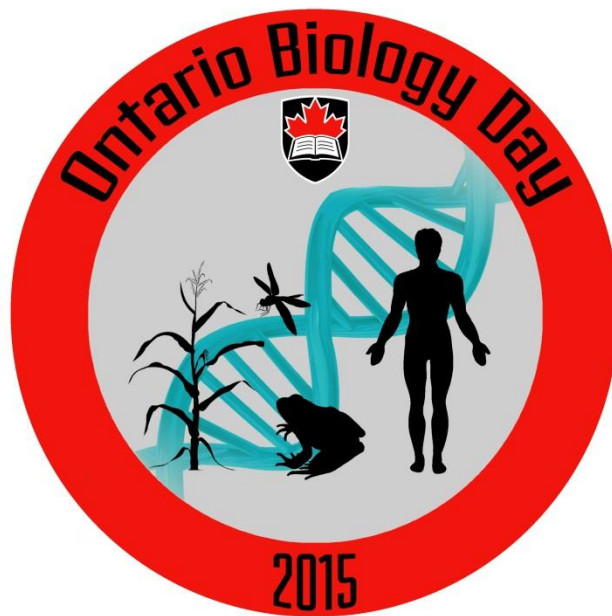


Ontario Biology Day

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Abstracts



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Abstract: 001

EFFECTS OF PRENATAL BYSTANDER STRESS ON OFFSPRING BEHAVIOUR AND ADOLESCENT SENSITIZATION TO NICOTINE IN RATS

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This study investigated the effects of prenatal bystander stress (BS) on offspring anxiety, working memory, locomotor behaviour and adolescent sensitization to nicotine. Sires were housed with their respective dams for the duration of gestation and subjected to elevated platform stress for thirty minutes bi-daily between G12 and G16. Analysis of pre-weaning BS offspring behaviour utilizing negative geotaxis, open field locomotion, and ultrasonic vocalizations revealed initial impairments in proprioceptive function and repetitive patterns of exploratory behaviour with no significant changes in vocalization frequency. However, BS offspring became indistinguishable from controls following subsequent testing periods of each paradigm. Juvenile behavioural analysis of BS offspring, consisting of the elevated plus maze, activity box, and novel object recognition tasks, showed reduced anxiety-like behaviour, male hyper-activity, and female hypo-activity with no significant alteration in temporal object memory. Adolescent nicotine sensitization was evaluated using subcutaneous injections of nicotine (0.3 mg/kg) once daily for fourteen days and monitoring of subsequent locomotor activity. Male BS offspring exhibited potentiated sensitization to nicotine while female BS offspring displayed an attenuated sensitization to nicotine over the fourteen day testing period. Overall, it appears that *in utero* emotional stress experienced by the mother has measurable and sex-dependent effects on offspring behaviour and response to nicotine.

Abstract: 002

EXPRESSION PATTERNS OF THE ACTIN BINDING PROTEINS, WDR1, CAP AND COFILIN DURING MONOCYTE DIFFERENTIATION IN THP-1 CELLS.

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Cellular differentiation is a main process involved in central areas of biology such as development, stem cells and cancer. A segment of the cell differentiation pathway involves reformation of the cytoskeleton via actin turnover which directs cell migration and movement and alters cellular adhesion to cellular substrates. The organization of actin is dependent on different classes of actin-binding proteins which act to drive actin turnover and rearrangement in response to cellular signals. Recently, it has been determined that an actin binding protein mammalian WD-repeat-1(WDR1) protein assists cofilin actin severing activity but little is known about how it does so or how it is regulated. One of the most studied differentiation processes is the change from monocyte to a macrophage. The differentiation of THP-1 monocytes to macrophages will be studied. These cells undergo differentiation to macrophages in response to PMA (a phorbol ester) addition. Levels of mRNA and protein for WDR1, its truncated isoform WDR1-Δ35, CAP1 and CAP2 (actin monomer binding proteins) and cofilin will be measured in THP cells in suspension and during various time points throughout their differentiation. Quantitative Real Time PCR will be used to quantify mRNA levels while Western blots will be used to measure protein levels in response to PMA induced differentiation. If the levels are altered during the differentiation process, THP-1 cells may serve as an important scientific model for cytoskeletal regulation involving WDR1, cofilin and CAPs.

Abstract: 003

DEVELOPING A STANDARDIZED METHOD FOR QUENCHING THE METABOLISM OF *ACIDITHIOBACILLUS FERROOXIDANS*

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The phenomenon known as acid mine drainage (AMD) is an environmentally and economically destructive process that is driven by its unique microbial community. One organism that is currently of interest is *Acidithiobacillus ferrooxidans* because it is suspected to supply the microenvironment with ferric iron that drives a larger scale reaction which in turn produces sulfuric acid contributing to environmental damage. To better understand how these processes work, meaningful metabolomics profiling can be used to provide a snapshot of this bacteria's physiology and fill in gaps of knowledge previously unidentified in literature. Obtaining this snapshot is proven difficult in this gram-negative species due to its thin cell wall that can be easily disrupted during quenching and extraction processes. To diminish this disruption, three quenching techniques are used to determine which technique provides the least amount of metabolite leakage, as determined by using liquid chromatography mass spectrometry. Developing a standardized method to quench the metabolism of *At. Ferrooxidans* will fast track the identification of metabolites within this species and begin the process of implementing possible remediation or controlling techniques.

Abstract: 004

MICROBIAL ROAD KILL: HOW ROADS IMPACT BACTERIAL ACTIVITY AND DIVERSITY

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Soil bacteria have an immense influence on ecosystem processes. They are responsible for cycling nitrogen and carbon, acquiring nutrients for plants and soil weathering and formation. Roads are linked to the emission of many pollutants originating from car tires, brake linings and fuel combustion, and also from the chemical composition of the road infrastructure itself. Storm runoff and dry deposition (a.k.a. dust) can cause the contamination of soils and impact ecosystem health. Heavy metal contamination has been shown to reduce rates of litter decomposition and soil respiration, bacterial nitrogen mineralization and fixation, and bacterial enzyme activity. The purpose of this study was to examine how road pollutants influence bacterial activity and diversity around roads of varying traffic volume, and determine their road-effect zone. We measured a variety of soil properties such as pH, bulk density, moisture content, soil temperature, metal and nutrient concentrations and organic matter content. We also measured the bacterial biomass and respiration rates and characterized the bacterial community composition using high throughput amplicon sequencing of biomarker genes. We predicted a gradient within the road effect zone in which microbial diversity, biomass, and activity decreased as sample plots approached the road edge. Regardless of road size, we found that the soils closest to the roads were very alkaline (pH~9-10) and had less moisture, higher bulk densities and very low nutrients and organic matter (i.e. both physical and chemical properties were impacted). The impact of roads on bacterial communities likely represents an important linkage between roads and broader terrestrial ecosystem functioning and sustainability.

Abstract: 005

TRACKING THE IMPACT OF GIANT MINE ON A SMALL LAKE NEAR YELLOWKNIFE, NT, CANADA USING LAKE SEDIMENTS

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Among the most detrimental anthropogenic influences on natural ecosystems are the impacts associated with resource extraction, which can be both direct and indirect. In Canada's Northwest Territories, resource extraction and activities associated with this industry have a long history, and represent one of the key contributors to overall economic development. The Giant Gold Mine, located in the capital city of Yellowknife, is one of the best known examples of both historical northern development, and unfortunately, the detrimental impacts of resource extraction. Operational between 1948 and 2004, Giant Mine produced over 200,000 kg of gold. However, one of the environmental impacts of gold extraction from arsenopyrite ore through roasting was the release of large quantities of toxic, water-soluble arsenic trioxide dust. It is estimated that remediation of the Giant Mine site will top \$1 billion, most going to managing the 233,000 tonnes of As₂O₃. My research is assessing the impact of development at Giant Mine on nearby aquatic ecosystems by examining a lake sediment core from Pocket Lake, a small site ~1 km from the NW tailings pond. I am assessing changes in both Chironomidae (the non-biting) and Chaoboridae (phantom) midge community composition over the recent past. These organisms are important indicators of changes in ecosystem health, and by assessing fluctuations in chironomid abundance and chaoborid assemblage composition I will infer whether contamination of this lake by activities at Giant Mine resulted in major changes to aquatic ecosystems, as well as whether any recovery has occurred to date.

Abstract: 006

PREVALENCE OF LATERALIZATION AND FLUCTUATING ASYMMETRY IN TELEOSTS.

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There are two popular types of asymmetry that are often investigated within scientific research, lateralization and fluctuating asymmetry, both of which allow testing of evolutionary and environmental drivers on development. We examined the occurrence of lateralization and fluctuating asymmetry in the eyes, otoliths and brains of spottail shiners (*Notropis hudsonius*), emerald shiners (*Notropis atherinoides*), round gobies (*Neogobius melanostomus*) and Chinook salmon (*Oncorhynchus tshawytscha*) to assess how common each metric is and to test possible roles of habitat on asymmetry. Previous research has shown significant left-hemisphere bias in neural processing, supporting our hypothesis for a larger left side bias. Each fish was weighed, measured, and dissected so that two saccular otoliths and the brain were removed. The area and perimeter of the left and right eyes, otoliths, and brain regions: telencephalon, optic tectum and cerebellum, were measured and compared using an ANOVA to test for the presence of lateralization or fluctuating asymmetry. Preliminary results show significant differences in area and perimeter measurements between left and right hemispheres within the cerebellum of the fish and a significant asymmetry in telencephalon perimeter in the telencephalon as well as species-level differences in body to brain mass ratio. These results suggest asymmetrical morphology differences within the brains of the fish, notably within the cerebellum as well as differential energy allocations in brain:body between species.

Abstract: 007

BEHAVIOURAL GUIDANCE OF LARGEMOUTH BASS USING LIGHT EMITTING DIODES

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² ATET-TECH Inc.

Freshwater ecosystems are threatened by the infrastructure related to hydropower development, irrigation and municipal water-taking as well as industrial cooling. This is especially apparent when taking into consideration the risk of entrainment or impingement that such infrastructure presents to fish. To minimize these threats, mitigative strategies have been developed which aim to direct fish away from “dangerous” areas to “safer” areas. Behavioural guidance systems provide a cost effective method by exploiting the sensory physiology of a species where stimuli can be used to manipulate their natural behaviour. These systems often incorporate light, as sight is one of the primary sources of information used by fish for interacting with their external environment. Previous studies that have taken this stimulus into account for behavioural fish guidance have primarily focused on flash frequencies with white light alone and are thus limited in regards to the light spectra being explored. The primary objective of this study was therefore to investigate the behavioural responses of largemouth bass in terms of orientation and movement to varying light spectra (red, orange, yellow & green) as well as flash frequencies (120 min⁻¹, 300 min⁻¹, 600 min⁻¹) as a potential form of behavioural fish guidance.

Abstract: 008

VISUALIZING THE TUBERIN-CYCLIN B1 INTERACTION USING LIVE CELL MICROSCOPY.

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The cell cycle is a complex process by which cells grow and proliferate. Progression through the cell cycle is controlled by checkpoints. The inappropriate regulation of these checkpoints results in cell growth and proliferation diseases. Recently, a novel cell cycle checkpoint has been discovered. This checkpoint occurs at the G2/M transition and is regulated by the Tuberin-Cyclin B1 complex. Tuberin is a tumour suppressor encoded by the TSC2 gene. Tuberin also forms a heterodimer with Hamartin to control cell growth through the mTOR pathway and cell cycle progression through the G1/S transition. Cyclin B1 is a mitotic cyclin that partners with Cyclin-dependent kinase 1 to regulate mitotic onset. My project aims to characterize the cellular timing and localization of the Tuberin-Cyclin B1 interaction. As a tool for these studies, I have constructed a variety of bimolecular fluorescence complementation (BiFC) mammalian expression vectors. The formation of a yellow fluorescent complex when the Tuberin-Cyclin B1 interaction occurs can then be monitored through live cell microscopy, providing details as to how these proteins regulate the cell cycle. Mutations in Tuberin lead to the development of numerous cancers as well as Tuberous Sclerosis, an autosomal dominant multisystem disorder characterized by the development of hamartomas. The success of using the BiFC system to analyze the Tuberin-Cyclin B1 interaction will allow for a greater understanding of the pathology of Tuberous Sclerosis and other cell growth and proliferation related diseases, like cancer, contributing to the development of effective treatments for patients with these diseases.

Abstract: 009

DEVELOPMENTAL STUDY OF BLOW FLIES AFTER EXPOSURE TO EXTREME TEMPERATURE

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The effect of exposure to heat shock at 42°C was investigated in two forensically important blow flies, *Lucilia sericata* Meigen and *Phormia regina* Meigen (Diptera: Calliporidae). Exposure to high temperature is known to influence development time, adult emergence, adult body size and mortality. Blow flies are utilized in forensic science as an estimate of time of death of homicide victims, ecology as a model to examine degradative succession, behaviour and species coexistence and medicine for maggot debridement therapy. Currently there is little known about the effects of heat shock on the development of blow flies. The 3rd instar wandering larvae of the two blow fly species were exposed to 42°C degrees for a period of two hours to measure the effect of high temperature on the rate of development and successful adult emergence. The results of this study will be used to understand the physical and physiological effects of high temperature on blow flies for use in future behavioral experiments.

Abstract: 010

FUNGAL ENDOPHYTES OF RED FESCUE (*FESTUCA RUBRA*) INCREASE HOST SURVIVAL BUT REDUCE PLANT TOLERANCE TO SIMULATED HERBIVORY

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Fungal endophytes – fungi that live within plant tissues – are ubiquitous in nature and can have profound effects on the ecology and evolution of plants. While they may improve host plant responses to both biotic and abiotic stresses, they can sometimes act as pathogens and often impose metabolic costs on hosts. Here we examine the role of fungal endophyte communities in mediating responses of the host grass red fescue (*Festuca rubra*) to salt and herbivore stress. We collected 4 red fescue genotypes from its native range on Akimiski Island, Nunavut, Canada, where it occurs in the intertidal region on the northern part of the island and is heavily grazed by nesting and brood-rearing Lesser Snow (*Chen caerulescens caerulescens*) and Interior Canada Geese (*Branta canadensis*). We then conducted a fully factorial greenhouse experiment where we crossed genotype (4 levels) with simulated grazing (clipped or unclipped), endophyte status (present or absent) and salinity (0, 32 or 64 ppt). Overall, the presence of endophytes reduced plant mortality by 42%, although this effect depended on salt concentration. On the other hand, endophytes reduced plant tolerance to simulated herbivory. Thus, our results suggest a potential endophyte-mediated trade-off in host plant survival and tolerance to herbivory and suggest this trade-off may be altered by stressful abiotic conditions.

Abstract: 011

EFFECT OF BASKING DISTURBANCES BY MOTORBOATS ON THE ENERGY BUDGET OF NORTHERN MAP TURTLES

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Behavioural thermoregulation is critical to the fitness of ectotherms that experience thermally challenging environments. For some ectotherms such as freshwater turtles, this important behaviour can be disrupted by human activities. Yet, to our knowledge the physiological cost of disrupting behavioural thermoregulation has never been quantified for any organism. We assessed the implications of basking disturbances by motorboats on the body temperature (T_b), and net energy retention (NER) of mature female northern map turtles (*Graptemys geographica*), a species that appears to be particularly vulnerable to boat disturbances. We quantified the effects of basking disturbances on T_b at various times during the northern map turtle's active season in Ontario, by modelling the difference in the daily T_b profile of turtles never disturbed from basking and turtles repeatedly disturbed from basking. The model was built using field observations of times to return to basking following disturbances and experimentally determined rates of change in T_b when basking and when submerged. Differences in T_b between disturbed and undisturbed turtles were transformed into differences in NER. The average reduction in daily NER caused by a single disturbance per hour is 50.0%, 7.0%, 1.5% and 0.4% during May 1-5, June 1-5, July 1-5 and August 1-5, respectively. Thus, from the simulation results, even relatively low rates of motorboat disturbance can have large effects on the ability of northern map turtles to maximize their energy retention using basking behaviour.

Abstract: 012

GABAergic MODULATION OF OLFACTORY BULB RESPONSES TO PHEROMONES AND AMINO ACIDS IN THE SEA LAMPREY (*PETROMYZON MARINUS*)

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Pheromones can elicit a behavioural change in the sea lamprey, *Petromyzon marinus*, via the olfactory system. Pheromones applied to the olfactory epithelium stimulate olfactory sensory neurons that send information to the olfactory bulb (OB). The OB processes the information and sends it to higher brain regions, leading to a behavioural change. Previous research showed the presence of chemotopy within the OB of the lamprey. Pheromones are strongly detected in the dorsal region. The response magnitude may be affected by modulatory pathways present in the olfactory system; one such path is the inhibitory GABAergic system whose fibers extend into all layers of the OB. Identifying how GABAergic modulation affects pheromone-evoked responses in the sea lamprey was studied by comparing dorsal OB local field potential responses to pheromones before, during, and after the local injection of gabazine (SR-95531), a selective GABA_A receptor antagonist. Upon gabazine injection, an increase in average maximum peak amplitude was observed in response to three different pheromones in the dorsal region. Recovery can be seen in all cases. Based on these findings we conclude that GABA modulates responses in the olfactory bulb by modifying peak amplitude of pheromone responses, and we propose that endogenous GABA modulates olfactory-mediated behaviours.

Abstract: 013

QUANTIFICATION AND IMMUNO-LABELING OF SOLITARY CHEMOSENSORY CELLS FROM MIGRATING AND SPAWNING PHASE SEA LAMPREY, *Petromyzon marinus*.

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Invasive to the Great Lakes, sea lamprey *Petromyzon marinus*, have chemosensory systems that aid in foraging and reproductive behaviours. Sea lamprey migrate towards spawning sites in response to chemical variations of pheromones produced by larvae and adults. In addition to the olfactory system, sea lamprey possess a diffuse chemosensory system composed of solitary chemosensory cells (SCCs) located on small papillar mounds in the mouth, gill and tail portion of the body. The papillae appear more developed, and the SCC cells appear more numerous during the end of the life cycle (migration and spawning) compared to earlier stages. The aim of this study is to quantify and compare the SCC abundance on gill papillae using scanning electron microscopy and to substantiate their identity as SCC through immunolabeling of proteins associated with chemosensory function - acetylated tubulin for neuronal labeling, and calretinin for the cytoplasm of lamprey taste cells. Our results suggest that spawning lampreys contain more SCCs than the earlier migratory specimens. Successful labeling of SCCs with a calretinin antibody was seen, and acetylated tubulin labeling showed neuronal fibers approaching the base of these SCCs. This study suggests that the SCCs are utilized by migrating and spawning phase sea lampreys.

Abstract: 014

DETERMINATION OF ENVIRONMENTAL FACTORS AFFECTING SOCIAL SPACE IN *DROSOPHILA MELANOGASTER*.

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In addition to genetic factors, several environmental factors contribute to social behaviour of *Drosophila melanogaster* seen within a social group. Social space is a measure that refers to the distance between a fly and its nearest neighbour. For a given population of *D. melanogaster*, this measurement is stable when testing under the same conditions. Therefore, social space can be used to study social interactions of flies within a group. The purpose of this research is to better understand the conditions that affect social space. This will allow researchers to predict which social behaviour variations can occur and why. To further investigate these variations, groups of Canton-S flies were used to understand the dynamic behind individual distance. Their social space was measured under various conditions. This chamber that was used is a straightforward social space assay specifically developed as a diagnostic tool to study this form of social behaviour. Unlike several other behaviours, the results have shown gender of the flies has no effect on social space. The parameters of the chamber, including size and orientation, do affect social space. Similarly, density of the group also affects social space. In addition, previous social isolation affects social space, as does the time of day when the interaction is tested. Knowledge of the variations these conditions create will allow the social space assay to become a better diagnostic tool. It will give researchers a reliable measure to study social behaviour, as well as eliminate any requirements for complex calculations and sophisticated equipment.

Abstract: 015

SEROTONIN SUPPRESSES LIPOLYSIS AND PROTEIN KINASE A SIGNALLING IN BROWN AND BEIGE ADIPOCYTES INDEPENDENT OF ITS METABOLITES

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The mitochondria of brown adipose tissue (BAT) contain uncoupling protein 1 (UCP1) and its activation has therapeutically relevant implications for obesity and type 2 diabetes. Serotonin is an ancient signaling molecule that has important functions in both the central nervous system and the periphery namely, the enteric nervous system and the bloodstream. Recently, peripheral serotonin has been shown to suppress the thermogenic function of BAT, a highly oxidative tissue. The recent association between BAT and peripheral serotonin requires further evidence that metabolites (5-hydroxyindoleacetic acid, 5-hydroxytryptophan, melatonin, L-tryptophan) of serotonin synthesis do not elicit the same suppressive effects. Additionally, white adipose can be converted to a phenotype resembling BAT, termed beige adipose tissue. The effect of peripheral serotonin on beige adipose tissue is unknown. The current study has found that the metabolites of serotonin synthesis do not suppress hormone sensitive lipase (HSL) mediated lipolysis or downstream protein kinase A (PKA) signaling in cultured brown adipocytes. Furthermore, serotonin significantly ($*P < 0.05$) decreases HSL-mediated lipolysis, PKA signaling and UCP1 gene expression in primary cultured beige adipocytes. The findings of this study have demonstrated that serotonin has a suppressive effect on adrenergic signaling independent of its metabolites in BAT. Furthermore, the suppressive effects of serotonin on beige adipose tissue lipolysis and PKA signaling are also observed. Inhibition of peripheral serotonin synthesis may serve as a novel treatment for obesity and its associated comorbidities.

Abstract: 016

GEOGRAPHIC VARIATION IN SOMATIC GROWTH RATE OF WOOD TURTLES (*GLYPTEMYS INSCULPTA*)

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Life history traits in turtles, such as body size and age at maturity, have been found to display intra-specific variation across a species' geographic range. Previous studies have examined body size variation of turtles among populations and found that body size increases with increasing latitude; an exception is the wood turtle (*Glyptemys insculpta*), which follows a non-linear relationship with larger body sizes at the two range extremes. Similarly, growth rates are influenced by climatic variables and should display geographic patterns reflective of different environments. Few studies have examined variation in somatic growth rate across the wood turtle's geographic range, and to date no studies have quantified somatic growth for the Sudbury District population, located near the species' northern range limit. The objective of this study was to quantify somatic growth in the Sudbury population, and to determine if the observed geographic variation in body size was the result of inter-population variation in somatic growth rates. In the Sudbury population, males grew faster than females during early life history stages, and overall grew to larger mean carapace lengths than females. Similar trends were observed throughout the geographic range. Populations at southern latitudes had higher somatic growth rates, and grew to smaller mean carapace lengths than those at the northern latitudes; this pattern was related to the number of frost-free days and mean active and annual temperatures. Understanding variation in species' life history traits is critical to understanding changes in population demography, which is important when managing populations that are in decline.

Abstract: 017

PREVALENCE OF *COXIELLA BURNETII* IN *DERMACENTOR VARIABILIS* AND *IXODES SCAPULARIS* TICKS, COLLECTED IN ENDEMIC REGIONS OF SOUTHERN ONTARIO

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Coxiella burnetii is a zoonotic obligate intracellular bacterium that causes Q fever in humans, and reproductive problems in ruminants, and wild and domestic animals. The common route of transmission is through inhalation of contaminated aerosols but little is known whether ticks serve as vectors. To assess the presence of *Coxiella burnetii* in two different tick species (*Dermacentor variabilis* and *Ixodes scapularis*) 85 ticks were collected in the summer of 2014 in southern Ontario. Using the tick dragging method, 15 *Dermacentor variabilis* and 70 *Ixodes scapularis* were collected. DNA was extracted using the Qiagen DNeasy® Blood & Tissue Kit and its procedures. DNA extracts from these ticks were tested using real-time PCR and sequencing. Out of the 85 ticks tested, 16 were positive for *C. burnetii*. All positive ticks were *Ixodes scapularis*, with 5 of 16 from Turkey Point Provincial Park, and remaining 11 from Murphy's Point Provincial Park. Results indicate that *Ixodes scapularis* have a higher prevalence of *C. burnetii* than previously thought. Future research should focus on determining the transmission routes of *C. burnetii* through ticks and whether transmission to humans can occur via ticks.

Abstract: 018

SEASONAL SITE FIDELITY OF FEMALE ELK (*CERVUS ELAPHUS*)

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Site fidelity is the tendency of an animal to return to a previously used site in following seasons or years. While the site fidelity patterns of many animals have already been established, those of North American elk (*Cervus elaphus*) are still relatively undocumented. We used radio telemetry and Geographic Information System (GIS) software to determine seasonal site fidelity of adult female elk from a population in north-central Ontario. We compared data collected weekly from 22 collared elk among three years, from January 2012 until December 2014. We found that the highest degree of site fidelity was during the calving and summer seasons (May through August), and the lowest degree of site fidelity was during the spring (March and April). High summer site fidelity is likely related to successful calving and rearing events in previous years. Low degrees of site fidelity during the spring may be related to changes in spatial dynamics among years as it is likely that snow depth could impede movement. Understanding the site fidelity patterns of the elk in north-central Ontario can help with management and conservation strategies, as well as with decisions involving land development.

Abstract: 019

TESTING CANDIDATE GENES FOR FEMALE SPECIES-SPECIFIC MATE PREFERENCE IN *DROSOPHILA SIMULANS*

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Behavioural isolation occurs when individuals mate with their own species and avoid mating with individuals from another species. This form of reproductive isolation is usually determined by female preference, but the genetic basis for female preference remains largely unknown. A previous genetic mapping study identified eight candidate genes for female species-specific mate preference that contribute to *Drosophila simulans* female rejection of *D. melanogaster* males. I will test these eight candidate genes using hybrids with various gene disruptions using 'no choice' mating assays. *transformer 2* appears to be a candidate gene, so it is expected that candidate genes involved in the sex determination pathway will play a role in female species-specific mate preference by causing females to display *D. simulans*-like female rejection behaviour. Further results will be discussed in the presentation. Overall, this study will help to identify genes responsible for female species-specific mate preference in *Drosophila*.

