can be used to characterize the regulatory regions of *FoxP2* using conservational comparative analysis that finds conserved motifs. PoRC Hunter takes two aligned homologous sequences and searches for conserved regulatory motifs that are chosen by the user. PoRC Hunter assigns a score to each location where the motif is found. The promoter sequences from 55 different vertebrate species were aligned pairwise to the human *FoxP2* promoter and used as input for PoRC Hunter. We hypothesize that the regulatory motifs in the promoter region of FoxP2 of two homologous sequences are more similar to each other then non-functional regions because they are conserved. Therefore PoRC Hunter will assign the regulatory motifs a greater score then non-functional motifs. This study provides a stepping-stone for future studies of regulation of *FoxP2* because it attempts to characterize the promoter of this very important gene. Knowing the different elements of the human *FoxP2* promoter as well as the *FoxP2* promoter of other species will provide a great basis for studying the regulation of this gene because it can be used to identify the specific transcription factors that regulate this gene. Studying the regulation of *FoxP2* may provide evidence necessary to explain the critical difference between the human *FoxP2* gene and *FoxP2* from other vertebrates that makes language a human-specific trait. In addition, PoRC Hunter can be generalized and used in future studies for the search conserved regulatory motifs in promoters of other genes.

CTCF In Chondrocyte Differentiation

K.L. Rabicki, Western University; J.R. Bush, Western University; F. Beier, Western University

Bone Development; CTCF; Chondrocyte Differentiation; Micromass Culture; Mouse Embryonic Fibroblasts

CCCTC-binding factor, better known as CTCF, is a highly-conserved zinc-finger DNA-binding protein that is involved in gene repression and insulation, and is suspected to be a key player in tissue-specific mechanisms of gene regulation in developing cartilage. Deregulation of chondrocyte (cartilage cell) physiology results in a number of diseases, including dwarfism and skeletal deformities during development, and osteoarthritis in the adult. However, the normal processes controlling chondrocyte physiology remain incompletely understood. Through use of *in vitro* mouse embryonic fibroblast (MEF) micromass cultures the phenotype of CTCF-knockout chondrocytes was characterized for the first time. I hypothesized that chondrocyte-specific loss of CTCF would cause increased expression of genes directly related to chondrogenesis. A comparison of CTCF-knockout and control micromass cultures was achieved through use of a collagen II-specific cre/*lox* system. *Cre*-positive heterozygote micromass cultures were stained with Alcian blue and LacZ to determine the timing of chondrocyte differentiation and cre protein activation. CTCF-knockout micromass cultures were grown alongside *cre*-negative cultures for 2-8 days, and stained by Alcian blue, Von Kossa stain, and alkaline phosphatase. Stains were quantified using imaging software, then quantitative real-time polymerase chain reaction was performed to examine *Sox9, ColII, ColX, VEGF,* and *CTCF* gene expression. Preliminary findings indicate that cre protein activation in micromasses begins before day 5 and Alcian blue staining increases over time, indicating an increase in proteoglycan production within the micromass cultures. Overall, comparison of wild-type and CTCF knockout micromass models allows a cell biology-based perspective which could not be so easily attained through an *in vivo* investigation.

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The Role Of Mitotic Cyclins In Drosophila Meiosis

S. Sinha, University of Windsor; A. Swan, University of Windsor

Drosophila Melanogaster; Meiosis; Cell Cycle; Cyclins

Meiosis is a specialized cell division for haploid gamete production while mitosis results in identical diploid cells. Regulation of both cell cycles is mediated through cyclin dependent kinases (Cdks) and their cyclin partners. In *Drosophila*, there are three mitotic cyclins; A, B, and B3 whose roles in meiosis are still not well understood. My project examines the roles of these cyclins in *Drosophila* meiosis. I have two main objectives; to examine the interaction of Cyclin B3 and PP2A and to study the redundancies between cyclins. Preliminary results indicate that Cyclin B3 interacts with PP2A, a protein phosphatase that removes phosphorylations from Cdk1 targets. Results demonstrate that knocking down *cycA* in the female germline results in a nuclear envelope breakdown delay. Interestingly knocking down *cycA* in a *cycB3* null background partially rescues this delay. I will also be discussing the effect of knocking down both *cycB3* and *cycB.*

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Variation in Distribution-Abundance Relationships in Waterbird Communities and Their Implication for Conservation

M.B. Ackert, University of Guelph; T.D. Nudds, University of Guelph; N. Rooney, University of Guelph

Waterbirds; Spatial Abundance; Distribution; Detection; Ecological Specialization

The relationship between distribution and spatial abundance has been considered one of ecology's few empirical rules. Hence, it has been used, for example, to infer species' risk of extinction or conservation status. However, few studies have examined the variation that exists in the strength and form of this relationship. I aimed to first characterize the relationship for waterbird species in wetland ecosystems across Central Ontario and then to examine two potential sources of residual variation. I hypothesized that residual variation could be explained by interspecific variation in (1) ecological specialization, such that, at similar spatial abundances, ecological specialists would have smaller range sizes than ecological generalists; and (2) detectability, such that at similar range sizes, species with low detectabilities would have smaller range sizes than species that were more readily detected. I measured ecological specialization as the diversity of wetland habitats used by species, and detectability as the proportion of fifteen recordings at a site in which species were detected. Range size and spatial abundance were strongly positively related ($F_{1,18}$ = 62.75, R^2 = 0.765, $p<0.001$). Species such as Greater Yellowlegs (*Tringa melanoleuca*) and Ring-necked ducks (*Aythya collaris*) tended to have greater ranges sizes, given their spatial abundances, than Alder Flycatchers (*Empidonax alnorum*) and Great Blue Herons (*Ardea herodias*), whose ranges sizes tended to be smaller at a given spatial abundance. However, interspecific variation in neither ecological specialization nor detectability accounted for significant residual variation in the distribution-abundance relationship in this system.

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Survival And Development Of Song Sparrows (*Melospiza melodia*) During The Post-Fledging Period

A.M. Chang, Western University; L.Y. Zanette, Western University; M.J. Clinchy, University of Victoria; M.C. Allen, Western University

Population biology; Survival; Avian biology; Radiotelemetry

Predicting wildlife population trends is important for conservation purposes and requires accurate survival

models. However, in small passerine species, survival estimates during the post-fledging stage are lacking due to difficulties in monitoring birds during a critical life stage in which they are especially inconspicuous. Using radiotelemetry to establish known fate, I monitored fledgling song sparrows (*Melospiza melodia*) for up to 11 weeks post-fledge, a time period beyond previous known fate studies of post-fledging survival in small passerines. This research was conducted in the Gulf Islands National Park, BC. Based on behavioural observations, I divided the post-fledging stage into periods of dependence and independence from parental care. Physiological measures taken in the nest and at fledging were used to predict survival to different periods within the post-fledging stage. Mortality was highest immediately following fledging with 75% of birds surviving past the first eight days out of the nest. Survival to the end of the study was 56%. In the nest, skeletal measures were predictive of survival, whereas at fledging, only fat reserves were. This suggests that individuals with higher growth rates are more likely to survive, but only if they have sufficient energy reserves to cope with reduced parental provisioning following fledging.

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Evaluating The Application Of Automated Recordings To Monitor Avian Species In Forest Fragments

K.A. Grittani, McMaster University; P. Chow-Fraser, McMaster University

Avian; Recording; Monitoring; Forest

The use of automatic recording devices (ARDs) as a method of species monitoring has become a popular alternative to the traditional point count. The ARD presents a convenient way to gather large amounts of information and can prove beneficial for consistent species monitoring in fragmented or damaged areas. In this study, we aim to apply ARDs to a monitoring setting to understand the limitations and advantages of the tool and suggest a methodology for application. We performed point counts and deployed automatic recording equipment in eight sights throughout four forest fragments in Southern Ontario, gathering data from two ten-minute point counts and ten days of audio recording for each site. We compared the species detection from point counts to lab analysis of recordings to determine the abilities and limitations of each method, based on four focal species. We also evaluated practical advantages of the ARDs, using the recordings to understand how best to apply them as the only method in a monitoring setting. Random recorded periods were analyzed for each site to determine the repeatability of species detections when choosing how to analyze the large amounts of recorded data. We narrowed the scope of the project further to focus on an important interior species, the ovenbird, systematically choosing several recorded periods for each site to analyze for ovenbird detection. We suggest that while the ARD method is a viable method of species monitoring, it may prove more effective with different species. In addition to this, more analysis is required for accurate representation of the presence of a less common species at a site and the features of the ARD method make it a more suitable option for such a situation.

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Evaluation of Range Expansion of Trumpeter Swans (*Cygnus buccinator*) Re-introduced in Southwestern Ontario

S.A. Handrigan, The University of Western Ontario; M.L. Schummer, Long Point Waterfowl; S.A. Petrie, The University of Western Ontario, Long Point Waterfowl

Cgynus buccinator; Range Expansion; Density-Dependence

Trumpeter swans (*Cygnus buccinator*) were extirpated from southwestern Ontario in 1886 due to hunting and habitat loss. From 1982 to 2006, 584 trumpeter swans were released in 42 different locations within southwestern Ontario by The Ontario Trumpeter Swan Restoration Group. The population abundance of southwestern Ontario trumpeter swans is believed to be increasing in size; however, little is known about the populations breeding range. As the abundance of trumpeter swans increases, an increase in breeding range is expected if there is density dependence. The potential for range expansion has not yet been investigated. If the population is failing to expand its range, there is potential for it to become a nuisance species, as occurred in the Alaskan population. Sighting locations of marked swans collected since 1982 are being used to calculate the area of the population's home range per year by using a kernel-density estimator in ArcGIS. In conjunction with this data, the average migration distance per year will be calculated. As analyses are ongoing, the average migration distance per year is expected to increase as nesting habitats are occupied.

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Vocal Flexibility In White-Throated Sparrows (*Zonotrichia albicolis*) As A Mechanism For Avoiding Acoustic Interference From Spring Peepers (*Pseudacris crucifer*).

A.K. Lenske, University of Guelph; T.D. Nudds, University of Guelph; A.G. McAdam, University of Guelph; V.T. La, University of Guelph

White-throated sparrows; acoustic interference; background noise; spring peepers; vocal flexibility

Songbirds sing to attract mates and defend their territories. Noise that interferes with successful song transmission, therefore, could potentially have a significant impact on a bird's fitness. Background noise is common and variable in the habitats of breeding birds and can come from a number of sources including other animals. Birds can avoid the masking of their songs by background noise using a variety of strategies including singing at different frequencies, and changing the timing of when they sing. Research suggests that the higher frequency of urban bird song is the result of short-term flexibility, but there has been very little investigation into whether birds make similar frequency adjustments in response to 'natural' background noise. Additionally, research on acoustic competition in birds has so far neglected to address the possibility of inter-order acoustic competition between birds and anurans.

I tested if white-throated sparrows (*Zonotrichia albicolis*) make short-term changes to their song, including frequency changes, when spring peepers (*Pseudacris crucifer*) are chorusing. If white-throated sparrows compete acoustically with spring peepers, then song structure and/or timing will differ in the presence versus

absence of calling spring peepers so that overlap in acoustic space is reduced. I predicted that when spring peepers are calling white-throated sparrows will: (1) sing songs with different structural characteristics, reducing spectral overlap, and/or (2) will change when they sing, reducing temporal overlap with chorusing spring peepers.

Using sound recordings from central Ontario wetlands, I examined the structure and timing of white-throated sparrow songs in areas with variable background noise. I found that white-throated sparrows sang shorter songs with higher minimum frequencies and smaller bandwidths when spring peepers were chorusing. Conversely they did not adjust the timing of their songs to avoid temporal overlap with the spring peeper chorus but were less likely to sing during background noise generated by cars and other birds. Sparrows also sang significantly longer songs when conditions were windy. These results demonstrate that white-throated sparrows make changes to their singing behavioural in response to the spring peeper chorus that are similar to those birds have been observed to make in the presence of anthropogenic noise. This suggests that the flexibility seen in the songs of urban birds may have developed in response to natural background noise rather than as a novel adaptation to anthropogenic noise. Additionally, the variability observed in white-throated sparrows' response to different sources of noise suggests that the adjustments birds make depend on the type of background noise, possibly due to differences in noise duration, persistence and variability.

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Investigating Offspring Competition in a Joint-laying Bird

E.H. Nagai, McMaster University; C.J. Dey, McMaster University; J.S. Quinn, McMaster University

Joint-nesting; nestling competition; microsatellite genotyping

In joint-laying birds, multiple females contribute eggs to a single nest and share in parental care. As such, hatching order and dominance relationships among chicks in a joint-nest are likely important in determining individual fitness of co-breeding females, yet we have a poor empirical understanding of offspring competition in these unique species.

This study investigated the relative competitive ability of offspring from co-breeding females in the joint-laying pukeko (*Porphyrio melanotus* melanotus). Behavioral data collected in the field on hatching order, growth rate, and survival was used to investigate differences between clutches within a shared nest. We found that the dominant female usually lays first and that these eggs hatch earlier than the secondary clutch. Earlier hatching eggs produce chicks that grow faster and are more likely to survive.

Unfortunately, not all chicks in joint-nests were able to be assigned to a specific female. As a result, DNA was extracted from blood samples acquired from the study population. Samples were genotyped using microsatellite primers in an effort to determine parentage. Some useful primers were identified during this project but further optimization is required before parentage can be determined with confidence.

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Factors underlying the effect of toxic metals on transpiration rates in radish *(Raphanus sativus L.*)

T.N. Degazio, Laurentian University; Supervisors Peter Ryser and Fallon Kirkey, Laurentian University

Plant; Transpiration; Metal; Contaminant

Transpiration rates in plants are known to be linked with metal contamination and it has also been shown that xylem conductivity is reduced with the presence of metals. Aquaporins are membrane proteins which can serve as functional water channels within a plant and these channels can be hindered by mercury and toxic metals. Research has noticed that these proteins may have the capability of regenerating after being affected by metal contaminants such as slag, however this capability has not been monitored yet over a period of time after a plant has been transplanted from a slag contaminated environment. The central question posed is whether the reduction of transpiration by metal contamination is a result of reduced hydraulic conductivity, or whether it is a result of blocked aquaporins which aid the plant in the uptake of water in the roots. As aquaporin blockage is reversible, a recovery of transpiration would be expected in the latter case, when plant is transplanted in non-contaminated conditions.

This question was tested in in Sparkler White Tip radish (*Raphanus sativus*) since they are very fast growers and respond quickly to change. The radishes were grown in sand contaminated with 0%, 1.5%, and 3% ground slag from a Ni smelter and were then transferred to test tubes with 5 treatments: distilled water, 50 umols/L NiSO4, 100 umols/L NiSO4, 200 umols/L NiSO4, 13.9 mg/L Hg (where the concentration of Hg was calculated from the HgCl2 used), and distilled H2O. The Ni-solutions were to continue the exposure of the growth conditions, and mercury is known to block aquaporins. Transpiration was measured over 2 days as weight loss of the test-tubes. The transpiration rate per leaf area was calculated. Repeated measures ANOVAs were conducted on transpiration rate data.

Plants grown in a contaminated substrate were much smaller than those on non-contaminated substrate. Plants grown on 3% slag transpired significantly more than plants grown in the other substrates. The solutions in the test tubs had no consistent effect on transpiration. One can conclude that metal contamination of the growth substrate caused structural damage where the transpiration rates of the plants did not recover during two days in uncontaminated solutions. The contamination concentrations utilized in the test-tubes were too low to have a consistent effect on transpiration.

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Does Functional Diversity Matter? Evaluating Community Responses of Collembola After Cadmium Contamination and Phytoextraction

J.C.J. Fraser, Western University; Z. Lindo, Western University; S.M. Macfie, Western University; M. Vankoughnett, Western University

Cadmium; Phytoremediation; Collembola; Functional diversity; Community ecology

High levels of cadmium inhibit many essential ecosystem processes. However, plants reduce cadmium bioavailability by taking it from the soil and incorporating it into their roots or leaves—that is, phytoextraction. This study used collembola as bioindicators to test community changes under cadmium and phytoremediation treatments. Field soil was contaminated to 100 mg/kg with 1.0 M $CdCl_2$ solution before *Brassica juncea* and *Lactuca sativa* were added as phytoextractors. Due to their different physiologies, the two plants created different soil cadmium concentrations. Cadmium levels were highest in the contaminated soil, lower in *L. sativa* soil, and lower still in *B. juncea* soil. The plants were harvested after 4 weeks and the soil was used for a reproduction test with *Folsomia candida,* and a four-week bioassay with natural collembola communities. Effects of cadmium on the collembola communities were evaluated for each treatment using species richness, abundance, community composition, and functional diversity (FD). Overall, all community measurements are predicted to be negatively correlated with cadmium concentration and FD is expected to be a more sensitive indicator to changes within community than traditional measures. If FD proves to be a more sensitive indicator, it has many applications in environmental management and conservation biology.

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Does Social Environment Affect Growth And Allocation In Paperwhite Narcissus (Daffodil)

A. Jennings, McMaster University; S.A. Dudley, McMaster University

Narcissus; Allocation; Social Environment; Competition

Recent studies show that plants are able to sense and respond to the relatedness of their neighbours through changes in morphology and allocation. Previous studies have examined allocation strategies in annual plant species and to date, none have looked at a perennial plant species. Perennial plants with belowground storage organs face several decisions during the growing season that may affect their allocation patterns. These choices include using stored carbon resources from the past year or photosynthate from the current year for growth, reproducing either sexually (seeds), asexually (offsets) or both, and storing resources for increased growth the following year. The paperwhite daffodil (Narcissus) represents an ideal species to investigate these allocation decisions because they have a short generation time and do not require cold stratification prior to planting. In this study, we used two different paperwhite daffodil cultivars to ask the following questions;

1. Do paperwhite daffodils exhibit phenotypic plasticity in either their above-or belowground morphology and allocation in response to neighbour identity?
2. Does offset production affect allocation to bulb growth?

3. Does bulb density affect growth and biomass allocation?

To answer these questions, I manipulated the social environment of paperwhite bulbs by planting them in alone, in same-cultivar pairs, in mixed cultivar pairs, and mixed cultivar groups of four. One-half the pots were harvested in February and allocation to above and belowground biomass was measured. The remaining pots will be allowed to complete their growth so that final allocation and seed production can be assessed.

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Molecular Analysis Of Red Maple (Acer rubrum) Populations From Areas Contaminated With Metals In The Greater Sudbury Region

K. Kalubi, Laurentian University; Dr. M. Mehes-Smith, Laurentian University; Dr.Omri, Laurentian University

Polymorphic loci, genetic variation, ISSR analysis

Many decades of sulfuric acid and other mineral emissions from smelters combined with water vapor from the atmosphere forming sulfuric acid causing acid rain has had a huge impact on the environment. The main objective of this study is to compare the levels of genetic variation among and within red maple populations from areas with different metal contents within the Greater Sudbury Region. Soil from contaminated and uncontaminated sites will be analyzed to determine the current levels of metal content in the ecosystems. There were higher levels of metals in soil samples from sites from Sudbury within 15km from the smelters compared to the control site. No significant differences in pH were found among the sites since all the values were typical of the Canadian Shield ranges. ISSR analysis revealed the level of polymorphic loci ranging from 58% to 75%. The level of genetic variation was higher in control sites than in metal contaminated sites. The populations analyzed were genetically closely related but two different groups were identified. The results of the present study indicate that red maple populations in the Greater Sudbury region are sustainable. Physiological analysis and further genetic studies suggested determining how red maple copes with metal accumulation in soil.

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Identifying Genetic Loci Involved in Kin Recognition in *Arabidopsis*

P.D. Langille, McMaster University; S.A. Dudley, McMaster University

Plants; Genetics; Ecology

Kin recognition has been observed in several plant species, and has been linked to the soluble molecules found in root exudates. Previous studies have done transcriptome analysis of *Arabidopsis* to identify genes involved mechanistically in kin recognition. In this study I attempted to find quantitative trait loci involved with

kin recognition in *Arabidopsis thaliana.* Recombinant inbred lines of *Arabidopsis* represent fixed chromosomal recombination between 2 different *Arabidopsis* ecotypes. A social environment was simulated by growing RILs in liquid media containing the root exudates of either a RIL of the same genotype, a parent ecotype, or an ecotype not involved in creating the RIL. Quantitative root traits were measured, and the longest lateral root length was found to be affected by social environment. Quantitative trait loci mapping was used to find genetic markers that have an effect on lateral root length depending on their social environment and which parent the marker was inherited from. Four such markers were identified and found to be located close to markers identified in a previous study by Loudet et al. (2003) to play a role in root growth using this RIL population, or close to genes identified by Biedryzcki et. al. (2011) in a transcriptome analysis of kin recognition in *Arabidopsis*. The genes located near these markers would be ideal candidates for future genetic studies of kin recognition in Arabidopsis.

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The Mapping Of Lichen Diversity On Street Trees In London, Ontario

A. Timoshenko, Western University; R.G. Thorn, Western University

Lichen; Traffic; London; Air pollution

Different species of lichens growing on tree trunks show differential sensitivity to some of the harmful compounds in polluted air, and so these epiphytic lichens have been used worldwide as biomonitors. As one of the main sources of air pollution, road traffic has been linked to lichen diversity. Although many studies relating lichen community composition to air pollution have been conducted throughout the world, no such study has been done in London, Ontario. Ten sampling units representing each of three traffic volume levels (in units of thousands of cars per day: low – 0-14.9, medium – 15-29.9, and high - 30+) were selected from across London. For each traffic volume level, five sampling units were on streets running East-West and five running North-South. From each sampling unit, five Norway maples (*Acer platanoides*) with a diameter of at least 20 cm were sampled, once from the side of the tree facing the road, and once from the side facing the sidewalk. Sampling involved taking a picture of a 10 x 15 cm quadrat of bark and recording the lichen species present in each quadrat and their respective percent cover. Bark pH measurements were obtained twice per tree as well, from the sides facing the road and facing the sidewalk. Tree diameter, distance from road and shading were also noted. A two-way ANOVA was conducted that explored the effect of traffic volume level and street direction on total lichen cover per quadrat. Simple main effects showed that high traffic volume sampling units had significantly less lichen cover than medium and low sampling units ($p=0.01$), but there were no differences in lichen cover between the different street directions. A paired t-test revealed no differences in bark pH between east- and west-, and north- and south-facing samples. The low lichen coverage on high volume street trees is likely due to variables caused by site location. Most of the high volume sampling units were located on the outskirts of the city, and often had very few trees and mostly low-rise buildings on them, which would increase the lichens' exposure to wind and sun, and remove any barriers from these elements. Traffic volume was expected to impact bark pH (due to road traffic emissions of as SO_2 and NO_x), but the effects may have been minimized by the Norway Maple's bark pH buffering capacity, and thus did not yield significant results.

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Testing The Feasibility Of Histologically Quantifying Gastritis Using Retrospective Necropsy Tissue

A.V.E. Colles, Laurentian University

Inflammation; Histology; Small Intestine; Feasibility; Ferret

Gastritis, the immune response of inflammation within the digestive tract, is commonly found among several captive species, ranging from house pets such as dogs (*Canis lupus familiaris)* and domestic ferrets (*Mustela putorius furo)*, to larger species such as cheetahs (*Acinonyx jubatus)*. The aim of this study was to test the feasibility of using archived histological sections of small intestine from necropsy material for quantifying the presence of gastric inflammation. Specifically, the reliability, accuracy, applicability, and potential for standardization of this methodology were investigated. Sections from domestic ferrets and black-footed ferrets were used; these sections were photographed and counts were taken, including mitotic figures, leucocytes, and fibroblasts. Investigation of current analytical methods for inflammation is important, as there is little standardization currently available. The ability to detect gastritis is essential for treatment of these animals in both home and zoo settings; additionally, it can be used to test hypotheses relating environmental changes to the immune system.

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Effects Of *Lactobacillus rhamnosus* And *Lactobacillus reuteri* On Loperamide-induced Constipation In BALB/c Mice

J.T.Y. Tam, McMaster University

Probiotics; Prebiotics; Lactobacilli; Constipation; Motility

The efficacy of probiotic treatments on constipation has shown inconclusive results in both animal and human models. Lactobacilli have been shown to potentially show beneficial effects on gut motility and stool evacuation when administered orally. We investigated the effects of *Lactobacillus rhamnosus* (JB-1), *Lactobacillus reuteri* (DSM), and prebiotic 2'fucosyllactose on loperamide-induced constipation in BALB/c mice. We also studied the potential involvement of vasoactive intestinal peptide or the mast cell stabilizing agent doxantrazole on such models. 2'fucosyllactose increased stool production in normal mice but not in constipated mice whilst DSM showed laxative effects on mice with loperamide-induced constipation. However, JB-1 did not show any significant effects upon administration. Furthermore, intraperitoneal injections of vasoactive intestinal peptide prior to treatment did not induce significant effects to stool production. These findings show that certain

probiotics can be effective in treating constipation and gut dysmotility although further research is needed on the detailed mechanisms by which this occurs.