Abstract: 020

FUNCTIONAL ANALYSIS OF HERC5 SINGLE NUCLEOTIDE POLYMORPHISMS AS PREDICTED BY A MODEL OF POSITIVE SELECTION AND THE IMPACT ON ANTIVIRAL ACTIVITY

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HERC5 is a novel antiviral protein that potently blocks replication of the human immunodeficiency virus (HIV) by multiple mechanisms. HERC5 contains an amino-terminal regulator of chromosome condensation 1 (RCC1)-like domain and a carboxyl-terminal homologous to the E6-AP Carboxyl Terminus (HECT) domain. The HECT domain functions as an E3 ligase capable of ISGylating target HIV proteins and disrupting their functions. HERC5 mediated ISGylation of retrovirus structural protein prevents virus from budding from the cell membrane. The RLD domain acts as a guanine nucleotide exchange factor capable and disrupts the Ran-GTP/Ran-GDP gradient across the nucleus, which is required for nuclear export processes. HERC5 blocks the export of HIV RNA, which is required for expression of key virus structural proteins. The RLD and HECT domains trap HIV inside cells and prevent the release of new virus.

The HERC5 protein is highly conserved among evolutionarily diverse mammals. We have recently shown that HERC5 is evolving under strong positive selection. We have used an evolutionary-guided functional approach to identify biologically relevant amino acid determinants of HERC5 activity. Using an HIV release assay, we showed that some amino acids exhibited differential abilities to inhibit HIV release. Depending on the particular allele of SNP, it was associated with either a full block or no block to HIV release. Together, our data shows that biologically relevant HERC5 polymorphisms in the human population can significantly impact the ability to block HIV release and highlight HERC5 SNPs as a potential new host factor that may impact HIV disease progression.

Abstract: 021

THE EFFECT OF PRENATAL ALCOHOL EXPOSURE ON LEARNING AND MEMORY VIA HIPPOCAMPAL GENE EXPRESSION.

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Prenatal alcohol exposure leads to deficits in spatial learning and memory with associated changes in gene expression in the brain during development. Previous gene expression microarray data showed significant expression changes of the candidate genes in the whole brain. Since the hippocampus is the main learning and memory formation center, the objective of this experiment was to investigate hippocampal gene expression that leads to the spatial learning and memory deficit using a mouse model. Candidate genes in this study include *bcl2*, *cul4b*, and *nsdhl*. These genes are part of a predicted network and are in the same gene ontology of developmental disorder. The experimental design involved DNA and RNA extraction, cDNA synthesis from RNA, and Real-Time PCR to confirm changes in gene expression in male mouse hippocampus. In the hippocampus, *bcl2*, *cul4b* and *nsdhl* genes, showed no significant expression changes between control and ethanol treatment groups ($p = 0.58, 0.49, 0.99$, respectively, n.s). The results suggest the expression of these genes in the hippocampus can be ruled out as candidate genes that affect the behavioural phenotype. More experiments are required using other genes in the hippocampus that affect learning and memory.

Abstract: 022

THE GROWTH OF OATS (*AVENA SATIVA*) ON REMEDIATED MINE TAILINGS AND THE EFFECTS OF N-VIRO™ SOIL AMENDMENT

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Mine tailings cover vast amounts of land, so it is beneficial when they can be used for productive purposes. At Glencore's nickel-copper mine in Onaping, northwest of Sudbury, Ontario, we have been successfully growing biofuel crops, and have begun experimenting with applications of N-Viro™, a soil amendment derived from alkalized bio-solids. Different sections of tailings were covered with compost over the course of three years (2012-2014), and planted with oats (*Avena sativa*) in spring 2014. About half of the compost covers were treated with 15 t/ha of N-Viro™. The goals of the study were to determine if younger or older compost made a better plant growth medium, if the N-Viro™ application significantly increased oat growth and increased the bioavailable concentrations of metals and nutrients in the compost. We collected 155 plants and composite soil samples from five 1x1 m quadrats in each of six sampling areas. Collected data included the shoot and root lengths and masses, the soil moisture loss, pH, and metal and nutrient analyses of the soil. N-Viro™ promoted a significant increase in shoot and root length in plants on 2013 compost, but not on plants from the 2012 compost. The older compost was a significantly better growth medium for plant height. Nutrients were often significantly lower in the tailings and 2013 compost, especially without N-Viro™, from 2012 and 2014. This study demonstrates the potential benefit of using N-Viro™ and compost in land reclamation.

Abstract: 023

INVESTIGATION OF FUNGAL ACTIVITIES IN TECHNOSOLS AT THE BARRICK GOLD MINE IN HEMLO, ONTARIO

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Mine site restoration at the Barrick Gold Mine in Hemlo, Ontario involves a combination of various strategies to restore the mine site to former conditions after mining operation cease. The presence and activity of fungal communities within the reclamation media manufactured from crushed rock and lumber mill wood waste (Technosols) on the mine site is largely unknown. Technosol samples varying in the amount of organic matter (low 40%; high 80%) and crushed waste rock were collected from the edge of twelve 4 m x 4 m lysimeters. Soil samples were also collected from the top 5 cm of a naturally re-vegetated site, a white spruce site and a mature upland spruce and poplar forest site adjacent to the mine. After plating 1 g of soil with 5 ml of water, the cultures were incubated for 4 days at 26°C, and at room temperature for 24 hours before being refrigerated. Fungal colonies appeared 24 hours after the initial incubation period with significant growth by 4 days, with visible differences in abundance and appearances of colonies. The differences observed in colonies reflect either the amount of organic matter in the Technosols or the nature of the samples from the vegetated sites. These results will aid in creating a database to guide future Technosol development for mining restoration practices in Boreal Shield regions of northern Canada.

Abstract: 024

BIRDS OF A FEATHER: BEHAVIOUR AND GENETIC RELATEDNESS OF ADULT HERRING GULLS (*LARUS ARGENTATUS*) ATTENDING UNUSUALLY CLOSE-TOGETHER NESTS.

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Herring gulls (*L. argentatus*) breed colonially and monogamously. Typically, nests are distributed throughout a colony at regular intervals of intermediate distance due to trade-offs between high levels of territorial aggression at close proximity and inefficient defense of both nest and offspring in larger territories. In a colony of herring gulls in Hamilton Harbour, some gulls were observed to attend nests located closer together than usual. Previously, closely paired nests in colonies in the Great Lakes region belonged to polygynous trios or female-female pairs which likely resulted from sex-ratio skews. We investigated alternative mating strategies and elevated levels of relatedness as potential factors resulting in nests which were close together. We assessed the behaviours of gulls at pairs of nests that were close together and control pairs of nests with normal spacing through video recordings. DNA which was extracted from blood samples that we collected during the breeding season and was amplified through microsatellite, multiplex PCR to evaluate relatedness through microsatellite genotyping. We observed no evidence of polygynous trios in any of the nests studied. Infrequent adoptions and similar levels of aggression occurred at both control nests and nests that were close together. Apart from one pair of nests, gulls at close-together nests did not share greater relatedness than gulls at control nests. Overall, the factors contributing to the unusually close together nests remains unclear; the results may be improved with larger sample sizes and longitudinal study.

Abstract: 025

DO EXERCISE AND AIR EXPOSURE AFFECT BOLDNESS AND ACTIVITY LEVEL IN THE BLUEGILL SUNFISH?

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Fish that are captured and then released may experience altered behaviour due to exposure to stress for some period of time upon return to the water. Variation in such behavioural responses between individuals is therefore important as such differences reveal how individuals will differentially respond to stressors as well as acquire food and avoid predators. In this study, we used field-caught bluegill sunfish (*Lepomis macrochirus*) and behavioural assays to determine the effects of catch-and-release on short-term behaviour. The fish were placed into individual opaque plastic holding tanks and were assessed for individual differences in refuge emergence, activity, and flight-initiation-distance (FID) as well as several estimates of stress and condition (e.g. opercular count, reflex-action-mortality-predictors [RAMP]) before they were exposed to various combinations of exercise and air exposure (or control) treatments. Fish were exercised for 0s, 30s, or 120s, and exposed to air for 0s, 60s, or 500s. Afterwards, the fish were left for one hour, and behaviour was reassessed. Contrary to initial expectations, the majority of bluegill did not exit the refuge or explore their experimental arena. Tests for opercular count, FID and RAMP were conducted on fish, regardless of the results of the refuge emergence test. Operculum count and RAMP were not effective predictors for fish behaviour. As well, we detected no differences in FID among air exposure and exercise treatments. This study provides novel insight into using simple behavioural and reflex indicators to evaluate the short-term consequences of catch-and-release stressors.

Abstract: 026

ENHANCED DEGRADATION OF THE BLUE GREEN ALGAE HEPATOTOXIN *CYLINDROSPERMOPSIN*.

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Blue green algae, also known as cyanobacteria or cyanophyta, are found in a variety of ecosystems throughout the world. This is a public health concern since there have been outbreaks of hepatoenteritis resulting from exposure to cyanotoxins released into water systems by cyanobacteria. The use of germicidal ultraviolet (UV) light has been shown to degrade cyanotoxins but the process is slow and toxin remains at the end of the process. It has been speculated that the use of titanium dioxide (TiO₂) as a photocatalyst alongside UV can increase degradation of the toxin through the generation of hydroxyl radicals. To investigate this water samples containing 100 µg/L of cylindrospermopsin, a hepatotoxin produced by a variety of cyanobacteria, were exposed to UV irradiation alone (control) as well as to TiO₂ with UV irradiation. Samples that contained suspended TiO₂ had significantly higher degradation rates than controls and the majority of the TiO₂ experiments resulted in final cylindrospermopsin concentrations of 0 µg/L. Experiments conducted with immobilized TiO₂ resulted in average degradation rates greater than -20 µg/L/min but there was a loss of immobilized TiO₂ highlighting an area where the process could be improved. However, these results suggest that TiO₂ and UV irradiation is a very promising treatment for water supplies contaminated with algal blooms. This approach could be coupled with existing water treatment and used to protect against outbreaks of hepatoenteritis caused by the presence of cyanotoxins in water.

Abstract: 027

GENETIC DIVERGENCE AND DIVERSITY BETWEEN WILD AND CAPTIVE POPULATIONS OF YELLOW PERCH (PERCA FLAVESCENS)

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Captive breeding and reintroduction programs are used as a conservation tool in an attempt to rescue threatened or overexploited species. However, captive breeding can be dangerous because small, isolated populations can lead to changes in inbreeding and decreased heterozygosity, which can be problematic when individuals are released back into the wild. With increasing conservation efforts it is important to research different methods to attempt to preserve genetic diversity of species while in captivity. In our experiment, three different treatments of yellow perch (*Perca flavescens*) were analyzed for genetic diversity within each population and genetic divergence among them. The treatments consisted of wild yellow perch (W), "wild-collected" fertilized eggs born in semi-natural ponds with no predators (WC), and yellow perch born and raised for multiple generations in semi-natural ponds with predators (MGC). We found that the MGC treatment showed significantly higher levels of heterozygosity, allelic richness, and no significant differences in inbreeding compared to W and WC treatment. Also, we found significant genetic divergence between the MGC treatment and both W and WC treatments. As urbanization increases and more species become endangered, conservation efforts such as captive breeding programs will increase. Our study is important because we examine the effects of captivity on inbreeding and heterozygosity levels to predict the potential ability of a population to survive upon reintroduction into the wild.

Abstract: 028

INVESTIGATING THE ACUTE ANTIDEPRESSANT EFFECTS OF KETAMINE AND GHRELIN IN THE FORCED SWIMMING TEST.

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The efficacy of N-methyl-D-aspartate receptor antagonist ketamine to alleviate depressive symptoms has catalyzed a wealth of research looking at the glutamatergic system as a therapeutic target for depression. The side effects of ketamine limit its clinical use; one potential way to ameliorate these effects is through combination therapy. Ghrelin has recently been found to protect against stress-based models of depression. This investigation aims to observe possible drug interactions between ketamine and ghrelin.

CD1 male mice (n=56) were given an intraperitoneal injection of ghrelin (80µg/kg) or saline, followed by a saline, ketamine low (5mg/kg) or ketamine high (10mg/kg) injection. The acute effects of each group were then tested using the Forced Swimming Test (FST). Statistical analyses revealed significant antidepressant effects of ketamine and ghrelin that were dose dependent. Specifically, the low dose of ketamine reduced immobility in the FST, an effect that was attenuated in mice getting both ghrelin and the low dose of ketamine. At high doses, ketamine was ineffective at decreasing immobility in the FST, but it did reduce immobility when injected in combination with ghrelin. Finally, ghrelin alone decreased immobility time to the same extent as the low dose of ketamine. These results show that acutely, there is an interaction between ghrelin and ketamine which works to increase immobility. The results also show that ketamine and ghrelin have antidepressant effects when administered independently. This suggests that ghrelin or its analogs could be potentially used as antidepressant medications alone or in combination with fast acting antidepressant drugs like ketamine.

Abstract: 029

SPERM PERFORMANCE IN THE AFRICAN CYPRINID *BARBUS APLEUROGRAMMA* FROM DISTINCT DISSOLVED OXYGEN REGIMES

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Low levels of dissolved oxygen (DO) or hypoxia can have detrimental effects on the reproductive biology of wild fish populations—especially for freshwater organisms. The effects of hypoxia on general body condition and sperm quality (i.e. swimming speed) of the freshwater African cyprinid *Barbus apleurogramma* were investigated by collecting samples from 2 normoxic and 3 hypoxic sites in Uganda. Because hypoxia is an endocrine disruptor, we predicted that sperm quality should be lower in males from hypoxic than in males from normoxic sites. We found significant differences in body length and body mass between DO levels, with fish from hypoxic sites being smaller than those from normoxic sites. However, males from hypoxic sites had relatively larger gonads than males from normoxic sites. Interestingly, we found no trade-off between estimated sperm longevity and sperm swimming speed, nevertheless, our results suggest that a trade-off exists between total gonad weight and sperm swimming speed independently of DO level. We proposed that because of their smaller size, males from hypoxic sites are at a positional disadvantage during spawning. As a result, relatively larger gonads and slightly faster swimming sperm were naturally selected in the hypoxic sites. Future studies should determine if *B. apleurogramma* males from hypoxic and normoxic sites within a region aggregate for spawning, and if males from hypoxic sites are at a positional disadvantage. To the best of our knowledge, this is the first study on the reproductive biology of *B. apleurogramma* and assessing the effects of hypoxia on a freshwater cyprinid.

Abstract: 030

ELUCIDATING THE GENETIC DETERMINANTS OF EXTREME HIGH-DENSITY LIPOPROTEIN PHENOTYPES USING NEXT-GENERATION SEQUENCING.

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Presently, there is a gap in knowledge regarding the genetic determinants for extreme levels of high-density lipoprotein cholesterol (HDL-C). We have clinically ascertained a large cohort of patients with extremely high or low HDL-C lipid profiles ($N=240$). We explored whether the extreme HDL levels in these patients were modulated by genetic variants in known lipid genes or genes not yet implicated in lipid pathways. Initial sequencing with LipidSeqTM, a targeted next-generation sequencing panel for dyslipidemias and related metabolic disorders, did not detect any promising genetic mutations and therefore, was unsuccessful in the diagnosis of the primary genetic basis of extreme HDL-C for >80% of the patients under study. This surprising observation led us to hypothesize that extreme HDL-C levels in our unique cohort are the result of a large-effect mutation in a not previously implicated in dyslipidemia; or is the result of the inheritance of multiple genetic variants with individually modest effects but a cumulative, large effect. Whole-exome sequencing, a large-scale, next-generation sequencing approach that targets protein-coding regions of all genes in the human genome was performed on 120 of these genetically undefined patients. Of the 120, my project focused on unraveling the genetic basis of five patients. Using a robust bioinformatics analysis that is optimized for complex traits, we are able to identify potentially pathogenic HDL-C modulating genetic variants. These findings will serve as the first step towards implicating a mutation and its gene as a novel determinant in extreme HDL-C concentrations, and further characterizing the HDL pathway.

Abstract: 031

APPLICATION OF SURFACE AMENDMENTS TO MANUFACTURED SOILS TO ENHANCE PLANT GERMINATION AND PRODUCTIVITY FOR RECLAMATION PURPOSES

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In order to minimize their post-mine closure footprint, Barrick-Hemlo Gold Mine requested the development of Technosols constructed from regional waste materials for use as sustainable covers for the waste rock structures on their sites. Building on previous studies of Technosols for mine land reclamation, the current study investigates applications of ten soil amendments to improve fertility and increase soil moisture retention to enhance seedling and transplant survival. The eight week biochamber growth study, programmed to the predicted 2030 Hemlo spring conditions, investigated the use of elemental sulphur, wood boiler ash, N-ViroTM, Ahg (from Humic Gleysol) and LFH (from Orthic Humo-Ferric Podzol) horizon amendments at various rates to improve soil quality to promote growth of annual ryegrass (*Lolium multiflorum*). The results demonstrated that applications of 10% Natural Soil and 80T/Ha wood ash enhanced the root:shoot ratio. Addition of Sulphur prills (1.5% w/w) produced the largest increase in above and below ground biomass. Soil monitoring probes demonstrated that N-ViroTM at 34 T/Ha had the highest soil solution electrical conductivity after simulated rainfall events. Chemical analysis of through-flow collected after a simulated storm (1 cm rainfall hr⁻¹) confirmed the results obtained with the *in-situ* sensors. Soil sample analyses illustrated that N-ViroTM applications provided the highest enrichment of soil nutrients.

Abstract: 032

TUBERIN PROTEIN LEVELS IN THE PRE-NATAL MOUSE BRAIN

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The tumour suppressor genes TSC1 and TSC2, which form a heterodimer, code for the proteins Hamartin and Tuberin, respectively. Mutations in either one of the genes can lead to many neurological disorders including the autosomal dominant disease Tuberous Sclerosis (TS). TS is characterised by the formation of benign tumours known as hamartomas in multiple organs of the body, the most detrimental ones being in the brain. Mutations in TSC2 are more severe and 5 times more common in sporadic cases of TS. TSC1 and TSC2 null mice are embryonic lethal, indicating that these proteins are necessary for development. It is known that these genes play a role in regulating cell growth and proliferation through the Mammalian Target of Rapamycin signalling pathway. The focus of this study is to quantify the Tuberin protein levels in different sections of the brain and at different time points. I compared the values in the Purkinje cell layer and Choroid plexus between two time points, embryonic day 14.5 and embryonic day 16.5. I hypothesize that the protein levels of Tuberin in the purkinje cell layer and choroid plexus will be higher during stages that are critical for neurological development. To test this, I performed immunohistochemistry on brain tissue from mice embryos and quantified the stain intensity. The results of this study can provide further insight on the processes and pathways that are affected in disorders such as TS.

Abstract: 033

WHOLE-MOUNT PREPARATION FOR VISUALIZING OLFACTORY SENSORY NEURONS IN CHINOOK SALMON ALEVIN, *ONCORYHYNCHUS TSHAWYTSCHA*.

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Olfactory cues mediate a multitude of behaviours in commercially important Chinook salmon, such as foraging behaviour and homeward migration to natal streams for spawning. Fish detect olfactory cues when the molecules, such as amino acids, bind to olfactory receptors of olfactory sensory neurons (OSNs). The axons of the OSNs project into the olfactory bulb where they terminate on regions known as glomeruli. These glomeruli are the functional units for odour discrimination, partly due to their stereotyped organization, where amino acid responsive microvillous OSNs project to the lateral olfactory bulb region. Whole-mount immunohistochemistry (IHC) techniques have been applied in model organisms to map the organization of these glomeruli and to test for neuroplasticity in response to sensory experience. My goal is to expand the current understanding of glomerular organization in Chinook salmon alevin by adapting whole-mount IHC techniques to visualize glomeruli. Olfactory bulbs were bleached to eliminate melanophores located in the dura, then labelled for calcium-binding proteins, to target microvillous OSNs and visualized under fluorescence microscopy. The establishment of this whole-mount IHC protocol revealed amino acid responsive OSNs are organized to identifiable glomeruli present at hatch. Future studies can test whether the development of the Chinook salmon olfactory system is sensitive to olfactory experience, as is required for imprinting, indicated by a change in corresponding glomerular volume.

Abstract: 034

GENE HUNTING: IDENTIFYING THE MOLECULAR BASIS OF HUMAN LIPODYSTROPHY THROUGH NEXT-GENERATION SEQUENCING

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Familial lipodystrophy is a rare Mendelian genetic disorder primarily characterized by variable loss of adipose tissue, as well as metabolic complications including diabetes and hypertriglyceridemia. Generally, when familial recurrence of a rare phenotype is present, the likelihood that the disease is genetically determined is high. With lipodystrophy, although many causative genes have been discovered, there are numerous, unexplained and severe cases of genetically undiagnosed patients. This investigation sought to establish whether the lipodystrophy in these patients is caused by genetic mutations in a novel gene, not previously associated with lipid abnormalities. Through a next-generation sequencing custom-designed panel, LipidSeq™, 37 patients were sequenced then analyzed using multiple bioinformatics pipelines. 30 genes with various mutations were implicated, with 6 of these mutations (in *APOB*, *APOA4*, *LMNA*, *LDLR*, *GCKR*, *SCARB1*) confirmed as causative, and 2 (*BLK*, *APOC3*) requiring confirmation in the 37 patients under study. For 2 patients, whole-exome sequencing (WES) was performed. From these WES data, candidate variants in novel genes were identified, which may explain their lipodystrophic phenotype. Our next steps would be to validate these newly identified variants using an independent technique such as Sanger sequencing. These results provide a stepping-stone towards the discovery of a new causative gene for lipodystrophy, which can lead to refined diagnosis, improved therapeutic intervention, personalized medicine, and potentially, disease prevention in the future.

Abstract: 035

PROOF OF PRINCIPLE INVESTIGATION: EVALUATING COMBINED SOX9 KNOCKDOWN AND WEIGHT SUPPORTED TREADMILL TRAINING FOR THE TREATMENT OF SPINAL CORD INJURY

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Despite being a gold standard in post spinal cord injury (SCI) gait rehabilitation, weight supported treadmill training (WSTT) results in minimal functional improvements for persons with severe SCI. We hypothesize that recovery after WSTT is limited by the action of chondroitin sulfate proteoglycans (CSPGs) in perineuronal net and glial scar, which inhibit axonal growth. Thus, we predicted that knocking down SOX9, a transcriptional factor involved in the up-regulation of CSPGs, would improve locomotor recovery after WSTT in an animal model of SCI. Three weeks after a 70 kdyne contusion SCI at thoracic level 9, *Sox9* knocked-down mice (n = 5) and control mice (n = 8) were exercised with WSTT for 10 min, five days a week, for four weeks. Untrained control (n = 9) and knock-down (n = 4) animals served as additional controls. Motor changes were monitored using: a 2 mm rod grasp test, to assess sensorimotor function; the Basso Mouse scale for hind limb function; locomotor activity boxes to analyse voluntary movement, and; a 1.2 cm² grid walk to assess complex, coordinated hind limb stepping. This study established a novel WSTT protocol for mice. Though statistically underpowered, this preliminary study provides intriguing trends suggesting that *Sox9* knockdown may enhance the effects of WSTT.

Abstract: 036

DETERMINING PHENAZINE SPECIFIC SECRETION BY CLINICALLY SIGNIFICANT EFFLUX PUMPS IN *PSEUDOMONAS AERUGINOSA* PA14

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Phenazines are redox-active metabolites that are synthesized and secreted by the Gram-negative bacterium *Pseudomonas aeruginosa*. These molecules function in quorum sensing, virulence, and iron sequestering. The most abundant phenazine is pyocyanin, as it is produced by 95% of all *P. aeruginosa* strains. Research suggests that phenazines are secreted into the local environment via the efflux pumps MexAB-OprM, MexCD-OprJ, MexEF-OprN, and MexXY-OprM. To our knowledge, there is no previous research on phenazine-pump specificity. This study therefore aims to determine if phenazines are selectively secreted by specific efflux pumps using the following techniques: Whole-Genome Sequencing, Drug Inhibition, analytical chemistry, and Quantitative RealTime-PCR (qRT-PCR). Sequencing has revealed mutations in the *nfxB* and *lasR* genes. The *nfxB* gene produces a repressor for the MexCD-OprJ efflux pump, while *lasR* is a global transcription factor. Drug inhibition has revealed one of two possibilities: either that the red pigments are secreted by a different set of efflux pumps, or the drug is time sensitive. Initial chemical tests have revealed that pyocyanin is being secreted by all mutants. We are measuring efflux pump mRNA transcript abundance using qRT-PCR. Taken together these data suggest that there is efflux pump-phenazine specificity, but not in the way originally hypothesized. The pump inhibitor PAßN appears to work in liquid culture, suggesting that there is a previously uncharacterized pump system specifically secreting the red pigments, and that the Mex pumps secrete pyocyanin. This information can be used to develop an *in vivo* drug to block the efflux of pyocyanin into the human body.

Abstract: 037

THE RESPONSE OF ANTARCTIC HAIR GRASS (*DESCHAMPSIA ANTARCTICA*) TO ELEVATED TEMPERATURE AND ATMOSPHERIC CARBON DIOXIDE

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Anthropogenic fossil fuel emissions are causing atmospheric CO₂ and global air temperatures to increase at an alarming rate. This change is most evident in high latitude areas such as Antarctica, where temperatures are increasing more rapidly than in the rest of the world. Elevated temperatures and CO₂ often allow for higher photosynthetic rates through increased Rubisco function and electron transport rates. Understanding the responses of these measures of photosynthetic capacity to increases in atmospheric CO₂ and temperature is important for accurately predicting how the Antarctic plant *Deschampsia antarctica* will respond to a rapidly changing environment. In this study *D. antarctica* was acclimated to six combinations of temperatures and CO₂ environments that simulate future environmental conditions in the Antarctic Peninsula and maritime Antarctica. We then assessed morphological changes and photosynthetic performance: stomatal conductance, net carbon assimilation rates (A_{Net}), and photosynthetic capacity (maximum Rubisco carboxylation capacity, V_{Cmax} and maximum electron transport rates, J_{max}). Growth at elevated temperatures increased A_{Net} and altered leaf morphology. Leaf width increased both with temperature and [CO₂] and the combined effect of both, but no response was seen in stomatal conductance, V_{Cmax} or J_{max} across the different growth temperatures and CO₂ conditions. Thus, *D. antarctica* does not appear to be negatively affected by predicted future climate conditions, implying that the species will be able to tolerate the warming and higher CO₂ expected over coming decades.

Abstract: 038

A STUDY OF VR23, A NOVEL ANTICANCER COMPOUND WITH PROTEASOME INHIBITION ACTIVITY

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VR23 is a novel proteasome inhibitor that preferentially kills cancer cells over non-cancer cells. Data obtained from our study with several different cancer cell lines including HeLa (cervical cancer) and MCF7 (breast cancer) cell lines demonstrated that VR23 causes centrosome amplification through the stabilization of ubiquitinated cyclin E, leading to eventual cell death by apoptosis. In contrast, VR23 neither causes the accumulation of ubiquitinated cyclin E nor apoptosis in non-cancer cells (MCF 10A), which is an extremely desirable property for an anticancer drug. One possibility of this differential effect of VR23 on cancer and non-cancer cells may be a difference in permeability between the two different cell types. To test this hypothesis, we analyzed by HPLC the amount of VR23 in cell lysates prepared from HeLa and MCF 10A that were treated with VR23. Preliminary results indicate that the non-cancer MCF 10A cells do not uptake VR23 while HeLa cells do. Our finding suggests that the selective killing of cancer cells by VR23 is, at least in part, due to its greater permeability to cancer cells compared to non-cancer cells. In addition to the selectivity, VR23 can kill glioblastoma brain cancer cells at least 40 times more effectively than Temozolomide, a “standard” brain cancer drug used in clinics. To develop an effective brain cancer treatment protocol, we are in the process of systematically examining the combinational effects of VR23, Temozolomide and other anticancer compounds.

Abstract: 039

DETERMINING THE GENETIC BASIS FOR INTERSPECIFIC MATE DISCRIMINATION IN *DROSOPHILA SIMULANS*

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Female choice in mates is believed to be the first factor isolating diverged populations, preventing gene flow and leading to the evolution of separate, distinct species. For example, female *Drosophila simulans* reject interspecific mates from a closely related species, *D. melanogaster*, forming reproductive isolation between the species. The genetic basis of female choice in *D. simulans* was previously mapped to large candidate regions on the right arm of the third chromosome. In order to fine map the chromosome, I performed deficiency mapping within these regions. Genotypes with and without deficiencies were compared for mating success through both behavioural assays and female dissections for the presence of sperm. If hybrid individuals inheriting a deficiency show reduced mating success compared to individuals without a deficiency, the region is said to be significant and may contain a candidate gene. A comparison of overlapping deficiency regions (both significant and non-significant) with previously found significant regions will reduce the number of candidate genes, allowing for subsequent testing of individual genes contributing to female *D. simulans* rejection of *D. melanogaster* males. Identification of individual loci for species-specific female mate preference will improve our understanding of the initial isolating factors involved in speciation.

Abstract: 040

THE ROLE OF P-eIF2 α ON PLACENTAL DIFFERENTIATION IN RCHO-1 RAT PLACENTAL CELLS

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Intrauterine growth restriction (IUGR) in humans is the inability of the fetus to grow to its full potential due to impaired placental development. Moreover, IUGR is associated with increased endoplasmic reticulum (ER) stress in the placenta. ER stress is the accumulation of misfolded proteins in the cell leading to abnormal cellular function, and in severe cases, apoptosis. Unfolded protein response (UPR) is activated to eliminate this harmful accumulation. While the UPR activates phosphorylation of eukaryotic initiation factor 2 α (P-eIF2 α) within the PERK pathway to attenuate protein translation and relieve ER stress short-term, the effects of P-eIF2 α on placental differentiation and function are unknown. We hypothesize that an increase in P-eIF2 α will lead to adverse effects on downstream target genes critical for trophoblast differentiation and placental function. We employed the drug Salubrinal in a RCHO-1 rat placental cell line to exclusively study the effects of P-eIF2 α on trophoblast differentiation and function. When RCHO-1 cells were treated with 75 μ M Salubrinal, ER stress was activated, as indicated with an augmented P-eIF2 α at 6 hours followed by an increase in CHOP expression at 24 hours. Trophoblast differentiation was suppressed as demonstrated by a decrease in the expression of placental lactogen-1 (PL-1). While the expression of placental glucose transporter-1 (Glut-1) was diminished by treatment of Tunicamycin (a global inducer of ER stress), it was unaltered by Salubrinal. Collectively, this suggests that activation of P-eIF2 α impairs trophoblast differentiation, but more severe ER stress is required to impact placental function (*e.g.* glucose transport).

Abstract: 041

METHANE OXIDATION DYNAMICS AND METHANOTROPH COMMUNITY STRUCTURES IN WETLANDS ACROSS A SULPHUR AND METAL DEPOSITION GRADIENT IN SUDBURY, ONTARIO.

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Methanotrophic bacteria aerobically oxidize methane (CH₄) as their sole carbon and energy source. Due to the potency of CH₄ as a greenhouse gas, oxidation of biogenic CH₄ emissions from wetland soils into the atmosphere by methanotrophs mitigates the global greenhouse effect. It is known that environmental copper (Cu) influences expression of different CH₄ monooxygenases, the enzymes which initiate CH₄ oxidation, and that sulphate (SO₄) inhibits soil methane producers, altering CH₄ efflux. Consequently, it is speculated that both factors modulate methanotrophic community structures and activity. In this study we examined the impact of legacy metal and sulphur loading from smelters on methanotrophs across ten poor to intermediate fen wetlands in Sudbury, Ontario, Canada. We predicted that methanotroph diversity and activity would be most severely altered near smelters. Concentrations of bioavailable inorganic elements (Cu, Ni, Fe, Ca, S, N) were analyzed in surface peat soils and porewater samples, and potential rates of methane oxidation were quantified in soil incubations. DNA was then extracted from peat ($n = 30$) and the methanotrophic community was characterized using molecular techniques targeting CH₄ monooxygenase (*pmoA* and *mmoX*) gene sequences. Variations in peat methane oxidation rates were observed across sites, but there was no clear correlation with distance from smelters. Additional data on chemical findings and diversity will be presented. Initial results may suggest that the relationship between Cu and S with methanotrophs is weak and methanotroph dynamics are more strongly related to other environmental factors.

Abstract: 042

DYNAMIC INTERACTIONS BETWEEN TYPE IV PILUS ASSEMBLY COMPLEX COMPONENTS IN *PSEUDOMONAS AERUGINOSA*.

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The Type IV Pilus is an evolutionarily conserved system found in a variety of microorganisms, including the opportunistic pathogen *Pseudomonas aeruginosa*. This intricate biological nanomachine possesses a diverse repertoire of functions; importantly, Type IV Pili mediate a form of motility known as "twitching". Recent studies have illustrated the complexity of this system by showing multiple pairwise interactions can occur between its components. A dynamic model for pilus extension and retraction has been proposed, wherein rotations of the secretin and alignment complex components mediate the transition between extension and retraction of the pilus fiber. Here, we focus on the dynamics of the PilP/Q interaction. We modelled this interaction upon existing co-structures in the homologous Type II Secretion System, using individual NMR structures for PilP and PilQ. We aim to use structure determination methods such as small angle X-ray scattering or X-ray crystallography to validate this model. The putative interface will be probed using cysteine mutagenesis and disulfide bonding analysis to effectively lock PilP/Q in a static conformation. Wild type *pilP/Q* genes were successfully deleted from the *P. aeruginosa* chromosome to accommodate complementation with PilP/Q cysteine mutants. Functional assays will be performed to determine the effect of disulfide bonding on Type IV Pilus function. Transient PilP/Q interactions may serve as good targets for protein-protein interaction inhibitors to disrupt Type IV Pilus function and reduce virulence in *P. aeruginosa*.

Abstract: 043

EXPLAINING BEHAVIOURAL VARIATION IN CAPTIVE-REARED ATLANTIC SALMON (*Salmo salar*) FOR RESTORATION EFFORTS

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Phenotypic variation in the form of behavioural flexibility amongst individuals are of great importance in restoration projects in conservation biology since this variation will influence the animal's ability to adaptively respond to new environmental conditions, and thus affect population persistence. The presence of bold and shy behavioural phenotypes can exist amongst individuals within a population, including fish, and is influenced by many intrinsic and extrinsic factors. This study set out to determine the mechanisms driving behavioural flexibility of captive-reared populations of juvenile Atlantic salmon (n = 86) that originated from two source populations. The mechanisms investigated were body size, genotype, and innate personality. A behavioural assay was designed that consisted of an open arena, a refuge area, a novel food item, and a model predator attack. Each fish's behaviour was recorded and quantified using the software JWatcher, and analyzed for behavioural consistency across these different trials. A general linear model was then used to examine the effect of body mass and genotype (population strain) on behaviour scores. The results of this study will contribute to the recognized importance of including behavioural assessments of individuals raised in captivity to gauge the probability of successful reintroduction.

Abstract: 044

THE IMPACT OF *COXIELLA BURNETII* INFECTION ON THE BODY CONDITION OF SMALL MAMMALS

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Zoonoses are infectious pathogens that can be transmitted between animals and humans. Sixty percent of all human pathogens are recognised as zoonoses. The interface between humans and animals has grown due to urban sprawl, large-scale domestication, industrialization and agriculture. In this context, zoonoses become a greater risk to public health. *Coxiella burnetii* is a zoonotic bacterium that is shed by small ruminants and causes Q fever in humans when the infectious bacterial spores are inhaled. Currently, we know the effect that *C. burnetii* has on both small ruminants (still-births, abortions, weak offspring) and humans (acute Q fever includes flu-like symptoms and chronic Q fever includes pneumonia, osteomyelitis and endocarditis). In Algonquin Provincial Park, the presence of this bacterium has been confirmed in wild populations of forest rodents; however the effect that infection has on these rodents is currently unknown. In this study, we have compared the body condition of Eastern chipmunks (*Tamias striatus*), raccoons (*Procyon lotor*) and deer mice (*Peromyscus maniculatus*) across Southern Ontario to determine if body condition is affected by *C. burnetii* infection. We have found that the deer mice and male raccoons' body conditions are impacted by infection status, while chipmunks and female raccoons' are not. Infected deer mice and male raccoons have a lower body condition in comparison to those that are not infected. Understanding the impact of *C. burnetii* on vertebrates can aid in effectively monitoring and controlling the spread of this pathogen.

Abstract: 045

SEASONAL DIET SHIFTING IN SONGBIRDS IS NOT CAUSED BY CHANGING NUTRITIONAL TARGETS.

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Dietary shifting, for example from insects to fruits, is a common mechanism used in migratory songbirds to accumulate fat to fuel migratory flights. I studied the underlying causes of dietary shifting in yellow-rumped warblers (*Setophaga coronata*) by comparing fall migrants to wintering birds. I offered three nutritionally controlled and distinct diets: a high carbohydrate diet, an intermediate diet, and a high protein diet. These diets were paired together to form three diet combinations, which were fed to the birds in a rotation during each season. I collected leftover food of the diets offered which I oven dried to determine cumulative energy and crude protein intake of each diet combination. I used the geometric framework of nutrition to compare cumulative total energy intake and crude protein intake of each diet combination within and between seasons. I also measured preference to determine which diet within a combination was favored during and between seasons. I found that yellow-rumped warblers maintain a constant total energy intake during migration regardless of diet combination. This suggests that yellow-rumped warblers consume macronutrients available to them without protein limitations to reach their total energy intake target. When given a choice, warblers consistently choose the diet with the most non-protein energy content throughout both seasons. My findings suggest that songbirds have the ability to shift their diet, resulting in high flexibility in their macronutrient requirements.

Abstract: 046

ATYPICAL TERATOID/RHABDOID TUMOURS (AT/RT): THE IMMUNOTHERAPEUTIC POTENTIAL OF NATURAL KILLER (NK) CELLS

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Atypical teratoid/rhabdoid tumour (AT/RT) is a rare, highly malignant paediatric brain tumour with a dismal prognosis. Natural killer (NK) cells have shown considerable promise for therapeutic use for a number of human cancers; however, their efficacy against AT/RT has not been described to date. The goal of this study was to assess the *in vitro* susceptibility of AT/RT to an NK cell line and *ex vivo* cytokine-activated NK cells. Major NK cell inhibitory and activating receptors/markers were assessed via flow cytometry for *ex vivo* IL-15 activated NK cells before and after 21 days of culture. IL-15 culture resulted in downregulation of inhibitory NK cell killer immunoglobulin-like receptors (KIRs) and upregulation of an activating receptor, NKp44. Other activating receptors/markers (NKp30, NKp46, DNAM-1, NKG2D, perforin and granzyme B) maintained high expression levels after culture. The surface expression of NK cell ligands on two AT/RT cell lines (CHLA-02 and CHLA-04) was also surveyed. We found that major NK cell activating ligands (Nectin-2, PVR and MICA/B) as well as a death receptor (Fas) were highly expressed by both AT/RT cell lines. NK-92 and IL-15 activated NK cells were assayed for functionality by performing fluorescence-based europium-TDA cell cytotoxicity assays against AT/RT. Both AT/RT cell lines were highly susceptible to killing by both *ex vivo* IL-15 activated NK cells and NK-92, although to a lesser extent, providing a rationale for further research into the immunotherapeutic potential of NK cells for AT/RT patients.

Abstract: 047

CHARACTERIZATION OF INSULIN-SECRETORY PROPERTIES OF GLP-1-TF FUSION PROTEIN

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The incretin hormone glucagon-like peptide (GLP)-1 is a promising new therapeutic option for treatment of type 2 diabetes. However, a major limitation to the therapeutic use of the native GLP-1 molecule is its very short plasma half-life due to rapid degradation by the ubiquitous proteolytic enzyme dipeptidyl peptidase-4 (DPP-4). Moreover, to have an effect, GLP-1 has to be given by injection, which is neither convenient nor patient friendly. To enhance its therapeutic efficacy and as well to develop an oral form of GLP-1, we have previously produced GLP-1 as a fusion protein with human serum transferrin (GLP-1-Tf) in transgenic tomato plants, and demonstrated that the plant-derived fusion protein retained its ability to stimulate insulin secretion from pancreatic beta cells *in vitro*. The aim of this study was to demonstrate the receptor binding of plant-derived GLP-1-Tf fusion protein to a GLP-1 receptor (GLP-1R) using Chinese Hamster Ovarian (CHO) cells transfected with human GLP-1R. Several binding assays were conducted and protein-receptor binding was not observed. This is most likely due to the fact that GLP-1-Tf does not bind to GLP-1R and instead mediates its effect through other receptor mediated mechanisms.

Abstract: 048

A THRESHOLD OF HSV-1 OR HCMV VIRAL PARTICLES MUST BE SURPASSED BEFORE INDUCING AN INTERFERON INDEPENDENT ANTI-VIRAL RESPONSE IN HEL CELLS.

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Over 90% of the world population is infected with a form of herpes virus. This family of viruses are well known for their latency throughout the host's lifetime. During times of physical and emotional stress the virus can be re-activated, leading to viral shedding and physical presentation.

Upon viral infection of cells, the host produces an innate and adaptive immune response. The current project is focused on the innate response, more specifically the interferon (IFN) independent anti-viral response. This response is triggered by viral perturbation of the cell membrane, leading to the production of IFN stimulated genes (ISG's) that shut down cell machinery preventing viral replication. Previous studies from the Mossman lab have found a 1000 fold variation in the multiplicity of infection (MOI) of two very similar viruses, HSV-1 and HCMV to induce an IFN independent anti-viral response. By taking into account that viral preparations consist of many particles that are inactive and incapable of replication, thus not accounted for in measurements such as MOI, the large variation can partially be explained. The current project aims to uncover the number of particles required of HSV-1 and HCMV to induce an IFN independent anti-viral response.

We found a similar number of viral particles of HSV-1 and HCMV were required to induce an IFN independent anti-viral response. Concluding a threshold of HCMV or HSV-1 particles must be surpassed before HEL cells mount an IFN independent anti-viral response.

Abstract: 049

EVIDENCE OF ANTHROPOGENIC EUTROPHICATION IN LONG LAKE

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Eutrophication is the input of excess nitrogen and phosphorous into a water system, which stimulates the growth of toxic cyanobacteria, which can jeopardise water quality and restrict use. Long Lake in Sudbury, Ontario has had frequent cyanobacterial blooms over the past decade despite being characterized as oligotrophic. The blooms affect the water supply of over 1000 shoreline residents. As the blooms are localized in particular bays, they may be the result of septic beds that are either over capacity or in poor repair. As a method to indirectly evaluate nutrient conditions in individual bays, epilithic algal samples were collected from the shorelines of several bays within Long Lake that have recurring blooms and several that don't. In order to test our ability to detect known blooms, we also sampled the shorelines of eutrophic lakes. Shorelines were classified as developed or undeveloped based on the presence of infrastructure. If eutrophication from residential wastewater treatment is responsible for algal blooms then eutrophic conditions should be associated with developed shorelines. Several metrics that indicate eutrophication were assessed. Total organic carbon (TOC) suggests that the highest algal growth is found within the residential shorelines of Long Lake. Results will be discussed from the Trophic Diatom Index (TDI), a standard method for evaluating productivity. If negative effects on lake habitats can be traced to nutrient loads from specific locations, this can be used to enforce management of household wastewater treatment.

Abstract: 050

LATITUDINAL GRADIENT OF ACANTHOSCELIDIUS ACEPHALUS AND SPODOPTERA EXIGUA PERFORMANCE ON OENOTHERA BIENNIS

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The Latitudinal Herbivory Defense Hypothesis (LHDH) proposes that plants at lower latitudes receive greater herbivory and thus exhibit higher levels of defense, yet recent studies report mixed support for this hypothesis. Many past studies of latitudinal variation in herbivory and defense have overlooked the potential importance of herbivore specialization and the contribution of clines in constitutive and induced resistance. Here we investigate whether there are latitudinal based clines in constitutive and induced resistance against generalist and specialist herbivores of *Oenothera biennis*. We conducted no-choice assays on 6 populations of *O. biennis* representing the North American latitudinal range with a specialist weevil, *Acanthoscelidius acephalus*, and a generalist caterpillar, *Spodoptera exigua*. Contrary to the LHDH, we observed no latitudinal pattern in constitutive or induced resistance to *A. acephalus* herbivory. By contrast, *S. exigua* herbivory and weight were significantly higher in the North than in the Centre and South, indicating a genetically based cline in resistance to this generalist caterpillar. We detected induced resistance to *S. exigua* herbivory and mortality, but this effect did not vary across populations. Currently, we are investigating whether varying phenolic content in *O. biennis* leaves can explain the observed latitudinal patterns of herbivore performance. Overall our results show latitudinal patterns in constitutive and induced resistance affect generalist and specialist herbivore performance differentially.

Abstract: 051**REGULATION OF PPAR TRANSCRIPTION FACTORS IN *RANA SYLVATICA* SKELETAL MUSCLE AND LIVER DURING FREEZING AND ANOXIA**

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When temperatures plummet below 0°C the wood frog (*Rana sylvatica*) freezes up to 65% of its body water in extracellular ice masses, showing no measurable brain activity, no breathing, and a flat-lined heart. To survive this extreme stress the freeze tolerant frogs retreat into a state of suspended animation characterized by global suppression of metabolic functions and reprioritization of energy usage to essential survival processes including selective gene expression. Peroxisome proliferator-activated receptors (PPAR) are transcription factors that regulate essential processes of carbohydrate and lipid metabolism, adipogenesis, and inflammation. PPARs are regulated through their interactions with their co-activator, peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1 α), which in turn is regulated through its interactions with myocyte-specific enhancer factor 2 (MEF2), an important transcription factor involved in muscle remodelling. Western blotting was used to measure relative levels of proteins in the PPAR pathway in frog skeletal muscle and liver. It was determined that these were differentially regulated when comparing 5°C controls with frozen (24 h at -3°C) and anoxic (24 h under N₂ gas at 5°C mimicking the oxygen deprivation component of freezing) conditions. Two PPAR isoforms, PPAR α and PPAR β , were generally significantly downregulated in response to freezing and anoxia. Moreover, skeletal muscle exhibited stronger suppression of most protein targets analyzed whereas liver showed upregulation of some targets. Understanding the role that PPARs play in the wood frog's ability to survive freezing and anoxia unscathed could contribute to developing novel approaches to medical problems such as organ cryopreservation and novel therapeutics for PPAR metabolic diseases.

Abstract: 052**TURN DOWN GENE EXPRESSION FOR WAT: ANTI-APOPTOTIC SIGNALING PROTECTS WHITE ADIPOSE TISSUE IN HIBERNATING GROUND SQUIRRELS**

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During hibernation, the metabolic rate of 13-lined ground squirrels (*Ictidomys tridecemlineatus*) can drop to just 1-5% of normal resting rate at 37°C; body temperature can plummet as low as 5°C and heart/breathing rates fall to low values. Energy saved by using hibernation allows squirrels to survive the whole winter without eating, living off lipid reserves in white adipose tissue (WAT). During hibernation some energy must be used to defend against conditions that would normally be damaging for mammals (e.g. low temperatures, hypoxia, ischemia) and that can trigger cell death via apoptosis. Life or death for cells is largely dependent on the relative amounts and activities of the pro- and anti-apoptotic Bcl-2 family of proteins that respond to stress signals. The objective of this study was to determine how anti-apoptotic proteins respond to protect WAT cells during hibernation. Relative levels of several pro-survival anti-apoptotic proteins were quantified in WAT via immunoblotting over six time points of the torpor-arousal cycle. These included anti-apoptotic Bcl-2 family members functioning at the mitochondrial level: Bcl-2, p-Bcl-2 (S70), p-Bcl-2 (T56), Bcl-xL, p-Bcl-xL (S62), Mcl-1, p-Mcl-1 (S159) as well as caspase inhibitors downstream of the mitochondria: x-IAP and c-IAP. Bcl-xL and c-IAP exhibited no changes over torpor-arousal, but significant increases in Mcl-1 and x-IAP proteins occurred during torpor compared to euthermic levels, and significant decreases in p-Mcl-1 (S159), Bcl-2, p-Bcl-2 (T56), p-Bcl-2 (S70), and p-Bcl-xL (S62) levels were noted. Generally, these data suggest an overall increase in white adipose tissue survival efforts during hibernation.

Abstract: 053**ROLE OF MICRO-RNA IN CARDIAC AND SKELETAL MUSCLES OF THE FREEZE-TOLERANT WOOD FROG, *RANA SYLVATICA***

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Wood frogs, *Rana sylvatica* have evolved various biochemical adaptations to cope with the severe environmental stress of freezing during winter months. Frogs are able to endure conversion of ~65 % of total body water into extracellular ice using mechanisms like altered expression of many genes. MicroRNAs are short RNA sequences that are ~22 nucleotides in length that affect the abundance of mRNA transcripts in virtually every cellular pathway. This study looks at elucidating the stress-induced microRNAs response in the freeze-tolerant wood frog. MicroRNAs of interest were measured using a modified protocol which employs polyadenylation prior to conversion to cDNA by reverse transcriptase and amplification by qPCR. Relative levels of 55 microRNAs were assessed in the frog's cardiac and skeletal muscles comparing control, frozen and thawed conditions. MicroRNAs showed tissue specific expression patterns. Twenty-two microRNAs were identified which decrease in the cardiac muscles during thawing conditions while 21 microRNAs increased during freezing stress in the skeletal muscle. These findings indicate that genes were activated in the heart tissue and deactivated in the skeletal muscle in response to freezing. Bioinformatics analysis using the DIANA miRPath program (v.2.0) predicted that the differentially expressed microRNAs in this study regulate cytoskeletal, cancer, and mitogen-activated protein kinase (MAPK) signaling pathways in heart tissue. Furthermore, cytoskeletal, PI3K-AKT, and MAPK signaling pathways were predicted to be regulated in skeletal muscle by microRNAs during freezing. These results add to a growing number of studies which suggest that microRNAs play important regulatory roles in animals coping with environmental stress.

Abstract: 054**DIFFERENTIAL SCANNING FLUORIMETRY TO DETECT CHANGES IN PROTEIN STABILITY UNDER SOLVENT ALTERATIONS**

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Differential scanning fluorimetry (DSF) is a sensitive technique used to detect changes in protein stability as the result of thermal denaturation. Data are gathered as changes in fluorescence as temperature rises and an important parameter, the melting temperature (T_m), of the protein is calculated; this denotes the temperature at which the protein is half-unfolded. This study used shifts in T_m to gain information on the effects of solutes on enzyme stability. The enzymes at the vanguard of this investigation were enolase from rabbit muscle tissue, hexokinase (HK) from *Saccharomyces cerevisiae*, malate dehydrogenase (MDH) from porcine heart, sorbitol dehydrogenase (SDH) from sheep liver and glucose-6-phosphate dehydrogenase (G6PDH) from *Leuconostoc mesenteroides*. Results showed that glucose increased the T_m , indicating that the sugar increased the stability of the protein against thermal denaturation while urea lowered the T_m indicating that it destabilized enzyme conformation. The effects of ionic strength on protein stability were also tested by varying NaCl and KCl concentrations in the solvent. The effects of enzyme substrates on T_m values were also analyzed showing both increasing and decreasing thermal shifts depending on the specific protein and substrate combination. Finally, DSF analyses were also performed in the presence of nanoparticles. Nanoparticles have some unusual properties that remain largely unknown in terms of affecting biological systems. This study determined that nanoparticles do have a slight influence on the stability of proteins, and this may have implications for their medical uses.

Abstract: 055

DEVELOPING A LINK BETWEEN ALTERNATIVE POLYADENYLATION & STRESS GRANULE ASSEMBLY IN *DROSOPHILA MELANOGASTER*.