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Examining The Temporal Patterns Of Irritable Bowel Syndrome (IBS) And The Association With Psychiatric Comorbidities And Low-Grade Intestinal Inflammation.

C. Welsh, McMaster University; P. Bercik, MD, Department of Medicine, McMaster University

Irritable bowel syndrome (IBS); Gut; Microbiota; Birmingham

Irritable bowel syndrome (IBS) is a chronic condition that greatly impacts quality of life. IBS is characterized by abdominal pain, altered bowel habits with diarrhea and/or constipation, which are frequently accompanied by psychiatric comorbidities, such as anxiety and depression. IBS affects an estimated 7-10% of the population worldwide and costs health care systems billions of dollars annually. Although its pathogenesis is not fully elucidated, low-grade gut inflammation is found in a significant proportion of patients with IBS. It is characterized by various polymorphonuclear and mononuclear cells in the intestinal mucosa as well as increased levels of pro- inflammatory cytokines. Recent studies have also found abnormal composition and instability of gut microbiota in patients with IBS. Bacteria are known to modulate gut function and affect the gut immune system and recent studies demonstrated that microbiota can also interact with the CNS. The aim of this study is to investigate relationship between changes in microbiota, gut symptoms and mood in patients with IBS. A cohort of 30 patients will be followed for 6 months and their symptoms and stool samples collected weekly. It is hypothesized that IBS patients will have bouts of dysbiosis (altered microbiota composition) accompanied by increased immune activation as assessed by human β-denfensin-2 (HBD-2), and/or anxiety/depression that will be directly proportional to the severity of IBS symptoms. This study is utilizing REDCap database software for data compilation and analysis. Preliminary analysis found that there are constant fluctuations in patients' IBS symptoms over the 25-week period. There appears to be a correlation between IBS symptoms severity, anxiety, depression and physical symptoms. This is the first study to link the temporal pattern of IBS symptoms to changes in gut microbiota, low-grade intestinal inflammation and psychiatric comorbidity. The results of this trial will likely advance our understanding of pathogenesis of IBS.

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A Gut Feeling about the Role of Kaiso and PPARγ in the Mammalian Intestines

D. Wojtal, McMaster University; R. Chaudhary, McMaster University; C.C. Pierre, McMaster University; J.M. Daniel, McMaster University

Recently, the nuclear hormone receptor peroxisome proliferator-activated receptor γ (PPARγ) was identified as a putative target of the canonical Wnt signalling pathway. The canonical Wnt signalling pathway plays a key role in homeostasis of mammalian intestines. However, when this pathway malfunctions, it gives rise to cancer. Currently, there is a discrepancy regarding the role of PPARγ in colorectal cancer. Studies have shown an increased number of polyps in $APC^{Min/+}$ mice when subjected to PPARγ agonists, implicating PPARγ in an oncogenic role. However other studies in human colon tumour cell lines implicated PPARγ as a tumour suppressor. In order to resolve this discrepancy, a possible mechanism of PPARγ's function in a mammalian colon cancer model must be determined. As a first step to elucidating a mechanism, we aim to determine if there is a correlation between PPARγ expression and Kaiso , a transcription factor which co-regulates a subset of Wnt/β -catenin that promote cell proliferation and invasion. Kaiso has been identified as a co-repressor in complex with SMRT, which has been implicated in the downregulation of PPARγ however PPARγ has not yet been established as a "*bona fide*" Kaiso target gene. In addition, PPARγ is of particular interest as it is the master regulator of adipogenesis, and we have recently found that intestinal-specific Kaiso overexpressing mice, $Kaiso^{TG/+}$, are smaller than Non^{TG} littermates and have less body fat. The same trend was observed when $Kaiso^{TG/+}$ were mated with the $APC^{Min/+}$ mice; their progeny $Kaiso^{TG/+}$*;* $APC^{Min/+}$ mice also had a decreased body size and weight. Misexpression of Kaiso in human colon cancer cell lines HCT 116 as well as in vivo in non transgenic mice, $Kaiso^{TG/+}$, and the cancer models $APC^{Min/+}$ and $APC^{Min/+}$*;* $Kaiso^{TG/+}$ suggests that Kaiso regulates PPARγ on the protein level as seen through immunoblotting and immunohistochemistry and not transcriptional level as observed though reverse-transcription PCR.

Sunday March 17th, Session 3, Room 1 – MDCL 1309, 9:00 – 10:30 am
Environment -

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The Effect of Drought Stress on Sphagnum CO_2 Exchange: Contrasting Hummocks and Hollows across a Hydrological Gradient

L.A. Breadner, McMaster University; P.A. Moore, McMaster University; J.M. Waddington, McMaster University

Sphagnum; drought stress; CO_2 exchange; species plasticity

With current concerns over global climate change peatlands are gaining increasing value as a potential carbon sink. However, peatlands also pose a risk as a possible source of atmospheric carbon as they are vulnerable to environmental factors that induce negative NPP in Sphagnum species. Drought is one such environmental factor, causing reductions in Sphagnum productivity. The aim of this study is to compare the effect of changes in water availability on hummock and hollow productivity as well as productivity in Sphagnum from sites of differing hydrology.

Three hummock and three hollow samples were taken from a wet site, a dry site and a site of intermediate water availability. All samples consisted of a mixture of *S. Capillifolium* and *S. Angustifolium*. CO_2 flux measurements were taken for each sample as they were drained on pressure plates at increasing tension increments. Draining was followed by a period of re-saturation during which CO_2 flux measurements were taken regularly over a period of 34 days.

The CO_2 flux response to increases in tension did not differ between hummocks and hollows or across sites. Within sites, only hummocks and hollows at the intermediate site expressed differences in GPP response to tension. These similarities in CO_2 flux occurred despite differences in capitulum density across sites and between hummocks and hollows. At -500mb tension, NPP values were surprisingly high at an average of 16% of a sample's pre-drying NPP flux. During the 34 day re-saturation period only hummocks from the intermediate site showed a significant degree of recovery. All other samples maintained constant NPP values roughly 10% to 25% of that expressed before the onset of drought stress. This study provides an example not only of Sphagnum's ability to resist drought stress at high tension values, but also species plasticity in *S. Capillifolium* and *S. Angustifolium*. The results presented here support the necessity for the incorporation of species adaptation in peatland predictive modeling.

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Cold Hardiness Strategy of the Western Bean Cutworm (*Striacosta albicosta*) and the Effects of Warmer Winters on their Eastward Range Expansion

P.L. Charlebois-Page, Western University; H. Udaka, Western University; J.N. McNeil, Western University

Western bean cutworm; Cold Hardiness; Freeze intolerance; Range expansion, Overwintering

In the past decade, Western bean cutworm (WBC) *Striacosta albicosta* (Smith) (Lepidoptera: Noctuidae), a univoltine pest of dry beans and corn, has expanded its range eastward in North America. It has been proposed that milder winters, as a result of climate change, may have allowed permanent populations to establish in the Great Lakes region, but currently nothing is known about the overwintering capacity of the WBC. In order to survive winter conditions, insects use one of two main cold hardiness strategies that involve significant physiological changes: freeze intolerance or freeze tolerance. A preliminary experiment shows that the WBC was freeze intolerant so this species would not be able to establish permanent populations when temperatures in overwintering habitats consistently fall below the temperature at which they freeze. I determined the supercooling point (SCP) values (the temperature at which freezing starts/ice crystals begin to form) of newly formed prepupae that had been reared throughout larval development under controlled laboratory conditions (20±1°C/15±1°C (L:D); 70 ±5% RH; 16L:8D photoperiod) or under natural field conditions. The mean SCP was -7.9°C for both treatments and thus larval rearing conditions did not significantly affect the SCP ($t=0.003$, 50, $p=0.998$).

However, there was considerable inter-individual variability in SCP values for both laboratory (Range: -2 to -14°C, n=29) and field (Range: -3 to -14°C, n=23) reared diapausing prepupae. Therefore, I examined the incidence of mortality when prepupae were cooled once at a rate of 0.1°C/min from 0°C to lower temperatures ranging from -7 to -15°C and then brought back to 5°C at the same rate. As expected, based on the SCP data, morality was <20% if the temperatures were above -6°C but >75% at those below -7°C. Furthermore, those who survived the initial cold shock at lower test temperatures died several days post treatment.

Currently there is no information where diapausing individuals overwinter, so in the fall a sample of newly formed prepupae were allowed to naturally burrow into the soil and form overwintering cells. At the same time a series of ibuttons were placed at 5 cm intervals from the soil surface to a depth of 30 cm to record soil temperature data. Larvae will be excavated in early April to determine the depths at which overwintering cells were formed and individual survival.

Combining the data from both the laboratory and field experiments I can estimate the probability of surviving prevailing temperature conditions as a function of the depth at which WBC prepupae form their overwintering cells. Consequently, I will test the hypothesis that warmer winters have played a role in the recent expansion of the WBC by comparing winter soil temperature data prior to and after the appearance of this insect (e.g.1990-2000 vs 2000 -2010) in the Great Lakes region.

Effects Of Prescribed Burning On Flowering Dogwood (*Cornus florida*) In Spooky Hollow Nature Sanctuary, Ontario

J.V. Marcaccio, McMaster University; P. Chow-Fraser, McMaster University

Ecology, G.I.S., Endangered Species, Management, Flowering Dogwood

Although rare in Canada, there is a large collection of flowering dogwood (Cornus florida), a native and endangered understory tree, present in the Spooky Hollow Nature Sanctuary, Norfolk County. Due to the anthracnose disease, this plant has been decimated across its entire range from Florida to Ontario. I analyzed the effects of two prescribed burns of the lower canopy vegetation around the flowering dogwood trees that was conducted by the Hamilton Naturalists' Club during early spring of 2012. Prior to the burn, field surveys were conducted to map the location of all 1300+ dogwood trees and health and environmental conditions were recorded. A large subset of those trees were analyzed for further post-burn information after the growing season. This includes trees within and outside the prescribed burn areas. When mapped, clusters of dogwood in good and poor health are easily distinguishable. A multivariate analysis shows that the burn was damaging to smaller trees, but beneficial for the medium to large trees in the population. These results indicate that a prescribed burn can be a good management strategy for larger dogwoods in the short-term. Future projects should focus on the long-term success of the burned population as well as the control trees.

A Natural History of Eriophyid Galling Mites from Canada's Northern Boreal Forests

D.K. Marshall, Wilfrid Laurier University; J. Baltzer, Wilfrid Laurier University; R. Patankar, Wilfrid Laurier University; F. Beaulieu, Canadian National Collection

Keywords: arthropods; parasite; wetland; diversity

Canada's northern boreal forests face unprecedented changes as a consequence of regional climate warming. In the Northwest Territories, permafrost thaw in response to warming is resulting in changes in forest composition and land cover, as patches of discontinuous permafrost (which provide the foundation for black spruce (*Picea mariana*)-dominated forests) are being converted to bog and fens. Shrub species are abundant in these areas, so knowledge of their interactions with environmental stressors including herbivory is vital. While the influence of large generalist vertebrate grazers on plants here have recently received increased attention, little is known about the diversity, distribution or impacts of specialist natural enemies such as gall-inducing arthropods. One such guild of specialist arthropods are gall-inducing mites (Acari; Eriophyiodae), which are microscopic arthropods that commonly induce galls on tree and plant species in diverse ecosystems worldwide. The primary objective of this thesis is to examine the diversity of gall-inducing mites on commonly occurring deciduous shrubs in a northern boreal wetland-forest complex, with an aim to provide the first detailed natural history account of eriophyid galling mites from Canada's northern boreal forests. Despite these arthropods being a prevalent source of herbivory on shrubs in northern ecosystems, next to nothing is known about their diversity and host-specificity. The proposed study will thus examine galling mite diversity from species of birches (*Betula*) and willows (*Salix*) that comprise the majority of deciduous shrubby vegetation in a wetland-forest complex. Further, gall morphological traits, most often a unique outcome of specific host-mite interactions, are characterized to determine the diversity of gall morphology visible across host plant species.

Two primary questions are examined in this study:

1) What species of eriophyid mites occur and induce galls on deciduous shrubs in northern black spruce-dominated wetland forests of Canada?
2) What is the diversity of galling structures occurring here and how does this change depending on the host?

The first question was explored by taxonomical identification using key morphological characteristics on

eriophyid mites collected from both *Salix* and *Betula* shrubs in Scotty Creek (61°18'N; 121°, 18'W), Northwest Territories (NWT). There were two species of eriophyid mite found, *Aculus niphocladae* on *Salix* and *Vasates oldfieldi* on *Betula* shrubs, indicating genus-level host specificity.

For the second question, gall morphological characteristics were quantified and analyzed using a principal component analysis. It was found that all species of *Betula* had remarkably similar gall morphology. In *Salix* shrubs, gall morphology had a high degree of variability not only between species but also within species depending on the habitat where they occurred (bogs, fens, or upland forests), as well as on the individual level, which indicates genotype-level variation in gall morphology. Such inter- and intra-specific differences in gall structure may be associated with different physiological costs for the plant host.

This study improves our ability to identify the species of mite based upon the species of host plant coupled with the gall morphological characteristics. It also gives a good basis for future studies on eriophyid mites which will become increasingly important as shrubs become the dominant flora in Canada's northern boreal forests.

A Survey of Mycorrhizal Fungi on Discontinuous Permafrost Habitats in the Taiga Plains of the Northwest Territories

B.J. Moore, Wilfrid Laurier University; J. Baltzer, Wilfrid Laurier University; K. Stevens, Wilfrid Laurier University

Keywords: mycorrhizae; climate change; wetlands; Northwest Territories; fungi

Climate warming is causing permafrost thaw and the widespread land cover change in the Taiga Plains region of the Northwest Territories. In the zone of discontinuous permafrost, a forest-wetland mosaic exists and warming is leading to the conversion of forested permafrost plateaus to wetlands (bogs and fens). At present, we know very little about the biotic interactions in these communities or how these are impacted by the variable conditions in these habitat types. Mycorrhizae are mutualistic fungal root symbionts that provide nutrients to plant hosts in exchange for carbon. Mycorrhizal relationships have yet to be explored in subarctic wetland ecosystems affected by climate change, and this project offers the first insight to the rates of mycorrhizal colonization of vascular plants distributed amongst these habitats.

The objectives of this study are to:

1. Determine the mycorrhizal status of the predominant vegetation of permafrost plateaus, bogs and fens in a subarctic peatland; and
2. Characterize patterns of mycorrhizal colonization among habitats.

The study was conducted at Scotty Creek, Northwest Territories (61°18'N; 121°, 18'W), where the landscape consists of discontinuous permafrost forming plateaus adjacent to flat sphagnum bogs and channel fens. Five different fungal relationships were explored across 30 plant species to determine how mycorrhizae interact with plants and to infer any relationships between peat-to-bog conversions that symbiotic fungi may have. Field collected samples were placed into ethanol and brought back for analysis at Wilfrid Laurier University. Roots were processed by staining material to observe mycorrhizal structures, or were directly observed underneath a dissecting microscope.

Of the 30 species assessed 16 formed mycorrhizal associations, while no indication of endomycorrhizal or

ectomycorrhizal structures were detected in the remaining 14 species. Four species formed arbuscular mycorrhizal colonizations as confirmed by the presence of internal hyphae, arbuscules and vesicles (*Rubus arcticus, Maianthemum canadensis, Potentilla palutris,* and *Cicuta maculata)*, which were found on varying habitats. Four species were identified as ectomycorrhizal by the presence of a fungal sheath around modified root tips (*Betula glandulosa, Betula occidentalis, Picea mariana,* and *Larix laricina*), with *B. glandulosa* having significantly higher colonization rates in bog over plateau habitats as well as higher colonization than *P. mariana* and *L. laricina*. Ectomycorrhizal rates were consistent for all four species when occurring on permafrost plateaus, but colonization rates on bog sites differed amongst species. Ericoid mycorrhizae was present on nine species, as evident by the presence of fungal coiling in hair root epidermal cells, and were significantly more present on plateau than bog habitats. Only one species was colonized by arbutoid mycorrhizae (*Arcostaphylos rubra)*. Dark septate endophytes (DSE) were observed on 28 species, but were not present on *Cicuta maculata* or *Drosera rotundifolia*. Both ericoid mycorrhizae and DSE colonization rates were reduced in wetlands (bogs and fens) when compared with permafrost plateaus.

These findings illustrate an important role of habitat type on the degree of mycorrhizal association in certain types of fungi, namely ericoid and dark septate endophytes. This study, in providing the first mycorrhizal survey of subarctic peatland complexes in the Northwest Territories, can contribute to our understanding of how mycorrhizae may be affected by permafrost thaw and increased conversion into wetlands.

Elevated Temperatures Prevent Photosynthetic Decline In Norway Spruce (*Picea Abies*) During A Simulated Autumn

J.R. Stinziano, Western University; L.V. Kurepin, Western University; D.A. Way, Western University; V. Hurry, Umeå University; G. Öquist, Umeå University; N.P.A. Hüner, Western University

Climate Change, Photosynthetic Capacity, Boreal Forest, Photoperiod, Growing Season Length

Global surface temperature at high latitudes is projected to increase more severely in the next century than lower latitude regions on Earth, which could have negative consequences on the boreal forest system. The boreal zone is one of the largest forested areas on Earth; any large changes to this region would have a notable effect on global carbon flux. Warming is likely to lead to changes in growing season length via changes in the winter dormancy of boreal species. Winter dormancy induction during fall is important for the overwintering survival of boreal tree species. We investigated the effect of elevated temperatures on Norway spruce *Picea abies* photosynthetic capacity during a simulated autumn with photoperiod and ten year average air temperature conditions for London, Canada, from August to November. We hypothesized that elevated temperature would slow or delay the fall decline in photosynthetic rate compared to ambient temperatures. Using a Licor-6400 to measure photosynthetic capacity, we found that photosynthetic capacity (maximum rates of Rubisco carboxylation (Vcmax) and electron transport (Jmax)) declined in trees exposed to ambient temperatures during the simulated autumn, and unexpectedly, photosynthetic capacity did not change for the elevated temperature-exposed trees. Further, no changes were observed in key photosynthetic proteins in either treatment. Our results suggest that elevated temperatures may lead to an extended growing season for Norway spruce by reducing or eliminating winter dormancy induction. Further research is needed to determine the relative importance of temperature and light in regulating winter dormancy induction, and the impact of changes to winter dormancy on overwintering physiology of boreal species.

Sunday March 17th, Session 3, Room 2 – MDCL 1110, 9:00 – 10:30 am
Physiology -

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Diversity Of Insect-Eating Bats As Illustrated By Bite Force

T.T. Chang, Western University

Bite force; Chiroptera; insectivorous; eco-morphology; feeding specialization

Over 1200 species of bats (Chiroptera) are distributed around the world. Bats represent approximately 20% of all mammal species and occupy a large number of niches. Most bat species are insectivorous but others eat plant products, animals, or blood. If diet underlies the diversity of bats, then the bite force that bats can exert should also reflect this diversity. Here I examine the bite force of bats that are primarily insectivorous, as well as bats from the family Phyllostomidae that include animal, blood, and plant-eaters. I used Freeman and Lemen's model to determine the relative bite force of approximately 600 bat specimens representing 464 species from 16 families. I found that size and bite force have a significant relationship (R^2 = 0.736). Size-corrected bite force is significantly different between families. However, a discriminant functional analysis showed that size measures for bite force are not particularly reliable predictors for family membership.

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Resource Selection of the White Rhinoceros (*Ceratotherium simum*) in Welgevonden Private Game Reserve, South Africa

C.S. Hart, University of Guelph

White rhinoceros; resource selection; preference; conservation; Ceratotherium simum

In an attempt to optimize their fitness, it is thought that individuals will select areas that contain sufficient amounts of desired resources. The quality and abundance of particular resources within a habitat will have an effect on its attractiveness to animals and will thus influence cases of preference or avoidance. Land set aside for the conservation of wildlife should aim to provide the animals with the appropriate habitats that contain favoured resources. The white rhinoceros (*Ceratotherium simum*) is under constant threat of poaching, thus the majority of their remaining population is forced to live within the confines of African reserves. If resource availability influences the presence or absence of animals in a particular habitat, then the white rhino in Welgevonden Game Reserve will locate themselves in the landuse types that will maximize their fitness, regardless of their availability. In addition, if the rhinoceros are selecting habitats based on the resources they offer, then those areas will include the resources which best meet their requirements. From June to August 2012, the GPS coordinates of white rhinoceros observed in Welgevonden Private Game Reserve in Limpopo, South Africa were recorded. Using GIS, these observations, along with previous sightings dating back to April

2011, were compared with various layers of reserve features, including landuse types, river systems, and vegetation cover. With the white rhino being a grazing herbivore, vegetation surveys were used to calculate the dominant grass species in each area. A resource selection function was used to compare the proportion of rhino within a landuse type to the proportion of area offered by that landuse type. Statistical testing found the rhino to be actively selecting areas defined as Plains, Valley Bottom and Old Farmlands, while they avoided Hill Slope and Crest. Presence in Riparian, Saddle and Plateau was equivalent to their available area. The dominant grass species in the selected areas were found to be of higher nutritional value than those in the non-preferred areas. When separated by vegetation cover, the rhino preferred the grasslands, over woodlots and rocky plateaus. Rhino abundance decreased as distance from water sources increased (R^2=0.60) with 75.4% of sightings occurring within 400m. Analysis on gender differences and cases of seasonality are currently in progress. The white rhino in Welgevonden were found to be actively selecting habitats based on the resources that they offer and this information could be used by management to prioritize landuse types and influence future decision-making. This better understanding of rhino preference could benefit conservational efforts by ensuring the provision of sufficient resources in supporting a healthy white rhino population.

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Relative Fuel Use Between High Altitude And Sea Level Sub-populations of Deer Mice (*Peromyscus maniculatus*) During Exercise in Hypoxia

D.S. Lau, McMaster University; G.B. McClelland, McMaster University

hypoxia; exercise; sub-maximal work intensity; indirect calorimetry; respiratory exchange ratio

The oxidation of fuel is essential for providing energy to drive cellular processes, exercise, and any other physical exertions by muscles that are required for daily tasks. In mammals, energy for exercise in the form of ATP is mainly derived from the oxidation of varying proportions of carbohydrates and lipids. Due to differences in their chemical composition and properties, carbohydrates are generally the preferred fuel for moderate to high exercise intensities while lipids are preferred at low to moderate exercise intensities.

For animals living in high altitude, exercise is preformed in an environment with unavoidably low O_2 availability. It is important to note that carbohydrate oxidation provides more ATP per mole of O_2 consumed. For this reason, it was previously postulated that low altitude animals acclimated to hypoxia would use more carbohydrates as a mechanism to save O_2. However, when these acclimated animals were compared at the same relative work rate, it was found that the proportion of fuel use across the mammalian taxa stayed constant.

Recently, striking evidence has added yet another element to this conundrum. By using a multispecies comparative approach with leaf-eared mice of the *Phyllotis* species, Schippers et al. found that mice native to high altitude environments used more carbohydrates during exercise in hypoxia compared to their low altitude counterparts. This was the first compelling evidence that high altitude mammals exhibited a metabolic adaptation to conserve O_2.

In our research, we hypothesized to see similar findings in our high altitude groups while using an intra-species approach of deer mice, *Peromyscus maniculatus*. We exploited their wide range of native altitudinal distributions and obtained two subspecies of deer mice, one from high altitude and one from low altitude. Half of the individuals from each subspecies were then kept in normoxia, and the rest were kept in hypoxia. We determined the proportion of fuel use during exercise at around 75% VO_{2max} in hypoxia by using indirect calorimetry to determine the respiratory exchange ratio (RER). It is hypothesized that during hypoxia, high

altitude deer mice will adjust their metabolism towards a greater use of carbohydrates as fuel compared to their low altitude conspecifics, and this will be indicated by higher RER values. With these findings, we hope to determine whether a higher proportion of carbohydrates are used in mice native to high altitude environments, indicating a true genetic adaptation to hypoxia. These outcomes can open doors to future research projects regarding the mechanism of fuel selection, and can even be applied to relevant issues such as altitude training and colonization.