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Alternative Polyadenylation (APA) is an RNA-processing phenomenon that generates diversity in transcript length through the preferential use of different polyadenylation sites within the same gene. While reported to be salient in neural tissue across species, deep-sequencing analysis of *Drosophila* cell lines revealed that APA-mediated 3' UTR extensions of several genes encoding RNA-binding proteins (RBPs) are also prevalent in epithelial cells. As extension of the 3' UTR exposes a greater number of cis-regulatory motifs for RBPs to bind to, our primary objective was to identify whether APA-extended isoforms of the same gene localize differentially within a cell. Using a combination of cell fractionation and qPCR on the *Drosophila* epithelial cell line, D17, we show that APA-extended isoforms of the genes *elav*, *rin*, *imp*, and *shep* are preferentially localized in the insoluble fraction. Double-stranded RNA-mediated knockdown against SHEP and PABP2 suggest that these RBPs are involved in the APA of *rin*, a putative marker of cytoplasmic mRNP granules that form under conditions of cellular stress. Stress induction by arsenite reduces the relative extended-to-total isoform abundance of *rin* transcripts and also interferes with the constitutive post-translational modification of the Rin protein, a process that might be involved in the assembly of stress granules. Our data provide a working basis for a model that can be extended *in vivo* and *ex vivo* to better elucidate a link between APA and stress granule assembly, processes that are both implicated in cancer and neuropathologies when disrupted.

Abstract: 056

WHY ARE UNGULATES IMPORTANT FOR THE WOLVERINE? FROM THE PERSPECTIVE OF THE YUKON TERRITORY

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The wolverine (*Gulo gulo*) is known as an opportunistic generalist predator that relies on carrion during the winter season for its dietary needs. Carrion in the form of ungulates has been documented with the highest frequency in the diet in Scandinavian and Canadian populations. In this study, we looked into the importance of ungulates for the wolverine's diet during the winter season in the Yukon Territory. The study included 617 specimens harvested from two ecozones over 8 consecutive years. As previous studies also documented a high frequency of snowshoe hare (*Lepus americanus*) in the diet, our first hypothesis was that there would be a negative relationship between the snowshoe hare and ungulates because of a dietary shift between two primary food resources, but we failed to detect any significant relationship between the two groups over time, although there was an apparent shift on some years. The ungulates are abundant and diverse in the Yukon, therefore we examined if any particular species showed a consistently higher frequency in the diet. The woodland caribou (*Rangifer tarandus caribou*) demonstrated the highest frequency in both ecozones. Future studies in the Yukon should document further the availability of caribou within ecozones, as well as the population levels in its predators.

Abstract: 057

ANTIBODIES AGAINST OXIDIZED PHOSPHOLIPIDS ENHANCE PULMONARY MACROPHAGE UPTAKE OF LIPIDS AND CIGARETTE SMOKE PARTICLES IN MICE

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Background The negative impact of cigarette smoke on health is well established; however, the processes underlying this effect are not completely understood. Recently, we generated data suggesting that smoking leads to disruption of pulmonary lipid homeostasis, initiating the recruitment and activation of macrophages and neutrophils. While pulmonary macrophages are instrumental in maintaining pulmonary lipid homeostasis, the adaptive immune system has also been shown to respond to lipid deregulation and damage. Therefore, we decided to investigate the humoral response against damaged lipids, specifically oxidized phospholipids, and their role in the pulmonary immune response to cigarette smoke.

Methods/Results Using a mouse model of cigarette smoke exposure, we found higher titers of pulmonary IgM and IgA antibodies against oxidized phospholipids (anti-OxPL) after 24 weeks of smoke exposure. To better understand the role of anti-OxPL antibodies, we delivered anti-OxPL IgM intranasally to C57BL/6 mice exposed to cigarette smoke for 4 days. We found that administration of the anti-OxPL antibody decreased the expression of some inflammatory cytokines and increased uptake of lipids by pulmonary macrophages. Furthermore, we observed that anti-OxPL antibodies increased the uptake of smoke particles by pulmonary macrophages when delivered during smoke exposure and following smoking cessation.

Conclusions/Significance Overall, the humoral response against OxPL, in the context of smoke exposure, appears to be beneficial and may help the innate immune system deal with damaged lipids and smoke particles. Further investigations are required to better understand the long-term effects of anti-OxPL antibodies as well as its therapeutic potential in helping restore pulmonary lipid homeostasis.

Abstract: 058

THE EFFECTS OF URBANIZATION ON THE DENTAL HEALTH OF THE NORTH AMERICAN RACCOON (PROCYON LOTOR)

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The North American Raccoon (*Procyon lotor*) is an exemplary opportunist that has been able to thrive in many types of habitats and landscapes across North America. Since the start of the twentieth century raccoons have become one of the most common species in North American cities; a part of this success is due to their ability to exploit anthropogenic resources. We hypothesized that raccoons feed predominately on human refuse (garbage), and that this food resource is of poor consistency, and has high carbohydrate and increased bacterial content. We predicted that this diet would cause poorer dental health in urban populations of raccoons than raccoons from rural/natural areas. To test this we compared the dental health of urban raccoons from Toronto, Ontario to rural raccoons trapped south of Lake Simcoe around Georgina, Ontario. Dental health was assessed using a calculi index and by measuring tooth wear, bone loss and bone changes, and presence of caries. Our results will provide insights into the costs of urban habitats for wildlife, particularly as a result of differences in diet.

Abstract: 059

25 YEARS OF RECOVERY OF FISH COMMUNITIES IN SUDBURY FOLLOWING EMISSION REDUCTIONS AT LOCAL SMELTERS.

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Major reductions in acid and metal emissions have been achieved from the smelters in Sudbury, Ontario, an area where air pollution has resulted in severe damage of fish and other aquatic biota. Substantial water quality improvements, in terms of increasing pH and declining metal concentrations, have accompanied the industrial clean up, but it appears that the recovery of fish and other aquatic communities is occurring at a much slower pace. The recovery of fish communities in Sudbury lakes over a 25 year period was assessed with regard to changes in; species richness, diversity, and fish biomass. A qualitative survey was conducted in 1990/1991. A quantitative fish community survey was conducted across 9 study lakes in Sudbury, and 9 reference lakes near Dorset, Ontario, in 2014, following the Nordic Index Netting Protocol. Results indicated that the Sudbury lakes have increased in species richness and diversity although they are still not comparable to the reference lakes in Dorset. However fish biomass was similar in Sudbury and Dorset. Catch per unit effort (CPUE) values were significantly higher in Sudbury recovery lakes, likely due to the lakes being largely dominated by acid tolerant small bodied fish, whereas Dorset reference lakes have greater numbers of large bodied acid sensitive fish species. Sudbury lakes appear to be progressing well towards recovery although they have not recovered fully.

Abstract: 060

THE SEX-RATIO OF JUVENILE FRESHWATER TURTLES KILLED ON ROADS IN ONTARIO.

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The magnitude of the effect of roads on populations will depend on the sex ratio of road mortality. Sex biases in juvenile dispersal is largely unknown in freshwater turtles. To determine juvenile dispersal and mortality risk for Snapping Turtles (*Chelydra serpentina*) and Painted Turtles (*Chrysemys picta*), we tested the hypothesis that the sex ratio of juvenile turtles killed on roads in Ontario is skewed because juvenile dispersal is sex biased. Sex ratios were obtained from dead juvenile turtles collected from roads in south and central Ontario, and sex was determined through identification of the gonads in dissected turtles. In total, 19 male and 19 female Snapping Turtles were identified, and 9 male and 2 female painted turtles were identified. There was no bias in the sex ratio for Snapping Turtles, indicating the dispersal is not sex biased. In Painted Turtles there is a trend toward a male biased sex ratio, possibly indicating male biased dispersal. Male biased dispersal in Painted Turtles could be due to male competition for mates to avoid competing with kin. These results suggest that at least in young Painted Turtles, mortality risk may be higher for males than females. Understanding mortality risks and dispersal patterns in juvenile turtles is important for determining population viability, especially in species at risk such as the Snapping Turtle, and for making informed mitigation decisions.

Abstract: 061

CROSS-HYBRIDIZATION OF NAKED MOLE RAT GENOMIC DNA TO PROBE SEQUENCES ON THE MOUSE DIVERSITY GENOTYPING ARRAY

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The Mouse Diversity Genotyping Array, targeting 493,290 SNP loci and 421,905 invariant genomic regions in the mouse, has enhanced investigations of somatic mosaicism and copy number variation. A comparative genomics study using the CanineHD BeadChip identified conserved SNPs between the seal and dog, and demonstrates the application of tools designed for model organisms to nonmodel organisms (Hoffman *et al.*, 2013, PLoS ONE 8: e68365). Although naked mole rats (*Heterocephalus glaber*) and mice (*Mus musculus*) diverged approximately 73 million years ago, a bioinformatics comparison of Mouse Diversity Genotyping Array probe sequences to the two published naked mole rat genomes identified significant sequence conservation. In the male naked mole rat genome, 709 SNPs and 45,832 IGPs are predicted to be conserved whereas in the female genome, 802 SNPs and 47,480 IGPs are predicted. Here, conserved sequences will be identified empirically by a cross-hybridization between naked mole rat genomic DNA and Mouse Diversity Genotyping Array probe sequences. Using tail samples from four naked mole rats (three litter mates and one of a different colony) and five C57Bl/6J inbred mice (a family project design: mother, father and three sons), genomic DNA was extracted and quantified. Hybridization of genomic DNA is in progress. Mouse array data will be analyzed using publically available bioinformatics software. A bioinformatics approach to identify cross-hybridization targets will be developed. Genic conservation will be summarized and target hybridization across naked mole rat samples will be compared. Identification of conserved sequences will provide insight into population structure, evolution and functionally important genes.

Abstract: 062

THE FUNCTIONAL ROLE OF THE C194-C200 DISULPHIDE BOND IN THE CARBOXYSOMAL γ -CARBONIC ANHYDRASE, CcmM

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Carbonic anhydrase (CA) is a ubiquitous enzyme that catalyzes the reversible inter-conversion between HCO_3^- and CO_2 . There are three distinct evolutionary lineages of CA found in living organisms, designated α , β and γ , found mainly in animals, plants, eubacteria and Archaea, respectively. In photoautotrophic cyanobacteria, CA is essential for photosynthetic CO_2 fixation. Unlike in higher plants, this enzyme is confined to a structure known as a carboxysome, which also houses the cellular complement of the Calvin cycle enzyme Rubisco. The carboxysomal HCO_3^- dehydration complex of the diazotrophic cyanobacterium *Nostoc* PCC 7120 contains a unique multi-domained γ -CA, CcmM that binds to Rubisco while at the same time delivering CO_2 to Rubisco's active site. CcmM also has a unique disulphide bond not found in other CAs that is apparently involved in the regulation of CA activity in the carboxysome. To understand the role of this disulfide bond, we have created a number of recombinant CcmM proteins with specific substitutions and/or deletions and evaluated their enzymatic activity. Wild-type CcmM209 Cys194-Cys200 was a moderately active CA with a k_{cat} of $2 \times 10^4 \text{ s}^{-1}$ and a k_{cat}/K_m of 4.1×10^6 . Removal of the C-terminal 8 amino acids did not have any appreciable effect, but CcmM196 and CcmM193 displayed no activity. The CcmM209 C194L had a k_{cat} of 8.59×10^3 and C194L-C200L displayed a k_{cat} of 1.33×10^4 . Substitutions of one or both cysteines with serine resulted in k_{cat} around 4.8×10^3 . The data indicated that the C194-C200 disulfide bond is required for maximum activity. Biological reducing agent GSH to CcmM209 results in a k_{cat} of 8.49×10^3 , while using a redox reagent like DTT resulted in no activity. We conclude that unlike most other enzymes is activated under oxidizing conditions and inhibited under reducing conditions.

Abstract: 063

MONITORING THE IMPACT OF URBAN ENVIRONMENTS ON MERCURY CONTAMINATION OF LOCAL AQUATIC ECOSYSTEMS USING *ORCONECTES PROPINQUUS* AS A BIOINDICATOR SPECIES.

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Mercury is a hazardous heavy metal that has considerable effects on humans, the environment, and local ecosystems. Its ability to travel long distances, as a gas, through the atmosphere makes it a global concern. It is important to know how urban environments affect local ecosystems. Some research has shown that urban environments have a higher concentration of mercury than rural environments. The effect on urban rivers however, is relatively understudied. In this study, I assess mercury contamination in an aquatic ecosystem upstream and downstream an urban center. I use the crayfish, *Orconectes propinquus* as a bioindicator species. It is abundant, easy to catch, area-bound, and bioaccumulates methylmercury, making it an accurate indicator of mercury conditions in its habitat. This study was performed in the Thames River in the city of London, Ontario, as it has no specific point source of mercury contamination. Crayfish samples were captured in two locations upstream and one location downstream of the city. They were then measured, dissected and freeze dried, to determine dry weight. Dry samples were then run through the Direct Mercury Analyzer to determine total mercury content. Statistical analysis and results are to be discussed. This study examines whether an urban environment can have an impact on mercury contamination in local aquatic ecosystems, using the crayfish, *O. propinquus* as a bioindicator species.

Abstract: 064

DETECTION OF SOMATIC MOSAICISM ARISING IN LUNG AND KIDNEY TISSUES OF C57BL/6J MICE USING THE MOUSE DIVERSITY GENOTYPING ARRAY.

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Mutation clusters are a recently discovered mutation signature in cancerous tissue that are problematic in cancer diagnosis, treatment and prognosis, and are poorly understood. The close proximity of multiple mutations in clusters suggests that these mutational events are not random or independent, but rather chronocoordinate mutational events contributing to somatic mosaicism within a tissue. Evidence has shown somatic mosaicism is a reoccurring event, not only in cancerous tissue, but also in normal tissue development. The Mouse Diversity Genotyping Array (MDGA) is a high-resolution single nucleotide polymorphism (SNP) array used to detect SNP's and CNV's across the mouse genome with a combination of ~4.8 million SNP probes and invariant genomic probes (IGPs) covering 915,195 unique loci. In this study, the MDGA is used to detect *de novo* SNP's and CNV's in the offspring of inbred C57BL/6J mice using parental mice as a reference to filter-out inherited mutations. It is hypothesized that somatic mosaicism occurs during development, thus creating distinct mutation profiles between lung and kidney tissues in the same mouse. Mutations in healthy tissues will not cluster, but rather the mutational events that do arise will likely contribute to overall function of the tissue. Preliminary results show high-quality genomic DNA has been extracted from both tissues, which will ensure accuracy of the results obtained from the array. This experiment will be the first to provide inter-tissue comparisons for genome wide-mutation profiles of kidney and lung tissues, and will bring us closer to understanding the mutational profiles that lead to cancer development.

Abstract: 065

VALIDATION OF NON-INVASIVE TECHNIQUES FOR ESTIMATING: EGG SIZE, OVARY SIZE, AND MUSCLE FAT CONTENT IN SEXUALLY-MATURE FISH.

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Female salmonids must balance a trade-off between energy invested into egg production and energy stored for fueling metabolism until death. This trade-off has never been tested within individuals because of a sampling limitation. At the individual level, non-invasive techniques must be used to measure energy stores and gonadal investment, because data is needed on reproductive lifespan. Ultrasound and fat meters have been used as non-invasive techniques for estimating fecundity and muscle fat content, respectively, in salmonids. However, these techniques have not been applied together in sexually-mature fish with enlarged ovaries and low fat content. We tested validity of using ultrasound image analysis to estimate egg diameter and ovary volume in brook trout, and the validity of Distell Fish Fatmeter for estimating muscle fat content in kokanee and lake trout. Estimates of egg diameter and ovary volume were compared to true values obtained from lethal sampling. Ultrasound image analysis estimated egg diameter with mean absolute error (E) of 0.095 mm and mean absolute percent error (%E) of 2%. Image analysis estimated ovary volume with much poorer accuracy (E = 25.1 cm³; %E = 30%). We compared estimates of percent fat to true values obtained from fat extraction of muscle samples. Distell Fatmeter estimated percent fat with low absolute error (~1.5% fat), but with variable absolute percent error (6% for kokanee; 24% for lake trout). With low fat in sexually-mature fish (2.1% fat in kokanee; 3.7% in lake trout), Distell Fatmeter does not have sufficient sensitivity for assessing muscle fat content.

Abstract: 066

METABOLIC PROFILES OF CARDIAC AND SKELETAL MUSCLE WITH COMPLEX I DYSFUNCTION IN HARLEQUIN MICE COMPARED TO WILD-TYPE MICE

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Respiratory chain complex I (RCCI) dysfunctions account for up to 30% of all mitochondrial disorders and approximately 50% of patients have no molecular diagnosis. Due to inter- and intra-patient variability and a lack of therapeutic approaches, disease origins and mechanisms require detailed study. Down-regulation of the *Apoptosis-inducing factor* (*Aif*) gene, in the *harlequin* (*hq*) mouse, is a useful model of RCCI dysfunction. Previously, *hq* cardiac tissue has not shown complex I deficiency, however, skeletal muscle tissue has demonstrated a 30% decrease in complex I activity and decreased subunit expression. As a result, it is proposed *hq* mice undergo a metabolic shift and use glycolytic metabolism for energy production instead of oxidative phosphorylation. This shift may be tissue-specific, which could explain the different pathological severities seen in different tissues and it may also underlie the *hq* phenotypes such as neurodegeneration. The goal of this project is to examine the associations with elevated feeding, low body mass, *Aif* expression and primary metabolite profiles in *hq* compared to wild-type mice. I hypothesize there will be tissue-specific quantitative differences in *Aif* expression associated with different metabolite profiles between *hq* and wild-type mice in cardiac and skeletal muscle at 2 and 6 months of age. RT-qPCR and liquid chromatography-mass spectrometry is used to assess gene expression and primary metabolite profiles, respectively. A feeding study provided evidence consistent with metabolic differences between *hq* and wild-type mice. Overall, the analysis of metabolic profiles will provide insight to the metabolic mechanisms occurring as a result of RCCI dysfunction.

Abstract: 067

EFFECTS OF TIME POST-EGG ACTIVATION ON FERTILIZATION SUCCESS IN CHINOOK SALMON

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The teleost egg is surrounded by an impenetrable membrane that contains a single narrow channel or ‘micropyle’ that allows a sperm cell to gain access and fertilize the egg. Once a sperm cell binds with the egg, the micropyle closes to prevent polyspermy (fertilization by multiple sperm cells) which could kill the egg. In oviparous fish species such as Chinook salmon (*Oncorhynchus tshawytscha*), river water causes physiological changes to the egg resulting in the eventual closure of the micropyle. Sperm cells surviving past this closure will not successfully fertilize the egg, suggesting that sperm longevity and time to micropyle closure are tightly linked. Although there is little known about Chinook salmon micropyle closure, it has been documented that their sperm cells remain active for about fifteen seconds post-activation. This study aims to investigate fertilization success of eggs activated in river water for varying periods of time (0,3,5,7 or 9 seconds) before sperm addition. We performed ten crosses (two females with five males) with two replicates per cross. Eggs were incubated until the eyed stage. All dead eggs were examined for fertilization success using an acetic acid assay. Preliminary data suggests no significant difference in fertilization success, suggesting that the micropyle closes sometime after nine seconds, but further analysis is required. Additionally, investigation of chinook eggs using scanning electron microscopy will be performed in order to verify micropyle physiology in this species.

Abstract: 068

ROLE OF THE RENIN-ANGIOTENSIN SYSTEM IN A FETAL PROGRAMMING MODEL OF HYPERTENSION

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The renal renin-angiotensin system (RAS) is an important physiological mechanism involved in blood pressure control, and alterations in gene expression of the components in this system have been shown in both a glucocorticoid-induced, fetal programming model (FP) and a genetic model of hypertension (spontaneous hypertensive rat – SHR). In the SHR, renal expression of Ace2 is decreased while Ace1, AT1R and (pro)renin receptor are increased. Sexual differences also exist for RAS components such as angiotensinogen (AGT). However, the exact RAS mechanism involved in FP model is still unclear. For the genetic study, kidneys were collected from male WKY and SHR adult rats. For the FP model, kidneys were collected from adult rats of each sex that were prenatally exposed to either the synthetic glucocorticoid, dexamethasone, or saline and from naïve control groups. Coronal sections were dissected through the hilus and total RNA extracted. Conventional RT-PCR analysis was performed for AGT, renin, AT1Ra, AT1Rb, Ace1, Ace2, Mas1 receptor, and (pro)renin receptor genes. Gene expression was quantified using densitometry and normalized to the 28s gene. Preliminary data show increased expression of all genes except renin in the SHR compared to WKY. FP rats showed no change in gene expression across all treatment groups but showed a characteristically lower expression of AGT, AT1Rb, and Ace2 in females. To conclude, the genetic model of hypertension shows upregulation of specific genes which are not observed in the FP model. This study highlights contrasting RAS-related mechanisms contributing to the development of hypertension between the two models.

Abstract: 069

EXAMINING THE FINE SCALE GENETIC DIVERSITY IN A GLACIAL RELICT BUTTERFLY (*LYCEANA EPIXANTHE*).

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Ecological and anthropogenic stressors can reduce the survival of a species and entire ecosystems. One of the most prevalent factors impacting populations' survival is habitat fragmentation. The isolation of one population from another through natural and artificial fragmentation processes inhibits gene flow, which can in turn lower the genetic diversity of populations. As well, isolated populations are usually smaller in size than the previously connected population. A smaller population is more susceptible to the effects of genetic drift, meaning allelic diversity is reduced and recessive alleles that have deleterious effects are more likely to manifest in homozygous individuals. These deleterious alleles impact the overall survival of a population and have the potential to drive a population to extinction. This study seeks to examine the level of genetic diversity in a population of geographically isolated glacial relict butterflies, *Lyceana epixanthae*, or the bog copper butterfly. Microsatellite fragment lengths were used to assess the effective population size (N_e) and mark-release-recapture studies were used to estimate the census population size (N). The ratio of these parameters (N_e/N) is important in understanding the relative level of genetic diversity in this population, which can highlight the susceptibility of this population to stochastic events. Results of this study will be forthcoming.

Abstract: 070

ARABIDOPSIS THALIANA* PROMOTER ANALYSIS OF AROGENATE DEHYDRATASES IN *NICOTIANA BENTHAMIANA

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Arogenate dehydratase (ADT) is an enzyme that catalyzes the final step in phenylalanine biosynthesis. The aromatic amino acid phenylalanine is an essential building block of proteins in plants and it is a precursor for aromatic secondary metabolites. ADTs catalyze decarboxylation and dehydration reactions to convert arogenate into phenylalanine. In *Arabidopsis thaliana*, there is a family of six ADT genes. These genes are differentially expressed in tissues. I hypothesized that the promoters of these genes have elements that enable transcription to be induced by different internal signals or under different environmental conditions. I proposed to clone the six ADT promoter sequences using PCR amplification. Gateway Technology was used to recombine PCR products into entry vectors. A Gateway recombination strategy was then used to move the promoter sequences into a destination vector. Successful transformants were selected with spectinomycin and the plasmids isolated and transformed into *Agrobacterium tumefaciens*. *Agrobacterium* containing the vector was then injected into *Nicotiana benthamiana* leaves. Green fluorescent protein (GFP) and β -glucuronidase (GUS) were used as reporters to visualize the ADT promoter properties in *N. benthamiana*.

Abstract: 071**CHARACTERIZATION OF NOVEL γ -LACTAM ANTIBACTERIAL COMPOUNDS AGAINST DRUG RESISTANT *E. COLI* AND *S. AUREUS* STRAINS**Alexis Fong^{*1,2}, Alison Douglas², James Knockleby², Viswas Raja Solomon² and Hoyun Lee^{1,2,3}¹Department of Biology, Laurentian University, Sudbury, Ontario, Canada, P3E 2C6²Advanced Medical Research Institute of Canada, Sudbury, Ontario, Canada, P3E 5J1³Medical sciences Division, Northern Ontario School of Medicine, 935 Ramsey Lake Road, Sudbury, Ontario P3E 2C6

According to the World Health Organization, the prevalence of antibacterial resistance is 83.3% in World Health Regions (as listed by WHO). Furthermore, the number of bacterial strains resistant to existing antibiotics increases every year, necessitating the development of a new classes of antibiotics. Toward developing a novel class of antibacterial compounds, we have created a chemical library by a hybrid approach using chloroquine as a basic scaffold. We then screened the chemical library for antibacterial activity. Of the 211 compounds screened, 27 compounds were effective against *Escherichia coli* and *Staphylococcus aureus*. Structural analysis of the 27 active compounds revealed the presence of an isatin group and thus being classified under the γ -lactam class of antibiotics. All of the 27 active compounds mimic the β -lactam-based antibiotics while displaying activity against resistant strains of bacteria. The average minimum inhibitory concentration of the 27 compounds was comparable to the well-established kanamycin and ampicillin as determined by a broth dilution method. All of the 27 compounds were also active against ampicillin-resistant, kanamycin-resistant and NDM-1 resistant *E.coli* strains as well as against methicillin-resistant *S. aureus*. However, none was effective against vancomycin-resistant *Enterococcus faecalis*. Further studies are being carried out to unravel the mechanism of action of the 27 novel compounds.

Abstract: 072**EFFECTS OF SPERM LONGEVITY ON OFFSPRING FITNESS IN CHINOOK SALMON**Sumeet Bhardwaj^{*1}, Jennifer Smith¹, Trevor Pitcher^{1,2}¹Department of Biological Sciences, University of Windsor, Windsor, Ontario, Canada, N9B 3P4²Great Lakes Institute for Environmental Research, University of Windsor, Ontario, Canada, N9B 3P4

Variation in numerous sperm quality metrics of a single male's ejaculate are linked to variation in offspring fitness. It has been shown in Atlantic salmon (*Salmo salar*) that sperm longevity affects offspring fitness in that longer lived sperm produce larger offspring on average but also have a lower fertilization success rate when compared to shorter lived sperm. In our study we examined the effect of sperm longevity on offspring fitness in Chinook salmon, *Oncorhynchus tshawytscha*. Sperm were activated and then used to fertilize eggs either immediately (zero seconds) or at ten seconds post-activation. We performed a total of 28 crosses (two females with seven males) and replicated each cross. Offspring were reared to the alevin stage. We measured yolk sac area, standard length and hatching success in order to assess offspring fitness. We found that sperm longevity had the greatest effect on hatching success; eggs fertilized with sperm ten seconds post-activation had a much lower hatching success than those fertilized immediately after activation. Overall, our results confirm previous data suggesting that longer lived sperm within a single ejaculate can have negative consequences on offspring fitness. Additional studies using a larger number of females are required to fully understand the effects of sperm longevity on offspring fitness.

Abstract: 073**DNA YIELDS AND PCR AMPLIFICATION SUCCESS USING DIFFERENT ANIMAL TISSUES, EXTRACTION PROTOCOLS AND ISSR PRIMERS: A COMPARITIVE ANALYSIS**Jimmy Ilunga^{*}, Frank Mallory

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The specific objectives of the present study were: 1) to compare DNA yields for different animal tissues; 2) to compare traditional (manual) DNA extraction protocols with commercial procedures; and 3) to determine the success of PCR amplification of ISSR loci in degraded animal samples. Liver, stomach, and muscle samples were extracted from coyote (*Canis latrans*), cow (*Bos Taurus*), and long-tailed weasel (*Mustela Frenata*) for this research. Manual protocols for DNA extraction were compared to commercial kit procedure (Qiagen kit). Genomic DNA in different states (intact, apoptotic and degraded) were amplified using a panel of ISSR primers. No DNA was recovered from coyote stomach samples using the manual extraction protocol. DNA concentrations in stomach and liver samples from coyote were 10.31 ng/ μ L and 15.8 ng/ μ L respectively using the Qiagen extraction kit. We found that muscle DNA was more stable than liver DNA for cow sample extracted via manual procedure. In general, the kit extraction procedure yielded more DNA than manual extraction procedure. Intact and apoptotic genomic DNA were successfully amplified by PCR resulting in a similar profile. Artificially degraded DNA showed partial amplification. Thus, the ISSR marker system is suitable for animal population genetics when only limited and/or degraded animal DNA is available.

Abstract: 074

MELANOCORTINS AS INDICATORS OF PHYSIOLOGICAL STRESS RESPONSE IN FISH

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The acute stress response has coevolved with the melanocortin system in vertebrates to regulate homeostatic balance and energy utilization following exposure to an acute stressor. Melanocortins are pleiotropic hormones that display phenotypic modulation in melanin synthesis and regulation in glucocorticoid production. In Atlantic salmon (*Salmo salar*) parr, there exists an association between melanin-based pigmentation and the neuroendocrine stress response that is regulated by the Hypothalamic-Pituitary-Interrenal axis, which ultimately releases cortisol as an adaptive physiological coping strategy in response to stressors in the environment. We examined relationships between stress-induced cortisol release and melanin-based pigmentation in juvenile parr from Sebago (n=22) and LaHave (n=23) populations, and also tested hybrid crosses (n=46) of those two allopatric populations. We non-invasively measured gill-secreted cortisol in water during a 30-minute confinement following a capture stress, and quantified melanin variation among individuals by i) objectively measuring colour reflectance of parr marks, ii) calculating the total area of parr marks, and iii) measuring the parr mark to skin colour contrast. Our preliminary results suggest that melanin concentration in parr marks is positively associated with higher cortisol concentration for only hybrid individuals ($P = 0.01$). Cortisol concentration is also higher in hybrids than in individuals from pure stocks ($P = 0.045$, one-tailed), despite the latter being larger in mass on average ($P = 0.05$). Our results suggest pigment density is correlated to cortisol concentration, indicating that melanin may reflect stress coping regulation at early life-history stages.

Abstract: 075

THE ROLE OF A CHANGING CLIMATE ON THE FREQUENCY AND SEVERITY OF LARCH SAWFLY OUTBREAKS IN HIGH LATITUDE BOREAL FORESTS

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High latitude climate warming is occurring rapidly and this is largely driven by increases in winter time temperatures. Warming winter temperatures enhance overwinter survival of insect pests, thereby increasing their population size as well as geographic distribution. Changes in pest pressure may have marked impacts on forest dynamic processes including tree growth, mortality, and recruitment. To determine frequency and severity of sawfly outbreaks and their correlation with climate change, multiple dendrochronological methods of pest outbreak detection are applied. One method includes studying tree core samples in tamarack for morphological signals of sawfly infestation, such as extreme growth reduction, pale latewood, or missing rings. Another method includes measuring the annual radial growth of rings and cross-dating samples to produce non-host and host chronologies. These chronologies are compared in OUTBREAK to disregard growth anomalies caused by other environmental factors. A third method used in this study includes using the pointer year equation to determine years with dramatic decreases in ring growth from infestation ("pointer years") by comparing each ring width with neighbouring ring widths. By comparing anatomical, pointer year equation, and OUTBREAK results, infestations are confirmed during the same time period in all three methods. Once the correlation between sawfly outbreaks and climate change is analyzed, records of outbreak dynamics can be documented and effects of climate variation can be assessed to provide a better understanding of drivers of decreased boreal productivity.

Abstract: 076

FOOD WEB POSITION & TROPHIC NICHE OF SLIMY SCULPIN RELATIVE TO OTHER SMALL BODIED FORAGE FISH IN TWELVE BOREAL LAKES.

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Freshwater sculpins are an ecologically important component of Boreal lake food webs. Sculpins are the primary group of small bodied fishes composing assemblages in deep offshore benthic habitats in Boreal lakes. Slimy sculpin (*Cottus cognatus*) is the most widespread sculpin species in boreal lakes with a ubiquitous distribution spanning from Alaska to Labrador. Slimy sculpins are believed to be a critical food item to sport fish emanating from a tendency to develop in high densities and overlap in home range with other large bodied piscivores. Despite their potential ecologic importance, our understanding of slimy sculpins remains limited. This is thought to be as a result of overemphasis of sport fish and commercially important species in fisheries management and an ineffectiveness of conventional sampling gear. We assessed the food web positions and niche sizes of slimy sculpin relative to other small-bodied forage fish species using an existing stable isotope data set for 14 boreal Canadian lakes ranging in size from small (~300 ha) to very large (Lake Ontario). An intensive sampling program directed at slimy sculpin was carried out in two of the 14 lakes in which gut contents in addition to stable isotopes were analyzed to assess both habitat use, in terms of depth, and prey items consumed.

Abstract: 077

TESTING CANDIDATE GENES AFFECTING CUTICULAR HYDROCARBON PRODUCTION BETWEEN *DROSOPHILA MELANOGASTER* AND *DROSOPHILA SIMULANS*

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Chemical communication plays a critical role in a wide variety of traits, including mate recognition. The surface chemicals of insects, called cuticular hydrocarbons (CHCs), serve a dual purpose: they limit desiccation and are involved as chemical signals of both intra- and interspecific interactions, including mate recognition. The CHC profiles differ between sexes and species, including between *Drosophila melanogaster* and the closely related *D. simulans*, where CHCs play an important role in mate recognition and female preference. Despite the importance of these CHCs in *Drosophila* behaviour, the genetic basis influencing CHC production is largely unknown. Previous studies identified regions on chromosome 3 containing candidate genes that influence the CHC profile observed in both *D. melanogaster* and *D. simulans*. Using deficiency mapping combined with gas chromatography, I analysed *D. melanogaster* and *D. simulans* females with regional genetic deletions or single gene disruptions in order to refine the previously identified regions and test individual candidate genes. Results will be discussed. Overall, this study will help to identify candidate genes involved in both CHC production and species divergence in *Drosophila*.

Abstract: 078

CHRONIC SOCIAL DEFEAT PARADIGM EFFECTS ON GHSR AND NR3C1 MRNA EXPRESSION IN THE PFC, HIPPOCAMPUS AND VTA OF C57/BL6 MALE MICE

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Ghrelin, a gut-derived peptide hormone involved in the regulation of energy balance also plays a role in the stress response. Receptors for ghrelin (the growth hormone secretagogue receptor or GHSR) are found in regions like the prefrontal cortex (PFC), hippocampus (HIPP) and ventral tagmental area (VTA). These regions are commonly damaged in depressed patients. Recent studies show that ghrelin is secreted during the stress response, has anti-depressant effects and protects areas like the HIPP and PFC. Furthermore, chronic stress impacts the ability of ghrelin to have these protective effects, presumably through a down-regulation of ghrelin receptors (GHSR) in these regions. To determine if this is the case, we examined the expression of glucocorticoid receptor (NR3C1) and GHSR mRNA in the PFC, HIPP, and VTA of control and mice that were placed in a chronic social defeat paradigm for three weeks, and sacrificed at the onset of the light or dark cycles. Our results show that stressed mice had lower levels of GHSR mRNA expression in the PFC ($p < 0.05$) and lower levels of NR3C1 mRNA expression in the HIPP ($p < 0.05$). Both NR3C1 and GHSR mRNA expressions were elevated in the VTA ($p < 0.05$), an area associated with reward seeking behaviors. These data support the idea that chronic stress decreases the ability of ghrelin to protect the HIPP and PFC, promoting the development of depressive like behaviors.

Abstract: 079

EFFECTS OF A GHSR KNOCK OUT MODEL ON MALE RAT SEXUAL BEHAVIOR

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The physiology of reproduction in animals depends on the organism's metabolic state and energy availability. The orexigenic peptide hormone ghrelin plays an integral role in energy metabolism and is known to have an inhibitory effect on reproductive physiology. In contrast, ghrelin and its receptor, the GHSR-1a, is also key for generating motivated behaviors, including sex behaviors. The aim of the current study was to investigate the effects of a GHSR knock out genetic manipulation animal model on appetitive and consummatory male sexual behavior using bi-level chambers. Male wild type (n=4) and GHSR KO rats (n=9) were exposed to sexually receptive females for 30 minutes inside bi-level chambers and were allowed to copulate. This protocol was followed every four days for a total of five sessions. During these sessions we measured appetitive (locomotor activity in anticipation of a female) and consummatory behaviors (latency to mount, intromissions, latency to ejaculate, and number of ejaculations). Results show that GHSR KO rats have a shorter latency to approach and mount a female during the initial exposure to the female, and this effect disappears by the fifth session. Interestingly, male GHSR KO rats displayed more anticipatory locomotor activity in the bi-level chambers than their WT littermates. These data suggest that ghrelin has an overall inhibitory effect on sexual consummatory behavior.

Abstract: 080

MATERNAL EFFECTS ON EMBRYONIC SURVIVAL IN WALLEYE SANDER VITREUS

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There are a number of factors that affect offspring quality which subsequently affect offspring survival. Variability in offspring survival has implications on year-class strength and population dynamics, and is a key area of research in fish culture. Walleye, *Sander vitreus*, is a primary species for both recreational and commercial fisheries. Literature suggests that maternal and ova traits may be the most influential factors accounting for variability in offspring quality and survival amongst female walleye. Understanding which traits are most influential to offspring survival is key to management of wild walleye populations, and can also be used to establish more efficient fish culture practices. I examined embryonic survival with respect to a suite of maternal and ova traits in Lake Nipissing walleye to determine which traits had the greatest effects. Females were analyzed for a host of traits most notably body size and condition, age, and egg size. Eggs were incubated in the laboratory and analyzed for lipid content, fatty acid composition, and mineral content. A subset of maternal and ova traits believed to account for the majority of variation in embryo survival were selected to generate numerous models. Models were ranked using AICc ranking to determine which traits accounted for the majority of variation in embryonic survival. I predicted that maternal age and size would have stronger effects than egg composition. Results to follow.

Abstract: 081

CHARACTERIZATION OF THE CALCIUM CHANNEL, VOLTAGE-DEPENDENT, β SUBUNITS IN *XENOPUS LAEVIS*.

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Voltage-dependent calcium channels (VDCC) are transmembrane ion channels that helps to facilitate influx of calcium (Ca^{+}) into cells in response to an action potential. This influx plays crucial roles in many processes such as muscle contraction, synaptic transmission and hormone secretion in excitable cells including muscle, neuronal and endocrine cells. VDCC β subunits especially are responsible for localization of the channels as well as modulating its electrophysical properties. These channels have been extensively studied in mammals such as mouse (*Mus musculus*), but nothing is known about the VDCC β (*cacnb*) subunits in *Xenopus Laevis*. Clones of the *cacnbs* were made and their expression patterns are characterized at different stages during embryonic development via *in situ* hybridization. The *cacnbs* are found to be expressed in a variety of different tissues including the heart, somite and neural tubes. These expression patterns suggest that there is tissue specific expression of *cacnbs* in *Xenopus laevis*, and their sequence homology would suggest that these channels have been conserved throughout evolution.

Abstract: 082

THE ROLE OF SOCIAL MEDIA, ACADEMIC DISCIPLINE AND ETHNIC BACKGROUND ON STUDENT PERSPECTIVES REGARDING MENTAL HEALTH ISSUES AT UNIVERSITY OF TORONTO MISSISSAUGA

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Although mental health is both neglected and stigmatized, its increasing prevalence among adolescents has created concerns about student academic success in postsecondary institutions. The objective of this research is to examine the impact of students' academic discipline, ethnicity and social media have on student help-seeking behaviour and perspectives on mental health. Surveys composed mainly of previously published questions were used to carry out this study. Students at the University of Toronto Mississauga (UTM) were invited to partake in online or interceptive surveys on a voluntary basis. In regards to academic discipline, it is predicted that students at UTM working towards an HBa or BCom are more likely to experience incomplete mental health and were found to be more apprehensive towards seeking help than those working toward an HBSc. In regards to ethnicity, it is hypothesized that differences regarding mental health perceptions among different student ethnic groups would be detected parallel to previous research that has shown that developing nations may see mental health issues as unimportant. In regards to social media, based on previously conducted studies, we expect that students are more likely to consult social networking sites in order to report depression, stress and access emotional support. Researching these specific aspects can help us gain a better understanding of mental health issues that pose a challenge to student success in postsecondary education and take appropriate steps to address them in our classrooms and institutions.

Abstract: 083

DEVELOPING TECHNIQUES FOR MANIPULATING YEAST COMMUNITIES IN THE GUT OF *DROSOPHILA MELANOGASTER*.

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The guts of animals host a community of microbial symbionts collectively known as the microbiome. These microbial symbionts influence host nutrient uptake, metabolism, and development. While the majority of microbiomic research has focused on bacterial symbionts, we know little of the role of symbiotic yeasts (the mycobiome). *Drosophila* spp. are hosts to a variety of yeast species in the microbiome, and we can easily measure an array of mycobiome-dependent phenotypes in these insects; therefore, *Drosophila* is an ideal model in which to explore the effects of the mycobiome on host physiology. To understand the host-mycobiome interaction, we must first generate germ-free (axenic) flies and flies whose microbiomes contain selected yeasts (gnotobiotic). I have developed techniques for rearing *Drosophila melanogaster* in a sterile environment, and explored techniques to inoculate the guts of larvae, pupae, and adult *D. melanogaster* with yeasts. Eggs that were surface sterilized with 70% ethanol and subsequently transferred to sterile food developed into axenic adult flies. Inoculating sterile food with *Saccharomyces cerevisiae* subsequently introduced the yeast into the gut. Inoculating a complete medium with a homogenized fly hindgut demonstrated the presence of microbes in the gut, but not in fecal samples. The methods I have developed to manipulate the diversity of yeast residing within the gut of *D. melanogaster* provides a foundation for research on elucidating mycobiome-dependent changes in development, cold tolerance, metabolic rates, and energy storage of *D. melanogaster*, in addition to potential coevolution between the mycobiome and the host.

Abstract: 084

THE PHYLOGEOGRAPHY OF *MAMMUT AMERICANUM* IN NORTH AMERICA

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The American mastodon, *Mammot americanum*, first appeared during the Pliocene and spread throughout much of North America. The study of the movement and distribution of mastodons is necessary to obtain a more holistic view of Pleistocene North America due to its role as a keystone species and food source for early human populations. With established ancient DNA techniques, we extracted and enriched for the mitochondrial genomes of 108 American mastodon specimens. We successfully obtained 20 complete and 9 partial mitochondrial genomes from sites throughout Canada and the northern United States. We observed a split between Great Lakes and Alaskan mastodons, which likely occurred during the Sangamonian interglacial, and propose an episodic model may have driven divergences between mastodon populations during repeated glacial periods. Finally, we examined a lineage which diverged approximately 3.5 million years ago, near the paleontologically determined appearance of *M. americanum*, and attempt to explain its position within our phylogenetic analysis.

Abstract: 085

EVALUATING THE COLOMAC REMEDIATION PROJECT: MYCORRHIZAL DENSITY AND INNOCULATION POTENTIAL OF SOILS OBTAINED FROM THE TRUCK LAKE CHANNEL

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The Colomac gold mine, located 225 km north of Yellowknife, was operational from 1994 to 1999. It is currently in the monitoring phase of an extensive re-vegetation effort. While the developing plant communities have been monitored for a three-year period, there have been no assessments of associated belowground fungal symbionts known as mycorrhiza. Associations with mycorrhiza are important for establishing healthy plant communities in disturbed sites, affecting seedling establishment and community composition. The objective of this study was to assess the presence, density and inoculation potential of mycorrhizae in soils from a reconstructed stream channel that had previously been infilled to accommodate mining activities within the Colomac lease boundary. Mycorrhizal spores were extracted from soils using the sucrose-gradient method and spore density quantified. To assess colonization potential *Epilobium angustifolium* and *Phalaris arundinacea* seedlings were grown in soils obtained from the site. Harvested roots were examined for fungal colonization by using light microscopy and quantified using established protocols. Fungal spores were present in all soil samples ranging from 2-136 spores/g of soil. Fungal colonization was observed in plants grown in all soil samples, ranging from 22%-90% for *E. angustifolium* and 6%-55% for *P. arundinacea*. Soils collected from the channel were analyzed by ICP-OES for concentrations of potassium and phosphorus. Among the sample sites, phosphorous and potassium ranged from 0-0.00126 g/g of soil and 0.00054-0.001646 g/g of soil, respectively. Relationships between seedling performance, fungal colonization, soil characteristics, spore density and sample site location will be discussed.

Abstract: 086

THE BIOENERGETIC REGULATION OF CANINE EMBRYONIC STEM CELLS

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Embryonic stem cells (ESCs) can be distinguished by the ontologically distinct pluripotent states, primed and naïve. Only naïve ESCs can form chimeras and differentiate into the cells the embryo and extra-embryonic membranes making them the more desired state. Through the use of transgenesis or molecular inhibition interconversion between pluripotent states can be accomplished. Primed ESCs are primarily glycolytic while naïve ESCs are bivalent having characteristics of both glycolysis and oxidative phosphorylation. Both pluripotent stages are present in mice, however recent human studies and unpublished raw data from our lab suggests that canine ESCs (cESCs) may adopt a naïve pluripotent state with extrinsic leukocyte inhibitor factor exposure and chemical blockade of GSK3 β and MEK signalling. This investigation compares discriminatory metabolic markers specific to glycolysis and oxidative phosphorylation using real-time qPCR and Western Blotting. Primed cESCs exhibit greater abundance of TFAM messenger transcripts relative to naïve cESCs. Conversely, primed cESCs exhibit an induction of proteins associated with elevated lactate dehydrogenase A in comparison with naïve cESCs. The immunoblotting findings indicate that naïve cESCs depend less on glycolysis than primed cESCs for bioenergenesis as seen in a ratio of pyruvate kinase M1 and M2. This is consistent to what has been described in mouse and human ESC lines. These contrasting results are potentially due to the short half-life of mRNA or the cell cycle stage of the cESCs. This investigation addresses the unmet need to more accurately portray human early embryonic physiology and development using canines as a large animal model.

Abstract: 087

ADAPTIVE RESPONSES TO POTASSIUM DEFICIENCY IN TWO NATURAL ACCESSIONS OF *EUTREMA SALSUGINEUM*.

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Potassium is an important macronutrient responsible for osmotic regulation and enzyme activation. We used the halophyte *Eutrema salsugineum*, a crucifer relative of *Arabidopsis thaliana*, to deepen our understanding of potassium homeostasis in a species adapted to highly saline soils.

We compared two natural accessions of *Eutrema*, one from the Yukon Territory, Canada and the other from Shandong Province, China, and their recombinant inbred lines (RILs). Using 0, 0.5 or 2.0 mM potassium media, Yukon *Eutrema* seedlings showed a dose-dependent increase in root and shoot biomass and total root length while Shandong seedlings had the highest root and shoot biomass and total root length with the intermediate 0.5 mM potassium medium. RILs grown on media lacking potassium showed a normal distribution of root and shoot phenotypes intermediate to the parental lines.

Root hairs on primary roots of Yukon seedlings grown without potassium were two-fold longer than those grown with high potassium. This phenotype was not evident for Shandong seedlings. In *Arabidopsis*, both potassium sensing and root hair growth are mediated by hydrogen peroxide (H₂O₂). Yukon and Shandong seedling roots were stained with 3,3'-diaminobenzidine to localize H₂O₂. As expected, H₂O₂ content was highest in the region proximal to the root tip in the zone of elongation with intense staining at root hair tips.

Future work will focus on the genetics underlying the differential growth of the *Eutrema* accessions and their RILs to potassium deficiency. Developing a model for potassium sensing and responses is a fundamental step in developing stress tolerant crops.

Abstract: 088

COMPENSATORY EVOLUTION IN ANTIBIOTIC-RESISTANT *PSEUDOMONAS AERUGINOSA*

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The initial acquisition of antibiotic resistance is often associated with a fitness cost when resistant strains are grown in the absence of the antibiotic. Previous studies have demonstrated that antibiotic-resistant pathogens commonly fix additional mutations that alleviate costs of resistance. This process, referred to as compensatory evolution, allows resistant pathogens to proliferate within clinical settings in the absence of antibiotic selection, as fitness costs incurred by resistance mutations are compensated by secondary-site mutations that increase fitness without compromising resistance. This study used replicate genotypes of antibiotic-resistant *Pseudomonas aeruginosa* (strain PA14) that differed in their starting resistance to antibiotic and fitness cost in the absence of antibiotic. After 50 generations of selection in the absence of the antibiotic, populations significantly decreased in resistance and increased in fitness. Significantly greater rates of fitness increase occurred in starting genotypes that had larger initial fitness costs. Targeted sequencing at the sites of ancestral resistance mutations demonstrated that compensatory mutations occurred outside of these resistance genes. Altogether, results indicate that compensatory evolution can ameliorate the fitness burden associated with antibiotic-resistant microbes. Thus compensatory evolution may be an important process whereby resistant pathogens are able to persist in clinical settings in the absence of the antibiotic treatment, while retaining some level of resistance.

Abstract: 089

NO EFFECT OF PATERNITY REDUCTION ON PARENTAL CARE IN THE PLAINFIN MIDSHIPMAN (*PORICHTHYS NOTATUS*)

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Parental investment theory predicts that parental effort should be adaptively adjusted in response to cues of changing parentage. Thus, when parentage is uncertain due to intrasexual competition, it is expected that caregivers will reduce their parental care. Despite a strong theoretical framework, empirical tests of this theory are generally lacking in fishes, especially in field settings. We conducted a field experiment using the plainfin midshipman (*Porichthys notatus*), an intertidal spawning teleost fish with an extended paternal care period and highly variable paternity due to intense male-male competition. We created artificial nests along a sheltered beach and transplanted offspring between nests to investigate if caregivers were able to perceive a loss of paternity and whether it affected parental care. We tracked parental residency time and offspring survival over a one-month period throughout offspring development. We did not find significantly decreased parental residency time or brood survival following transplants as compared to controls. Our results suggest that midshipman guarding males either lack mechanisms to assess paternity using direct cues from offspring or they may be choosing to not react to direct cues and instead rely on predispositions for decision making regarding parental investment.

Abstract: 090

THE ROLE OF GHRELIN IN A CHRONIC CORTICOSTERONE MODEL OF OBESITY

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Continuous exposure to glucocorticoids is associated with the development of obesity and metabolic syndrome. The mechanisms with which chronic exposure to glucocorticoids mediate weight gain are not well understood. Ghrelin is an orexigenic peptide hormone secreted in the gut that is involved in stress, modulating weight gain and appetite. Circulating ghrelin levels increase with stress, presenting a potential mechanism in which glucocorticoids mediate the development of obesity through the activity of ghrelin. We hypothesized that the obesogenic effects of a chronic corticosterone (CORT) treatment would be regulated by the presence of ghrelin. To test this, we exposed mice to either 1% EtOH in water or corticosterone (100mg/ml) mixed in this solution for a period of 28 days. Half of these mice also received chronic infusions of the growth hormone secretagogue receptor (GHSR) antagonist JMV2959 (100µg/day), while the other half received vehicle. Results show that animals placed on chronic CORT treatment had significant higher body weight, food intake, and water intake compared to control animals, and this effect tended to be attenuated by treatment with the GHSR antagonist although this effect was not statistically significant. CORT treatment led to hyperleptinemia, an effect that was significantly attenuated by co-treatment with the GHSR antagonist. Plasma ghrelin levels were attenuated, whereas glucose levels were significantly higher in CORT treated animals compared to controls and regardless of whether they received the GHSR antagonist or not. These data suggest that ghrelin mediates some endocrine changes associated with the chronic corticosteroid model of obesity.

Abstract: 091

DEFINING A MODEL OF FETAL ALCOHOL SYNDROME IN *XENOPUS LAEVIS*.

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Maternal consumption of ethanol during pregnancy results in a condition known as Fetal Alcohol Spectrum Disorder (FASD). Despite the awareness of the toxic effects ethanol has on the developing embryo, approximately 8000 children are diagnosed with FASD each year in the United States alone. Our lab is currently investigating the potential of *Xenopus laevis* as a model for this disease as the exact mechanism of ethanol's teratogenic effects are unknown. *Xenopus laevis* embryos were dermally treated with 0.3%, 1%, 1.5% and 2.5% ethanol (EtOH) prior to gastrulation at stage 8.5 of their development (mid-blastula transition). At stage 40, embryos were observed to have morphological defects when treated with ethanol concentrations of 1.5% and 2.5%. These morphological defects include shortening along the anterior-posterior axis, ventral edema, and a reduction in eye size. These defects were less severe in embryos that were removed from the ethanol solutions following gastrulation and neurulation. Whole-mount *in situ* hybridization was performed to look at the expression of *otx2* in ethanol treated embryos. *Otx2* is a gene involved in the development of the forebrain and midbrain. Compared to the control, embryos treated with 2.5% EtOH appear to exhibit a reduction in *otx2* expression as well as differential expression patterns.