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Le régime alimentaire hivernal du carcajou (*Gulo gulo*) au Yukon

A.M. Levac, Université Laurentienne

Carcajou (Gulo gulo); régime alimentaire; piégeage au Yukon; conservation; disponibilité de proies

L'analyse du régime alimentaire de n'importe quel animal est importante en regard du statut trophique et l'écologie des populations. On peut en sorte faire de meilleures pratiques pour gérer les populations chassées ou piégées, particulièrement lors de fluctuations de la disponibilité des proies. Cette étude porte sur la diète du carcajou au Yukon pendant les hivers 2007-2008 et 2008-2009. Elle rendra possible la compilation des résultats de trois études à ce sujet pour en avoir une série complète de 6 ans consécutifs. Deux études antérieures par Kotipelto (2009) et par Blais (2012) ont rapporté que les proies les plus fréquentes étaient le lièvre d'Amérique (Lepus americanus) et les ongulés (incluant l'orignal (Alces alces) et le caribou (Rangifer tarandus)). Si ces proies sont primaires tel que suggéré par ces auteurs, elles devraient être les proies les plus importantes dans la diète du carcajou pendant les 2 années à l'étude. L'analyse des contenus stomacaux en utilisant diverses catégories de proies a rendu possible la détermination des fréquences de proies dans le régime, en plus de calculer leur proportion (en volume, mL). Les aliments inclus dans la diète ont été analysés et comparés entre les deux années, les périodes hivernales, les sexes et les âges. Une vérification par le calcul du Chi-carré avec le correctif de Yates et du test t de Student n'ont pas déterminé de différences significatives des fréquences et des proportions de proies entre aucun des groupes comparés. Toutefois, en comparant la fréquence avec la moyenne des proportions, il a été possible de déterminer l'importance de proies diverses dans le régime. Finalement, en utilisant l'index de Schoener, un chevauchement non-significatif ($S < 0.6$) entre la diète du mâle et de la femelle a été démontré. Le lièvre d'Amérique et les ongulés étaient encore une fois des proies primaires dans la diète hivernale du carcajou au Yukon. Le lièvre d'Amérique a démontré l'importance la plus grande dans la première année comparativement à la deuxième, et l'inverse est vrai pour les Ongulés. Ce phénomène est peut être dû au cycle de population du lièvre, mais ceci demande à être vérifié. Les mâles et les femelles ont démontré une différence dans le régime alimentaire global, peut être en raison du dimorphisme sexuel de la taille corporelle et aux sélections de proies de la femelle. La présence de proies secondaires et accessoires dans le régime alimentaire du carcajou supporte la notion que l'animal est polyphage et opportuniste. Ces traits sont importants en temps d'une baisse des populations du lièvre. Cette étude ajoute deux années d'observations au programme d'aménagement du carcajou au Yukon. Avec la 7e année (étude en cours), ceci rendra possible une analyse temporelle longitudinale, ainsi que des analyses spatiales en relation aux conditions variables de disponibilité de proies, ce qui devrait permettre de mieux gérer les populations exploitées de carcajou au Yukon.

The Effect of Hypoxia Acclimation on Maximal Oxygen Consumption and Muscle Capillarity in Deer Mice from High- or Low-Altitude Ancestry

M.A. Lui, McMaster University; T.S. Elogio, McMaster University; G.R. Scott, McMaster University; J.F. Storz, University of Nebraska-Lincoln

Hypoxia, high altitude, acclimation, VO2max, capillary density

Endotherms have demonstrated the capacity to adjust physiological systems, namely the organs involved in oxygen transport, in order to compensate for low oxygen tension experienced at high altitude. These adjustments may be based on genetic adaptations accrued over many generations as a result of natural selection, or as a result of phenotypic plasticity which develops according to the conditions experienced over the lifetime of an individual.

The model organism used in this study are deer mice (*Peromyscus maniculatus*), a species well known for its ability to thrive throughout a broad altitudinal range. Specifically, we focused on the differences between mice with high-altitude ancestry and mice with low-altitude ancestry (abbreviated ME and LN mice, respectively). Our main objective is to measure changes in maximal oxygen consumption ($VO_{2\ max}$) after long-term hypoxia acclimation (stimulating 4300 m). Additionally, we will use histological information to investigate the specific physiological mechanisms that drive changes in whole-animal aerobic performance.

VO_{2max} is known to be a plastic indicator of an individual's aerobic capacity. Results show that there is a decrease in $VO_{2\ max}$ in both LN and ME mice when exercised under hypoxic conditions. All mice acclimated to hypoxia for 6 weeks tend to have a greater $VO_{2\ max}$ than individuals of the same population acclimated to normoxia. When exercising in hypoxia, ME mice always have a higher VO_{2max} compared to LN mice, regardless of acclimation condition. After a 6 week hypoxia acclimation, both populations of mice have increased hematocrit and hemoglobin concentration. Alkaline phosphatase staining was performed in order to count muscle fibres and capillaries in deer mouse gastrocnemius muscle. We compared LN and ME mice from two different environments; wild-caught and F1 progeny raised in the lab. The ME mice had a greater capillary surface density and capillary/fibre surface density ratio regardless of the environment. This suggests that increased capillary density is an important subordinate trait which allows ME mice to attain a greater $VO_{2max.}$

Investigating the changes in whole-animal performance and muscle phenotype in mice from high- and low-altitude ancestry will allow us to elucidate the contribution of genetic adaptation versus phenotypic plasticity in compensating for low oxygen availability in hypoxia.

Does Logging Affect The Long-Term Stress of Eastern Chipmunks (*Tamias striatus*)?

H. L. McCurdy-Adams, Laurentian University; A. I. Schulte-Hostedde, Laurentian University

stress; cortisol; selection logging; chipmunk

Stress hormones, such as cortisol, increase an animal's heart rate, breathing rate, and blood pressure to allow them to quickly react to an immediate threat to their survival. This response is usually beneficial in the short-term, but chronic elevated stress have a negative effect on fitness. Logging activities have caused widespread habitat destruction and disturbance for a longer period of time at a higher intensity than more common stressors, such as predation risk. Eastern chipmunks (Tamias striatus) are common in both logged and natural forests. Does logging affect their cortisol levels? We used mark-recapture techniques and enzyme immunoassays to collect and measure cortisol levels of chipmunks in Algonquin Provincial Park, Ontario. We tested the hypothesis that cortisol levels will be higher in logged sites compared to natural sites. Sex, age, reproductive condition, and body size measurements were recorded for each chipmunk caught. Scrotal males, and pregnant females were removed from analysis. There was no effect of logging on either faecal or hair cortisol concentrations. Female chipmunks had lower hair cortisol concentrations than male chipmunks. Chipmunks in one natural site had lower faecal cortisol concentrations than in any other sites (natural or logged). Chipmunks in low density populations had lower faecal cortisol concentrations than chipmunks in high density populations. Future studies should sample chipmunks and other small mammals in forests undergoing different types of logging practises. Characteristics of each forest's structure may also be beneficial to measure. Stress levels of chipmunks are affected by complex processes, but selection logging alone does not seem to have a negative effect.

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Identification and Characterization of *hypH*, a Putative Alpha-Ketoglutarate Semialdehyde Dehydrogenase in *Sinorhizobium meliloti*

J.M. Boudreau, McMaster University; T.M. Finan, McMaster University

Hydroxyproline; alpha-ketoglutarate semialdehyde dehydrogenase; citric acid cycle; Sinorhizobium meliloti; root-dwelling;

Hydroxyproline is a common amino acid in plant and animal cells. Its availability in soil and root cell exudates makes it a potentially significant source of carbon and nitrogen for soil and root dwelling bacteria. The ability to catabolize hydroxyproline has been identified in a number of bacterial species, including the root-dwelling, nitrogen-fixing, legume endosymbiont *Sinorhizobium meliloti.* A putative pathway for hydroxyproline catabolism in *S. meliloti* has been developed and is in the process of being characterized. In the putative pathway, *trans*-4-hydroxy-L-proline is converted over the course of four enzymatic reactions to α-ketoglutaric acid (KG), a significant substrate in the citric acid cycle. This project investigates one of these putative reactions, the conversion of α-ketoglutarate semialdehyde (KGSA) to KG by an α-ketoglutarate semialdehyde dehydrogenase (KGSADH), putatively coded by the gene *hypH* in the hydroxyproline catabolism cluster on the pSymB megaplasmid. Strains that are *hypH* mutants retain the ability to grow with hydroxyproline as a sole carbon and nitrogen source, likely due to the complimenting activity of another KGSADH, *araE*, found in the arabinose catabolism cluster. Strains that are mutant for both *hypH* and *araE* show greatly diminished ability to grow with hydroxyproline. Complementation of the double deletion mutant with *hypH* restores the hydroxyproline growth phenotype. The conversion of KGSA to KG is measurable by the accompanying conversion of NADP+ to NADPH in a 1:1 ratio, which produces a measureable change in absorbance at 340nm. Such a change has been observed for HypH in the presence of KGSA. Enzymatic characterization is ongoing as is the characterization of the substrate and product with mass-spectrometry. Demonstrating the function of *hypH* and characterizing the activity of its enzyme product will increase the understanding of the widely present hydroxyproline catabolic pathway in bacteria.

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Investigation of Differentially Regulated Genes and Salicylic Acid Biosynthesis Pathways During Age-Related Resistance in *Arabidopsis*

M.I. de Vries, McMaster University; R.K. Cameron, McMaster University

Arabidopsis; ARR; pathogen; salicylic acid

As plants mature they develop an increased resistance to certain pathogens, which is referred to as age-related resistance (ARR). The ARR response in *Arabidopsis* shares similar characteristics with another form of resistance termed systemic acquired resistance (SAR). SAR is an induced response to pathogen infection that confers systemic resistance to a broad range of pathogens after a local "immunizing" infection. Both ARR and SAR require accumulation of the plant hormone salicylic acid (SA). A microarray performed by our laboratory identified 231 genes that are differentially regulated during ARR. The involvement of an up-regulated gene, *WHIRLY1 (WHY1*), and a down-regulated gene, *XYLOGEN PROTEIN 1 (XYP1*), in ARR were examined using mutant studies. An ARR assay comparing young and mature plants was performed using *why1* and *xyp1* mutants. Our results show that *why1* is ARR-deficient, while *xyp1* is ARR-competent and shows no significant difference from the wild type. These findings suggest that *WHY1* is involved in ARR and *XYP1* is not. The SA biosynthesis mutant *salicylic acid induction deficient 2* (*sid2*), commonly used as a negative control in ARR experiments, occasionally displays an ARR-like response. In *sid2*, the enzyme ISOCHORISMATE SYNTHASE 1 (ICS1), which catalyzes the first step in converting chorismate to SA, is mutated. Our research examines the possibility that *sid2* plants occasionally use an alternate pathway to synthesize SA by comparing it to another SA-deficient mutant (NahG) in an ARR assay.

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Characterizing Protein Interactors For Select AROGENATE DEHYDRATASES In *Arabidopsis thaliana*

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Keywords: Arogenate dehydratase; Arabidopsis thaliana; protein-protein interaction, yeast two-hybrid screen, phenylalanine biosynthesis

The arogenate dehydratase family in *Arabidopsis thaliana* has six members (ADT1 to ATD6). These enzymes catalyze the final step of phenylalanine biosynthesis. In addition to a catalytic and regulatory domain, ADTs have an N-terminal transit peptide. These peptides direct ADTs to the chloroplast, where they predominately localize to stromules. Two of the ADTs have additional localization patterns. ADT2 localizes in a ring-like structure around the chloroplast while ADT5 has been observed in the nucleus. Such unexpected localization patterns suggest that ADT2 and ADT5 have additional non-enzymatic roles. I propose that ADT2 has a role in chloroplast division and ADT5 functions as a transcription factor in the nucleus. Furthermore, ADTs have been shown to interact with each other to form homo- and heterodimers. Two-hybrid screens have also shown an interaction between ADT1 and other chloroplast proteins. I hypothesize that ADT2 and ADT5 have unique protein interaction profiles and interact with chloroplast division and nuclear proteins, respectively. A yeast two-hybrid approach was used to identify two putative interactors for ADT2 and 28 for ADT5. For this project, the sequence and identity of these ADT interactors was determined. It will be discussed how the identity of these putative interactors may give insight for the function of ADT2 and ADT5.

The Relationship Between Cytokinin Dehydrogenase And Nodulation In Low-Nodulating Mutants Of Pea

A.P. Macdonald, Wilfrid Laurier University; F.C. Guinel, Wilfrid Laurier University

Nodulation, pea, cytokinin, cytokinin dehydrogenase

Legume plant species are able to form symbiotic relationships with atmospheric nitrogen-fixing *Rhizobium* bacteria, in which the plant receives a source of nitrogen for nucleotides and amino acids, and the bacteria are granted protection within root organs called nodules. Plant development is regulated by the hormone cytokinin, which is responsible for lateral root production, chlorophyll stability, and the initiation of nodule production. Cytokinin can be irreversibly degraded by the enzyme cytokinin dehydrogenase (CKX). Recently, it has been found that pea mutants which are defective in cytokinin perception form delayed and/or fewer nodules compared to the wild-type. I am using (*Pisum sativum* L. cv Sparkle) as a model organism to investigate the relationship between *PsCKX1* and nodulation in five lines of low-nodulating, pleiotropic mutants (E2, E107, E132, E151, and R50). It is predicted that R50 will express *PsCKX1* in larger amounts compared to Sparkle, and that the relative levels of *PsCKX1* may help to explain the phenotypic traits in these lines of mutants.

Short-fragment Assembly and Annotation of the Genome Sequence for *Phaseolus vulgaris* cv. 'OAC-REX'

S. Munholland, University of Windsor; C. Dinatale, University of Windsor; A. Platts, McGill University; K.P. Pauls, University of Guelph; W.L. Crosby† University of Windsor

Phaseolus spp., Genome annotation, Genome assembly, Bioinformatics

Phaseolus vulgaris (common bean) is an agricultural crop of importance in diverse regions around the world. Most cultivars currently in use derive from two ancestral land-races of Andean and Mesoamerican origin. In oreder to better understand the genetic foundation for desirable agronomic traits in this and related crop species, we have undertaken to generate a short-fragment assembly and draft genome sequence of the *P.vulgaris* variety 'OAC-REX'. OAC-Rex is a derivative of Andean origin that has been crossed with *Phaseolus acutifolius* as a source of resistance alleles to Common Bean Blight (CBB) of importance to the Canadian agricultural context. CBB is a bacterial pathology that is endemic to Ontario and is responsible for significant crop losses both before and after harvesting. In order to shed new light on the genetic foundation oc CBB resistance in the cultivare, a short-fragment assembly of the OAC-REX genome was generated in several datasets.

These short sequence reads were subsequently filtered and trimmed based for quality, prior to generation of a

contiguous sequence assembly. The collection of contigs was submitted to higher-order scaffolding followed by submission to a computational annotation pipeline. This *de novo* assembly and annotation process was undertaken using a variety of computational tools in order to generate, quality-check, and verify the results, a summary of which will be presented.

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Expression of Fatty Acid ω-Hydroxylases (*GmFAωH*) Genes in Soybean (*Glycine max*) Roots

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soybean, Phytophthora sojae, suberin, resistance, GmFAωH

Soybean (*Glycine max* [L.] Merr.) is worldwide crop that is highly susceptible to infection by *Phytophthora sojae*, and thus increased resistance to this pathogen is required to aid in reduction of global crop loss. Root suberin levels are positively correlated with partial resistance to *P. sojae*, suggesting increased suberin deposition will result in increased resistance. Fatty-acid ω-hydroxylases (FAωHs) catalyze the production of key elements of poly(aliphatic) component of suberin and are encoded by six putative *GmFAωH* genes in soybean. It is hypothesized that *GmFAωH* gene expression will be tissue specific, with a subset being expressed in roots where suberin is deposited. *GmFAωH* gene expression pattern both wild-type and hairy roots of soybean are being established using gene specific primers and RT-PCR. Establishing the *GmFAωH* gene expression patterns in hairy roots will facilitate development of a model system for studying the role of suberin in *P. sojae* resistance using an *Agrobacteriium rhizogenes*-based gene transformation system to manipulate soybean root suberin levels. Results indicate that *GmFAωH-5* and *GmFAωH-6* are expressed in wild-type roots, while other GmFAωH genes are not. This suggests that *GmFAωH-5* and *GmFAωH-6* are involved in root suberin deposition and therefore are potential target genes for manipulation of root suberin levels.

Sunday March 17th, Session 3, Room 4 – MDCL 1102, 9:00 – 10:30 am
Ecology (in Fish) -

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Stable Isotope Analysis of the Diet Array of the Alternative Life Histories of Chinook Salmon in Lake Ontario

H. Alhasan, University of Windsor

Isotopes, Chinook Salmon, Alternative Life Histories, Diet Array

Stable isotopes of carbon and nitrogen were used to investigate the diet array and trophic positions of the alternative life histories (Jack and Hooknose) of Chinook salmon in Lake Ontario. The two main objectives are to test for any significant variation in the diet array of Jacks and Hooknoses and to investigate if these two tactics differ in their trophic levels and in their niche breadth. Muscle tissue samples were collected from 10 Jacks and 14 Hooknoses. The data generated was then analyzed using various statistical method and the results will be further discussed. .

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The Effect Of Ovarian Fluid Concentration Gradients On Sperm Velocity In Alternative Reproductive Tactics Of Chinook Salmon, *Oncorhynchus Tshawytscha.*

A. Forest, University of Windsor

Salmonids; reproduction

Previous experiments study fish sperm metrics and fertilization success in river water even though ovarian fluid is present during spawning in the wild. Studies by Flannery et al. (2011) show that ovarian fluid has an effect on the performance of sperm. Sperm velocity is a major determinant of fertilization success in salmonids. In the alternative reproductive tactics of Chinook salmon, Jacks outcompete Hooknose males in river water while there is no difference in respective paternity success in 50% ovarian fluid. During my honours thesis project, I set out to determine if there is a switch-point in ovarian fluid concentration at which Hooknoses gain an advantage in sperm velocity over Jacks. The results of this study could help gain insight into the ovarian fluid concentrations experienced in the wild by sperm of alternative reproductive tactics of Chinook salmon and help tailor future studies in order to replicate this natural environment.

Social Memory in African Cichlids

S.D. Potvin, McMaster University; C.M. O'Connor, McMaster University; A.R. Reddon, McMaster University; S. Balshine, McMaster University

Cichlids; Social; Memory; Object; Conspecific

The social brain hypothesis posits that organisms that live in groups have bigger brains with more developed cognitive abilities in order to deal with the complexities of social life. To test this idea, we compared co-operatively breeding cichlid fish *Neolamprologus pulcher* (a highly social species) with their closest pair breeding relatives, *Telmatochromis temporalis* (a non-social species). We conducted two experiments with individual members of these two species. First, we tested if there was a preference for familiar objects over unfamiliar objects and whether that preference varied across species or sex. Second, we tested whether fish showed a preference for familiar conspecifics over unfamiliar ones and again explored how sex and species influenced preferences. The fish preferred to spend time with an object, but there was no preference demonstrated with respect to familiarity, and there were no species or sex differences. Similarly, there were no species or sex differences in preferences for familiar versus unfamiliar conspecifics. However the *N. pulcher* spent more time socially interacting than did *T. temporalis,* and in particular displayed more social and aggressive behaviours towards the familiar fish than towards the unfamiliar fish. These findings suggest that interaction assays (that include potentially costly behaviours) provide a better window through which to study social brains than assays that merely measure duration spent near a particular stimulus.

Morphological Variation Among The Sexes In Spawning Chinook Salmon (*Oncorhynchus tshawytscha*)

Prableen Singh, University of Windsor

Morphology, Variation, Hooknoses, Jacks, landmarks

Secondary sexual characteristics such as large humped backs and elonged snouts play a vital role in the reproductive fitness in the Chinook salmon (*oncorhynchus tshawytscha*) mating system. I sampled 15 females and 29 males (13 guard type males – hooknoses and 16 sneak type males – jacks). Different selection pressures are hypothesized to lead to different optima for body shape and morphology of males from the alternative life histories and females. I predict jacks to be more streamlined for sneaking fertilization giving them the ability to quickly dart into nesting areas and hooknoses to be larger males because they participate in dominance-based hierarchies. Also, the females should have a larger body cavity due to fecundity selection on the ovaries. Various body measurements as well as homologous landmarks were placed. The analysis of the warp scores will be assessed using discriminate function analysis, which will either confirm or reject my predictions.

Long-Term Changes in Muskellunge (*Esox masquinongy*) Nursery Habitat in Georgian Bay, Lake Huron: Implications for Habitat Availability

J.D. Weller, McMaster University; P. Chow-Fraser, McMaster University

muskellunge; nursery habitat; GIS; Georgian Bay

In 1981, twenty coastal wetlands in Severn Sound (SS), Georgian Bay (GB) were surveyed and found to have young-of-the-year (YOY) muskellunge (*Esox masquinongy*). In 2012, we repeated the survey method in 16 sites, and could not find any YOY. We hypothesize that the absence of YOY is due to loss of critical nursery habitat as a result of water-level declines and shoreline alterations. Satellite imagery and historic air photos were obtained for 7 time periods between 1931 to 2009, which included both low and high water-level scenarios. For each site, we delineated available aquatic habitat and calculated dock density (#docks per shoreline length (km)). Data before and after 1981 were grouped and compared; mean dock density was significantly higher in the latter period. We also found a significant decline in cover of submerged aquatic vegetation (SAV) with declining water levels. We suggest that the absence of YOY in recent times in Severn Sound is due to loss of aquatic habitat because of low water levels as well as the negative impact of increased dock densities. We contrast these results to another study being carried out in Beaverstone Bay, where there has not been any significant increase in dock density, and where YOY are still found during a 2012 survey.

Sunday March 17th, Session 3, Room 5 – MDCL 1009, 9:00 – 10:30 am
Genetics -

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Analysis Of A New Population Of Hawaiian Endemic Yeast To Determine If Genetically Intermediate From Previous Studies

L.M. Bishop, University of Western Ontario

Metschnikowia hawaiiensis, yeast populations

The Big Island of Hawaii contains an endemic species of yeast, *Metschnikowia hawaiiensis* that is haploid and heterothallic. The yeast is found living in the gut of the beetle *Prosopeus subaeneus*, which lives inside morning glory flowers. Morning glories are found growing in areas of forest that have been disturbed such as forest clearings or roadsides. Forests of the Big Island are separated by lava flows, forming islands of forests known as kipukas that are also isolated by the geography of the Mauna Loa volcano. Morning glories constitute discontinuous habitats due to both human disturbance and lava flows. The beetles, being small and poor flyers, are limited in the distance they can travel. Dispersal events such as human interference and high winds caused by extreme weather could account for genetic similarities between populations. In previous studies, *M. hawaiiensis* has been found in two localities situated 70 km apart. A new population of morning glories, approximately mid-point between the two previously studied sites and containing *M. hawaiiensis* has been discovered in an area called Wood Valley. Seven strains have been isolated from the Wood Valley location. Due to the discontinuous environment of the forests caused by lava flow and human activity, it is hypothesised that a new population recently discovered at the approximate mid-point will be genetically intermediate to the two previously studied genetically different populations. This can be determined by characterising 14 loci, which have been shown to be polymorphic at ten loci in previous studies on *M. hawaiiensis.* Loci have been characterised using both SSCP electrophoresis and sequencing of selected alleles. The Wood Valley locality has been shown to share alleles that have been found in the previously studied localities but new alleles have been identified as well, demonstrating that the new population is genetically distinct.

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Soil Fungi Diversity And Abundance In Limed And Unlimed Areas Contaminated With Metals In The Greater Sudbury Region

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Fungi; Lime; Metals; Diversity; Abundance

This study is aimed to assess the fungi species and their abundance occurring in limed and adjacent unlimed areas contaminated by metals in the Greater Sudbury Region. Soil samples were collected from layer 1 of the soil for the following areas: Wahnapitae Hydro-Dam, Daisy Lake, Kingsway and Hagar (St-Charles specifically) as a control site. In regards to Wahnapitae Hydro-Dam soil samples, pH ranged from 6.34-6.75 for limed substrate and 3.56 - 3.82 for unlimed substrate. Concerning Daisy Lake soil samples, pH ranged from 4.05 - 4.12 for limed substrate and 3.87 – 4.04 for unlimed substrate. For Kingsway soil samples, pH ranged from 4.35 - 4.67 for limed substrate and 2.35 - 3.87 for unlimed substrate. Pertaining to Hagar soil samples, pH ranged from 3.23 – 3.50; soil samples from control site were unlimed. Total and bio-available concentration of nutrients and metal elements in the limed and unlimed organic surface horizons (LFH) of soils from the Sudbury region sites were determined in regards to Al, As, Ca, Cd, Co, Cu, Fe, K, P, Pb, Mg, Mn, N, Na, Ni, S, Sr and Zn. The fungi species have been isolated from the soil samples via the soil dilution method on Sabouraud Dextrose Agar (SDA) and Malt Extract Agar (MEA). Results showed a total of 34 fungi species belonging to 23 genera growing on Petri dishes. The most common species found were: *Cryptococcus neoformans, Mycoderma aceiti, Penicillium fellutanum, Rhizopus stolonifer, Sporotrichum schenkii, Trichophyton mentagrophytes and Trichophyton tonsurans.* The present study suggests that greater species diversity is present in the limed areas compared to the unlimed areas for each of the 3 studied sites when grown on SDA media. In general, fungi abundance on SDA media followed this trend. However, when grown on MEA media, species diversity was slightly greater in unlimed areas compared to limed areas with the exception of the Daisy Lake site. Fungi abundance also followed this trend but to a lesser degree then in the case of the SDA media.