Abstract: 092

PHYLOGENOMICS OF THE HAWAIIAN *METSCHNIKOWIA* SPECIES

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Recent advances in DNA sequencing technology now allow us to infer the best possible phylogenies. I investigated the phylogenetic relationships of six Hawaiian endemic yeast species belonging to the genus *Metschnikowia* and for which draft genome sequences are available. Previous work elicited three variant gene tree topologies (A, B, and C). I used bioinformatic approaches to explore several hypotheses and ultimately generate an accurate species phylogeny. Bootstraps and posterior probabilities for nearly 1000 gene trees were analyzed, and the results identified a topology that paired *M. mauinuiana* with *M. hamakuensis* (topology B) as being the most accurate representation of the species tree. I constructed 20 additional trees from intergenic regions to rule out the possibility that topology B might be the result of selective pressure. Eighteen of these trees added strong support to topology B. Genes that yielded well-supported trees with alternative topologies (A or C) were examined to see if adjacent sequences always agreed with those topologies. They did not. Most adjacent sequences supported topology B, indicating that the alternative phylogenies are not the result of introgression. As evidence was not found for gene-specific selection nor for horizontal gene transfer, the most likely explanation for the conflicting topologies is incomplete lineage sorting.

Abstract: 093

ASSESSING THE RELATIONSHIP BETWEEN TEST SCORES AND THE TEACHING OF SCIENCE PROCESS SKILLS IN CANADIAN PROVINCES

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Science process skills are broad transferable skills ranging from observation, experimental design, data analysis, and result interpretation, to model formulation. This study aims to explore the possible relationship between the timing of science process skill introduction and the reinforcement with standardized test scores across the Canadian provinces. The science curricula and standardized test scores were examined for the 10 provinces and 3 territories. According to the test scores, Alberta consistently performed above the Canadian average, whereas majority of the provinces performed below the average. The Northern territories were then excluded from the study due to lack of sufficient data. The data suggested that Central and Western Canadian provinces introduced concepts up to elementary level, whereas Atlantic Canada introduced concepts up to the middle school level, resulting in less time for reinforcement of the skills. The results suggest that earlier introduction of science process skills, combined with prolonged reinforcement, will lead to higher standardized test scores. In addition, this study implicates the need for provincial science curriculum documents to be updated and an increase of studies that observe the efficacy of these documents.

Abstract: 094

IMPROVING THE STUDENT LEARNING EXPERIENCE IN ANATOMY AND PHYSIOLOGY COURSES AT THE UNIVERSITY OF TORONTO MISSISSAUGA THROUGH IDENTIFICATION OF MISCONCEPTIONS AND THRESHOLD CONCEPTS

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In large undergraduate classes, students bring with them varying backgrounds and experiences, which may be accompanied by misconceptions in fundamental understanding of a given concept. The goal of this project is to identify common misconceptions in second year anatomy and physiology courses at the University of Toronto Mississauga and to address those areas that might be problematic for student learning and performance. Once addressed, such areas have potential for improving student learning experiences, deepening student understanding and thus may be used to identify the threshold in the understanding of the topic, to further improve student understanding or change their perspectives. We used previously published diagnostic pre- and post- test surveys and case studies in order to assess student understanding of respiratory and cardiovascular systems, particularly oxygen dissociation from hemoglobin and venous return. Student performance on tests and exams from previous years was also analyzed and misconceptions relating to topics other than respiratory and cardiovascular systems, were recognized. We identified excretory and reproductive systems as problematic areas. For example, we noted that physiological events requiring diffusion/osmosis or event sequence explanations were challenging for students. In particular, we found that students struggle with the integration of knowledge, such as coalescing information from more than one system or interdisciplinary topics that depend on the basic concepts of physics, chemistry and biology to truly understand the mechanics of a system.

Abstract: 095

THE STUDY OF VARIEGATION IN *NICOTIANA TABACUM* LEAVES USING VARYING LIGHT INTENSITY AND PLASTOQUINOL TERMINAL OXIDASE (PTOX) INHIBITION.

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Most plant leaves are dark green in colour due to the presence of chlorophyll and other carotenoid pigments. In the genetic mutant IMMUTANS, plant leaves display white sectors or variegation due to the accumulation of the carotenoid phytoene. The gene IMMUTANS codes for plastoquinol terminal oxidase (PTOX) which is involved in photosynthetic electron transport and carotenoid biosynthesis. Variegation is thought to occur in these plants from a combination of photoinhibition, due to defects in carotenoid biosynthesis, and a hyper-reduction of the photosynthetic electron transport chain due to PTOX dysfunction. Variegated plants are valuable in the Canadian horticultural and landscaping sectors, but are currently generated using time consuming traditional breeding methods. This study explores the hypothesis that variegated plants can be generated using a combination of light regime and a chemical inhibitor of PTOX. In this experiment, *Nicotiana tabacum* seeds were subjected to four light intensities and five PTOX inhibitor concentrations to determine the optimal combination of light and inhibitor leading to variegation. Our results indicate that several light levels and inhibitor levels in combination are capable of causing variegation in tobacco within 30 days of treatment.

Abstract: 096

ONE SIZE DOES NOT FIT ALL: THE INFLUENCE OF ANCESTRY AND ENVIRONMENT ON DIFFERENCES IN NEWBORN WEIGHT AMONG MOTHERS OF GUYANESE ORIGIN

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Population-based birth weight curves are used to determine whether an infant is of appropriate weight for gestational age. This method is limited given a tendency to misclassify newborns by over-reporting the number of small for gestational age (SGA) infants and under-reporting the number of large for gestational age (LGA) infants, particularly those born to South Asian mothers. Both SGA and LGA are associated with various complications and negative health outcomes. There is growing evidence to suggest that customized curves adjusted for characteristics such as maternal ethnicity yield a more sensitive method of detecting pathologically SGA and LGA infants. This holds relevance to Canadian-Guyanese people, in that the degree to which distinct aspects of their identities influence their health, remains unclear. We will be using a provincial database to characterize birth weights for the following ethnic groups: Indo-Guyanese, Indian (South Asian), South American and Caribbean. Smoothed birth weight curves will be generated for females and males in each group and statistical analyses will be used to calculate the degree of variance between groups. Results from a similar study looking at infants born to Filipina mothers showed significant differences in birth weights compared to infants born to Canadian-born women and other East Asian-born women. We anticipate the results of our analyses will provide information about the utility of customized curves for Guyanese women in Canada and insights into whether these women are more similar to South Asian women due to genetic factors, or to the South American/Caribbean populations due to environmental factors.

Abstract: 097

TOWARDS CHARACTERIZING THE ENERGY LANDSCAPE OF A DISORDERED PROTEIN USING FORCE PULLING EXPERIMENTS

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Intrinsically disordered proteins (IDPs), which lack a well-defined tertiary structure, have become a focus of recent studies due to their impact on signalling and regulation within cells. Sic1 is an example of an IDP responsible for the transition between growth and DNA replication phases in yeast. Through multistate phosphorylation, Sic1 changes conformation and is targeted for degradation, advancing the cell to S-phase. Structural information and transition states of Sic1 can be determined to obtain energy landscapes by use of Single Molecule Force Spectroscopy (SMFS). Pulling experiments where Sic1 is immobilized between an Atomic Force Microscope (AFM) tip and a surface yields force distance curves. These force curves can quantify motifs and the magnitude of the forces that maintain a given protein conformation, which relates to the boundaries defining an energy landscape. To obtain the energy landscape, a method to attach the protein on the surface and tip must be established. Silicon AFM tips were modified for Sic1 attachment by adding amine groups via silanization and a cross-linker molecule, which could bond to terminal cysteine residues present on the Sic1 construct used in this investigation. Surrogate surfaces were treated in the above manner to permit verification of the surface derivatization chemistry through AFM and fluorescence imaging. SMFS will be calibrated by force induced denaturation of a well characterized double-stranded oligodeoxyribonucleotide. After verifying the system, a 90 amino acid construct representing the disordered region of Sic1 will be studied with the goal to obtain data to interpret the energy landscape of Sic1.

Abstract: 098

FINE-MAPPING OF THE ASSOCIATION WITH OBESITY AT THE *FTO* LOCUS IN A MULTIETHNIC POPULATION

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Fat Mass and Obesity Associated (*FTO*) intron 1 gene variation is the more important contributor to body mass index (BMI) variation in Humans. Fine-mapping studies restricted to African populations failed to identify a causal variant accounting for this association. This prompted us to analyze the contribution of 298 single nucleotide polymorphisms (SNPs) at the *FTO* locus in 17,423 individuals from the EpiDREAM study. The participants are at risk for obesity / type 2 diabetes and represent 6 ethnic groups. The 298 SNPs were genotyped using a 50K cardiovascular gene-centric array. Anthropometric measurements were obtained using a standardized protocol. Eighteen SNPs were associated with BMI after a correction for multiple testing ($P < 1.7 \times 10^{-4}$) and the best hit was rs3751812 SNP ($\beta = 0.518 \pm 0.066$, $P = 1.1 \times 10^{-14}$). Pairwise conditional regressions confirmed that the rs3751812 SNP was more significantly associated than the other SNPs. The Akaike information criterion also identified the rs3751812 SNP as providing the best quality association model (AIC=108888.8). Finally, the rs3751812 SNP displayed the higher posterior probability to be the causal variant (60.38%) using a Bayesian analysis. The frequency of the BMI-increasing rs3751812 T allele was 0.417, 0.331, 0.313, 0.225, 0.169 and 0.107 in populations of European, South Asian, Latino American, Native North American, East Asian and African ancestry. In summary, our data highlight the rs3751812 SNP as the likely causal variant for the association with BMI at the *FTO* locus, and represent the first multiethnic fine-mapping effort at the *FTO* locus.

Abstract: 099

GERMLINE MUTATIONS IN C57BL/6J MICE ASSAYED WITH THE MOUSE DIVERSITY GENOTYPING ARRAY

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Single nucleotide polymorphisms (SNP) and copy number variants are forms of diversity that can be detected using the Mouse Diversity Genotyping Array, which targets 493,290 SNP and 421,905 invariant genomic probes. By comparing SNP variation in a C57BL/6J (B6) mouse family, differences to the reference B6 genome can elucidate inherited mutations, while differences between and within family members of different generations will reveal germline *de novo* mutations, which are heritable unlike those in somatic tissue. Baseline *de novo* mutagenesis that arises due to meiosis contributes to intraindividual germline mosaicism, and is assayed for by comparing the genotypes of pre-meiotic spermatogonia with those of post-meiotic spermatids. Genetic variation between and within sperm stem cells and post-meiotic spermatids are expected to reveal mutational patterns that differ in frequency and type. High purity spermatogenic populations are isolated through fluorescence-activated cell sorting based on characteristic differences in DNA content and Hoechst 33342 efflux capacity. DNA extraction involving chaotropic agents that account for nuclear protamines is currently in progress. Subsequent hybridization to the microarray will be followed by bioinformatics analysis using publicly available reference sequences and software. Future comparisons of inter- and intraindividual genetic variation, such as those between cells of different germ layers that diverge early on in development, will offer glimpses into cell lineage and genome evolution. Determining the level and nature of germline mutations and mosaicism provides a baseline for future monitoring of environmental germline mutations as well as for understanding mechanisms behind clinical phenotypes as varied as male infertility and achondroplasia.

Abstract: 100

CAN THE PREFERENCE FOR INBREEDING EVOLVE?

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When a pool of potential mates includes closely-related individuals, the decision with whom to mate may become more complicated. On one hand, mating with a close relative (e.g. a sibling) can result in offspring suffering from inbreeding depression, due to the increased expression of deleterious recessive alleles, and/or the absence of competitively superior heterozygotes. Alternatively, inbred offspring might gain a fitness advantage via the preservation of co-adapted gene complexes. Numerous surveys of species and populations have found varying attitudes with respect to sibling mating (tolerance, indifference, and a preference for inbreeding have all been observed). This suggests that this mate preference trait may be evolving in response to selection. For this trait to be able to evolve, additive genetic variation must also be present in a population's gene pool. Here, using the model species *Drosophila melanogaster*, we employ cytogenetic cloning techniques in order to investigate the genetic basis of female preference for their hemiclone "brothers" (sharing 50% of the same genome). We find that there is considerable variation with regards to the amount of time a female spends in close association with a male hemiclone. About 10% of this phenotypic variation can be attributed to differences between *D. melanogaster* clone lines, indicating that there is substantial additive genetic variation upon which selection might be able to act.

Abstract: 101

EFFECT OF SODIUM SALICYLATE ON THE EXPRESSION OF GABA_B RECEPTOR SUBUNITS IN THE RAT'S AMYGDALA

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Tinnitus is a phantom auditory perception in absence of external acoustic stimulation, and can be induced by salicylate-containing drugs such as aspirin. Neural mechanisms responsible for tinnitus are poorly understood. It is believed that this phantom sensation is likely due to hyperactivity in auditory neurons. Research in our lab has revealed that sodium salicylate can enhance the activity of auditory neurons through modulating inhibition mediated by the gamma-aminobutyric acid (GABA) receptors. Furthermore, the drug can reduce the level and distribution of the type-B GABAergic (GABA_B) receptors in neural structures including the auditory midbrain and the cochlear nucleus. These effects of the drug on inhibitory neurotransmission can lead to enhanced neural activity in the auditory system. Neurons in the major auditory pathways receive inputs from other brain regions such as the amygdala, which are implicated in the induction of tinnitus. Therefore, it is important to examine how sodium salicylate can change the activity of neurons in auditory structures by affecting the neurophysiology and neurochemistry in these non-auditory structures. We hypothesize that systemic application of sodium salicylate can reduce the level of the GABA_B receptor expression in the amygdala. Immunohistochemical experiments were conducted by using the rat as an animal model to examine the distribution of the two GABA_B receptor subunits, GABA_BR1 and GABA_BR2, in the structure. Our preliminary results indicate that the level of the two GABA_B receptor subunits was reduced in the amygdala three hours after an intraperitoneal injection of sodium salicylate.

Abstract: 102**CORRELATION BETWEEN 3D VOLUME PROJECTIONS CAPTURED DERIVED FROM 3D ULTRASOUND AND DIGITAL SUBTRACTION ANGIOGRAPHY**

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Atherosclerosis in the carotid artery is an indicator of cardiovascular health and risk of cerebral stroke. Currently, Digital Subtraction Angiography (DSA) is the industry standard for determining plaque burden in the carotid. This technique, however, has low throughput, requires specialized equipment, and exposes the patient to significant x-rays. This study was conducted to demonstrate that 3D volume projections generated from 3D ultrasounds can qualitatively and quantitatively correlate to the DSA images as an indicator of plaque burden. 24 neck side 3D ultrasound and DSA scans were taken from 15 patients in Beijing Anzhen Hospital who were previously screened for the presence of atherosclerotic lesions. The ultrasonic images were captured using the Vasqscan 3D ultrasound system, processed to generate 3D views, and projected into 2D views to equate to their DSA counterparts. DSA images were zoomed and cropped to visualize the equivalent area. Four blinded comparisons were conducted in which clinicians scored the levels of similarity in different anatomical areas in both image types, as well as randomized controls. Clinicians also measured levels of linear stenosis in each sub-vessel in both image types. The results were then statistically analyzed to determine the similarity between the 3D ultrasound projections and DSA images. The results demonstrated a significant correlation between 3D volume projections and DSA images taken from the same patient irrespective of vessel orientation or equivalent area size (Fischer Exact Test, $p < 0.01$) in contrast to randomly paired images. Further, there was no difference between the two modalities when contrasting anatomical sub-regions. The results suggest that volumes derived from 3D ultrasound are visually similar to those derived by DSA. All together, clinicians are successfully able to visualize non-equivalent perspectives of 3D volumes when derived from the same patient. This suggests that the volumes derived from 3D ultrasound may be an inexpensive alternative to DSA in evaluating carotid stenosis.

Abstract: 103**THE EFFECTS OF OIL SANDS PROCESS WATER ON HSP70 IN *CHIRONOMUS RIPARUS*.**

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Hsp70 is a highly conserved heat shock protein chaperone which helps in protein folding. Hsp70 is known to be up-regulated under stressful conditions such as increase in temperature or exposure to polluted environments. Such polluted environments are found in the oil mining landscape of Northern Alberta. The method of crude bitumen extraction from oil sands deposits result in a waste by-product of tailings, which consist of naphthenic acids, polycyclic aromatic hydrocarbons and metals. These tailings are stored in wetlands, which allow for the introduction of the toxic by-products into the ecosystem, with naphthenic acids being of primary concern. The objective of this experiment was to determine whether increasing levels of naphthenic acids in oil sands process water (OSPW) increased Hsp70 regulation in *Chironomus riparus*, a freshwater midge common in northern wetland ecosystems. I hypothesized that higher levels of naphthenic acids, indicating high toxicity in a wetland, would induce a greater response in Hsp70 of *C. riparus*. For the experiment, cultured third and fourth instar larvae of *C. riparus* were exposed to varying concentrations of naphthenic acid (10mg/L, 18 mg/L, 29 mg/L, 48 mg/L, 80 mg/L $n=4$) in OSPW for 24 hours with . Hsp70 protein levels were then quantified using western blotting. Actin was used as an internal control to quantify the intensity of band expression. The results of this study may provide insight into the stress response of primary consumers to OSPW in wetlands in post-mining landscapes. It can also be used to assess whether *C. riparus* would be an adequate bio-indicator of stressed wetlands.

Abstract: 104**USING A PARTICIPATORY KINAESTHETIC DNA SIMULATION TO OVERCOME STUDENT MISCONCEPTIONS IN A LARGE BIOLOGY CLASS**Caitlyn D'Souza^{*1}, Tamara Kelly², Fiona Rawle¹¹Department of Biology, University of Toronto Mississauga, Mississauga, L5L1C6.²Department of Biology, York University, Toronto, M3J 1P3

There exist a set of common ingrained misconceptions of basic biology concepts, which when left unaddressed can limit further understanding of biology. For example, biology students often struggle with the integrated concepts of chromosome ploidy, chromosome structure, and DNA structure. Research has shown that active learning strategies are effective in correcting misconceptions regarding fundamental biology concepts; however, the implementation of active learning can be challenging in large classes and the literature highlights the difficulty of keeping students engaged in active learning when not using an audience response system (e.g., clickers). This study evaluated the impact of a novel in-class activity on students' understanding of DNA structure. The study was conducted in Fall 2014 in a large first year introductory evolution course, with 860 students over two lecture sections, at the University of Toronto Mississauga. Pre and post-tests consisting of multiple true/false assessments were used to evaluate students' understanding of DNA structure. The activity was a physical illustration exercise and involved all class members using their bodies to "become" DNA nucleotides, with the entire class forming one long single-stranded DNA molecule. The class formed this strand with outstretched "hand-back-elbow interactions" to simulate the 3', 5' phosphodiester bond in the DNA backbone. Sixteen students were then invited to the front of the room to form a double-stranded DNA molecule with eight students comprising each strand. Analysis of pre-/post-test results revealed that most students had overcome the former prevalent misconceptions. Controls for learning gains included questions on related misconceptions that were not targeted during the activity. This active learning intervention will be demonstrated during the oral presentation.

Abstract: 105**A MATTER OF PERSPECTIVE: DIFFERENCES BETWEEN PROFESSOR AND STUDENT VIEWS ON BARRIERS TO ACADEMIC SUCCESS IN THE SCIENCES**Ashna Khanna, Wadia Forrester, Caitlyn D'Souza, Ting Xi (Cassandra) Zhao, Silvio Lioniello, Steven Chatfield, and Fiona Rawle
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Difficulties in the transition from high school to higher education for students in science programs have been well documented in the literature. The purpose of this study was to examine differences between faculty and student perspectives on barriers to academic success in the sciences. Data was gathered via the following methods: (1) Faculty members in several science departments at the University of Toronto Mississauga were interviewed, (2) Introductory Biology students completed a perspective survey, and (3) participated in focus groups. Qualitative data was coded and analyzed through the identification of common themes and key words. Quantitative data consisted of perspective survey questions answered using the Likert scale. The primary barrier, indicated by faculty during interviews, was student motivation, such as students studying for a grade rather than studying to learn material. The number two barrier, as identified by faculty, was lack of time management skills. Student perspective survey responses showed that the primary barrier was time management. When asked what advice students wish they had at the beginning of term, the most common response was "study skills and time management skills". When asked what the best way would be to get this advice to students, the most common response was "from an upper year student". Student focus group responses revealed that science students experience particular challenges due to increased class sizes, increased independence and decreased academic support relative to high school. Our overarching goal is to use the data from this study to inform the development of transition programs and strategies to increase science student success.

Abstract: 106

COMBINED STRESSOR EFFECTS OF THERMAL, CHEMICAL, AND RADIOLOGICAL TREATMENTS ON A RAINBOW TROUT (*Oncorhynchus mykiss*) GONADAL CELL LINE

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Combined stressor models allow for the investigation of complex interactions between stressors that cannot be studied in single compound exposures, and the utilization of appropriate controls allows for the elucidation of single stressor effects. The effects of acute temperature changes and morpholine, a chemical corrosion inhibitor utilized in industrial processes, in modifying the response of a the RTG-2 rainbow trout (*Oncorhynchus mykiss*) gonadal cell line to ionizing radiation were studied. A characteristic dose-response relationship to ionizing radiation was established utilizing the cytokinesis-block micronucleus assay. Morpholine concentrations of 10 and 100 mg/L appeared to have minimal effects on MN formation both alone and in combination with a 10 Gy radiation dose. A $\Delta 6^{\circ}\text{C}$ heat shock similarly did not appear to have an effect alone or when treated prior to a 10 Gy dose, however, a $\Delta 9^{\circ}\text{C}$ heat shock treatment alone caused MN formation in $26.8 \pm 7.7\%$ (mean \pm SD) of binucleate cells compared to $2.3 \pm 0.6\%$ MN formation in control flasks. The $\Delta 9^{\circ}\text{C}$ heat shock treatment additionally appeared to have additive effects when combined with a 10 Gy dose with MN formation increasing from $51.6 \pm 4.6\%$ to $69.3 \pm 5.4\%$ in the 10 Gy irradiation and the combined $\Delta 9^{\circ}\text{C}$ heat shock and 10 Gy irradiation treatments respectively. These results are relevant when considering the types of stressors present at low levels in thermal power generation outflow waters, which similarly occur simultaneously. This results in the need for the investigation of potential interactions between these various types of stressors.

Abstract: 107

ASSESSMENT OF SUITABILITY OF SLIDE-BASED PLANT GROWTH APPROACH FOR USE IN WATER QUALITY MONITORING IN FIELD DEPLOYABLE ASSAYS.

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This study examined the suitability of a vertical, slide-based growth approach to be used in a novel field-deployable assay (FDA) using wetland plant species as bio-monitors. The in-situ assay approach is being developed as an alternative to current toxicity testing methods, which focus on lab-based approaches that involve sampling of test sites and subsequent analysis of those samples in the lab. The impacts of horizontal and vertical growth orientation were compared in *Lactuca sativa* and *Phalaris arundinacea* seedlings, with slight increases in root length identified in seeds grown vertically. *L. sativa* plants grown horizontally had a mean root length of 2.87cm, while two separate vertical growth approaches yielded mean root lengths of 3.68 and 3.18 cm respectively. Standard deviation was 1.33 cm for horizontal growth, with values of 2.73 and 1.48 cm for vertical growth. *P. arundinacea* roots had a mean length of 5.88cm for horizontal growth, with 8.34 and 9.65 cm for vertical growth. Standard deviation for horizontal root growth was 2.84, and vertical growth produced standard deviations of 2.81 and 3.44 cm. *L. sativa* seedlings were also exposed to several concentrations of arsenic, a known toxicant, in order to assess their performance within the slide-based assays. The results of this study will contribute to the development of the in-situ FDA, which may reduce cost and time-inputs incurred by existing test methods while providing a more accurate reflection of real-world conditions at test sites.

Abstract: 108**DOES FISH CONDITION AFFECT SPERM SWIMMING VELOCITY IN *BARBUS NEUMAYERI*?**Dominic Hamelin¹ and mery Martínez¹¹Department of Biology, Laurentian University, Sudbury, Ontario, P3E 2C6

Anthropogenic activities can dramatically affect water pH, toxicity and water dissolved oxygen levels. These environmental factors can have a direct effect on fish survival and reproduction. In this study, we explore whether variations in sperm swimming velocity of the wide spread African cyprinid *Barbus neumayeri* (Fischer, 1884) exist. Fish were sampled across three sites with divergent dissolved oxygen levels (DO), from hypoxic swamps to intermittent normoxic streams. We examine the possible relationships between habitat type, condition index (K), Gonadosomatic Index (GSI), testes mass and their possible correlation with sperm swimming velocity. Among sampling sites, significant differences were found in fish total body length ($P = 0.0122$), standard body length ($P = 0.00013$), somatic body mass ($P = 0.0185$), right testes mass ($P = 0.000197$), total testes mass ($P = 0.0006$), and GSI ($P = 0.0500$). Meanwhile, the total body length ($P = 0.00096$), the standard body length ($P < 0.0001$), and K ($P = 0.00295$) significantly differed between levels of DO. Interestingly, the average path velocity (V_{AP}), the curvilinear velocity (V_{CL}), and the straight-line velocity (V_{SL}) of sperm ($P < 0.0001$ for all velocities) significantly varied among sampling sites. However, sperm swimming velocities did not significantly differ between levels of DO. Opposite to our expectations, K was not significantly correlated with sperm velocities, while GSI was correlated with V_{AP} ($P = 0.0111$), V_{CL} ($P = 0.00896$), and V_{SL} ($P = 0.0102$). Since K did not affect the sperm swimming velocity, this could suggest that *B. neumayeri* individuals may be experiencing trade-offs between growth, survival and fitness. Future studies should evaluate the correlation between sperm morphology, testes tissue composition, and sperm velocity of *B. neumayeri* from divergent habitats such as rivers, swamps, and streams during both the dry and the wet season.