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Extensive Genetic Variation In *Geomyces destructans* From Canada

J. Khankhet, McMaster University; J. Xu, McMaster University

White-nose syndrome; emerging infectious disease; bats

Geomyces destructans is the cause of an emerging infectious disease that threatens the bat population in North America. The fungus was first observed in 2006 and has been estimated to have caused the mortality of over five million bats. The disease caused by *G. destructans* is white-nose syndrome, which is characterized by the growth of the fungus on the nose, ears or wings of the bats. Relatively little is known about the genotype of *G. destructans*. Two previous studies have found that the North American population of *G. destructans* is clonal however their sample size was small and only included isolates from the United States. Multilocus sequence typing examining eight gene fragments in 16 Ontario and 28 New Brunswick isolates showed no variation in gene sequences. However, DNA fingerprinting using $(GACA)_4$ and M13 primers revealed extensive genetic variation.

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Identification of *Cantharellus* Species of Newfoundland and Labrador Using Phylogenetic Analysis

J.I. Kim, Western University; R.G. Thorn, Western University

Cantharellales; Taxonomy; Molecular systematics; Newfoundland and Labrador

Species of C*antharellus*, commonly known as chanterelles, are prized edible mushrooms that are wild harvested worldwide. The application of molecular phylogenetics has elucidated the taxonomy of *Cantharellus* and led to the description of additional species. In eastern Canada only one species, *Cantharellus cibarius*, is widely known. The goal of this study is to identify the chanterelles of Newfoundland and Labrador through phylogenetic analysis of partial sequences of the large subunit region of ribosomal DNA. Genomic DNA was obtained from collections using a bead-beading technique, followed by amplification of the target region using PCR, then sequencing. A phylogenetic tree was constructed using the obtained sequences along with reference sequences from GenBank. Results revealed three species of chanterelles in Newfoundland and Labrador including *Cantharellus formosus* and *Cantharellus roseocanus* which were originally described from Pacific Northwest, and European *Cantharellus amethysteus*. The phylogeny demonstrated that none of the Newfoundland and Labrador specimens were conspecific with European *Cantharellus cibarius*.

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Macrofungal Diversity Of The Ailao Mountains By ITS Barcoding

N. Pun; JP Xu, Department of Biology, McMaster University

fungal diversity, ITS barcoding, biodiversity

Fungal diversity is a key component of overall biodiversity. Fungi are involved in many key ecosystem processes like nutrient cycling, decomposition and are part of key interactions with plants and animals. Among the diverse groups of fungi, mushrooms are the most conspicuous and the most extensively surveyed group of fungi. However, relatively little is known about mushroom diversity in many parts of the world, with over 15 000 estimated unknown species in tropical and subtropical Asia alone. This study aims to sample to macrofungal diversity of the Ailao mountains in Yunnan, China using the ITS barcode region as a species identifier. Over 300 samples of mushrooms were collected and likely comprise only a portion of the fungal diversity in this area. A large majority of these species have ITS sequences that are <97% in similarity to any published sequences which implies that they are novel species.

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The Sequencing And Annotation Of The Genome Of The Large-Spored Yeast, *Metschnikowia hawaiiensis*

B. A. Stanford, The University of Western Ontario

The large spored yeast *Metschnikowia hawaiiensis* can be distinguished from other yeasts primarily by the large needle-shaped spores generated during meiosis, which are associated with pathogenic behaviour. However this yeast also has an abnormal meiotic cycle wherein only two final meiotic products are generated despite the transient appearance of four. The evolution of these distinct characters is a mystery, in part because the current phylogeny of the large spored *Metschnikowia* clade of yeasts is based solely on data from the rRNA gene cluster, and these data are not statistically robust enough to generate a reliable phylogeny. As next generation sequencing platforms have dramatically lowered the cost of sequencing for small independent labs and more genome data has become available for other yeast species, I sought to contribute a first step towards a robust *Metschnikowia* phylogeny by generating whole genome data for *M. hawaiiensis*.

M. hawaiiensis genomic DNA was extracted using a standard Qiagen blood and tissue kit and was sequenced at the Robarts Research Institute using the Illumina Miseq. The resulting short read data were assembled with the genome assembly software programs SOAPdenovo 1.05 and ABYSS 1.3.4 over a wide range of kmer values (29 to 79). Using data from the SOAPdenovo assembly and genome data from *Candida albicans*, genes were predicted using the software program AUGUSTUS 2.6.1.

The program BLAST+ was used to align the putative amino acid sequences to possible homologues in the non-redundant protein sequence database using the algorithm blastp, with an expected value cutoff of 10^{-5}. To expedite the annotation process and ensure reliable annotation of each gene, a second alignment of the putative genes was conducted under the same conditions except at an expected value cutoff of 10^{-100}. Based on currently available sequence data from the *Candida albicans* clade, and using the terminal command "grep", I searched the BLAST+ output files for gene families that are enriched in the pathogenic species of the *C. albicans* clade as well as for mating-type loci (MTL) genes so as to identify the MTL in *M. hawaiiensis*.

Based on annotations currently in progress and relative to its closest sequenced relative, *Candida (Clavispora) lusitaniae, M. hawaiiensis* has an expanded number of reductases, dehydrogenases, amino acid permeases, and major facilitator transporters, which likely increase the yeast's ability to adapt to more hostile host environments. Interestingly, *M. hawaiiensis* lacks any GPI (glucose-6 phosphate isomerase) family 18 genes suggesting the *C. lusitaniae* homologues have been mutated or deleted over time. Mating type loci proteins were more difficult to identify and required use of the lower expected value cutoff alignments. Neither the a-factor nor the alpha-factor could be identified, as none of the *C. albicans* clade homologues produced significant alignments in any of the putative genes. However, homologues of other genes involved in the mating process were identified (e.g. STE2, a pheromone response and conjugation protein in mating-type a cells, among others) and these could potentially be used to identify other associated mating pathway genes in future studies.

I thank Dr. T. Hsiang for completing the assembly process, and for generating the predicted genes used in the annotation process.

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Tuberin Expression and Function in Mammalian Brain Development

Bashaer Abu Khatir, Mallika Somayajulu-Nitu, Elizabeth Fidalgo da Silva and Lisa A. Porter, Biological Sciences, University of Windsor, ON

Tuberous Sclerosis, Tuberin, Hamartin, hamartomas, mTOR

Tuberous Sclerosis (TS) is a neurological autosomal dominant multisystem disorder that affects about 1 in 6,000 births. A key feature of TS is the formation of benign tumours called hamartomas which can form in different organs of the body including the brain. These lesions form as a result of mutations in at least one of two tumour suppressor genes. TSC1 and TSC2 are the tumour suppressor genes which encode the proteins Hamartin and Tuberin, respectively. Mutations in the gene for Tuberin are associated with more severe phenotypes including those associated with severe neurological hamartomas. Hamartomas in the central nervous system are found early in life and are thought to be the result of a developmental defect. Indeed TSC2 null mice are embryonic lethal due to improper closure of the neural tube. Despite this important data the normal role for Tuberin in brain development is not known. The primary objective of this project is to determine how Tuberin, and Tuberin effector proteins, are regulated and localized during early embryonic development of the brain.

Using mice collected at different embryonic time points we conducted extensive immunohistochemistry on a number of developing brain structures. We quantified the levels and relative localization of Tuberin and the effector proteins, p27 and pS6K, at each time point. I hypothesize that Tuberin levels and activity will be elevated at stages and in regions critical for neurological development. This work will provide valuable information regarding the normal function of the protein Tuberin; results may shed light on the processes that can become abrogated in diseases such as TS.

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Relevance of the Tuberin/Cyclin B1 interaction in Tuberous Sclerosis

S.L. Botsford, University of Windsor; E. Fidalgo da Silva, University of Windsor; and L.A. Porter, University of Windsor

Tuberin; Tuberous Sclerosis; Cyclin B1

Tuberous sclerosis (TS) is a multi-organ disorder characterized by the formation of benign tumours called hamartomas. Mutations in the tumour suppressor proteins, Tuberin and Hamartin, are implicated in this disease. In addition to its role as a negative regulator of mTOR, our lab has shown that Tuberin also binds and

regulates the G2/M cyclin, Cyclin B1 (CycB1). To help test the hypothesis that Tuberin binding to CycB1 lengthens G2 phase of the cell cycle, I have developed Tuberin mutants and fluorescent systems that will allow us to study the Tuberin-CycB1 interaction in real-time.

To date, I have constructed a CFP G2/M sensor to accurately time the length of G2 and I have also generated Tuberin-CycB1 bifluorescence complementation vectors that will indicate when and where in the cell these proteins interact. Furthermore, I have begun to construct vectors that express mutant forms of Tuberin in a cell without the complications of wild-type Tuberin expression. Cells will be manipulated using combinations of these vectors and the effects on the timing of G2 phase will be recorded with live cell microscopy. These results will help us conclusively determine the physiological importance for this novel interaction and it will further test the effects of mutations known to occur in patients with TS.

Assessing The Extent Of Biopsy Core Involvement By Prostate Cancer And Its Impact On The Selection Of Patients For Active Surveillance: Is Eyeballing Accurate Enough?

O. Mahamud, University of Toronto; T.H. van der Kwast, Department of Pathology, University Health Network; M.R. Downes, Department of Pathology, University Health Network; and A.J. Evans, Department of Pathology, University Health Network.

Prostate; Active Surveillance; Percent Core Involvement; Needle Core Biopsy

Background: Active surveillance (AS) is an established management option for patients with low-risk prostatic adenocarcinoma (CaP). Biopsy criteria for AS include Gleason score $\leq$ 6/10, involvement of $\leq$ one third of cores in a biopsy set and $\leq$ 50% core involvement in any positive core. There is no standardized method for assessing % core involvement and a variety of methods can be used including estimation, or eyeballing (EB), and direct measurement. In this study, we assessed the concordance between EB and measurement in a typical cohort of positive needle biopsies.

Design: As part of a prospective digital pathology validation project in our department, digital slides from 95 sets of prostate biopsies were available for review, 43 (45%) of which had least one core with CaP. CaP was annotated on the digital slides by two experienced urological pathologists. Open source java script software (Image J, http://rsbweb.nih.gov/ij) was used to measure the linear extent of CaP in relation to total core length. Image J permits precise length measurements of non-linear objects such as prostate cores. The results, expressed as % core involvement, were compared to values obtained by EB of the digital slides. A sub-set of cores determined to have 40-60% involvement by EB was identified to assess biopsies particularly relevant to the issue of AS patient selection.

Results: A total of 192 cores contained CaP in the 43 positive cases. There was no significant difference between EB and Image J in terms of mean % involvement when considering all positive cores (EB - 32.7 % $\pm$ 29.0 (SD) versus Image J - 31.4 % $\pm$ 26.4 (SD), p=0.08). There were 38/192 cores (20%) determined to have 40-60% core involvement by EB. In this sub-set there was a significant difference between the two methods (EB - 54.8 % $\pm$ 13.4 (SD) versus Image J - 47.0 % $\pm$ 11.6 (SD), p= 0.0004), with EB tending to overestimate % involvement by an average of 12%. Issues complicating the assessment of % core involvement by either method included: core contour (straight, folded or bent), core fragmentation, discounting rectal mucosa or

periprostatic soft tissue from total core length and discontinuous core involvement by CaP.

Conclusion: There was excellent overall concordance between EB and Image J, however direct measurement from digital images may be more robust when assessing % core involvement for AS patient selection. Our study also raises the question of a need for standardization for determining extent core involvement given its importance in the setting of AS.

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Endogenous Expression Of Tuberin (TSC2) In Neuronal Cell Lines

K.J.L Osmon, University of Windsor; M. Somayajulu-Nitu PhD., University of Windsor; D. Lubanska, University of Windsor; Dr. L. Porter, University of Windsor

Tuberin; Cell growth; Mutation; Cancer

Tuberin and Hamartin bind as a protein complex to negatively control cell growth and proliferation by inhibiting the mTOR pathway. Mutation in the gene product, or aberrant protein regulation, of Tuberin or Hamartin are known to cause Tuberous Sclerosis, an autosomal dominant disorder characterized by lesions, called hamartomas, throughout the central nervous system. Recently, Tuberin has also been implicated in a number of human cancers including a subgroup of medulloblastoma (MB). Evidence from our lab and others suggests that Tuberin may play an essential role in regulating fate in neural precursors. My research focuses on characterising the role of Tuberin in cell fate decisions over the course of differentiation in two different cell lines. Using gene and protein expression assays I will determine how endogenous Tuberin is regulated during neural fate decisions. I will also determine the consequence of manipulating Tuberin levels on neural precursors using a variety of cell fate assays. The results from this work may have implications on the initiation and progression of several forms of brain cancer, including that of MB.

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Relationship Amongst SCF^{Skp2}, APC^{Fzr} & CycA In The Regulation Of Diploidy

S. Palazzolo, University of Windsor

Mitosis; Polyploidy; Tumor suppressor; Ubiquitination

Polyploidy is a known characteristic of cancer cells. The mitotic cell division cycle prevents polyploidy through an ordered cyclic proteolysis catalyzed by E3-ubiquitin-ligases. SCF^{Skp2} and APC/C^{Fzr} E3 complexes regulate cell replication and division by tagging specific proteins for degradation. Skp2 SCF activator protein is a known proto-oncogene in mammals. In *Drosophila,* however, S*kp2* seems to act as a tumor suppressor as its loss

generates polyploidy. Many studies have analyzed the oncogenic role of S*kp2* in promoting polyploidy, but it is unknown how *Skp2* acts in polyploidy prevention. We have observed similar polyploidy phenotypes in flies with mitotic *CyclinA* knockdown, as well as in flies overexpressing *Fzr*; leading us to propose a functional relationship amongst SCF^{Skp2}, APC^{Fzr} and CycA in maintaining diploidy. We predict SCF^{Skp2} protects CycA against degradation by APC^{Fzr}. I will present the results from the genetic methods utilized in testing this model.

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Identification and Functional Implications of Soluble Bone-Derived Factors on Breast Cancer Migration and Growth

G.M. Pio, Western University, A. Allan, Western University

Breast cancer, cancer stem cells, metastasis, bone marrow niche

Breast cancer represents 26% of newly diagnosed cancers in women and is ranked second in mortality, contributing to 14% of cancer-related deaths. Patients succumb to their disease primarily because of metastasis, a condition for which there is currently no cure. Cancer is considered metastatic when a subpopulation of cancer cells detach from the primary tumor and migrate to other tissues in the body via the blood or lymph. Clinical studies show that metastatic cells preferentially migrate to certain tissues, a phenomenon termed organ tropism. Recent research suggests that the cells that do migrate have stem-like properties (termed “cancer stem cells”), such as the combined ability to self-replicate and differentiate into other cells that compose a heterogeneous tumor. Breast cancer stem cells are identified by their high aldehyde dehydrogenase activity (ALDH) and/or $CD44^{+}CD24^{-}$ phenotype. It is unclear whether properties of the cancer cell or the organ microenvironment (or both) facilitate organ tropism. This study will use whole population and $ALDH^{hi}$ $CD44^{+}CD24^{-}$ breast cancer cells to investigate mechanisms behind the breast cancer cell’s affinity for the bone microenvironment by attempting to identify soluble factors in bone marrow that influence metastasis. It is hypothesized that at least 1-2 protein(s) bone-specific soluble factors will facilitate breast cancer cell growth and migration. Bone-conditioned media generated from athymic nude mice will be analyzed for the expression of different soluble factors. Soluble factor(s) of interest (1-2) will be depleted from bone-conditioned media using immunoprecipitation, and growth/migration of whole population and $ALDH^{hi}CD44^{+}CD24^{-}$ cells will be assessed.

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Assessing Genomic Variance In Families With Monozygotic Twins Discordant For Schizophrenia

Z. Awamleh, Western University; C.A. Castellani, Western University; M. Melka, Western University

Schizophrenia; copy number; Genome wide association; software packages

Schizophrenia is a mental disorder that generally affects individuals 16-34 years of age. Patients diagnosed with schizophrenia suffer a spectrum of psychotic symptoms and cognitive impairments. These include hallucinations, lack of motivation, social withdrawal, disorganized thinking and lack of concentration. With limited treatment options and poor understanding of the disorder, further studies are required to untangle the complexity of schizophrenia. Many psychiatric diseases, including schizophrenia, have been attributed to both environmental factors and genetic predisposition. A genome wide association approach will investigate genetic variation between affected subjects and non-affected subjects (control), in order to find a link between the genetic variation and the disease phenotype. To understand the underlying genetics of schizophrenia, recent studies have focused on the analysis of copy number variations in schizophrenic patients. Copy number variations are a type of genetic structural variation where large sections of DNA are deleted or duplicated. Copy number variations can range in size from one kilobase to several megabases.

The current genome wide association study being conducted focuses on copy number variation in six pairs of monozygotic twins discordant for schizophrenia. DNA samples were analyzed for copy number variations using the Affymetrix® human SNP 6.0 array™. Fluorescent signals were detected and a data file was generated for each subject. The data files generated were then input into a comprehensive analysis software package to further analyze copy number variations. Analysis softwares packages are used for genotyping DNA samples, but more importantly they are used to estimate copy number variations. This project uses Partek® Genomics Suite™ to assess and estimate copy number variation in all subjects. In addition, the output from Partek® Genomics Suite™ is compared with the outputs from two other software packages to increase sensitivity and reproducibility of copy number calls. The other software packages are Affymetrix® genotyping console™ (analysis completed by Melkaye Melka, Post-Doctoral Fellow) and Golden Helix SVS Suite 7.0 (analysis completed by Christina Castellani, PhD Candidate).

Partek® Genomics Suite™ detected 462 copy number variable regions (CNVRs), compared to Affymetrix® genotyping console™ which detected 563 CNVRs and Golden Helix SVS Suite 7.0 which detected 2241 CNVRs. Using a ≥ 50% reciprocal overlap rule, it was determined that Affymetrix® genotyping console™ shares 172 CNVRs and 5 CNVRs with Partek® Genomics Suite™ and Golden Helix SVS Suite 7.0 respectively. Meanwhile, Partek® Genomics Suite™ and Golden Helix SVS Suite 7.0 share 17 CNVRs. Several commonalities and differences have been identified between twin pairs and some CNVRs have also been found to overlap functionally important coding regions within the genome. Next steps will include confirming some of the detected CNVRs using traditional methods such as quantitative polymerase chain reaction.

Identification of Polymorphisms in the Protein C and Endothelial Protein C Receptor (EPCR) Genes that Impair PC-EPCR binding and Contribute to the Risk of Unprovoked Venous Thromboembolism

T.N. Nguyen, McMaster; L. Pepler, McMaster; P. Liaw, McMaster

Coagulation, Protein C, EPCR, APC, Venous Thromboembolism

Unprovoked Venous Thromboembolism (VTE) is characterized by the formation of blood clots that will obstruct the blood flow in the veins of the legs, called deep vein thromobosis (DVT) or in the lungs known as pulmonary embolism (PE). The occurrence of VTE is due to a disruption of hemostasis, which is the balance of procoagulant and anticoagulant pathways, where VTE is a result of a hypercoagulation state. The protein C (PC) anticoagulation pathway is an important regulator of the coagulation pathway. In the presence of thrombin bounded to thrombomodulin (TM), PC will bind to endothelial protein C receptor (EPCR) and become activated. We hypothesize that genetic mutations found in either PC or EPCR genes that impair the binding of the two proteins may be a risk factor of unprovoked VTE. We sequenced exon 3 of PC and 3 of EPCR in 209 patients with unprovoked VTE enrolled in the DODS study and college controls that have no history of VTE. Any mutations found in these exons may lead to an impairment of the PC-EPCR binding interactions leading to an impaired anticoagulation response. We will also characterize the effects of a mutated EPCR protein on APC activation in mice. Our data suggests that any genetic mutations that affect the interactions between PC and EPCR may increase the VTE risk.

The Mouse Diversity Genotyping Array Determines Copy Number Variable Regions In Mice With Implication To Man.

S.E Sherman, Department of Biology, The University of Western Ontario

CNV- Copy Number Variant; MDGA- Mouse Diversity Genotyping Array; HD-CNV– Hotspot Detector for Copy Number Variants

The genome, by nature, is constantly subject to mutational change brought about by various mechanisms, and even subtle aberrations in sequence can have dramatic effects on phenotype. There are alterations which enable individuals to confer resistance against diseases, or even be subject to life threatening diseases. Many of these alterations are associated with copy number variants (CNVs) throughout the genome. CNVs are alterations which are greater than 500 base pairs in size and can result from deletions or duplications which can happen through many mechanisms. Interestingly, areas of the genome that are dynamic in CNV activity appear to happen in a non-random fashion in functionally relevant regions correlating to fitness. For example, there are individuals that are naturally resistant to infectious diseases like HIV/AIDS associated with CNVs in immune regions. Other species, like the dog, also display CNVs in immune regions which suggest the non-

random characteristic of these CNV events. Mice have served as model organisms for human disease as about 99% of genes in the mouse have a human homolog. However, limited information is available for CNV data from the whole genome as studies are limited to low-resolution platforms. Because CNVs are associated with many modeled diseases like cancer, Huntington's, schizophrenia, and intellectual disabilities it is quite appropriate to probe the whole mouse genome. Ideally, both the least and most extreme ends of genomic variation in mice are of interest because they represent both the most likely regions to be inherited (inbred strains) and the regions that are prone to *de novo* CNVs (Wild Caught strains). To do this, the most comprehensive SNP-based array for the mouse, the Mouse Diversity Genotyping Array (MDGA), is being used. This platform rivals the human SNP 6.0 array as it contains approximately 1 million probes that span both functional exons and SNPs throughout the genome.

In this experiment, DNA samples from the tail tissue of 336 mice were prepared and hybridized on the MDGA. Various software programs were used to interpret the fluorescence data obtained from the array. Affymetrix Genotyping Console was first used to determine the genotype of the samples. The output was used by a software program called PennCNV to determine the copy number state of each loci. Each of the identified CNVs were then grouped and merged based on their overlap by a newly developed software program called HD-CNV. This allows other software programs to be used to visualize graphically how regions between mice share CNV events, warranting further investigation.

In investigating CNV regions common to the inbred and wild caught mice strains, HD-CNV identified regions containing recurrent CNVs on chromosomes 4, 11, and 17. These regions in the mouse contained CNVs that overlapped both the most homozygous strain of mouse, C57BL/6J, and the most diverse mouse strains being the wild caught. Within the identified recurrent CNV events, genes involving immunity, neuronal responses, and metabolism were identified. The identified genes are homologous to genes in the human which also contain CNV events according to the Database of Genomic Variants. This brings to light there are recurrent CNV events that occur non-randomly throughout the genome and can also be found in other species. The findings presented have helped to identify CNV 'hotspots' that are ideal for future study as these CNVs associated with genes involved environmental interaction. This phenomenon may have implications from evolutionary concepts to diagnosis of various potential conditions in a medical setting.

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The Effects Of CCCTC-binding Factor On Alternative Splicing In The Brain

E.P.Yu, Western University; N.G. Bérubé, Depts of Pediatrics and Biochemistry, Western University

CTCF, alternative splicing, rtn4, nrp2, kif1a

CCTCF-binding factor (CTCF) is a ubiquitously expressed and highly conserved transcription factor. Recently, CTCF binding was found to increase stalling of RNA polymerase II during transcription leading to increased inclusion of alternative exons. As alternative splicing is common in the central nervous system, we investigated alternative exon inclusion in murine brain null for CTCF. The purpose of this project is to determine whether there is a change in exon inclusion in neural development genes chosen due to preliminary findings from RNA-sequencing and CTCF-ChIP-sequencing data.

Mice null for CTCF in the rain were generated by Nestin promoter driven Cre Recombinase expression in CTCF flox/flox mice. Preliminary findings were obtained from RNA sequencing of RNA extracted from the whole brain of E14 littermate pairs generating a list of genes with alternatively expressed exons. Contrary to

reported findings, our data showed increases in alternative exon inclusion in CTCF null brain compared to wild type. Three genes with this expression pattern and CTCF binding at the site of the specific exon were selected for validation and further investigation. Neuropillin-2 (*nrp2*) is involved in neuron migration and axon guidance, kinesin-like motor protein 1a (*kif1a)* is responsible for transport of axonal synaptic vesicles and reticulon-4(*rtn4*) has a role in regulating neurite outgrowth. Exons sixteen of nrp2, two of kif1a, three and five of rtn4 were investigated with two or one sets of primer pairs by quantitative real time (q-RT)-PCR. Results for relative expression (expressed as average fold change± SE) as compared with the housekeeping gene *GAPDH* are as follows:

- Nrp2: 2.06±1.15(n=4) and 1.05±0.23 (n=5)
- Kif1a: 1.14±0.05(n=5) and 1.18±0.14 (n=5)
- Rtn4 exon 3: 0.852±0.107 (n=5)
- Rtn4 exon 5: 0.996±0.14 (n=5) and 0.896±0.09 (n=4)

A one-tailed, paired Student's t-test was performed and *kif1a* was observed to have significant exon inclusion differences between CTCF null and wild type brains ($p=0.01$, $p<0.05$). No other exon showed significant differences, thus the preliminary findings are largely not supported. CTCF binding appears to have a more complex effect on exon inclusion than expected as results indicate some exons may be included in the absence of CTCF as is the case with *kif1a*.

Sunday March 17th, Session 4, Room 2 – MDCL 1110, 10:45 – 11:45 am
Ecology (Behaviour) -

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Fear On The Brain: Using Immediate-early Gene Expression To Identify The Avian Fear Centre

E.C. Hobbs, Department of Biology, The University of Western Ontario, London, Ontario, Canada; L. Zanette, Department of Biology, The University of Western Ontario, London, Ontario, Canada

Predator-prey ecology; Predator stress; Population dynamics; Nucleus taenaie of the amygdala; cognition

Predator-induced stress is now recognized as a force that can result in lasting effects on the demographics of prey populations, affecting behaviour and reproduction. Focusing on predators as direct killers may greatly underestimate their impact, as exposure to predator cues can result in lasting effects on prey populations and demography. Recent studies have shown that songbird reproduction is significantly decreased upon exposure to auditory predator cues in the absence of direct predation. It is unknown whether predator-induced stress alters the avian brain, resulting in these impacts on reproduction. The mammalian amygdala is known to be a part of a network relating to emotions, motivation and fear learning. Whether there is an analogous network in the brains of wild birds is not well understood. Recent studies have shown activation in the nucleus taeniae of the amygdala (TnA) in birds in response to indirect predator cues. This brain area may be analogous to the mammalian amygdala. Testing of immediate-early gene (IEG) expression in the avian brain is essential to confirm lasting neurological changes due to exposure to predator stress. This study quantifies IEG expression of the genes *c-fos*, *fosB* and ZENK in the TnA and other brain regions in Black-capped Chickadees (*Poecile atricapillus*) using immunohistochemistry following exposure to auditory predator cues. ZENK and *c-fos* are rapidly expressed and down regulated upon exposure to predator cues, providing a short-term signature in the brain. *fosB* can remain elevated for days after predator exposure in mammals. Therefore, the quantification of the expression of these genes in the brains of Chickadees exposed to predator threat is essential in order to determine if gene expression is altered over time in the avian brain in response to auditory predator cues. This study is the first to investigate the use of *fosB* for IEG study in the avian brain. Results obtained lay the groundwork to understanding predator-induced stress and its short- and long-term impacts on the avian brain and provide a link between neurological changes and prey demography.

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Predator-Prey Evolution In A Novel Evolutionary Simulation, Ecosim: Effects On Energy & Fitness

D. Murariu, University of Windsor; M. Khater, University of Windsor; R. Gras, University of Windsor

Ecological individual-based modelling, Fitness, Energy tradeoffs, Nonlethal effects, Predator-prey models

A longstanding question in ecosystem biology is how predators exert population control on lower trophic levels. The literature divides the study of mechanisms of such control into two groups: direct effects, characterized by direct consumption of prey, and indirect effects, in which prey modify their behaviours in response to predation risk. The latter comes at the cost of reduced food intake, especially during periods of high risk, and this strains energy allocation to growth and reproduction. In this study the long-term impact of variable energy uptake on prey fitness is investigated over an evolutionary time scale. We used an individual-based simulation already developed, EcoSim, to follow the evolution of prey in three conditions: prey coevolving with predators, prey evolving without predators, and a removal scenario in which predators were removed after a substantial period of coevolution. The simulation is based on a tritrophic system of primary producers (grass), herbivores (prey) and predators which shows predator-prey population dynamics. In addition, it allows complex behavioural interactions both between and within trophic levels. Preliminary results revealed that prey show a tradeoff between foraging behaviours and predation risk, and this affects their energy. Also, predation pressure appears to impact prey fitness in terms of offspring productivity, parental investment of energy in their offspring, and likelihood of dying from low energy. Our results thus far suggest that predation pressure influences prey behaviour both through indirect and direct effects and that this influences fitness and population demographics.

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An Evaluation Of Alternate Predation Rate Models In A Zooplankton Microcosm

C.M. Prokopenko, University of Guelph; J.M. Fryxell, University of Guelph; K. Turgeon, University of Guelph

Functional response; predator interference; feeding motivation; Cypriconcha ingens; Brachionus Calyciflorous

Functional response (*i.e.* the consumption rate of a predator) was originally thought to be influenced by prey density alone (*i.e.* prey-dependent). However, there is a recent debate regarding the addition of predator density in predation rate models (*i.e.* predator dependence and ratio dependence). In this study, we compared the ability of alternative predation rate models; prey dependent, predator dependent and ratio dependent, to predict feeding rate in a simple zooplankton microcosm. Feeding experiments were completed in a microcosm using ostracods (*Cypriconcha ingens*) as predators and rotifers (*Brachionus calyciflorous*), as their prey. Consumption rate was measured at various predator and prey densities. The change in prey density was assessed at regular intervals during one hour feeding trials. Starved and fed ostracods were used in the feeding trials to determine the effect of feeding motivation on consumption rate.

We compared nine models:

- Null
- Holling Type I prey dependent
- Holling Type II prey dependent
- Type I Arditi-Ginzburg ratio-dependent
- Type II Arditi-Ginzburg ratio-dependent
- Type I Beddington-DeAngelis predator dependent
- Type II Beddington-DeAngelis predator dependent
- Type II Arditi-Akcakaya predator dependent
- Type II Tyutyunov predator dependent

In starved conditions, both prey and predator density influenced ostracod consumption rate. Overall, the functional response exhibited a Type II relationship, where the rate of consumption increased non-linearly with increasing prey density and was depressed as predator density increased. The three predator interference models, Beddington-DeAngelis, Arditi-Akcakaya, and Tyutyunov, fit most parsimoniously with the observed feeding rates compared to the prey dependent and the ratio-dependent models. However, there was little to separate between the explanatory power of these three models.

Data analysis currently in progress includes:

- Model evaluation of short-term consumption rates for predators in the fed condition.
- Model evaluation of long-term consumption rates for predators in both fed and starved conditions
- Comparison of the predator consumption rates between the fed and starved conditions.

The highest predator densities used in this study (4 ostracods/5mL) were far below those occurring in the study population (29 ostracods/5mL). Therefore, predator interference is likely to occur in ostracod populations and influence their consumption rates, in addition to prey density.

The Use of Pegboards to Reduce Train/Wildlife Collisions

S. Reid, Laurentian University; M. Hall, Ministry of Natural Resources; J. Hamr, Cambrian College; J. Litzgus, Laurentian University

Pegboard; Ungulate; Ethogram, Railway Mortality

Trains can be a major cause of mortality for many wildlife species, especially for ungulates. Ungulates are often attracted to railways by food and water availability on right-of-ways. Studies throughout North America and Europe have documented large numbers of annual train/Moose mortality. In all studies mortality was

higher during winters with deep snow because ungulates use plowed railbeds as low energy travel corridors. It is a common flight response for ungulates to run along the railbed to try to out-distance trains instead of jumping to the side, resulting in collision. Mitigation is particularly important in the Greater Sudbury area because it is home to a small and slow growing population of reintroduced Elk (*Cervus elaphus*). Our study tests a new method of mitigating train/ungulate collisions using pegboards.

The pegboards we are testing are modified from a design used in Banff National Park to deter grizzly bears from the railway. The idea is that when animals step on the pegboards during flight response from an oncoming train, the pegs will cause discomfort resulting in the animal jumping off of the railway. Pilot tests took place at an Elk/Red Deer farm just north of Sudbury. Data were collected using motion-activated cameras and animal behaviours related to the pegboards were scored using Ethograms. Behaviours scored included; sniff, vigilant, walk between, step on, and walk away from the boards. Chi-square tests found that there were significant differences in observed behaviours and that animals habituated to the pegboards presence.

The pegboards are currently being tested at two predetermined high kill areas on the Canadian National Railway south of Sudbury. Camera data and Ethograms will again be used to examine animal interactions with the pegboards. To date, Moose and Deer have been observed walking alongside the railway; however, animal interactions with the pegboards have not yet been observed. During the study period, trains in the surrounding area have killed 2 Elk and at least 5 Moose, therefore it is important to develop new ways to mitigate these collisions.

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Transient Expression Of VP2 And P22sTsp Recombinant Proteins In *Nicotiana Benthamiana,* Using Four Unique Constructs

S. Miletic, Western University; S. Kohalmi, Western University; R. Menassa, Agriculture Canada

Plant Bioreactor; Vaccine; Bacteriophage: Hydrophobin; Elastin-like Polypeptide

Plants have been naturally producing compounds utilized by humankind for centuries. The idea of using plants to produce foreign proteins for pharmaceutical or industrial purposes originated in the 1980s with the first demonstration of transforming DNA into plant cells using *Agrobacterium tumefaciens.* Since then, research into using plants as bioreactors has blossomed and several proteins have been expressed.

Like other eukaryotes, plants can correctly assemble, fold, and modify recombinant proteins. Also, plant expression systems are safer, cheaper and easier to scale up than traditional expression systems such as bacteria, yeast, and insect cell cultures. Consequently, several recombinant proteins have been expressed in plants; however, accumulation levels were too low for large-scale production.

Several different technologies have been employed to increase protein yield. The use of fusion tags, including elastin like polypeptides (ELPs) and hydrophobins (HFBI), has been shown to enhance protein accumulation and purification. It is speculated that fusion tags increase protein solubility and thus, enhance stabilization.

The focus of this ongoing study is to transiently express two recombinant proteins, VP2 and P22sTsp, in *Nicotiana benthamiana.* VP2 is a porcine parvovirus (PPV) structural protein and is a potential subunit vaccine for PPV in swineherds. P22sTsp is a bacteriophage tail-spike protein, which reduces *Salmonella* colonization in infected chickens. These proteins will be fused to peptides targeting the chloroplasts, the endoplasmic reticulum (ER) and to the ER fused with either an ELP or a HFBI tag. It was hypothesized that VP2 and P22sTsp protein will be expressed in *N. benthamiana* and that protein yield will be highest when fused to an ELP or HFBI tag.

VP2 and *P22*s*Tsp* genes were recombined into four pCAMgate vectors and agroinfiltrated into *N. benthamiana.* Plant tissue was extracted 3,4,5,6, and 7 days after infiltration and protein was extracted using sodium dodeclysulphate-polyacrylamide gel electrophoresis and visualized by immunoblotting No protein was detected for the VP2 constructs. Repeating the infiltration along with employing different extraction techniques failed to further detect VP2 protein. However, reverse transcription (RT) PCR analysis revealed mRNA expression for each of the constructs. It is suspected that the VP2 recombinant protein is unstable and is being degraded after translation.

Conversely, protein of the expected size was visualized for the P22sTsp constructs targeted to the ER, ER fused with an ELP and ER fused with a hydrophobin, but not when targeted to the chloroplasts. Expression was highest five days after infiltration and when the protein was fused to either the hydrophobin or ELP tag.

Further research should focus on expressing VP2 and purifying P22sTsp in *N. benthamiana*. This research shows promise in terms of using a new expression system to efficiently produce recombinant proteins to potentially reduce viral and bacterial infection in animals.

Elucidating the Structure and Function of *Streptomyces hawaiiensis* ClpP proteases

P. K. Patel, McMaster University

Bacteria; Protease; Antibiotic; Resistance; Medicine

Proteases are involved in virtually every aspect of the bacterial physiology, including timing cell division cycle and responses to heat shock and other stresses. Proteases are also key contributors to the maintenance of protein homeostasis. They are critically important for the overall fitness of bacteria and contribute to virulence. One such key protease that is present in several bacterial species is caseinolytic protease P (ClpP), which resides in the cytoplasm of the bacteria and mediates degradation of proteins. ClpP is capable of degrading small peptides by itself but it requires the association of ATPases in order to degrade larger substrates. Until recently, the ATPases were considered to be absolutely required to activate the proteolytic function of ClpP. However, a new class of antibiotics, the acyldepsipeptide (ADEP) molecule, has allowed the ClpP proteases to become activated without the presence of its cognate ATPases, resulting in unregulated degradation of polypeptides. Binding of these compounds to ClpP overrides the protease inhibitory mechanisms and indiscriminately increases its activity. ClpP then targets newly synthesized proteins for degradation, which dramatically impacts bacterial growth and kills bacteria. ADEP is produced by *Streptomyces hawaiiensis,* a gram-positive, spore forming bacteria and it possesses six ClpP genes of its own. Previous studies of ClpP in *Escherichia coli, Bacillus subtilis* and other bacterial species have indicated that the bacteria in unable to survive in the presence of this ADEP molecule due to dysregulation of ClpP activity. However, it is uncertain as to why *S. hawaiiensis* is able to survive the deleterious effects of ADEP. My research focuses upon uncovering the structure and function of the ClpP proteases of *S. hawaiiensis* and its method of resistance to this lethal ADEP molecule. Structurally, it was found that ClpP assemble in a similar yet a different tetradecameric complex than those studied in other bacterial species. The functions of three of the ClpP proteases, ClpP-ADEP, ClpP1 and ClpP2 of *S. hawaiiensis* were tested against various substrates in the presence and absence of the ADEP molecule and ClpP1ClpP2 was found to be highly active in the presence of ADEP. Its mode of resistance against this antibiotic was discovered within one of its key ClpP proteases, ClpP-ADEP, which is absent in other bacterial species.

Expression Of The Alternative Oxidase In The Alpha-proteobacterium *Caulobacter segnis* In Response To Environmental Stress.

K.A. Shelton, Wilfrid Laurier University; A.E. McDonald, Wilfrid Laurier University.

Alternative Oxidase (AOX); Bacteria; Caulobacter; Respiration; Protein.

The alternative oxidase (AOX) protein is a haem-independent alternative terminal electron acceptor that has been found in the respiratory chain of a wide range of taxa including all higher plants, some fungi, some animals, algae, various protists and Eubacteria. The AOX proteins within angiosperm plants have been extensively investigated revealing that the protein participates in the inner membrane mitochondrial electron transport chain, branching at the point of ubiquinone, thereby bypassing the two of the three proton pumping complexes associated with the cytochrome c (COX) pathway. The protein itself pumps no protons across the inner mitochondrial membrane, but instead catalyzes the conversion of ubiquinol and molecular oxygen to ubiquinone and water. As a result, the AOX pathway is more energetically wasteful than the traditional COX pathway, producing less ATP; however unlike the COX pathway, AOX is said to be “cyanide-resistant” since it has been shown to be insensitive to all known cytochrome c oxidase (Complex IV) inhibitors. We have investigated the presence of AOX within the alpha-proteobacterium *Caulobacter segnis*. *Caulobacter* species are nearly ubiquitous upon the surface of the earth and are known to degrade alkane compounds within cold soil environments where AOX activity has likely been underestimated. Samples of genomic DNA were obtained from cultures of *C. segnis* and were subjected to PCR amplification with three different sets of AOX primers thereby confirming the presence of the AOX gene. Samples of total RNA were obtained from liquid culture samples of *C. segnis* in laboratory control and cold stressed conditions. The primers were then used to amplify AOX mRNA transcripts from DNase treated total RNA samples using reverse transcriptase PCR suggesting that the AOX genomic sequence is actively transcribed. Outer membrane protein samples were also obtained from *C. segnis* to allow for future confirmation of AOX protein expression through western blotting. By investigating how the transcription and expression of the AOX protein may change in *C. segnis* in response to environmental stress (such as low temperature, external oxidative stressors and low phosphate levels), further hypotheses about the physiological function and regulation of bacterial AOXs can be generated. Furthering our metabolic understanding of these bacteria may have an impact in the fields of microbial ecology and biotechnology and may aid in the development of novel bioremedial strategies to purify hydrocarbon contaminated soils. Furthermore, by establishing the tools and protocols necessary to study the AOX protein of *C. segnis,* this research can be expanded to the study of AOX homologs found within other bacteria.

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Effects of Inorganic Contaminants on Nutrient Reserves and Molt in Autumn Migrant Red-Necked Grebes (Podiceps grisegena) at Georgian Bay

K.L. Holman, Western University

Red-necked Grebes; Environmental Contaminants; Body Composition; Great Lakes

Red-necked grebes (Podiceps grisegena) are aquatic birds that breed on freshwater lakes in north-western Canada and winter on the Atlantic coast. Large numbers of red-necked grebes stage on Georgian Bay, Ontario where they replace their flight feathers. They are high trophic predators and are considered good indicators of ecosystem contamination. Over the past few decades, studies have highlighted health and reproductive effects due to bioaccumulation of environmental contaminants in fish, birds, mammals and reptiles, but data on contaminants in red-necked grebes are limited. The objective of this study is to determine if autumn migrant red-necked grebes are bioaccumulating environmental contaminants, and if so, are elevated concentrations affecting their fat and protein mass reserves and molt progression. It is hypothesized that nutrient reserves will vary negatively with contaminant burden. Furthermore, since molt is an energetically expensive event it is hypothesized that there will be a positive correlation between nutrient reserves and autumn molt intensity.

Proximate analysis was conducted on a sample of 21 autumn migrant red-necked grebes to determine fat mass and protein mass in these grebes. Liver samples were analyzed by the Environmental Analytical Laboratories at Laurentian University in Sudbury, Ontario to determine concentrations of aluminum (Al) arsenic (As), cadmium (Cd), copper (Cu), chromium (Cr), iron (Fe), lead (Pb), magnesium (Mg), manganese (Mn), mercury (Hg), nickel (Ni), selenium (Se) and zinc (Zn). Linear regression was used to determine the relationships between contaminant burdens and nutrient reserves. Sex was removed from the model because neither body condition –after being corrected for size using principle component analysis (PC1)– nor contaminant concentrations differed by sex. The model showed that eight contaminants were significantly related to fat composition: As, Cu, Fe, Hg, Mg, Ni, Pb, and Se; and five contaminants were related to protein mass composition: Al, Ca, Fe, Hg and Zn. Fe and Hg were the only contaminants to demonstrate significance for both fat mass and protein mass.

Molt score was determined by estimating molting feather coverage in 20 feather tracts to the nearest 5%. Feather tracts were then grouped into four regions: 1) head and neck, 2) back, 3) abdomen and 4) wings. Correlation analysis was conducted to evaluate the relationship between molt intensity and lipid or protein content adjusted for body size (PC1). The results showed that the only significant relationship was a negative correlation between adjusted fat and abdomen molt.

The results of this study will contribute to our understanding of how contaminants could affect red-necked grebes and other top predators staging on the Great Lakes.

The Effect of Perceived Predation Risk on Flight Performance in Brown-Headed Cowbirds

D.A. Landsman, Western University; L.Y. Zanette, Western University

Birds; Predation Risk; Stress; Flight Performance;

Increased predation risk results in a reduction to an individual bird's body mass — an important adaptation which improves take-off performance, maneuverability in flight and therefore escape success. A potential trade-off exists in escape success and flight endurance through the reduction of fuel reserves. I manipulated perceived predation risk in brown-headed cowbirds (*Molothrus ater*) to examine its effect on body composition and take-off ability. Fat and lean muscle mass was measured using quantitative magnetic resonance (QMR) body composition analysis and take-off ability was evaluated in a specially-built flight chamber. This research was conducted at the Advanced Facility for Avian Research (AFAR) at the University of Western Ontario.

Differences In Molt Intensities In Long-tailed Ducks (*Clangula hyemalis*) During Autumn And Winter

A.M. Payne, Western University

molt intensity; Long-tailed Duck; energy budgeting; molt suspension; migration

Feather molt and migration represent two major energetic events for waterfowl species that migrate bi-annually throughout North America. Waterfowl with low protein and lipid levels often delay molt until after migration due to nutrient constraints. This molt delay can be observed through individual differences in molt intensities. The purpose of this study was to make within-season comparisons of molt intensity and body condition of autumn migrating and wintering Long-tailed Ducks (*Clangula hyemalis*), hereafter LTDU. Molt scores, lipid levels and protein levels were determined for 79 birds salvaged during autumn migration and 255 birds collected during winter. I predicted a positive correlation between molt intensity and body condition, such that individuals in better condition would have higher molt scores. It was hypothesized that LTDU interrupt molt during migration and thus, molt intensity would be low during autumn migration as a direct result of lower lipid and protein levels. It was also hypothesized that due temporal differences in reproductive commitments, males will molt less intensively than females and juveniles within-seasons. A negative correlation was detected between molt intensity and both lipid and protein, with the exception of a slight positive relationship between protein level and molt score in wintering LTDU. Adult males and females molted at similar intensities in autumn but females and juveniles molted at higher intensities than adult males during winter. Therefore, LTDU do appear to at least partially suspend molt during autumn migration and adult males molt at lower intensities during winter, possibly due to the additional molt time afforded them after their reproductive effort.

Does Heterozygosity or Locally Common Genotypes Better Correlate with Estimators of Physiological Condition in Song Sparrows

N.E. Westwood, University of Western Ontario; E.A. MacDougall-Shackleton, University of Western Ontario

Heterozygosity; Locally common genotypes; H:L ratios; Song Sparrow

The fitness of an individual is assumed to be very dependent on their genetic make up. Previous hypotheses suggest that individual fitness may be increased through heterozygote advantage, as well as locally common genes. To test these hypotheses, I examined the heterophil:lymphocyte (H:L) ratios of 32 individual adults song sparrows (*Melospiza melodia melodia*) to estimate physiological condition and compared it to the estimated heterozygosity and genetic origin of each individual using seven loci. Results will be discussed.

Sunday March 17th, Session 4, Room 5 – MDCL 1009, 10:45 – 11:45 am
Genetics -

541

Identification And Characterization Of Novel Loci In *Phormia regina*

K.M. Farncombe, McMaster University

Microsatellite; Phormia regina; Post Mortem Interval

Blowflies disperse to locate a new food source during their larval stage, and attracted by the odour of decomposing carrion, quickly colonize the body of recently deceased organisms. The ability to identify the species and developmental stage is a useful tool for the forensic entomologist in establishing the time of death or post-mortem interval. A multitude of ongoing projects use microsatellites, amplified fragment length polymorphisms, and next generation sequencing in the interest of being able to distinguish between similar species such as *Lucilia illustris* and *Lucillia casesar*. These two species inhabit corpses at different rates despite their likeness; hence, a misidentification leads to the establishment of an inaccurate time of death. This project seeks to develop a microsatellite profiling system for *Phormia regina* through PCR amplifications, ABI runs, sequencing on a 3730 DNA Analyzer and computer analysis using microsatellite toolkit, GenAlEx, and GENEPOP. The identification and characterization of novel loci will allow for faster and easier identification of *P. regina* samples among the blowfly population inhabiting corpses.

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Evolutionary And Behavioural Variation In Sequential Mating Decisions In *Drosophila melanogaster*

H.J. McLeod, Wilfrid Laurier University; T.A.F Long, Wilfrid Laurier University

Sexual Selection; Sperm Competition; Experimental Evolution; Drosophila melanogaster, Female Preference

In species where females mate multiply, decisions with whom to mate may be influenced by a number of different factors related to the direct and indirect costs and benefits of choosing one male over another. For an individual female, weighing these costs and benefits may be influenced, not only by the the qualities of her current prospective mates, but also by the characteristics of her earlier mates. At a broader scale, these costs and benefits may also vary between species or populations due to differences in their specific evolutionary trajectories. In this study, I sequentially exposed females from replicate experimentally evolved populations of *Drosophila melanogaster* (the fruit fly) that had experienced divergent selection on female preferences for male body size, to either large or small-bodied males. Frequencies, of mating, remating, and offspring production were measured in

order to test whether the outcome of female mating decisions could be influenced by individual mating histories, as well as by selective histories.