Abstract: 109**PRIMARY STRESS RESPONSE LEADS TO OXIDATIVE STRESS IN SMALL MOUTH BASS**

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Vertebrates of all kinds elicit a phenomenon known as the stress response when faced with a homeostasis-threatening stimulus. The initial stress response is characterized by a neuroendocrine cascade, involving the release of cortisol from the adrenal glands. The Centrarchidae family of fishes, including small mouth bass (*Micropterus dolomieu*), are an ideal group for studying the effects of stress, as these fish undergo a variety of life-threatening situations throughout their spawning season. Long-term exposure to cortisol is shown to be detrimental to numerous organisms such as these, but the extent of these detrimental secondary effects is unknown. It is logical to assume that one such secondary effect may be oxidative stress, as this is commonly associated with many diseases and other metabolic disturbances in the body. 8-hydroxy-2-deoxy guanosine (8-OH-dG) is a marker of oxidative stress and can be measured in various tissues. Here, we quantified the levels of 8-OH-dG in samples of blood collected from 28 male small mouth bass during spawning season in order to determine the extent of oxidative stress in these organisms. Fish were treated with combinations of cortisol injection in order to imitate the primary stress response, and food in order to mitigate the primary stress response. Oxidative stress was highest in fish that had experienced cortisol injection alone and lowest in fish that were treated with just food. Our results suggest that the primary stress response leads to oxidative stress in small mouth bass, which further validates the detrimental impact of the primary stress response.

Abstract: 110**AN EVALUATION OF THE STATUS AND FUNCTION OF GPSM2 IN PRIMARY HUMAN GLIOBLASTOMA CELLS.**

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Cell polarization and spindle orientation play important roles in the division of stem cells. The coordination of cell division is a tightly regulated process involving multiple intra and extracellular signaling pathways. In recent years an increasing body of evidence has suggested that certain human cancers, such as glioblastoma multiforme, may arise through disturbances to these signaling pathways resulting in improper cell division and the production of highly-proliferative daughter cells. The signaling modulator protein GPSM2, regulates extracellular signal transduction through the modulation of cell surface receptor G-proteins and is known to play a role in the establishment of mitotic spindle orientation during cell division. Disruption of *GPSM2* functioning during stem cell mitosis could contribute to the genesis of glioblastoma multiforme. The purpose of this study was to evaluate the status of GPSM2 in primary human glioblastoma cells by Western blot analysis and siRNA knockdown as well as evaluates it's role in glioblastoma malignancy through reintroduction of a functional *GPSM2* gene by lentiviral transduction.

Abstract: 111

IS THE PROBOSCIPEDIA PHENOTYPE IN *DROSOPHILA MELANOGASTER* DUE TO ACTIVATION OF ARGOS EXPRESSION INHIBITING CELL PROLIFERATION?

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Proboscipedia is a homeotic gene responsible for patterning of the anterior-posterior axis of head structures during development in *Drosophila melanogaster*. However, the mechanism of PB function is still unknown. Recent studies have shown that the vestigial maxillary palp phenotype of pb mutant *D. melanogaster* can be rescued by expression of ligands that act positively on the epidermal growth factor receptor pathway. With this information we propose a model in which PB inhibits Argos expression. AOS is an antagonistic ligand to the epidermal growth factor receptor pathway. In pb mutants it is expected that there will be increased *aos* expression and as a result an increase in EGFR pathway activity. If this model is accurate, pb mutant flies should show decreased amounts of cell division in developing maxillary palp regions relative to wild type flies. Confocal time-lapse microscopy was used in an attempt to compare relative amounts of cell division between pb mutant and wild type flies. Mitosis occurred at a quicker rate than was feasible to quantify by confocal microscopy, and as such a relative comparison could not be done. However, the quick progression through mitosis appears to be unique and warrants further investigation.

Abstract: 112

MORPHOMETRIC ANALYSIS OF THE SUBSYNAPTIC RETICULUM IN WILD-TYPE AND MUTANT *DROSOPHILA* LARVAL NEUROMUSCULAR JUNCTIONS

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The synapse is a highly specialized structure that is essential for transmission between a pre-synaptic neuron and its target cell. The *Drosophila* neuromuscular junction (NMJ) is a commonly used model system for synaptic transmission because of the wealth of genetic tools that can be used and the ease of access for imaging and electrophysiological techniques. Through this project, we studied an elaborate system of post-synaptic plasma membrane folds, called the subsynaptic reticulum (SSR). Despite being present at the NMJ, little is known about the structural and functional role of the SSR. Thus, we studied the extent of SSR and its distribution in mutant *Drosophila* larvae by assessing the effects of differential SSR size on synaptic activity and development. Using genetic variants that affect SSR development, we conducted a comparative analysis of synaptic size and cross-sectional SSR thickness on muscles 4 and 6 of the *Drosophila* larvae. Furthermore, in order to decipher the cellular components that constitute the SSR, we determined the localization of potassium shaker channels on the membrane. Further work will pair these structural changes with electrophysiological experiments of synaptic activity. Ultimately, the results of this project will help to advance our understanding of the role played by the SSR in synaptic transmission and plasticity.

Abstract: 113

DOES THE STRESS OF SUBLETHAL METAL EXPOSURE ALTER METABOLIC ACTIVITY IN *DAPHNIA PULEX-PULICARIA*?

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Lakes in the Sudbury area have been contaminated by emissions from local mines and smelters. Emission reductions over the last 30 y have resulted in recovery of many lakes but nickel (Ni) and copper (Cu) remain as elements of concern which have caused a stress to aquatic life, particularly sensitive invertebrate species such as *Daphnia*. To understand the ongoing recovery of these lakes, it is important to understand the effects of metals, such as Ni and Cu, on the physiology of sensitive species. The goal of this study was to assess the effect of sublethal metal exposure on metabolic rate in *Daphnia pulex-pulicaria* in soft water. Using micro-oxygen sensing technology within flow-through chambers of 0.75 mls the effect of Cu, Ni and Cu+Ni mixtures on oxygen consumption rate was studied in exposures of up to 24 h. Low and high exposure treatments for Ni and Cu were chosen to be relative to concentrations that are associated with acute toxicity. Exposure to 1.5 mg Ni/L causes an increased oxygen consumption rate in *D. pulex pulicaria*, indicating the induction of stress. Copper exposures (5 to 20 µg Cu/L) resulted in no change in oxygen consumption, even in solutions that caused toxicity. The effect of Cu + Ni mixtures was studied to see if the effects are additive, synergistic, or antagonistic. This research project was funded by the NSERC CRD program with local support at Laurier from FOSSA and LIWS.

Abstract: 114**GENETIC VARIATION FOR FEMALE MATE BODY-SIZE PREFERENCES IN *DROSOPHILA MELANOGASTER***Mireille Golemic*¹ and Tristan A.F. Long¹¹Department of Biology, Wilfrid Laurier University, Waterloo Ontario, Canada, N2L 3C5

While traditional models of sexual selection assume, either implicitly or explicitly, that females all exhibit the same preferences for male characteristics, in reality females may often vary in their attraction to different males. We set out to quantify the extent and genomic location of the genetic basis for variation in female *Drosophila melanogaster* preference for male body-size, a trait with both beneficial and harmful effects on female fitness. Using cytogenetic cloning techniques, we were able to estimate the amount of additive genetic variation for mate preference behaviour in females from clone lines sharing either a whole haploid genome or only an X chromosome. This allowed us to explicitly test whether the X chromosome is a “hotspot” for said genetic variation as predicted by some theory. We found substantial genetic variation for female preference for male body-size, but this variation seems to be predominantly located on the autosomes, and not on the X chromosome. The evolutionary impacts of the results are discussed.

Abstract: 115**A COMPARISON OF qPCR AND MULTIPLEX METHODOLOGIES**

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Polymerase chain reaction (PCR) is a method in which DNA is amplified according to primers designed for a specific sequence. In real-time or quantitative PCR (qPCR) a single set of primers is added to a single tube, resulting in the amplification of a single gene. In multiplex PCR multiple sets of forward and the corresponding reverse primers may be added. These genes may be visualized separately through the use of a fluorescent dye added to a probe within the sequence. Multiplex may be advantageous due to limited handling of the reactants and simultaneous analyzing of samples consisting of both the gene of interest and the reference gene. To determine the usefulness of multiplex PCR methodology in fish reproductive endocrinology studies, hepatic vitellogenin (VTG) mRNA expression by qPCR and multiplex were compared in two separate studies. In one study, male fathead minnow (*Pimephales promelas*) were exposed to 10ng/L of EE₂ and control water in a laboratory for a period of 15 days. In a second study, female walleye (*Stizostedion Vitreum*) from Tathlina and Kakisa Lakes in the Northwest Territories were collected across different time points of their spawning cycle. In male fathead minnow, hepatic VTG mRNA expression by qPCR was significantly increased 1574 fold following exposure to EE₂, while female walleye hepatic VTG mRNA expression significantly increased 12 fold in pre-spawning compared to post spawning periods. Analysis and comparison of the multiplex PCR results is ongoing.

Abstract: 116**UTILIZING *NOTROPIS HUDSONIUS* (SPOTTAIL SHINER) AND *NOTROPIS ATHERINOIDES* (EMERALD SHINER) AS A SMALL-BODIED FISH BIOMONITORING SPECIES IN NORTHERN CANADIAN WATER SYSTEMS.**

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Brett Pomeroy*, Deborah MacLatchy, and Andrea Lister

Aquatic environments, such as the Slave River, NT, can be influenced by anthropogenic contaminants that impact ecosystem health. Portions of the Slave River are downstream of various industrial (e.g., oil sand extraction) and municipal (e.g., sewage treatment) activities which discharge waste water into the river. There is growing concern from residents in the area of Fort Smith, NT about the ecological integrity of their portion of the Slave River, which is a vital drinking water and food resource for the community. Small-bodied fish species with small home ranges are useful in environmental monitoring to assess site-specific impacts. *Notropis hudsonius* (spottail shiner) and *N. atherinoides* (emerald shiner) were collected by seine netting upstream, downstream, and directly adjacent to a sewage discharge exposed site on the Slave River near Fort Smith in September, 2014. Morphometric endpoints (gonadosomatic(GSI), liver somatic indices(LSI) and condition factor(CF)) indicated that no considerable changes were observed in GSI and CF in either species, however emerald shiner LSI was significantly higher at the sewage exposed site in relation to the other sites. Ovarian estradiol steroid levels were assessed to determine reproductive status and indicated that estradiol levels were higher at the downstream site relative to the other sites in emerald shiner. This study is part of a longitudinal study to develop baseline data for shiners as small-bodied fish biomonitoring species in the Fort Smith portion of the Slave River.

Abstract: 117**CHARACTERIZATION OF A FIC/DOC TOXIN-ANTITOXIN-LIKE SYSTEM IN *SINORHIZBIUM MELILOTI***

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Toxin-antitoxin (TA) systems are addiction modules typically found on plasmids but also present in prokaryotic genomes. The genome of the N₂-fixing alfalfa symbiont *Sinorhizobium meliloti* has >200 predicted TA systems, however the roles of these systems is often unclear. The TA locus *sma2105* contains a protein coding gene, annotated as a hypothetical Fic/DOC (abbreviated as fido) protein, as well as a ~500bp upstream region. Based on a previous RNA-seq study the 5' upstream region was predicted to contain a ~150bp antisense transcript which overlaps with the 5'UTR of *sma2105*. We found that deletion of a 60-kb region ($\Delta A131$) containing *sma2105* reduced cell viability unless the entire *sma2105* locus or the 500bp upstream region was present *in trans*. By generating smaller deletions of the *sma2105* locus, we show that the *sma2105* is a toxin, as its removal permits recovery of $\Delta A131$. Additionally, we demonstrated that the upstream antisense transcript may be a cognate antitoxin, as it complemented the 60-kb deletion when present *in trans*. Structural prediction of the Sma2105 protein indicates similarities to AMPylation proteins of the fido protein family: post-translational modification (PTM) proteins that are ubiquitous in nature yet diverse in function, with several shown to be secreted effectors associated with host-microbe interactions. This work suggests that Sma2105 is a toxin and likely exerts its toxic properties through aberrant PTM that may disrupt normal modification and result in cell growth inhibition.

Abstract: 118**INVESTIGATING THE EFFECT OF NOTOCHORD-SPECIFIC *Ccn2* KNOCKOUT ON GENE EXPRESSION IN THE INTERVERTEBRAL DISC**

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The intervertebral disc (IVD) confers flexibility to, and absorbs load along the spine. IVD degeneration is characterized by a number of aberrant processes including increased fibrosis, matrix degradation, and inflammation. CCN2 is a matricellular protein implicated in IVD biology. Our previous studies demonstrated that knockout (KO) mice with notochord-specific loss of *Ccn2* (*Noto^{Cre/+}; Ccn2^{-/-}*) exhibit accelerated age-associated IVD degeneration. However, the specific pathways by which loss of CCN2 promotes disc degeneration remain unclear. We predicted that IVDs from *Ccn2* KO mice will show increased expression of markers of tissue fibrosis, matrix degradation, neural in-growth and inflammation compared to age-matched wild-type (WT) controls, recapitulating human disc degeneration. Real-time PCR was used to quantify gene expression in IVDs from WT and KO mice at 6, 9, 14 or 19 months-of-age. The expression of extracellular matrix genes (*Acan*, *Col2a1*) decreased with age in wild-type mice; however no difference in their expression was detected between genotypes. Although no change was detected in expression of the matrix-remodeling enzymes *Mmp13*, *Adamts-4* or *Adamts-5*, *Mmp3* was upregulated in KO mice at 19 months-of-age. Interestingly, expression of inflammatory cytokines *Tnfa* and *Il6* was upregulated in KO mice. Furthermore, the expression of *Gap43*, a marker of neural growth, was upregulated in KO mice at 6 months-of-age. The expression of *Bdnf*, a neurotrophin implicated in maintenance of nociceptive neurons, was upregulated in KO mice at 19 months-of-age. This study has further validated the notochord-specific *Ccn2* knockout mouse as a model to study disc degeneration, implicating CCN2 as a potential therapeutic target.

Abstract: 119

STRUCTURE-FUNCTION CHARACTERIZATION OF BCSE PROTEIN INVOLVED IN CELLULOSE BIOSYNTHESIS BY *ESCHERICHIA COLI*.

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Most bacterial infections observed in the developed world utilize biofilms which consist of a cluster of microorganisms fixed within a bacteria-synthesized exopolymer matrix, such as cellulose. Biofilms enhance surface adherence and protection from temperature fluctuations, immune detection, and chemical or mechanical trauma during colonization. Bacterial cellulose biosynthesis is thought to occur via a multi-step process, involving the bacterial cellulose synthesis (Bcs) multi-protein complex which spans the inner membrane, peptidoglycan layer and outer membrane of the bacterial cell. The genes *bcsABZC* are essential for cellulose biosynthesis, and in enteric pathogens, such as *E. coli* and *Salmonella spp.*, *bcsEFG* are critical for optimal cellulose formation. This project utilizes biological and chemical methods to express, purify, and initiate structural characterization of the BcsE protein. The expressed protein was engineered with histidine tags so that, after large-scale production, it could be purified using immobilized metal affinity chromatography. Initial crystallization trials were conducted, however, BcsE has proven unstable over long periods and quickly precipitated out of solution in these trials. Binding substrates often improve protein stability in solution. One BcsE substrate, cyclic-dimeric-guanosine monophosphate (c-di-GMP) is not commercially available, so we have begun using WspR from *Pseudomonas aeruginosa* to enzymatically synthesize it. Once obtained, c-di-GMP will be added to new BcsE preparations, and crystal trials will continue. In conjunction with this structural work, we have also performed a set of experiments exploring the hypothesized protease activity of BcsE. Together, these results will lay the groundwork for understanding the role of this protein in bacterial cellulose synthesis.

Abstract: 120

ASSESSING SOCIAL BEHAVIOUR IN *FoxP* MUTANT *DROSOPHILA MELANOGASTER*

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The *FOXP2* gene is significant in not only speech and language development, but is also a behavioural requirement during social interactions. Vertebrate models have demonstrated that disrupted *FOXP2* results in impaired courtship songs and poor motor coordination. The invertebrate model fruit fly (*Drosophila melanogaster*) has been identified as possessing an orthologous gene to *FOXP2* known as *FoxP* important for proper complex social interactions such as courtship. But a simple form of interaction is the social spacing during group formation. Deciding proper social distance is a crucial step necessary before other complex behaviours. The normal *D. melanogaster* social distance is measured as 1-2 body lengths away from their closest neighbour within a group, and is therefore a quantifiable measure to analyze normal or abnormal social behaviour. It is hypothesized that the *FoxP* gene is necessary during social interactions and that an altered *FoxP* gene will lead to abnormal behaviours seen as deviation from the 1-2 body length distance expected during these interactions. We used a social space assay to observe social behaviours of interacting flies of both wild type *FoxP* and mutant *FoxP* in a 2-dimensional chamber. We show that male flies with a disrupted *FoxP* isoform exhibit abnormally closer distance to the nearest neighbours, but also form one large aggregate rather than smaller disbursed groups. Furthermore, mutant *FoxP* flies have been observed to have a loss-of-righting-reflex (LORR), the ability to re-orient oneself from supine position to prone. These results demonstrate a conserved role for *FoxP* in behaviour during social interactions.

Abstract: 121

DOES GROUP COMPOSITION IMPACT GROUP SCORES IN TWO-STAGE COLLABORATIVE EXAMS?

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Recently, two-stage exams were introduced in two large enrolment second year genetic courses at two different universities. The two courses follow similar formats and use course learning outcomes, activities and materials developed by both instructors. Two-stage exams are those in which students first write an exam individually, followed immediately by a second stage in which they write the same, or similar, exam as part of a small group. Exam grades comprised 85% individual mark and 15% group mark. Overall, student exam grades were improved by the group portion of the exam, however, the extent of score improvement varied between groups, and for several teams, group scores were lower than the members' individual scores. As we try to provide effective collaborative learning experiences through two-stage exams, the goal of this project is to identify factors that may improve or hinder gains [(group score – average of members' individual scores)/(100%-average of members' individual scores)] while students work in groups. To determine group-composition factors that impact team performance, we compare individual and team scores from midterm and final exams, categorizing groups as being fixed (group members stayed together throughout all 3 exams) or dynamic (composition of group members changed in at least one exam). Results thus far suggest that the effect of group composition is not the same when predicting exam scores vs performance gains [(group score – average of members' individual scores)/(100%-average of members' individual scores)]. We hope to use this opportunity to discuss different approaches to assess factors impacting performance on collaborative exams.

Abstract: 122

ANTIBIOTIC SUSCEPTIBILITY AND HORIZONTAL GENE TRANSFER POTENTIAL OF ANTIBIOTIC RESISTANT GENES OF AQUACULTURE AND HUMAN PATHOGENS

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As the use of antibiotics increases, the potential for antibiotic resistant strains of bacteria emerging is a major concern. The use of antibiotics in the aquaculture industry could potentially allow for resistance to be favoured among aquaculture pathogens. The selective pressure of antibiotics may encourage resistant bacteria to persist for a prolonged periods of time, which may permit their release into the broader environment. The goal of this research is to better understand the antibiotic resistance profiles of aquaculture pathogens. Furthermore, the research promotes the understanding of how resistance may change as a result of continual exposure, as well as the potential for horizontal gene transfer (HGT). *Aeromonas* spp. and *Yersinia* spp. aquaculture pathogens as well as potential human pathogens such as *Enterococcus* spp. were isolated. The resistance profiles of the aquaculture pathogens were determined and originally demonstrated minimal resistance. However after continual exposure to the antibiotics it was demonstrated that these bacteria were resistant to Vancomycin (VA 30 µg), Tetracycline (TE 5 µg) and Sulfadiazine (SD 250 µg). In the absence of selective pressure a previously resistant bacterial strain became more susceptible to the antibiotic. In the case of *Aeromonas* spp, the resistance for VA decreased by more than 44%. The resistance profiles of *Enterococcus* spp. were determined to be more susceptible to the antibiotics that were being tested. Preliminary attempts for horizontal gene transfer (HGT) have been unsuccessful using solid media. Subsequent tests will be conducted under high stress in attempt to optimize the potential for HGT.

Abstract: 123

RISK AVERSION AND LIFE HISTORY OF *AULACORTHUM SOLANI* AND *APHIS NERII* UPON EXPOSURE TO NECROMONES (OLEIC ACID)

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Aphids are one of the most destructive pests in agriculture and is estimated to cost hundreds of millions of dollars in damages annually. But wide scale management and control of aphids have generally been limited to insecticides which have serious environmental impacts. In this thesis, we investigate necromones (oleic acid) as a potential alternative in aphid control by looking at its effects on aphid's behaviour (risk aversion) and development (wing induction) as well as any effects on mortality. We have developed numerous protocols in order to observe the effects of necromones on aphids including three types of bioassays (leaf-dip, filter paper and whole-plant). Currently, we have conducted three risk-aversion experiments on two species of aphids (*Aulacorthum solani* and *Aphis nerii*) and have consistently found significant differences in aphid distribution between control (ethanol) and treatment (oleic acid) groups. In addition, we also found that the application of necromone causes higher nymph mortality and significantly reduced reproduction. However, wing induction has not been successfully observed in any of the studies conducted so far. These findings indicate necromones as a likely alternative in controlling and managing aphid population in the agriculture sector.

Abstract: 124

EXERCISE-INDUCE PERIPHERAL FAT UPTAKE IS REGULATED BY PGC-1 α -DEPENDENT MYOKINE, VEGF-B.

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Skeletal muscle is a mitochondria-enriched, highly adaptable tissue that undergoes robust metabolic adaptations in response to endurance exercise. It is also becoming increasingly recognized as a secretory tissue that mediates organ cross-talk and regulates whole-body metabolism. PGC-1 α is a transcriptional co-activator that is recognized as a “master regulator” of mitochondrial biogenesis and activated in response to a number of metabolic signals including exercise, fasting, and oxygen depletion. We have shown here that acute and chronic endurance exercise increases expression of PGC-1 α in skeletal muscle as well as circulating VEGF-B levels in both mice and human serum. Treatment of an endothelial cell line with conditioned media from myotubes overexpressing *PGC1 α* caused an increase in expression of fatty acid transporter proteins, *FATP3* and *FATP4*, which was abrogated with siRNA knock-down of *VEGFB*. Trans-endothelial flux of 4,4-difluoro-5-methyl-4-bora-3a,4a-diaza-s-indacene-3-dodecanoic acid-labelled saturated long chain fatty acid analogue (BODIPY-FA) is also regulated by myotube secretion of VEGF-B *in vitro*. Furthermore, *in vivo* uptake of BODIPY-FA in heart and muscle following oral gavage in mice is modulated by circulating VEGF-B. The physiological role of VEGF-B is controversial in the literature, and to our knowledge, we are the first group to decipher the role of VEGF-B as an endurance exercise-mediated PGC-1 α -dependent myokine regulating the uptake of fatty acids into muscle through a trans-endothelial route. We suggest that VEGF-B may be effectively utilized as an adjunctive therapy with endurance exercise to improve metabolic health in patients with mitochondrial myopathies or other secondary mitochondrial disorders.

Abstract: 125

SEARCH AND DISCOVER: PROTEOBACTERIAL ALTERNATIVE OXIDASE GENES IN CANADIAN METAGENOMES.

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Alternative oxidase (AOX) is a protein component of the electron transport chain found in many types of organisms including some plants, animals and prokaryotes. Little information is available for prokaryotes containing AOX genes, however it is speculated that proteobacteria are the only bacteria that carry AOX. Metagenomes carrying bacterial genes were collected from the arctic tundra and boreal forest these metagenome libraries were then cultured into more manageable sub-libraries. Primers were designed based off of known bacterial AOX sequences and used to amplify potential proteobacterial AOX genes within each sub-library. The viability of the primers was tested and the DNA that was amplified by these primers was sent off for sequencing. Using a combination of degenerate primers, PCR, gel purification, ligation and cloning, areas in the boreal forest and arctic tundra were sampled for potential AOX alpha-proteobacterial genes.

Abstract: 126

A SYSTEMATIC REVIEW OF SYNDROMIC OBESITY

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The obesity epidemic is a major healthcare concern due to its multiple negative impacts on physical wellbeing. Obesity exemplifies a strong model of a disease that results from an interaction between genes and the environment. The key environmental factors include diet and physical activity. Genetic causes of obesity can be divided into three major categories: non-syndromic monogenic obesity, syndromic monogenic obesity and polygenic obesity. Syndromic monogenic obesity follows Mendelian patterns of inheritance and involves features other than obesity, such as mental retardation, dysmorphic features, organ-specific abnormalities and hyperphagia. The objective of this project is to conduct a systematic review of literature on syndromic forms of monogenic obesity, as owing to their rareness and poor elucidation of genetic causes, no comprehensive and systematic review has yet been conducted on all forms of syndromic obesity. Literature search of seven online databases yielded 13,719 papers. After title and abstract screening, 289 records were considered for full text review. Relevant information from those records was extracted, analyzed and compiled. This review provides researchers with a summary of obesity syndromes found through the search process, which will enable them to focus on those whose genetic cause is unknown. Identification of the disease genes and their function will serve to improve the diagnostic tools and treatments for the disease, as well as its management. The review also reveals the challenges that were faced while dealing with scientific literature, such as discrepancy in the names of syndromes, and proposes ways to resolve such issues.

Abstract: 127

EFFECTS OF WNT ACTIVATION AND INHIBITION ON THYMIC SIGNALLING

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The effects of canonical Wnt signalling activator, BIO and Wnt signalling inhibitor PKF 118-310 on gene expression of *Foxn1*, *Dll4*, and a small subset of other functionally relevant markers in mouse adult TEC was studied. The use of thymic slices for study of gene expression was also tested. Thymic slices were cultured with 1 or 5 μ M of BIO, 0.1 or 1 μ M PKF 118-310, DMSO, or a media control, then purified and sorted by FACs before pre-amplification and RT-qPCR of results. ddCt method was used to analyze results, although no biologically relevant trends were seen for most markers. *Dll4* did show a slight increase in gene expression after activation with BIO, and decrease in gene expression after inhibition with the compound PKF118-310. Furthermore, *Foxn1* also showed a slight increase in change of gene expression after activation with BIO, although change in gene expression after addition of PKF 118-310 was also increased, greater and beyond that due to BIO activation. In both gene expressions, levels were increased beyond untreated, uncultured adult TEC controls. It is suggested that perhaps a balance in canonical Wnt activation and inhibition is required to regulate thymic gene expression. It was also shown that thymic slices were indeed able to support the study of gene expression, although various opportunities to improve how thymic slices can be used was also provided.