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A Test for Genetic Influences on Caste Differentiation in the Eastern Subterranean Termite

D.A. Scaduto[1]; G.J. Thompson[1], Department of Biology, Western University, London, ON, Canada

Population genetics; microsatellite; caste differentiation; invasive; Reticulitermes flavipes

Invasive pests such as termites represent one of the most economically important insects in the world. Although caste differentiation in eusocial insects is thought to be primarily influenced by environmental factors, there is growing evidence that genetic factors may also contribute. In this study I use population genetics to test for associations between microsatellite genotype and caste phenotype in the Eastern subterranean termite (Reticulitermes flavipes). Specifically I use microsatellite DNA analysis to assess whether certain genotypes are more likely to develop into reproductive or non-reproductive castes. Termites (n=320) collected from three unrelated populations across Ontario were sorted by caste (soldier, worker, nymph), sexed, and genotyped at ten polymorphic DNA loci. Pair-wise exact tests for genetic differentiation between castes within a single colony revealed a strong association between genotype and caste at one particular locus. This effect is significant across multiple colonies and does not appear to be sex-linked. Thus, along with environmental influences these results reinforce the notion that genetic factors may also contribute to caste differentiation.

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Effects And Interactions Of Genetic Identities On Maternal Investment Patterns In *Drosophila melanogaster* Mating Pairs

E.E. Sonser, Wilfrid Laurier University; T.A.F. Long, Wilfrid Laurier University

Maternal Investment; Genetic Identity; Differential Allocation; Sexual Selection; Drosophila melanogaster

In species where multiple mating is common, members of both sexes may be selected to invest differentially into their (costly) offspring depending on the identity (and associated qualities) of their current mate, in order to maximize individual lifetime reproductive success. In many species of insects, this differential allocation might be manifested as changes in the number and/or size of eggs laid following copulation between two individuals. In order to better understand phenotypic variation in maternal investment patterns, and whether genetic variation for this plasticity exists in males and/or females (and whether or not their specific genetic identities interact), we conducted a series of assays using 12 hemiclonal lines derived from a large, outbred population

of fruit flies, *Drosophila melanogaster;* a model species with strong sexual selection and substantial sexual conflict. Using cytogenetic cloning techniques, we generated hemiclonal male and female individuals (each sharing the same haploid genome) from each line, mated replicate hemiclonal males and females from different lines, and then counted, photographed and measured the volume of eggs laid. This approach allowed us to quantify the contributions of each sex, both individually and in combination, to short-term variation in post-copulatory offspring investment strategies..

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Zoobenthic Community Composition Trends In Constructed Boreal Wetlands – Residual Effects of Oil Sands Mining By-Products

H.L. Bagnall, University of Windsor; K.E. Kovalenko, University of Windsor; J.J.H. Ciborowki, University of Windsor

Wetlands; Invertebrate; Community; Reclamation; Oil Sands

Oil sands open-pit mining near Fort McMurray Alberta is an important economic resource for Canada causing extensive disturbance to the landscape. Once mining is completed, the companies must reclaim the landscape to pre-mining levels of productivity, diversity, and abundance. One reclamation strategy involves constructing wetlands using waste materials (clay-rich tailings and process waters). To better understand whether use of mine materials is an effective reclamation strategy we tracked changes in invertebrate richness, abundance and composition in oil sands material and reference constructed wetlands. Zoobenthos were collected from colonization tiles and enumerated and identified in the lab. Preliminary analysis suggests that mine-affected wetlands support equivalent abundance and slightly lower richness than reference constructed wetlands, but that there are persistent differences in community composition. This study will help to determine whether or not these approaches to wetlands constructions will ultimately support acceptable zoobenthic communities in the reclaimed landscape after reclamation is completed.

712

The Effects Of Water Quality On Representative Bacteria Present In The Rhizosphere

C. S. Baschuk, Wilfrid Laurier University.

Bacteria, Wetland, Wastewater, Antimicrobial

Wetlands provide many valued services including nutrient cycling, carbon sequestration, water purification, and habitat for many organisms. Many wetland services are provided by or facilitated by soil bacteria, particularly bacteria present in the rhizosphere of wetland plants. Municipal wastewater treatment plants (WWTPs) release many compounds derived from pharmaceutical and personal care products into the environment. Among these are antimicrobial compounds found in consumer products ranging from shampoos and detergents, to cooking utensils and athletic supplies. These compounds are not completely removed by conventional WWTP processing and are discharged into watersheds at low concentrations. Due to their

nature, I hypothesize that areas receiving WWTP discharge will have a reduction in soil bacterial communities. This study was conducted to determine if water obtained from locations upstream and downstream of a WWTP influences bacterial communities in the rhizosphere of the emergent wetland grass *Phalaris arundinacea*. Six plants were obtained from a low water quality site and high quality site, placed in pots and grown in growth cabinet (16/8 light dark cycle, 27C/20C temperature). Plants were watered regularly with water obtained from the site of harvest. The buried slide technique was used to sample bacterial populations followed by heterotrophic plate counts and viewing with bright field microscopy to characterize bacterial communities. Contrary to our hypothesis, preliminary data suggests that low water quality has higher CFU/mL values (3.16×10^8 CFU/mL ± 7.06×10^8) compared to the high water quality treatment (8.65×10^7 CFU/mL ± 1.04×10^8). Further analysis is being conducted to examine relationships between water quality parameters and CFU formation.

713

Smooth-Billed Ani (*Crotophaga ani)* Adults Do Not Synchronize Provisioning

C. Bobbie, McMaster University; L. Grieves, McMaster University; J.S. Quinn, McMaster University

Provisioning; Nestling Competition; Ecology; Animal Behaviour

The Smooth-Billed Ani (*Crotogphaga ani)* is a joint nesting and cooperatively breeding cuckoo found abundantly in Florida, the Caribbean and South America and form groups of up to 17 individuals. Unrelated females lay eggs in a communal nest and all adults then rear offspring cooperatively. This unique reproductive strategy may increase nestling food competition due to the low levels of relatedness among nestlings. As such, we proposed that adults may synchronize provisioning to ensure that all offspring receive relatively equal amounts of food. However, we found no evidence of this synchrony and propose that other methods of ensuring provisioning equality may be practiced by the smooth-billed ani.

714

Most Forest Ecosystem Variables In Yellowstone NP Are More Stable Than In Adjacent Logged Forests

S.S. China, McMaster University; M.P. Hammond, McMaster University; J. Kolasa, McMaster University

Aggregate Variability; MODIS; Yellowstone NP; Ecosystem Stability; Clearcutting

Variability of a quantity or process can be related to its spatio-temporal pattern using the following

mathematical model: Aggregate Variation = ΣSpatial Variances + ΣTemporal Covariances - ΣSpatial Covariances. This model implies that landscape stability can be reduced by spatial differences among sites and can be increased by the persistence of those differences in space. As the measured variation is often scale-dependent, the perception of stability is sensitive to changes in scale. Our study aims to both test the model in a large natural system (forested landscape) and to determine the patterns of sensitivity of ecosystem variables to temporal scales of observation. To accomplish this, we used satellite data from NASA's MODIS spacecraft to examine the spatio-temporal pattern of net primary productivity (NPP), gross primary productivity (GPP), evapotranspiration, land surface temperature (LST), leaf area index (LAI), normalized difference vegetation index (NDVI), enhanced vegetation index (EVI), and fraction of photosynthetically active radiation (FPAR). The area comprises a 41x21 km^2 landscape in Targhee National Forest (Yellowstone) that has a distinctive divide between a forested area and a clear-cut area. We found that the forested area was more stable for 7 of the 8 variables (r-squared values: 0.379-0.611). Furthermore, at both short- and long-term scales, slopes of regression lines between variation in space and stability of individual variables were steeper for the disturbed landscape, with a greater difference arising in the long-term. These patterns imply several processes: 1) variables in the disturbed landscape undergo a dynamic shift in conditions as a result of being dominated by regional (environmental) and local (disturbance) factors; 2) natural landscapes show greater spatial diversity associated with greater persistence of local differences; and 3) the negative effects of anthropogenic disturbance become more prominent at longer time scales.

715

Factors That Influence The Stability Of Rock Pool Microcosms

S.A. Esteves, McMaster University; M.H.L. Kaas, McMaster University; J. Kolasa, McMaster University; M. Hammond, McMaster University

Stability; Community; Functional Groups; Microcosms; Invertebrates

The aggregate variability of a system as it relates to the variability of community stability, and functional group richness was explored. The aggregate variability of 49 rock pool communities from Discovery Bay, Jamaica were calculated using their spatial and temporal variability as well as species abundances. Feeding trials were conducted to determine feeding preferences for common zooplankton species. This information, combined with past literature and isotopic analysis, was used to classify rock pool invertebrates into one of five functional groups: water column feeders, benthic feeders, deposit feeders, large predators, and small predators. Functional groups were determined based on food source, method, and location of feeding. Functional group stability was then compared to stability calculations for aggregate number of species. It was found that more generalist communities were unstable while more specialist communities were more stable. However, the significant driver behind this observation was environmental variability, where the degree of specialization of a community did not significantly impact community stability. Additionally, there is a significant relationship between functional group richness and stability. Pools with more functional groups are more stable. However, there is also a significant relationship between increasing number of species and stability. Functional group classifications may provide a new way in which to determine the diversity of an aquatic community and result in a more accurate understanding of how biodiversity influences the stability of a system. These results are significant to conservation biology as they make it possible to infer variation of a community from the variation of species abundance, and give rise to the indirect ability to predict the risks of the extinction or loss of certain species from a system

Effect of Predation on Snail Shell Chirality

W.G. MacIntosh; D. Rollo; J. Stone

Predation; Snail; Speciation; Snake

Snail populations are characterized predominantly by the dextral shell chirality phenotype, while the sinistral chirality phenotype, often encoded by recessive alleles, is rare due to reproductively isolating mechanisms. However, fully sinistral populations not only occur in the wild, but do so with a greater frequency than expected from mutation and genetic drift effects. Reports in the literature suggest that other factors, such as predation, may explain this discrepancy. The snake species *P. iwasakii* is equipped with extra teeth in its right mandible. Research has shown that these right-jawed snakes feed primarily on dextral snails and exhibit difficulty handling sinistral snails. In the wild, the frequency of sinistral populations is greatly increased when *P. iwasakii* is present. The goals of this computational experiment were two-fold: first, to use Mathematica computer simulation software to show that selective predation against dextral snails increases the rate of sinistral fixation and, second, to try to replicate circumstances observed in wild populations.

It was found that including selective predation as a factor significantly increased the rate of sinistral fixation, and we were able to replicate the circumstances found in the wild.

Migratory Trends In White-Throated Sparrows (*Zonotrichia ablicollis*) Dependent On Hematozoon Infection

R.J. Mistry, Sir Wilfrid Laurier University

White-Throated Sparrow; Migratory pattern; Hematozoons; Blood smears; Isotope Analysis

Fitness levels of migratory birds can be dependent on their ability to maintain their metabolic rate by fighting off external environmental stimuli such as viruses or pathogens (Caldwell and Mills, 2006). If individuals internal conditions are impaired, they must find alternate ways to compensate, which could shift their migratory behaviours (Webster *et al*., 2012). Short distance migratory birds provide the best examples for studying the relationship between condition and migration (Caldwell and Mills, 2006). White throated sparrows are short distance migratory birds, that breed in central Canada, and travel to wintering grounds in southeastern America (Webster *et al*., 2012). Migratory patterns within both gender and morphed (white-stripe & tan-striped) white-throated sparrows can be greatly influenced based on the individuals conditions. Females tend to migrate further south to avoid harsh wintering conditions, while both morphs of males migrate shorter distances to strategize arriving earlier to breeding grounds for mate selection (Jenkins and Cristol, 2002). Questions therefore arise to whether migratory behaviors will change due to the individual's conditions. The objective of the study is to determine if increased parasite loads within white throated sparrows can cause

alterations in their migratory behaviours. Blood and crown feather samples were obtained from 152 male and female sparrows breeding in Algonquin Park, Ontario (2006-2010). Blood smears were collected and examined for the prevalence of Haemoproteus and Leucocytozoons, while their corresponding crown feathers underwent feather isotope analysis which will provide the migratory distances an individual traveled during wintering destinations. Parasite loads may range from 0 to 335 infections among 10 000 cells examined. Understanding the relationship among condition and migratory behaviours can help suggest alterations in population dynamics among avian species.

718

Preventing Ring-billed Gull Collisions With Vehicles on Eastport Drive

S.M. Salim, McMaster University; J.S. Quinn, McMaster University

Gulls; Ecology; Vehicle; Collision

Hamilton Harbour supports a large breeding colony of ring-billed gulls (*Larus delawarensis*) at the Pier 27 area. Each year, gulls have been flying onto a road adjacent to the colony and colliding with vehicles. We tested strategies to reduce the number of gull-vehicle collisions. We installed a wire on a section of the Jersey barrier that separates the colony from the road to test whether preventing the gulls from perching on the barrier would reduce the number of gulls flying onto the road. We also covered a section of the nesting site with plastic tarps to reduce the number of nesting gulls. Our study shows that the number of nests was lowest in the plastic section and was significantly lower than three of the five other sections. The wire reduced the number of adult gulls perching on the barrier and crossing the road. However, there was no significant effect of treatment on the number of gull chicks perching on the barrier or crossing the road. Moreover, there was no significant effect of treatment on the number of dead gulls collected from the road. As a result, we recommend that the speed limit be reduced from 80 kph to 50 kph for the section of road adjacent to the nesting site to allow gulls and vehicles greater ability to avoid collisions.

719

The Response of *Mus Musculus* to Exposure to Various forms of Death Signals

Gurjot Toor, McMaster University; C.David Rollo, McMaster University

Death Signal; Death Extract; hypothalamic-pituitary-adrenal axis; corticosterone

The exposure of various signals of death has previously been studied in insects and invertebrates. In particular, insects such as cockroaches, and fish such as sea lampreys are repelled by the presence of a death extract in their environment. The reaction of mammals and vertebrates to death has not been extensively investigated. Using *mus musculus* as an animal model, this study will investigate the reaction of

mammals to death. Mice are social animals with sophisticated olfactory systems, which justify a prediction of a reaction to death. The presence of death in the environment can also cause the release of excess corticosterone in mice, which upregulates the hypothalamic-pituitary-adrenal axis (also known as the stress axis). The stress axis further affect various biological mechanisms, such as the antioxidant system and DNA repair mechanism. The reaction of mice to whole body dead conspecifics, death extract of same and opposite sex conspecifics, and blood of conspecifics will be observed.

Saturday March 16th, Poster Session, MUSC Atrium – 5:00 – 6:30 pm

Physiology -

730

Frequency Dependent Male Aggression In Guppies *(Poecilia reticulata)*

G.H.Y. Cho, University of Toronto; A. Li, University of Toronto; F.H. Rodd; University of Toronto

negative frequency dependent selection; colour polymorphism; rare male morph

Male guppies exhibit widely variable colouration patterns. It has been proposed that this variation is maintained by negative frequency dependent selection; this is a kind of natural selection where individual fitness is correlated with its relative frequency in the population. In natural populations, the males with rare phenotypes survive better than males with common phenotypes. However, the reason behind this pattern is unclear. My project examines whether aggressive behaviours shown in male guppies are frequency dependent. In order to do this, I manipulated male colour pattern ratios in an aquarium and recorded aggression between the males. If aggression is frequency dependent and if aggression puts some males at higher risk of attracting predators, this may be a possible explanation for the frequency dependent survival found in natural populations.

731

The Influence Of Metal Chelators And Polluted Sediment On The Longevity Of The Cricket, *Acheta Domesticus*

Dhami S.S., McMaster University; Rollo C.D., McMaster University

Metal Chelators; FOXO; Stress Resistance; Hamilton Harbour; Reactive Oxygen Species

Current theories of aging suggest that the formation of reactive oxygen species (ROS) and other free radicals can cause DNA damage. Overproduction of ROS can lead to oxidative stress, which in turn activates the forkhead box O (FOXO) pathway. FOXO is responsible for regulating stress resistance genes and is turned on in models displaying an extension in longevity. Metals have a significant effect in producing ROS and activating the PI3K/Akt pathway, which inhibits the FOXO pathway. Metal chelators are chemical compounds that bind and decrease metal ions in cells, which counteracts the PI3K/Akt pathway and further extends longevity by inducing the FOXO pathway. Furthermore, Hamilton Harbour is highly contaminated with metals at toxic levels. In this study, *Acheta domesticus* (crickets) will be used to examine the effects of metal chelators and the Hamilton Harbour sediment on longevity. The four treatments used in this study include the control diet, the sediment diet, the chelator diet, and the chelator-sediment combined diet. Survivorship, food consumption, growth rate,

weight, free radical processes, and metal content in crickets will be monitored and compared between different treatment groups to determine the effects of metal chelators and the sediment from Hamilton Harbour on the organisms.

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Impact Of Metformin And A Combination of Metformin & Aspirin On The Longevity And Behaviour Of *Acheta Domesticus*

Hans H.K., McMaster University; Rollo C.D., McMaster University

AMPK; FOXO; Stress Resistance; Growth Rate; Free Radical Processes

A large body of literature illustrates the effects metformin and aspirin have on longevity and cancer. Both drugs phosphorylate the energy sensor, AMPK, which is responsible for inducing Forkhead transcription factor (FOXO) activation. FOXO translocates to the nucleus and activates stress resistance genes, thereby enhancing an organism's stress resistance and hindering growth. A decrease in growth rate can increase the longevity of animals. However, the majority of the studies are conducted on mammals or mammalian tissue. The current study examines the effects of metformin and aspirin on the invertebrate species, *Acheta domesticus* (cricket).The cricket is a good model for aging studies because it shares the same pathway regulation as mammals. Crickets also have a relatively short lifespan, which allows longevity studies to be completed within a relatively short period of time, and they can be obtained in large numbers at an economically feasible price. Longevity, growth rate, food consumption, behaviour, and free radical processes will be examined to determine any significant differences between the control, metformin, and combination of metformin and aspirin treatments.

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The Effect Of Nutritional Status On Mate Selection In *Mus Musculus*

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Nutritional Status; Mate Selection; Vomeronasal Organ, Volatile Signals

Being nocturnal, mice have developed a dependence upon their olfactory system to survive. A mouse's olfactory system aids in foraging for food, marking territory, identifying and characterizing conspecifics, selecting mates, and in social communication. Pheromonal information that is released by mice through urine and exocrine glands allows individuals to investigate specific characteristics of conspecifics. In particular, the vomeronasal organ (VNO) in the mouse olfactory system specializes in the detection of various pheromones and urinary proteins. This information plays a vital role in the choosing of potential mates by a mouse.

Information such as what type of immunological genes a male may possess, allows female mice to make an informed mate choice that will maximize the fitness of their future offspring. Essentially healthier males are associated with being more active, larger, and being able to produce more intense mating calls compared to conspecifics, and females tend to choose males with these traits as mates**.** Thus suggesting that an individual's nutritional status and overall health may directly correlate with mate choice. Very little literature has focused on the mate selection of both females and males together, as well as the impacts of nutritional supplements and overall nutritional status on mate recognition. This study will test the hypothesis of whether or not individuals of one sex prefer a mate that is healthier due to being fed a supplemented diet or a mate with normal nutrition on a regular diet**.**

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The Influence Of Acetylsalicylic Acid On Life History Features And Longevity Of The Cricket *Acheta domesticus*

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Aspirin, Aging, Longevity, AMPK, Acehta domesticus

Acetylsalicylic acid, commonly known as Aspirin is a salicylate drug. Salicylate is derived from plants and has been in use as a curative agent since ancient times. Aspirin helps prevent cellular and functional decline, specifically by inflammatory and oxidative agents, and also aids in the reduction of stroke and heart attack. Inflammatory and oxidative damage has also been demonstrated to contribute to aging. It has been shown in a recent study that following ingestion of aspirin at high doses, and appropriate plasma levels of salicylate being reached, the adenosine monophosphate-activated protein kinase (AMPK) is activated. AMPK is involved in the regulation of cell growth and metabolism, cellular energy homeostasis, biogenesis of mitochondria and regulation of SirT1 proteins. Additionally, both AMPK and SirT1 have been reported to directly influence PGC-1α activity through deactylation and phosphorylation, further aiding in mitochondria biogenesis and longevity. It was hypothesized that lifelong supplementation of aspirin in a diet would result in an increased lifespan, using the house cricket *Acheta domesticus* as a model. Prior studies have shown that longevity in these crickets is strongly linked with a slower growth and delayed maturation; hence these life history features amongst others were carefully monitored.

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Swimming Performance and Body Movements in Lake Trout (*Salvelinus namaycush*)

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[1]Authors contributed equally to this work.

Ecology; Kinematics; Lake Trout; Swimming speed

Lake trout are predaceous fish found in cold and highly oxygenated waters between 12 to 15˚C (Plumb and Blanchfield, 2009). During summer stratification they prefer the cooler pelagic zone, but when food is scarce, they visit the littoral zone to feed (Scott and Crossman, 1973). There is conceivably an energy tradeoff in travelling between zones to feed. In order to better predict the energy costs of lake trout behaviour and thus where they will forage, it is important to understand the kinematics of lake trout swimming. Research on related fish species have found that amplitude of tail movement is body size dependent and for some species, it increases with speed until it reaches a constant (Bainbridge, 1957). We are interested in fine-scale movement (body amplitude) of lake trout, which has not been previously studied. The experimental lake trout were video recorded while exercising in a swim chamber at known speeds. Photos were captured to express the complete range of motion of the caudal fin and peduncle. We used ImageJ (Schneider et al, 2012) to calculate the amplitude of this range and found a significant relationship between swimming speed and amplitude of caudal fin, peduncle, and dorsal fin movement. The average change in amplitude was highest for the caudal fin, and decreased toward the front of the body. Combined with the knowledge that swim speed is positively correlated with the frequency of tail beats, our results seem to indicate that tail movement is more important for speed regulation than movement of the middle or front of the body.

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Impact of Melatonin and Valerian on the Longevity and Behaviour Of *Acheta Domesticus*

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Mitochondria; GABA; MAPK; mTOR;; Free Radical Processes;

Various animal studies have shown melatonin and valerian, two common sleep aids, to have an effect on longevity and behaviour. Melatonin, an indolamine produced in the pineal gland is a very powerful antioxidant and acts in a plethora of ways to counteract oxidative stress. Several of these methods include: acting on signaling pathways including MAPK, ERK, and mTOR; as a direct scavenger; and through its metabolites. Valerian, a root plant, influences the neurotransmitter GABA by effectively increasing its production when normal production is low during the aging process. The impact of these compounds on various aspects of the mammalian system to increase longevity and behaviour is a fairly novel topic of study. Previous studies conducted with these compounds have been on mammalian species. The current study will be looking at the effects of melatonin and valerian on the invertebrate species, *Acheta domesticus,* a species that shares many of the regulatory pathways as mammals. *Acheta domesticus* provides a simple and inexpensive method to study the aging process in a short period of time. Three treatments: valerian, melatonin and a valerian/melatonin combination will be analyzed to examine and compare the effect each diet has on survivorship, growth rate, food consumption, and behavior of these species.

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Sodium Transport In The Gut Of Rainbow Trout

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Epithelial sodium channel; pharmacological blocker; gastrointestinal sac technique

With rainbow trout (oncorhynchus mykiss) as the model system, this study focused on the role of the gastrointestinal tract in Na^+ uptake processes. By using the in vitro gastrointestinal sac technique and a 1 hour flux period, concentration dependence of Na^+ uptake was examined to determine whether carrier-mediated transport occurs in the gastrointestinal tract. Phenamil, a pharmacological blocker was used to target ENAC-like channel. 10^{-4}M of phenamil was dissolved in 0.1% DMSO and added to mucosal saline (Na^+=150mM & Na^+=10mM) with radioisotope ^{22}Na to compare Na^+ uptake pathway under high and low concentration of Na^+.10^{-4}M of phenamil reduced the Na^+ uptake rates in blood space of all intestinal sections in 150mM Na^+ mucosal saline, this strongly suggests the presence of ENAC-like channel in the gastrointestinal tract. Future study will investigate the other Na^+ transport pathways under low concentration of Na^+ by pharmacological treatments including EIPA, Bafilomycin, Thiazide and Furosemide.

Mechanisms of brain swelling in Rainbow Trout and ammonia tolerant goldfish.

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NMDA Receptors; MK801; High External Ammonia (HEA), Reactive Oxygen Species (ROS) Glutamate Dehydrogenase (GDH), ornithine-urea cycle (O-UC), hyperammonemia

Ammonia exists in two forms; it can exist as an un-ionized form (NH_3), or as an ionized form (NH_4^+) [$NH_3 + H_2O \leftrightarrow NH_4^+ + OH^-$]. Production of ammonia involves excess proteins undergoing proteolysis into smaller peptides and amino acids. Amino acids are converted to amino acid glutamate via transaminase enzymes, which is deaminated via oxidation reactions by glutamate dehydrogenase (GDH) enzyme. The final products yielded are NH_3 and α-ketoglutarate (Mommsen and Walsh 1991; Wright 1995) all other by-products are then used in a C-skeleton for energy production. Although, ammonia is required for the building processes of amino acids and nucleic acids, excessive amounts can have toxic effects. Mammals are able to regulate ammonia build up by converting into urea, which is less toxic via ornithine-urea cycle (O-UC) (Wright 1995; Wright and Wood 2009). Most fishes excrete the un-ionized form of ammonia by passive diffusion across the gills. Ammonia productions in aquatic environments are a result of industrial effluents, sewage discharge, agricultural run-offs and degradation of organic compounds. Increase ammonia (hyperammonemia) can arise due to liver failure, pre-hepatic portal hypertension, inborn genetic errors of ornithine-urea cycle (O-UC), and alcoholism (Ip *et al.* 2001; Felipo and Butterworth 2002; Randall and Tsui 2002; Wood 2001). Increased ammonia toxicity can lead to brain edema, which causes irreversible brain damage and death in most vertebrates. Ammonia toxicity also affects the nervous system, including over-excitation of NMDA receptors, generation of reactive oxygen

species (ROS), brain swelling and decrease ATP productions (Walsh *et al.* 2007; Wilkie *et al.* 2008). However, the mechanism(s) by which HEA causing brain swelling is still poorly understood. It is unclear how ammonia over-activates the NMDA receptors researchers suspect that it depolarizes neurons promoting the release of Mg^{2+} (Fan and Szerb 1993). Long exposures to ammonia leads to hyperammonemia where swelling of the brain astrocytes occur. Thus, using *in vivo* and *in vitro* approaches, the goal of this thesis is to determine the underlying mechanism that allows the goldfish to adjust to all these challenges and compare these results to the far more sensitive trout. By exposing ammonia sensitive trout (*Oncorhynchus mykiss*) to high external ammonia (HEA) we will determine if brain swelling occurs and compare it to ammonia-tolerant goldfish (*Carassius auratus*). This study will provide further insight as to how the ammonia exposure can lead to brain swelling. The experiment performed exposed groups of n = 72 trout to different concentrations of high external ammonia (HEA) in 100L-150L re-circulating system. Ammonia sensitive rainbow trout were exposed to nominal concentration of 0.5mmolL, 0.75mmolL, and 1mmolL of ammonia (NH_4Cl) for a period of 72hrs. Recent experiments conducted allowed for recovery after exposure. Brains were carefully removed blotted and dried. Blood was extracted and used to measure ammonia concentration. Under control conditions trout's brain water content averaged 4.35mL water/g dry tissue, 0.5mmol/L exposure averaged 4.69mL water/g dry tissue, 0.75mmol/L exposure averaged 4.98mL water/g dry tissue, and 1.0mmol/L exposure averaged 6.07mL water/g dry tissue. Data presented as the mean $\pm$ SEM n=15 controls, n=10 0.5mmol/L, n=6 0.75mmol/L and n=5 1.0mmol/L for each treatment. At 1mmol brain tissue water content increased by a substantial amount of 39.5%. At 1mmol plasma ammonia content increased substantially indicating the Rainbow trout's loss of equilibrium and abilities to regulate internal ammonia levels. As plasma ammonia concentration increases brain tissue water also increases. It is clear that trout's do have the ability to regulate their internal ammonia concentration up to a threshold of 0.5mM/L then loses all ability to maintain plasma ammonia concentrations. At 0.75mmol/L high external ammonia we observe that brain tissue water increases correlating with blood plasma. Rainbow trout are capable of regulating plasma ammonia up too 700uM before blood plasma levels rapidly increase. Brain water content is directly correlated to plasma ammonia concentrations. For recovery experiment trout's were exposed to 1mmoL/L HEA and allowed to recover in ammonia-free water. Brain water content levels were restored to normal control conditions. Therefore it is evident that increase plasma ammonia does affect brain water content.

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Meta-analysis Of Combination Antifungal Therapy: *In Vitro* And *In Vivo* Studies

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Combination therapy; antifungal; mycoses; invasive aspergillosis; cryptococcal meningitis; azoles

Invasive fungal infections are a serious cause of high morbidity and high mortality among the immunocompromised population. Existing repertoire of antifungals are impressive yet optimal therapies for invasive fungal infections remain unknown. There has been a growing research and clinical interest for combination antifungal therapy as a solution. However, *in vitro* and *in vivo* evidence is often conflicting and clinical trials in this area are sparse. Also, the methods used to assess combined antifungal effects *in vitro* and in animals are poorly standardized. In this study, we review data from *in vitro*, animal models and clinical trials to expand current insights and bridge the gaps in our knowledge surrounding combination antifungal therapy. We aim to evaluate the efficacy of the most marketed antifungals in combination, comparing *in vitro* and *in vivo* studies. Ultimately, the goal is to provide readers a comprehensive and useful review in the clinical potential of combination antifungal therapy and suggest novel antifungal combinations to widen the possibilities in decreasing the global burden of human mycotic infections.

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Art In Science: An Exploration Of Relationships Between Fine Art And Biology

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Art, Bacteriart, Bio-Art, Medical Illustration, Neuroaesthetics

Bio-Art involves a new branch of art practice that incorporates fine art and the biological sciences, usually with cells, genes, and other organic material as its media. It is a new and developing practice in the art world, becoming increasingly renowned for its introduction of scientists and biologists in the studio space, and vice versa. Its efforts exist to bridge the gap between two seemingly unrelated fields of academia, promoting collaboration and companionship between the arts and the sciences.

The main purpose of this study was to explore various facets of Bio-Art and to raise awareness on the intersections between fine art and biology. As an extension of the research, an artist portfolio was created to

reflect the artistic processes and practices of various kinds of Bio-Art.

The number of categories and “art movements” within the field of Bio-Art is immense, so this study is focused on a select few that best broadcast the intersections between the art and biological worlds. They include:

- medical illustration
- gene-aesthetics
- neuroaesthetics
- bio-photography
- art in nature
    a) animal art
    b) environmental art

This research project is intended to serve as a review on current Bio-Art initiatives and to inspire transdisciplinary study amongst the fine arts and sciences. The portfolio is intended to serve as a visual aid, complementing Bio-Art research and expressing the possibility of (and potential for) practicing Bio-Art in a university setting.

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Assessment of a Scientific Literacy Intervention in Introductory Biology

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Scientific Literacy; Concept Assessment; Learning Outcomes

Scientific Literacy is defined as the ability to comprehend, apply and critically evaluate information from scientific sources and many scientific topics. However, many current science courses do not properly teach adults the skills to critically assess the quality of scientific claims. Recent redesign of the introductory biology course, BIO152, at the University of Toronto Mississauga will emphasize and promote skill development. To improve scientific literacy skills in students, the course instructors used an intervention, the Scientific Literacy (S.L.) Assignment. The purpose of the study is to assess this intervention on student learning gains. A tool was needed to assess the impact of this assignment. We thus developed and validated a Scientific Literacy Concept Assessment (SLCA), and administered it to the BIO152 students. A perspective survey was also administered to assess student opinions of the assignment and course. Students improved on mostly all of the questions in the posttest SLCA, suggesting an improvement in skill development. According to the perspective survey, students found the S.L. assignment and workshops helpful. Further analyses will examine individual learning gains and specific learning goals on the SLCA.

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Impacts Of Thermal Agents On Blood Flow, Skin Temperature And Pain Perception As Individuals Age

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Thermal agents; blood flow; skin temperature; pain perception; aging

Background

Thermal agents are commonly used to treat musculoskeletal injuries. As individuals age, physiological changes may result in differences in blood flow, skin temperature and pain perception responses to temperature stimuli compared to younger adults. Thus, physical therapies may need to be modified in duration, intensity or modality to better suit these changes.

Methods

Participants with no hand pathology were divided into two age groups: 18 to 59 years old and 60 years and over. Simulating a “home application”, hot and cold gel packs were applied in separate applications for 10 minutes to the wrist area. Superficial microcirculation of the palm was analyzed using the TiVi600 Polarization Spectroscopy Camera to quantify red blood cell concentration. Images were taken pre-application of each thermal agent as a baseline measurement and directly post-application in 10 second intervals for 10 minutes. Skin temperature was measured using an infrared thermometer, before the thermal agents as a baseline and after in 1 minute intervals for 10 minutes.

Results

Immediately following the application of cold, an average increase of 5.23% in red blood cell concentration in the palm was observed in the younger age group and an average decrease of 6.96% in the older age group. Temperature was found to decrease by 4.76% in the younger group and by 5.33% in the older group on average immediately post-cold application. Post-heat application, red blood cell concentration showed an average increase of 23.28% in the younger age group and 2.66% in the older age group. Mean temperature was found to increase by 5.29% in the younger group and by 4.16% in the older group.

Conclusions

Further research with prolonged applications of thermal agents is required to determine longer term responses. There is potential to extend this study to include patients with pathologies such as arthritis or fibromyalgia.

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Frequency And Origin Of The *HARS* Y454S Mutation Of Amish and Non-Amish Populations And Its Effect On Total Protein Synthesis

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aminoacyl-tRNA synthetase; haplotype; total protein synthesis; HARS

Aminoacyl-transfer ribonucleic acid synthetases are crucial enzymes for protein synthesis as they attach specific amino acids to their respective tRNA molecules. A single missense mutation in a gene encoding histidyl-tRNA synthetase, *HARS* Y454S, has been discovered in an Old Order Amish population. This disease, named Usher-like syndrome type 3B, is associated with visual hallucinations and deafness accompanied with acute encephalopathy. Here we determine the mutation frequency in the Old Order Amish population and assess the effect of *HARS* Y454S on total protein synthesis. Population disease frequency and restriction fragment length polymorphism analysis were performed using polymerase chain reaction and restriction enzyme digests. In the Old Order Amish population, 16 individuals out of 100 were found to posses the *HARS* Y454S mutation, 15 heterozygous and 1 homozygous, resulting in a disease allele frequency of 8.5%. None of the 100 non-Amish individuals possessed the *HARS* Y454S mutation. Short tandem repeats, from the patients possessing the mutation, were also identified using fluorescently labeled probes for linkage analysis. The Old Order Amish population is experiencing a founder effect since the mutation was derived from one ancestral allele. Total protein synthesis in patient fibroblast cells were monitored using [^{3}H]histidine to test the hypothesis that the *HARS* Y454S mutation impacts protein synthesis at the translational level.

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Saturated Fats Activate An Immune Response In Muscle

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Saturated fat; skeletal muscle; inflammation; insulin resistance

Type 2 diabetes mellitus affects more than 285 million people worldwide and constitutes a severe health epidemic and economic burden. The most common factors involved in the eventual development of type 2 diabetes are obesity, sedentary lifestyles, and high fat diets. There is also increasing evidence for the role of inflammation and the innate immune system in obesity-induced insulin resistance that leads to type 2 diabetes. Previous studies in adipose tissue of mice subjected to high fat feeding showed an increase in infiltrating macrophages in expanding adipose tissue, causing inflammation and subsequently leading to insulin resistance. However, it is important to note that the insulin responsive state of skeletal muscle is a primary determinant of whole-body insulin resistance. Yet the role of muscle inflammation and the interaction between muscle cells and immune cells remains largely unknown. Interestingly, evidence also shows inflammatory macrophages in muscle during high fat feeding in mice. Does muscle emit signals to attract immune cells? Do they secrete cytokines and chemokines in response to the increased presence of macrophages?

We hypothesize that high fat levels, specifically saturated fats, will induce low-grade inflammation in muscle. Saturated fat may activate several different signalling pathways in the cells, including the transcription factor NFkB, a master regulator of cytokine expression involved in the inflammatory response. Activation of these pathways may in turn lead to the recruitment of immune cells. We chose a cell culture system to explore this hypothesis, due to its suitability to examine cause-effect relationships.

L6 myotubes were cultured for 18 hours with the saturated fatty acid palmitate (PA) and the monounsaturated fatty acid palmitoleate (PO). Signaling pathways were analyzed by western blotting and cytokine and chemokine expression by qRT-PCR. Supernatant (SN) from treated myotubes was collected to test its ability to attract immune cells using THP-1 monocytes in transwell migration assays.

Our initial findings show that PA can both activate and impair several different pathways in muscle cells. For instance, the insulin-signaling pathway was marginally reduced in PA-treated myotubes, as shown by a slightly decreased AKT phosphorylation indicating a low level of insulin resistance. Additionally, PA induced a significant activation of the classical NFκB inflammatory pathway, revealed by a dose-dependent decrease in the NFkB repressor IκB. NFkB controls the expression of most pro-inflammatory cytokines and consistently, there was a significant increase in the gene expression of inflammatory cytokines (IL6) and chemokines (MCP-1, CCL2). Interestingly, we found that supernatant from such PA-treated myotubes was able to attract monocytes, suggesting that muscle cells treated with saturated fat release factors affecting the immune system. Moreover, inhibition of the NFkB pathway with parthenolide (30 μM) inhibited SN-PA induced migration, demonstrating the importance of this pathway in lipid-induced muscle-cell autonomous inflammation.

These findings suggest that saturated fat induces activation of inflammatory signals within skeletal muscle cells, and that factors released by these cells attract monocytes. This strengthens our understanding of how high fat diets, especially diets rich in saturated fat, may induce inflammation and contribute to muscle insulin resistance. Such findings may help to potentially identify steps to promote healthy insulin action in normal and insulin resistant individuals.

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High Fat Diet and Age Exacerbate Acute Renal Injury Following Cecal-Ligation Puncture Induced Sepsis in a Murine Model

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Sepsis; Kidney; Obesity; Aging; Organ Failure

Recent epidemiological data within humans points towards age and diet in increasing susceptibility of sepsis and exacerbating the pathology. One particular organ of interest within sepsis research, the kidney, has been associated with increased mortality following injury or Acute Kidney Injury (AKI). The objective of the study was to analyze renal changes caused by age and a high fat diet within a murine model of sepsis.

Thus, using three different groups (Normal diet, 6 week; Normal diet; 15 week; High fat diet, 6 week) of male CD57BL/6 mice we induced sepsis using a surgery known as cecal-ligation and puncture (CLP). The time course of the study lasted six hours with two fluid resuscitation regimens of Ringer's Lactate. At endpoint, kidneys were harvested from mice and analyzed using a Hematoxylin and Eosin staining to identify AKI. AKI was determined through the use of qualitative scoring analyzing tubular and glomeruli damage, vascular occlusion and epithelium detachment.

The severity of AKI was slightly higher within high fat diet and aged mice than normal age mice with normal diet. Observations showed that aged mice and mice with a high-fat diet showed similar pathologies between the damage of kidneys. In addition, increased vascular occlusion was observed within high fat diet mice when compared to normal diet. Mice appear to show increased sensitivity to bacterial infection as age increases and consumption of a high-fat diet, exacerbating the pathology of AKI and sepsis.

Evaluating Anxiety and Depression Prevalence: A Comparison Between Simple And Complex Inflammatory Bowel Disease Clinic Patients At the McMaster University Medical Centre Digestive Diseases Ambulatory Clinic

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Keywords: Inflammatory Bowel Disease; Gastroenterology; Digestive Diseases Ambulatory Clinic; Short Form-12 (SF-12); Hospital Anxiety and Depression Scale (HADS)

Background: Many patients with inflammatory bowel disease (IBD) face multiple problems, including medical, surgical, nutritional, social, and psychiatric issues related to their disease and require a holistic approach to care. As a result, it is common for IBD patients to develop psychological disorders associated with their disease, most commonly in the forms of clinical anxiety and depression. We wished to compare physical and mental health and the prevalence of anxiety and depression between the following two groups: simple and complex IBD clinic patients.

Methods: Interdisciplinary team members consisting of gastroenterologists, a surgeon, psychiatrists, a nurse practitioner and a dietitian provide prompt access holistic care for patients with IBD. All patients with IBD were assessed using a Short Form-12 (SF-12) and the Hospital Anxiety and Depression Scale (HADS). An SF-12 composite score of <50 was considered below average health status, while a HADS score of >11 was considered an anxiety or depression case.

Results: The mean age of the 319 (205 female, 114 male) simple IBD clinic patients was 42.1±16.5 years. The mean age of the 67 complex IBD clinic patients (44 female, 23 male) was 35.7±13.0 years. Analysis of the SF-12 scores indicated that the mean±SD physical health composite score (PCS) was 41±11 in complex IBD patients and 46±10 in simple IBD patients. The mental health composite score (MCS) was 43±12 in complex IBD patients and 48±10 in simple IBD patients. The PCS score was below average in 71.9% and 56.0% of the complex and simple IBD clinic patients respectively. The MCS scores were below average in 65.6% and 45.3% of the complex and simple IBD clinic patients respectively. HADS scores in complex IBD patients indicated that 29.2% were anxiety cases, while 32.3% were depression cases. HADS scores in simple IBD patients indicated that 18.5% were anxiety cases, while 15.9% were depression cases.

Conclusion: Patients with complex IBD have a higher prevalence of anxiety and depression in comparison to patients with simple IBD. As a result, patients with complex IBD need to be more actively evaluated for the presence of underlying anxiety or depression.

Gender-Based Differences in Collaborative Learning

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Collaborative learning, online asynchronous forums, gender

Pedagogues are continually striving to enrich instructional strategies in order to convey information in an exciting and memorable fashion.

Hypothesis

It was hypothesized that collaborative learning within a face-to-face environment would lead in significantly higher tutorial quiz scores compared to a lecture-paradigm of instruction. In the second part of this study, it was hypothesized that female Biology 2C03 students participating in online asynchronous forums in all-female groups would have a higher incidence of core members, a lower thread: post ratio, more positive affect and stronger social networking ties and this would be correlated to academic success when compared to mixed-gendered groups and all-male groups. Academic success will be measured by determining performance on specific questions on the Biology 2C03 midterm which were associated with the online assignment given in the asynchronous forums.

Methodology

173 Life Science 2A03 students were randomized into a lecture based-tutorial (n=97) or a collaborative learning tutorial (n=76) via a randomized cluster design, where the topic of the tutorial was vitamin D deficiency with respect to health, disease and stem cell research. In order to extend this collaborative paradigm into the online domain, 291 Biology 2C03 students were categorized into online asynchronous forums in Avenue to Learn across 18 all-male, all-female or mixed groups, with 15-16 members in each group. Students were given a tutorial assignment on aneuploidy in conjunction with discussion questions and were asked them to discuss them in the online forums. The participation was voluntary and participation was given an extra-credit mark.

Results

No statistically significant result was found between the mean lecture-based quiz scores (73 ± 8 %) and the collaborative treatment (70 ± 9 %). Females performed equally well in the collaborative (72 ± 9%) and the lecture (73 ± 9%) treatments. However, males fared slightly better in the lecture-based learning (72 ± 9%) compared to the collaborative learning (67 ± 10%).

Collaborative learning is a resource-efficient method of teaching because it requires a lower preparation commitment by instructors and TAs. Significantly more teaching resources and time were invested in the lecture-based learning, however, the academic performance was comparable in both contexts for females. Efficient teaching methods should be further investigated for males in order to match them to their ideal learning environment, perhaps one that stimulates inherent male-male competitive behaviour.

In the online collaborative asynchronous forums, all-female groups had a higher number of posts (7.0 ± 3.3), followed by the mixed-gender groups (5.2 ± 2.2) and all-male groups 2.7 ± 2.6 posts. All-female groups also had the lowest member to post ratio (0.64 ± 0.19 females/post), indicating more interactions between female members. Females had stronger social networking ties as represented by a higher number of in and out degrees of centrality. Lastly, female groups had higher word counts and more core members compared to the

mixed and all-male groups. It is being investigated how test performance (with respect to questions derived from the discussion forum assignment) varied with group category.

Significance

Using collaborative learning is a time and cost-efficient way to teach female life science students. Furthermore, female biology students took advantage of online collaborative environments more than their male counterparts (data analyses are underway to determine if this affected academic performance). Enhancing collaborative skills has the potential to improve professional skill development, with respect to leadership, collaboration and communication skills which are important for the work force.

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Investigating Cell Surface Nucleolin Expression In Major Paediatric Respiratory Illnesses

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Respiratory Syncytial Virus; Nucleolin; Cystic Fibrosis, Asthma

Background: Human Respiratory Syncytial virus (RSV) is an enveloped non-segmented negative-sense RNA virus whose major site of replication *in vivo* is the ciliated columnar airway epithelial cell. It is the most common viral agent of lower respiratory tract infection (LRTI) in infants around the world. Children who have chronic lung disease, such as asthma and cystic fibrosis (CF), develop more severe RSV infections than otherwise healthy children, but the mechanisms responsible are poorly understood. Recently, our laboratory has identified nucleolin, expressed on the apical aspect of ciliated columnar airway epithelial cells, as a functional cell surface receptor of RSV. To determine whether children with asthma or CF have different levels of cell surface expression of nucleolin as a possible mechanism of increased severity of RSV infections, we compared cell surface nucleolin protein expression in columnar airway epithelial cells obtained from healthy children and from patients with asthma or CF.

Methods: Non-bronchoscopic bronchial brushings were collected from individuals in three different groups (normal n=10, asthmatic n=4, CF n=3), later spun onto slides, and subject to immunocytochemistry for nucleolin. Our project involved digital image acquisition from ciliated columnar cells and the quantification of the surface nucleolin expression on the apical aspect of cells using Image Pro Plus Software.

Results: This Table shows results of nucleolin quantification on the apical aspect of ciliated columnar epithelial cells in the three groups studied:

Table : Nucleolin quantification

Group	Nucleolin Expression (Mean ± Standard Error)	*P* value
Normal	41039 ± 3554	NS*
Asthma	29698 ± 6600	

CF	39458 ± 2369	

*NS : $P > 0.05$ by ANOVA

Conclusions: Nucleolin protein expression on the surface of ciliated columnar airway epithelial cells is similar between healthy children and children who have asthma or CF, such that other mechanisms are likely to be important in accounting for differences in the severity of RSV infections observed clinically in children who have chronic lung disease.

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Investigating Mouse Gut Microbial Populations In Response to Prebiotic Treatment

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Prebiotic; probiotics; Citrobacter rodentium; FISH; inflammatory bowel disease

Background: Prebiotics are defined as non-digestible food ingredients that selectively stimulate the growth of beneficial bacteria in the gut. Prebiotics, unlike probiotics (live bacteria that confer a health benefit to the host) which have been shown to provide protective effects in Inflammatory Bowel Disease (IBD), are less well characterized. Previous studies have demonstrated that prebiotic treatment increases the relative levels of *Lactobacillus* and *Bifidobacteria* spp. in the colon, which in turn have beneficial effects for the host.

Aim: This study examines whether mice fed prebiotics exhibit changes to the microbiome prior-to and following pathogen-induced infectious colitis.

Methods: C57BL6/J mice were given free access to sterile water or 10% prebiotics (inulin or short-chain fructo-oligosaccharide) in sterile drinking water for 7 days prior to an orogastric infection (10^8 CFU) with the murine pathogen *Citrobacter rodentium* (an animal model of IBD). Prebiotic treatment was continued throughout the study protocol and mice were sacrificed on day 10 after infection. Colonic tissue samples were collected in Carnoy's solution, paraffin embedded and mounted onto slides. The microbiota was subsequently examined using fluorescence *in situ* hybridization (FISH), a method used to detect, localize and quantify target microbial populations within tissue. General probes such as EUB338 were tested using Cy5, Cy3, and Alexa fluorescent tags to detect bacteria within the colonic sample against tissue autofluorescence. Results were visualized using confocal laser scanning microscopy. Specific probes with a Cy3 tag will target *Lactobacillus* and *Actinobacteria.*

Results: Qualitative evaluation of confocal images revealed that Cy3-tagged probes produced the best signal to noise ratio followed by the Cy5- and Alexa-tags. The location of colon-associated microbiota within the mucosal surface of infected mice was determined.

Summary: This analysis provides insights into the composition and location of colon- associated microbiota.

A Study of Yeast Flora including *Candida tropicalis* Associated with Tree Hollows in Hamilton, Ontario, Canada

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yeast; environmental isolates; Candida tropicalis

Species of *Cryptococcus* and *Candida* are the most devastating human yeast pathogens which have caused a high mortality rate. For instance, *Cryptococcus neoformans* inhabits worldwide and brings out various diseases in those who are immunocompromised, especially AIDS patients. The study of North Europe provides an idea that sampling environmental yeast by using selective media may be able to isolate strains of *C. neoformans.* The other disease causing genus *Candida* has predominated in mycotic infection, contributing significantly to nosocomial diseases. An environmental yeast survey done in 2008 around Hamilton reported that multiple isolates of *Candida spp.* were identified. However, the potential impact of environmental *Candida* on humans remained to be determined. In this study, 35 of 205 yeast positive samples swabbed from tree hollows were isolated using caffeic acid ferric citrate test agar (CAFC medium) and identified to contain 15 yeast species. There was no positive strain of *Cryptococcus* isolated at the sampling site, indicating *C. neoformans* and related *Cryptococcus* spp. associated with tree hollows might not exist in Hamilton, Ontario. There have been temporal species shifting of environmental yeast flora occurred based on survey results. Furthermore, a high percentage of *Candida* spp. was identified, particularly *C. tropicalis*. The DNA fingerprinting analysis predicts that *C. tropicalis* strains associated with tree hollows do not show potential pathogenic impacts on human health.

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The Role Of *SCO3875* And sIHF-TopA Interaction In The Development And Secondary Metabolism Of *Streptomyces coelicolor*

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Streptomyces*; sIHF; actinobacteria; synteny*

A tremendous number of medically useful compounds, particularly antibiotics, are secondary metabolites of *Streptomyces* bacteria and their close relatives. Many of these soil saprophytes also possess “cryptic” gene clusters, which appear to direct the biosynthesis of metabolites that have not yet been observed in a laboratory environment (Bentley *et al*., 2002). More distant relatives of *Streptomyces* include some important human pathogens, such as fellow actinobacteria *Mycobacterium tuberculosis* and *Corynebacterium diphtheriae (*Hopwood, 2007). Thus, there may be considerable medical utility in elucidating the regulatory systems that govern gene expression in *Streptomyces.* One potential nucleoid-associated protein and transcriptional regulator, present in the model actinomycete *Streptomyces coelicolor*, is sIHF (SCO1480). sIHF binds DNA in a sequence-independent, length-dependent manner, and may interact with *S. coelicolor’*s topoisomerase gene (SCO3543 or *topA)* Deletion of *sIHF* causes aberrations in antibiotic production, as well as abnormal spore morphology associated with defects in chromosome segregation (Swiercz *et al*., 2012). The region surrounding *sIHF* and its orthologs shows striking synteny in Gram-positive bacteria; the positions of many neighbouring genes are conserved in distant relative *Bacillus subtilis (*Kunst *et al*., 1997). *B. subtilis* does not, however, have an ortholog of *sIHF*; the gene in the equivalent position is *remA,* without which *B. subtilis* cannot form biofilms normally. A similar phenotype is seen with the deletion of *remB*, which also lacks an ortholog in *S. coelicolor* (Winkelman, Blair, & Kearns, 2009). However, *remB*’s neighbouring genes show highly conserved synteny, and the *SCO3875* gene of *S. coelicolor* is in the equivalent location. This project is intended to explore the roles of *SCO1480* and *SCO3875* in the metabolism and chromosome organization of *S. coelicolor.* Its major goals are to produce a knockout of *SCO3875* (comparing its phenotype to that of a SCO1480 knockout) and to use a bacterial two-hybrid assay to determine whether sIHF interacts directly with topA.

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Gut Fate Specification of Human Embryonic Stem Cells

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Human embryonic stem cells; Definitive endoderm; Gut; HNF4α; CDX2

During embryogenesis, the development of the gut first requires differentiation of cells in the early embryo to a germ layer called the definitive endoderm. The definitive endoderm will undergo further differentiation to form the foregut, midgut, and hindgut, each of which will undergo further specification. Following ectopic expression in pluripotent human embryonic stem cells (hESC), the transcription factor SOX17 was shown to be sufficient to produce stable definitive endoderm progenitors. As transcriptional regulators of embryonic endoderm differentiation in mouse, HNF4α and CDX2 have been shown to play important roles in early gut development. Manipulating gene expression by generating transgenic human embryonic stem cells will enable us to test the ability of these transcription factors to specify gut fate. We hypothesize that controlled expression of either CDX2 or HNF4α in SOX17-definitive endoderm progenitor cells will result in gut-specific differentiation.

To assess the ability of ectopic gene expression to specify gut fate, we will use transgenic tools to deliver sequential activation of either SOX17 and CDX2 or SOX17 and HNF4α in human embryonic stem cells for directed cell differentiation. Following transgene-mediated cell differentiation, gene expression of markers for pluripotency, definitive endoderm, primitive gut tube, foregut, and hindgut will be analyzed by real-time polymerase chain reaction.

Overexpression of CDX2 in SOX17-definitive endoderm progenitors promotes acquisition of a hindgut phenotype, whereas overexpression of HNF4α in SOX17-definitive endoderm progenitors resulted in induction of primitive gut tube, foregut, and hindgut markers, suggesting a mixed phenotype.

Our ability to regulate the differentiation of pluripotent human embryonic stem cells using transgenic technologies for ectopic expression of lineage determining transcription factors may allow development of protocols for directed stem cell differentiation.

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Analysis of the Structural Alterations in the Genomes of the C57BL/6J Inbred Mouse Line and 19 Wild Caught Mice.

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Copy number variant(CNV), isogenic

New genomic technologies have revealed that there is much more plasticity in genomic structure than previously seen. Structural variations like duplications, deletions, inversions, and translocations, occur throughout the genome and contribute to alterations in gene dosage and expression. Together, duplications

and deletions of regions greater than 500Kb are labeled copy number variants (CNVs).

CNVs are well studied in humans and while this is ideal, it has limitations. For example, obtaining tissue samples, especially brain, may be difficult. Also, it is not possible to control for environmental factors with humans and selective breeding or genetic modification cannot be done. In these cases model organisms, mice in particular, are utilized**.** In order to be useful as a model for studying CNVs it is necessary to understand copy number variation in the context of the mouse. Here, the nature of CNVs in 8 C57BL/6J inbred mice and 19 wild caught mice from diverse populations was investigated. The inbred mice have undergone artificial selection through extensive inbreeding, while the wild caught mice are subjected to natural selection in the wild. It is therefore expected that the CNV profiles will differ with respect to location, merged events (at least 40% overlap), unique events (less than 40% overlap) and genes affected. To identify CNVs, genotyping arrays are utilized. The CNVs for this analysis were previously identified using the Mouse Diversity Genotyping Array (MDGA) which contains 916,269 probes for calling CNVs. Duplications of one or two extra copies are called as 3 or 4, respectively and deletions of one or both copies are called as 1 or 0. A call of 2 indicates no CNV.

In the C57BL/6J mice, there was much more variation than expected for an isogenic line. There was an average of 29 events per mouse, of which the majority were involved in merges with events in other samples, and fewer unique events were observed. These shared CNV events are expected since the mice are inbred. In the wild caught mice there was an average of 68 events per sample. The majority of events were singletons, which is expected for wild caught mice, however there was still merging of events throughout the samples indicating genomic hotspots or common origin. The wild caught CNVs are also enriched for genes involved in sensory perception, olfactory and pheromone receptor activity, and the MHC protein complex, while the B6 mice CNVs are enriched for MHC protein complex, but not sensory perception or olfactory and pheromone receptor activity.

Inbred mice like the C57BL/6J are useful in scientific research due to their isogeneity, so it is important to identify structural variations present within inbred genomes. It may be useful to take these into account when breeding or selecting mice for research. Inbred mice, however, are unable to capture the genetic diversity seen in humans. For this, wild caught mice which have been subjected to generations of natural selection, may present a more accurate model for studying structural variants and their contribution to complex phenotype.

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Regulation Of the Response to Hypoxia By C/EBPβ In Chicken Embryo Fibroblasts

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C/EBPβ; ERK; p20K; CHOP; Hypoxia

During cellular quiescence, a reversible non-proliferating state, growth arrest-specific (Gas) genes are up-regulated. Fielding (M.Sc., 2011) previously demonstrated that some Gas genes, including the p20K lipocalin, are induced by hypoxia. Transcriptional activation of p20K depends on the CCAAT enhancer binding protein beta (C/EBPβ) but is also controlled by multiple inhibitors. The C/EBP Homologous Protein (CHOP) represses transcription of p20K by binding and inhibiting C/EBPβ in cycling chicken embryo fibroblasts (CEF). During hypoxia, when CHOP levels are low, C/EBPβ is able to form homodimers and induce p20K transcriptionally through the quiescence-responsive unit (QRU) of the p20K promoter (Fielding, M.Sc., 2011). CHOP is an ER stress (endoplasmic reticulum stress) inducible factor but is also expressed at low levels in proliferating cells,

where it contributes to the repression of p20K. In addition, Extracellular signal-Regulated Kinase 2 (ERK2) is able to transcriptionally repress p20K by binding to the p20K QRU during cell proliferation (Athar, M.Sc., 2011). This project investigates the regulation of these two known inhibitors of p20K, CHOP and activated ERK, in response to ER stress or decreasing concentrations of oxygen. Western blotting analysis indicated that p20K expression decreases as CHOP levels increase during ER stress. A co-immunoprecipitation revealed that C/EBPβ association with CHOP is also favored during ER stress, supporting the model that CHOP binds to C/EBPβ to inhibit p20K expression. Cell proliferation assays showed a significant decrease in cell proliferation during hypoxia. Finally, Western blotting analysis using proliferating CEF determined that levels of phosphorylated ERK are also markedly reduced during hypoxia, a condition where p20K is strongly activated (Fielding, M.Sc., 2011). ERK1/2 is an important regulator of cell proliferation and thus may have a role in the decrease in proliferation seen in hypoxia. It is concluded that p20K is regulated predominantly by the expression of two inhibitors, CHOP and activated ERK. These inhibitors are regulated by different factors in the cell (ER stress or hypoxia) and may affect cell fate by regulating proliferation.

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Visualizing Neural Crest Cell Migration Exploiting Melanophores in *Xenopus laevis*

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melanophores; melanocytes; neural crest cells; melaninin

Although *Xenopus laevis* and its sister taxa are commonly used models in the field of cell biology, relatively little is known of their neural crest migration. The neural crest is a transient component in the early stage of vertebrate embryos, which migrates to differentiate into one of three cell types: peripheral nervous system, facial skeleton, and pigment cells. Because neural crest formation has implications for dysfunctional cell population, such as melanoma, understanding the molecular mechanisms involved is important for our knowledge of human diseases.

In this study, we will use *X. laevis* embryos to visualize neural crest cell migration via melanophores in the living embryos up until the mid-tadpole stage. Previous investigations suggest that melanophores take dorsolateral pathway to reach their destinations. To test this, we will track the migration of melanocytes on DL-DOPA treatment in their early stages of development. DL-3,4-Dihydroxyphenylalanine (DL-DOPA) will be administered to the melanophores at different time points to initiate a series of chemical reactions in a tyrosine pathway, which lead to the production of melanin. On production of melanin, melanophores become visible. Thus, DL-DOPA forces melanophores to prematurely differentiate into melanocytes, and, with this aforementioned rationale, the routes that the neural crest cells take can be observed in timely fashion as the vertebrate develops.

To reiterate, understanding neural crest migration will benefit researches in providing solutions to abnormalities related to improper differentiation of the neural crest cells. Improper differentiation can be defined as premature differentiation in an unsuitable environment, traveling via inapplicable route, or failure to differentiate at all. With aid of this study, the pathways of the neural crest cells (in fate of becoming melanophores) can be tracked down, and examined from what point in their migratory pathway.

Functional Characterization Of The HopF Effector Family Using Arabidopsis Plants

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The HopF family, type III secretion system, effector function, resistance response in Arabidopsis plants

The coevolutionary arms race between pathogens and their plant hosts have prompted diverse pathogen virulence strategies and have modified plant surveillance and defense systems. The plant host must defend itself against pathogenic virulence in order to survive, while the pathogen has to evade or suppress host immune response in order to proliferate. *Pseudomonas syringae* is a Gram-negative, rod-shaped bacterial pathogen, used as a model for the study of specific host-pathogen interactions. Genome-wide protein analysis has revealed near 50 confirmed or predicted type III effectors in plant pathogen *P.syringae* pathovar *tomato* (Pst) DC3000 [Boch *et al*, 2002]. Here in this paper, we present the functional analysis of one family of type III effectors – the HopF family, in order to gain insight into their pathogenic mechanism. We examined if different effectors in the HopF family have a function in resistance or disease in *Arabidopsis* plants. Previous studies have worked on HopF2 PtoDC3000 showed a virulence function in tomato. In this study, we used the same effector HopF2, but from different pathogen strain PtoT1.We demonstrated that HopF2 from strain *PtoT1* induces a resistance response in *Arabidopsis* plants and is not dependent on its catalytic residues R72, D174, and the positive selection site S123 (correspond to T123 in $HopF1_{PtoT1}$). Furthermore, we also show that $HopBB1_{PavBpIC631}$ does not appear to have a function in *Arabidopsis* plants.

Subcellular Localization Of Isoflavone Synthase 1 And 2 In Soybean

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Isoflavonoids; Isoflavone synthase; Phytoestrogens; Subcellular localization

Isoflavonoids are plant natural products found almost exclusively in legumes. Due to their health benefits, it is of interest to understand the biosynthesis of isoflavonoids, enabling the manipulation of isoflavonoid levels in food products. Isoflavone synthase (IFS) channels flavonoid pathway intermediates to isoflavonoid biosynthesis pathway and is legume specific. IFS localizes to the endoplasmic reticulum (ER) membrane, forming a metabolon with other downstream enzymes. Recent studies have found other pathway enzymes, chalcone isomerase (CHI) and chalcone synthase (CHS), localized to the nucleus as well as the ER in non-legumes. It is therefore hypothesized that IFS1 and IFS2 will localize to the ER membrane and nucleus in tobacco and soybean. *IFS1* and *IFS2* were cloned and sequenced and *Agrobacterium*-mediated transformation of *N. benthamiana* leaves and soybean hairy roots resulted in expression of IFS-YFP proteins,

which were visualized through confocal microscopy. Preliminary results for IFS1 show localization to the nucleus when IFS1-YFP is stably expressed in the hairy roots of *G.max.* IFS2 has been shown to localize to the ER membrane in the leaves of *N.benthamiana*. The presence of IFS at the ER membrane further supports the presence of a metabolon structure with other key enzymes in the pathway for substrate channeling in the process of isoflavonoid biosynthesis. Nuclear localization of IFS could indicate a role for the enzyme outside of isoflavonoid biosynthesis, or potential for biosynthesis in the nucleus itself.

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The Bacterial Expression of Biologically Active ProNGF

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Nerve growth factor, ProNGF, TrkA, MAPK

Nerve growth factor (NGF) is a neurotrophin that promotes neuronal survival and maintenance. The study of proNGF, the predominant form of NGF in the brain, is critical for understanding neurodegenerative diseases. For this research to be possible, large quantities of proNGF must be easily and affordably obtainable. However, when expressed in bacteria, proNGF forms non-functional aggregates that deposit into inclusion bodies. Our aim is to express **r**ecombinant proNGF in a bacterial system in a soluble and biologically active form. Four proNGF cDNA constructs were generated using Gateway Cloning Technology and were expressed in *E. coli* Origami B pLysS cells. Each construct produced soluble proNGF. ProNGF with a C-terminal histidine (His_6) tag was purified using nickel affinity chromatography. The purified proNGF was tested for activation of its receptor (TrkA) and the MAPK signal transduction cascade using PC12 cells. TrkA and ERK phosphorylation were observed following addition of purified proNGF-His_6 to PC12 cells. These preliminary data suggest that purified proNGF-His_6 is biologically active; a cell survival assay and a neurite outgrowth assay will be done to further confirm that the recombinant proNGF is biologically active**.**

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Elucidating the Role of the Transcription Factor Kaiso in the $APC^{Min/+}$ Model of Intestinal Cancer

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$APC^{Min/+}$; Kaiso; Colorectal Cancer

Several studies have implicated the POZ-ZF, methyl-binding protein Kaiso, in tumourigenesis. However, a definitive role for Kaiso in tumourigenesis has yet to be determined. Although Kaiso's localization and expression (or misexpression) has been correlated to tumour grade and patient prognosis, the exact

mechanism with which Kaiso affects tumourigenesis has yet to be determined. The canonical Wnt signalling pathway is dysregulated in many human tumours, and it has been suggested that it is through this pathway that Kaiso regulates tumourigenesis. The difficulty faced in determining Kaiso's mechanistic role is due to conflicting data from *Xenopus* and mammalian models. In *Xenopus*, Kaiso was found to be a negative regulator of the Wnt pathway, whereas in the $APC^{Min/+}$ mouse model of colon cancer, Kaiso was implicated as a positive regulator. To further study Kaiso's role in the mouse model system, an intestinal-specific Kaiso transgenic mouse was created using the *villin* promoter. $Kaiso^{Tg}$ mice were subsequently crossed with $APC^{Min/+}$ mice to generate $APC^{Min/+}Kaiso^{Tg}$ mice. We observed that mice harbouring the $APC^{Min/+}Kaiso^{Tg}$ genotype exhibited smaller, but larger amounts of polyps in comparison to the $APC^{Min/+}$ mouse. Future studies will assess the expression levels of the proliferation marker Ki67 and the apoptosis protein caspase 3. The results of this study will help to elucidate further the role in which Kaiso plays in intestinal tumorigenesis and possible targets for therapeutic intervention.

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Crystal structure of heme-trafficking protein PhuS in *Pseudomonas aeruginosa* and its functional implications

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Cytoplasmic heme-binding proteins; iron acquisition; heme degradation; heme trafficking; heme oxygenase

The scarcity of biologically available iron leads to competition between the human host cell and the infectious bacteria such as *Pseudomonas aeruginosa*. This pathogen has evolved several ways to steal iron from the host to maintain proper growth during infection, largely through the uptake and breakdown of heme. The cytoplasmic heme-trafficking protein PhuS shuttles heme to PigA, a heme oxygenase, and is homologous to several other bacterial heme-binding proteins including the heme oxygenase ChuS in *Escherichia coli* O157:H7. The structural elucidation by X-ray crystallography of PhuS with and without heme bound reveals possible explanations for the difference in function of PhuS and ChuS, including the latter's function as a heme oxygenase.

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Reduced Levels Of Integrin and Integrin Related Proteins Leads To Changes In The *Drosophila Melanogaster* Heart

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Integrin; Talin; temperature sensitive Gal80; Actin filament arrangement; Embryonic and Larval Viability

Cardiovascular diseases are the leading cause of adult mortality in both Western and South Asian populations. Infants are also affected by cardiovascular problems, with structural and functional cardiovascular defects being a leading cause in childhood morbidity. As the occurrence of congenital heart disease and cardiomyopathies are on the rise, it is important to understand the underlying genetic basis, in order to improve diagnosis and therapeutic strategies, as well as reduce the financial burden on the healthcare system. Integrin is a heterodimeric adhesion receptor made up of an alpha and beta subunit that is involved in signal transduction, cell migration, cell adhesion and the proper formation of the heart. Within mammals, cardiovascular diseases, such as cardiac hypertrophy and dilated cardiomyopathy, are known to be affected by Integrin signaling and the level of Integrin within the ventricle.

In contrast to the complex mammalian heart, *Drosophila melanogaster* have a simple tubular heart that can be used to study cardiac disease and heart development, due to the conservation of several cardiac genes and Integrin structure and function. The conserved genes are able to be genetically manipulated using multiple methods, such as loss of function mutations and the GAL4/UAS system. To examine how the reduction of Integrin and Talin protein levels affect heart morphology and viability of *Drosophila melanogaster progeny,* transgenic flies containing, either a temperature sensitive allele or normal allele of Gal80 under the control of the cardiac-specific driver *Hand*-GAL4, were crossed with either *mys*(beta-integrin)- or Talin-RNAi lines. Progeny of these crosses were then collected and assessed for viability. Third instar larvae were also dissected and stained, to visualize the heart.

Despite the changes in Talin protein levels, the remaining Integrin was localized at the constamere junction between the actin filaments in adjacent cardiomyocytes. Progeny of crosses between Talin-RNAi and *Hand*-Gal80^{ts} were able to complete development up until pupation without a decrease in viability, pointing to an increased effect of the reduction of Talin levels later in the development of the heart. Imaging of stained third instar larvae from Talin-RNAi and *Hand*-Gal80^{ts} crosses revealed minor perturbations in heart structure when Talin levels are reduced. There is a more severe affect on adult heart morphology characterized by a retraction of the contractile apparatus, which is exacerbated when *Hand*-GAL4 is used as the driver. Reduction of Talin levels in the heart also reduced adult viability, suggesting that metamorphosis and adult function is more sensitive to levels of Integrin signaling.

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Profiling the Biofilm Formation, Composition and Antibiotic Resistance Capabilities of Escherichia coli and Salmonella spp. Environmental Isolates

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Bacterial biofilms; Antibiotic resistance profiling; Exopolysaccharides; Cellulose; Curli fimbriae

Greater than 65% of bacterial infections are due to bacteria that prefer to grow in biofilms. These biofilm-embedded bacteria are a prominent health-related problem because the biofilm matrix provides increased non-specific protection from a variety of antimicrobial agents. In addition to complicating antibiotic treatment in a medical setting, biofilm matrices also increase the persistence of bacteria in the environment. In some cases, like with Escherichia coli and Salmonella spp., the same biofilm is produced by bacteria in both chronic infections, as well as the natural environment. It is our hypothesis that these biofilms are direct contributors to the amount and survival of these pathogens in the environment. The specific goals of this study were to

assess the biofilm forming capacity and biofilm components of various environmental isolates of E. coli and Salmonella species and attempt to correlate these results with antibiotic susceptibility tests. The biofilm forming capabilities of the isolates were screened using tube and microtiter plate assays, after which the presence of curli fimbrae and cellulose (two prominent biofilm components produced by E. coli and Salmonella spp.) was detected by observing the colony morphology of the isolates with specific dyes for each biofilm component. Antibiotic resistance patterns of the strains that were strong biofilm formers were determined through the disk-diffusion method using four antibiotics (streptomycin, oxytetracycline, cefotaxime, and amoxicillin). Overall, the presence of curli fimbriae and cellulose was found to be directly related to the biofilm formation of many of these environmental isolates at multiple temperatures. This suggests that the isolates produce these polymers to survive situations that mimic both the host and the natural environment. However, a direct correlation between biofilm formation and increased antibiotic resistance could not be definitively established with our initial, limited antibiotic susceptibility trials. Further experiments are currently being performed to expand our biofilm resistance profiling to other antibiotics so that we can begin to understand the non-specific nature of resistance that the biofilm layer provides and how this relates to the pathogenic potential of environmental reservoirs of bacteria.

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Mycorrhizal Phenotype of *Pisum sativum* Mutant, E2

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Arbuscular mycorrhiza; Root symbiosis; Pea mutant; sym 5

Although detrimental to our environment, agriculture relies heavily upon the use of fertilizers in order to supply the soil with limiting resources. However, most land plants are capable of forming a mutualistic symbiosis with arbuscular mycorrhizal (AM) fungi. This symbiosis supplies phosphorus to the plant in exchange for sugars without the need for destructive fertilizers. Given its short germination and growth times, *Pisum sativum*, also known as pea, is a model organism for studying AM. E2, mutated in the *sym* 5 gene, was used as a tool in order to help to investigate the differences in mycorrhizal symbiosis among lines of pea. The mutant, E2, and its wild-type Sparkle were grown in soil containing the fungus *Glomus irregulare*, after which individual plants were harvested at different time periods. After having been cleared and stained, the roots were observed under bright-field optics and notes were taken regarding the presence, size, and morphology of infection events (extraradical hyphae, intraradical hyphae, hyphopodia, abuscules, and vesicles). It was found that E2 formed less intraradical hyphae, hyphopodia, arbuscules, and vesicles at 21 and 28 days after planting, as compared to Sparkle. In contrast, after 35 days of growth, E2 formed many more arbuscules than did Sparkle. These findings suggest that E2 has a delayed, myc^{++} phenotype. The results from the E2 mycorrhizal characterization can be used as a tool in future research in order to further characterize the AM fungi symbioses with pea.

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