Abstract: 128

IDENTIFICATION OF TYR65 AND TYR77 AS POSSIBLE PHOSPHORYLATION SITES WITHIN NFE2L1 THROUGH SITE DIRECTED MUTAGENESIS

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Nuclear Factor, Erythroid 2-Like 1 (NFE2L1, Nrf1) is a transcription factor that is localized in the endoplasmic reticulum (ER) in its inactive form, p120. NFE2L1 regulates the transcription of oxidative stress-related genes associated with antioxidant response element (ARE) promoters. NFE2L1 is negatively regulated by its N-terminal domain (NTD), which is tethered to the ER. There have been sub-domains identified within NTD including N-terminal homology box 2 (NHB2), single peptide associated sequence (SAS), cholesterol recognition amino acid consensus sequence (CRACs), and N-terminal homology box 2 (NHB2). Previous bioinformatics analyses identified two possible overlapping phosphorylation motifs and CRACs between the residues 62 and 70, and 74 and 82. The CRAC motif is defined by L/V-X₁₋₅-Y-X₁₋₅-K/R with Tyr residue being integral for the binding to cholesterol and/or sphingolipids also being the possible phosphorylation site. Through the phosphorylation of the Tyr residues within the CRACs subdomains leads to the deglycosylation and dissociation of p120 NFE2L1 from ER to generate its active form, p95. Through site directed mutagenesis, we produced Y65F and Y77F mutants of NFE2L1. These mutants have increased transcriptional activity using the glutamate cysteine ligase catalytic luciferase (GCLC-Luc) reporter plasmid compared to the wild type. Additionally, HEK293 cells transfected with mutants have decreased cellular viability compared to that of the wild type.

Abstract: 129

IMPACT OF LAMPRICIDE TOXICITY IN A REPRESENTATIVE NON-TARGET FISH, THE RAINBOW TROUT

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The lampricides 3-trifluoromethyl-4-nitrophenol (TFM) and 2,5-dichloro-4-nitrosalicylanilide (niclosamide) are used by the Great Lakes Fisheries Commission (GLFC) for the selective control of larval sea lamprey (*Petromyzon marinus*), which feed on economically important fishes of the Great Lakes during their parasitic life stage. TFM is commonly used alone during stream treatments, but niclosamide is added as a granular powder (at 1% of the TFM concentration) in lentic areas and deeper waters, to increase lampricide treatment efficiency. While TFM and niclosamide have been used for over 50 years, their interactions in mixtures and their effects on non-target fishes are not yet entirely understood. Therefore, the goal of this study was to investigate the interactions between these two chemicals in mixtures, in a representative non-target fish, the rainbow trout. To this end, we used a toxic unit (TU) approach, where 1 TU = 12 h LC₅₀ of each lampricide alone, and in a TFM/1% niclosamide mixture. Experiments revealed that niclosamide is approximately 100-fold more toxic than TFM (WRITE THE LC₅₀s for both), while the toxicity of TFM was increased by 3-fold after addition of 1% niclosamide (LC₅₀). Exposures to mixtures of various permutations of the TU of each chemical revealed that TFM and niclosamide have a synergistic (i.e. greater than additive) effect in rainbow trout. Although it is thought that TFM and niclosamide have a common mechanism of action (uncoupling of mitochondrial oxidative phosphorylation), the current findings suggest that they may also have other, independent modes of action, which may exacerbate their toxicity.

Abstract: 130

A COMPARISON OF STUDENT PERSPECTIVES AND IMPACT ON COMPREHENSION AND RETENTION OF PAPER VERSUS E-READERS

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The ongoing advance of technology has allowed students easy access to materials on electronic devices; as a result, “e-books” have become increasingly common. This study evaluates the impact of electronic tablets on student reading comprehension and retention. Data from 43 university students were used in the analysis. A perspective survey was implemented to collect data about demographics, and the use and perception of electronic devices. 56% of students preferred reading on electronic devices in general, while 44% of students stated they preferred to use an electronic device for reading an academic textbook. Participants were then randomly divided into one of two groups, electronic or paper, and were given a validated passage to read, in a controlled laboratory setting. In order to evaluate the reading comprehension of the participants, students were presented with a multiple-choice quiz about the passage, which they all answered on paper. One week and four weeks post-comprehension, participants were given similar questions regarding the same passage and were assessed on their retention of information. Participants of the paper group answered correctly, on average, more often than those of the electronic group, when being tested for both reading comprehension and retention. The long-range goal of this study is to apply the results to inform the ways that students both learn and demonstrate their knowledge.

Abstract: 131

AROGENATE DEHYDRATASE: IMPORT INTO *ARABIDOPSIS THALIANA* CHLOROPLASTS

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Plastids, especially chloroplasts, are essential organelles to the plants in which they reside, as well as all aerobic life on Earth. As the site of photosynthesis, they provide the O₂ and organic molecules that support virtually all life. Plastids are also the site of other essential biochemical processes, including the biosynthesis of essential amino acids such as L-Phenylalanine, which is produced by the enzyme Arogenate Dehydratase (ADT). There are six ADT isozymes in the model plant *Arabidopsis thaliana*. In previous studies, fluorescently labeled ADT isozymes were shown to localize to chloroplasts using microscopy and were found in isolated soluble sub-chloroplast fractions. There has not yet been a study on the import pathway used by ADTs. It is hypothesized that ADT isoenzymes 4 and 6 from *A. thaliana* use the Toc-Tic pathway to gain entry to the chloroplast and that they are targeted using a transit peptide which is cleaved following import to the stroma. Using ³⁵S-Methionine labeled ADT enzymes and freshly isolated *A. thaliana* chloroplasts, in vitro protein import assays were performed to confirm that ADT4 and ADT6 are imported into chloroplasts. SDS-PAGE was used to confirm the import of the proteins and to confirm that the transit peptides were cleaved, which was evident by a molecular weight shift of the protein to a lower molecular weight, mature form.

Abstract: 132

TWO PACEMAKER ACTIVITIES IN THE MOUSE SMALL INTESTINE; DISCOVERING THE UNDERLYING MOTOR PATTERNS BY PHASE-AMPLITUDE COUPLING

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Motor patterns within the gut have been described as either segmentation or propulsion. Segmentation has been hypothesized to involve two electrical myogenic pacemaker cells, the omnipresent slow wave activity that originates from the Interstitial Cells of Cajal (ICC) of the myenteric plexus (ICC-MP) and the stimulus-dependent rhythmic transient depolarizations originating from the ICC of the deep muscular plexus (ICC-DMP). However, little is known regarding the ICC-DMP network. It is believed that the wave parameters of ICC-DMP interact with ICC-MP in a variety of different manners, resulting in different electrical recordings. Within this study, a phase-amplitude coupling model was used to test this hypothesis and increase knowledge of the ICC-DMP network. The waves obtained through the model were compared to the original electrical recordings to determine if the wave properties of ICC-MP and ICC-DMP were interacting. Results demonstrate that the phase of ICC-DMP can modulate the amplitude of ICC-MP forming an altered wave pattern. In addition, evidence may demonstrate that at high temperatures, ICC-DMP may act in a dominant manner, thus allowing the phase of ICC-MP to modulate the amplitude of ICC-DMP. Additional studies must be conducted at high temperatures to further test this hypothesis.

Abstract: 133

DISCOVERY OF NEW ANTIBIOTICS AGAINST GRAM-NEGATIVE BACTERIA

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Antibiotic resistance is an inevitable consequence of the over-prescription and misuse of medicinal antibiotics. The usage of naturally produced antimicrobial compounds, such as bacteriocins and essential oils, that are able to provide effective treatment with limited side effects are ideal and generally underexploited. Here, we screened the sensitivity of 6 strains of ciprofloxacin-resistant *Escherichia coli* to compounds with known antimicrobial activity, as well as nisin, a bacteriocin used as a natural food preservative with known activity against gram-positive bacteria. Most of the *E. coli* strains showed similar sensitivity to these compounds as the wild type control. Nisin presented a strong inhibition when it was combined with buffered citric acid, a membrane disturbing chelator despite the typical resistance of gram-negative bacteria. To further explore the mode of action of nisin against gram-negative bacteria, we completed a chemical-genetic screen that revealed 4 supersensitive strains with mutations in genes involved with DNA replication. An *E. coli* plasmid replication assay was used as a proxy to test whether nisin perturbed chromosomal DNA replication. No evidence of DNA replication inhibition was observed using this assay, but additional flow cytometry-based experiments are underway to directly test for effects of nisin on DNA replication. This research provides evidence that nisin has increased antibiotic activity against specific ciprofloxacin-resistant bacteria, and reveals the existence of a novel mode of action of nisin against gram-negative bacteria. With increasing knowledge of the antimicrobial activity of this natural compound, its therapeutic potential as a supplement to conventional antibiotics may be exploitable.

Abstract: 134

MECHANISMS OF AMMONIA EXCRETION IN AN ANCIENT VERTEBRATE, THE SEA LAMPREY (*P. MARINUS*)

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Sea lamprey (*Petromyzon marinus*) are a jawless fish that belong to the superclass Agnatha. Although they are an ancient species of vertebrate, little is known about how they excrete toxic nitrogenous waste products such as ammonia. In the present study, the mechanisms of ammonia excretion were studied in larval sea lampreys by measuring excretion rates at different water pHs, or in the presence of different pharmacological agents. Exposure to amiloride resulted in a decrease in ammonia excretion, which suggests that ammonia excretion may be linked to sodium uptake, possibly via a Na^+ - NH_4^+ exchange mechanism in sea lamprey. The results from the exposure to the pH6 water suggest that NH_3 may also be trapped as NH_4^+ following the excretion of NH_3 across the gills. However, the lack of effect of acetazolamide exposure suggests that carbonic anhydrase-mediated acidification of the water crossing the gills does not likely play a role in ammonia excretion. These findings suggest that the most ancient jawless vertebrates excreted nitrogenous wastes in a mechanism similar to current model for fish nitrogenous waste excretion. Further studies with alkaline pH and sodium-free water will allow further elucidation of the mechanism by which these phylogenetically ancient vertebrates excrete ammonia.

Abstract: 135

ASSESSING BEE GENUS ASSEMBLAGES CORRESPONDING TO ELEVATIONAL GRADIENTS IN THE ATACAMA DESERT, CHILE

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Bees play a key role in pollination of diverse regions, one of which is arid and mountainous ecosystems. Often such regions have highly diverse pollinator communities, with adaptations to enable effective pollination even when climatic conditions are erratic. In order to assess how bee genus assemblages change, samples were collected at 500m altitude intervals from 500m to 4500m from the Atacama Desert in Chile. Samples were collected after both winter and summer rainfall periods in 2013. This study involved the identification of approximately 520 bee samples to the genus level for two transects Chañaral and Taltal. I hypothesized that with increasing elevation and rainfall, the number of genera found would also increase. The results concluded that the highest numbers of genera were found at 2000 m a.s.l and 3000 m a.s.l for Chañaral and Taltal correspondingly. The lowest numbers of genera were found at 3000 m a.s.l and 4000 m a.s.l for Chañaral and Taltal respectively. Overall, there was no strong correlation between increasing elevation and number of genera. The bee samples must be resolved further and identified to species level to test the hypothesis. Our future work includes a principle component analysis and ANOVA to test whether winter rainfall species have their closest relatives in summer rainfall regions and vice versa.

Abstract: 136

ELUCIDATING THE GILL'S ROLE IN ION-REGULATION AND ACID/BASE EQUILIBRIA WITHIN STENOHALINE FRESHWATER-ADAPTED AMAZONIAN RAYS, *POTAMOTRYGON* SPP.

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The purpose of this section of work with Amazonian freshwater ray (family: *Potamotrygonidae*) iono-regulation is to investigate how modification in gill structure/function has facilitated the successful adaptation of these elasmobranchs (class: Chondrichthyes) to an ion- poor, freshwater environment, and to investigate what role the gills play in the freshwater stenohaline strategies that these animals display. It is hypothesized that apical gill $\text{Cl}^-/\text{HCO}_3^-$ & Na^+/H^+ antiporters and gill basolateral Na^+/K^+ and H^+ ATPases will exhibit active ion uptake similar to freshwater Teleostei (bony fish) in response to a hypotonic environment, however the structure of the *Potamotrygon* gill is hypothesized to have been relatively conserved in relation to the marine elasmobranch cousins of this genus, *Dasyatis sabina* (Ballantyne & Robinson, 2010). This is proposed because gill-filament protein-function would be thought capable of displaying a more rapid response to environment than structural changes/replacement/removal of these transmembrane proteins from an evolutionary standpoint, hence suggesting that structural similarities may exist between these freshwater species and marine or estuarine rays. This study consists of immunolabelling the proposed conserved gill membrane ATPases and antiporters mentioned above under acidic, basic, sham HCl, and control conditions, to elucidate the role the gill is playing in these unique organisms with regards to ionoregulation and acid-base equilibrium.

Abstract: 137

ASSESSING MYCORRHIZAL SPORE DENSITY AND INOCULUM POTENTIAL OF SOILS OBTAINED FROM STEEVES LAKE SHORELINE: A REMEDIATION SITE AT THE COLOMAC MINE, NWT

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An open pit gold mine active throughout the 1990s, Colomac has since been closed and subject to remediation efforts. Currently, monitoring of the site focuses on above ground plant communities and no attention has yet been given to the belowground organisms, specifically mycorrhizal fungi. The purpose of this study is quantify arbuscular mycorrhizal fungi (AMF) and dark septate endophytes (DSE) along the Steeves Lake shoreline. This shoreline was built during remediation efforts in order to contain and filter hydrocarbons that were entering Steeves Lake from historic spills. In order to determine if AMF were present, fungal propagules were isolated and bioassays were used to measure the viability of soil propagules, using *Phalaris arundinaceae* and *Epilobium angustifolium* as trap plants. Abiotic factors including soil nutrient levels (K, P), distance of the samples from historic spills, and soil moisture were considered as explanatory variables. The presence of AMF propagules has been confirmed in all soil samples with spore densities ranging from 2 to 338 spores per gram of dry soil. All plants were colonized by AMF, DSE, or both. In the colonized roots of both trap plants hyphae, arbuscules and vesicles were observed. In *P. arundinaceae* AMF colonization ranged from 1 to 78%. In *E. angustifolium* AMF colonization ranged from 5 to 98%. Plant performance (shoot and root biomass), root surface area, and length were determined and their relationship with AMF levels and site abiotic factors assessed.

List of Presentations by Abstract Number

#	Presenting Author	Affiliation	
001	Fraunberger, E.	McMaster	Talk
002	Thrasher, C.	Windsor	Talk
003	Bieniek, A.	Laurentian	Talk
004	Pichette, J.	Laurentian	Talk
005	D'Addario, A.	Carleton	Talk
006	Cervi, S.E.	Windsor	Talk
007	Sullivan, B.	Carleton	Poster
008	Market, M.	Windsor	Talk
009	Okpara, P.	Windsor	Talk
010	Santangelo, J.	Toronto	Talk
011	Jain-Schlaepfer, M.R.	Carleton	Talk
012	Scott, K.	Windsor	Talk
013	Nader, G.	Windsor	Talk
014	McNeil, A.	UWO	Poster
015	Yabut, J.	McMaster	Poster
016	Marchand, K.	Laurentian	Talk
017	Bastien, C.M.	Laurentian	Talk
018	Fauvelle, C.	Laurentian	Talk
019	Bielaska-Da Silva, J.	UWO	Talk
020	Kochar, D.	UWO	Talk
021	Rethi, S.	UWO	Talk
022	Nicholls, K.	Laurentian	Talk
023	Engyel, M.	Laurentian	Talk
024	Hwang, M-H	McMaster	Talk
025	Hadden, C.I.L.M.	Carleton	Poster
026	Violin, A.	Laurentian	Talk
027	DiStefano, S.	Windsor	Talk
028	Landrigan, J.	Carleton	Poster
029	Varela, B.	Laurentian	Talk
030	Dron, J.S.	UWO	Talk
031	Serré, J.	Laurentian	Talk
032	Sameem, M.	Windsor	Talk
033	Zygowska, A.	Windsor	Talk
034	Ho, R.	UWO	Talk
035	Kurtzke, Y.J.G.	UWO	Talk
036	Kazmirchuk, T.	Carleton	Talk
037	Harris, N.	UWO	Talk
038	Kosiw, J.	Laurentian	Talk
039	Knoops, P.	UWO	Talk
040	O'Brien, M.E.	UWO	Talk
041	Aulakh, G.	Laurentian	Talk
042	Marko, V.	McMaster	Talk
043	Mehdi, H.	Windsor	Talk

044	Wilkes, S.	Laurentian	Talk
045	Marshall, T.J.	UWO	Talk
046	Fairhead, C.	McMaster	Talk
047	Samani, A.M.V.	UWO	Talk
048	Pathmarajah, M.	McMaster	Talk
049	Dykstra, M.	Laurentian	Talk
050	Chen, W.	Toronto	Talk
051	Hadj-Moussa, H.	Carleton	Poster
052	Logan, S.	Carleton	Poster
053	Bansal, S.	Carleton	Poster
054	Nguyen, T.D.	Carleton	Poster
055	Vaz, B.	McMaster	Talk
056	Rancourt, K.	Laurentian	Talk
057	Thayaparan, D.	McMaster	Talk
058	Loney, C.L.	Laurentian	Talk
059	Paishegwon, R.D.	Laurentian	Talk
060	Noble, N.	Laurentian	Talk
061	Rose, C.D.	UWO	Talk
062	Paquette, A.J.	Toronto	Talk
063	Edgar, N.	UWO	Talk
064	Hawley, Z.	UWO	Talk
065	Tieu, B.	UWO	Talk
066	Bentley-DeSousa, A.S.	UWO	Talk
067	Zubic, A.	Windsor	Talk
068	Williamson, C.	Laurentian	Talk
069	Smith, R.	UWO	Talk
070	Bui, J.	UWO	Talk
071	Fong, A.	Laurentian	Talk
072	Bhardwaj, S.	Windsor	Talk
073	Ilunga, J.	Laurentian	Talk
074	Power, S.	Windsor	Talk
075	MacDonald, J.	Wilfrid Laurier	Talk
076	Montgomery, J.	Laurentian	Talk
077	Gray, R.D.	UWO	Talk
078	Park, S-B	Carleton	Poster
079	Hyland, L.	Carleton	Poster
080	Harry, A.J.	Laurentian	Talk
081	Renwick, M.G.	UWO	Talk
082	Hafeez, A.	Toronto	Talk
082	Hanke, R.	Toronto	Talk
082	Tahir, Z.	Toronto	Talk
083	Brown, M.A.	UWO	Talk
084	Karpinski, E.	McMaster	Talk
085	Ehrenfellner, W.	Wilfrid Laurier	Talk
086	Dierolf, J.	UWO	Talk

087	Merwin, N.	McMaster	Talk
088	Cameron-Dermann, L.	Ottawa	Talk
089	Kou, H.H.	McMaster	Talk
090	Hay, R.	Carleton	Poster
091	Willoughby, P.M.	UWO	Talk
092	Hurtado, E.	UWO	Talk
093	Nguyen, F.	Toronto	Talk
094	Dias, K.	Toronto	Talk
094	Ahmadli, F.	Toronto	Talk
095	Péon, H.E.	Wilfrid Laurier	Talk
096	Ma, J.H.W.	McMaster	Talk
097	Rajic, M.	Toronto	Talk
098	Pansuriya, S.	McMaster	Talk
099	Baassiri, H.M.	UWO	Talk
100	Mak, A.	Wilfrid Laurier	Talk
101	Ghai C, M.	Windsor	Talk
102	Dialani, N.	UWO	Talk
103	Hinge, M.	Windsor	Talk
104	D'Souza, C.	Toronto	Talk
105	Khanna, A.	Toronto	Talk
106	Sreetharan, S.	McMaster	Talk
107	Szponarski, K.	Wilfrid Laurier	Talk
108	Hamelin, D.	Laurentian	Talk
109	Thuswaldner, J.	Carleton	Talk
110	Yaworski, R.	Carleton	Talk
111	Kissoondoyal, A.	UWO	Talk
112	Arain, U.	Toronto	Talk
113	Hammoud, M.	Wilfrid Laurier	Talk
114	Golemiec, M.	Wilfrid Laurier	Poster
115	Jones, L.	Wilfrid Laurier	Poster
116	Pomeroy, B.	Wilfrid Laurier	Poster
117	Hsieh, D.	McMaster	Poster
118	Veras, M.A.	UWO	Poster
119	Brenner, T.	Wilfrid Laurier	Poster
120	Jolley, S.N.	UWO	Poster
121	Sabourin, S.	Ottawa	Poster
122	Seaver, K.	Wilfrid Laurier	Poster
123	Ong, F.	McMaster	Poster
124	McPherrin, B.	McMaster	Poster
125	Siracusa, L.	Wilfrid Laurier	Poster
126	Kaur, Y.	McMaster	Poster
127	Yan, R.	McMaster	Poster
128	Hovey, O.	Carleton	Poster
129	Hepditch, S.	Wilfrid Laurier	Poster
130	Grabka, C.	Toronto	Poster
131	Wilcox, H.	Wilfrid Laurier	Poster

132	Maurice, Y.N.	McMaster	Poster
133	Graf, M.	Carleton	Poster
134	Glennie, M.F.	Wilfrid Laurier	Poster
135	Satti, S.H.	York	Poster
136	Rossi, M.	Wilfrid Laurier	Poster
137	Hamp, R.	Wilfrid Laurier	Poster

List of Presentations by Presenting Author

#	Presenting Author	Affiliation	
094	Ahmadli, F.	Toronto	Talk
112	Arain, U.	Toronto	Talk
041	Aulakh, G.	Laurentian	Talk
099	Baassiri, H.M.	UWO	Talk
053	Bansal, S.	Carleton	Poster
017	Bastien, C.M.	Laurentian	Talk
066	Bentley-DeSousa, A.S.	UWO	Talk
072	Bhardwaj, S.	Windsor	Talk
019	Bielaska-Da Silva, J.	UWO	Talk
003	Bieniek, A.	Laurentian	Talk
119	Brenner, T.	Wilfrid Laurier	Poster
083	Brown, M.A.	UWO	Talk
070	Bui, J.	UWO	Talk
088	Cameron-Dermann, L.	Ottawa	Talk
006	Cervi, S.E.	Windsor	Talk
050	Chen, W.	Toronto	Talk
005	D'Addario, A.	Carleton	Talk
102	Dialani, N.	UWO	Talk
094	Dias, K.	Toronto	Talk
086	Dierolf, J.	UWO	Talk
027	DiStefano, S.	Windsor	Talk
030	Dron, J.S.	UWO	Talk
104	D'Souza, C.	Toronto	Talk
049	Dykstra, M.	Laurentian	Talk
063	Edgar, N.	UWO	Talk
085	Ehrenfellner, W.	Wilfrid Laurier	Talk
023	Engyel, M.	Laurentian	Talk
046	Fairhead, C.	McMaster	Talk
018	Fauvelle, C.	Laurentian	Talk
071	Fong, A.	Laurentian	Talk
001	Fraunberger, E.	McMaster	Talk
101	Ghai C, M.	Windsor	Talk
134	Glennie, M.F.	Wilfrid Laurier	Poster
114	Golemiec, M.	Wilfrid Laurier	Poster
130	Grabka, C.	Toronto	Poster
133	Graf, M.	Carleton	Poster
077	Gray, R.D.	UWO	Talk
025	Hadden, C.I.L.M.	Carleton	Poster
051	Hadj-Moussa, H.	Carleton	Poster
082	Hafeez, A.	Toronto	Talk
108	Hamelin, D.	Laurentian	Talk
113	Hammoud, M.	Wilfrid Laurier	Talk

137	Hamp, R.	Wilfrid Laurier	Poster
082	Hanke, R.	Toronto	Talk
037	Harris, N.	UWO	Talk
080	Harry, A.J.	Laurentian	Talk
064	Hawley, Z.	UWO	Talk
090	Hay, R.	Carleton	Poster
129	Hepditch, S.	Wilfrid Laurier	Poster
103	Hinge, M.	Windsor	Talk
034	Ho, R.	UWO	Talk
128	Hovey, O.	Carleton	Poster
117	Hsieh, D.	McMaster	Poster
092	Hurtado, E.	UWO	Talk
024	Hwang, M-H	McMaster	Talk
079	Hyland, L.	Carleton	Poster
073	Ilunga, J.	Laurentian	Talk
011	Jain-Schlaepfer, M.R.	Carleton	Talk
120	Jolley, S.N.	UWO	Poster
115	Jones, L.	Wilfrid Laurier	Poster
084	Karpinski, E.	McMaster	Talk
126	Kaur, Y.	McMaster	Poster
036	Kazmirchuk, T.	Carleton	Talk
105	Khanna, A.	Toronto	Talk
111	Kissoondoyal, A.	UWO	Talk
039	Knoops, P.	UWO	Talk
020	Kochar, D.	UWO	Talk
038	Kosiw, J.	Laurentian	Talk
089	Kou, H.H.	McMaster	Talk
035	Kurtzke, Y.J.G.	UWO	Talk
028	Landrigan, J.	Carleton	Poster
052	Logan, S.	Carleton	Poster
058	Loney, C.L.	Laurentian	Talk
096	Ma, J.H.W.	McMaster	Talk
075	MacDonald, J.	Wilfrid Laurier	Talk
100	Mak, A.	Wilfrid Laurier	Talk
016	Marchand, K.	Laurentian	Talk
008	Market, M.	Windsor	Talk
042	Marko, V.	McMaster	Talk
045	Marshall, T.J.	UWO	Talk
132	Maurice, Y.N.	McMaster	Poster
014	McNeil, A.	UWO	Poster
124	McPherrin, B.	McMaster	Poster
043	Mehdi, H.	Windsor	Talk
087	Merwin, N.	McMaster	Talk
076	Montgomery, J.	Laurentian	Talk
013	Nader, G.	Windsor	Talk
093	Nguyen, F.	Toronto	Talk

054	Nguyen, T.D.	Carleton	Poster
022	Nicholls, K.	Laurentian	Talk
060	Noble, N.	Laurentian	Talk
040	O'Brien, M.E.	UWO	Talk
009	Okpara, P.	Windsor	Talk
123	Ong, F.	McMaster	Poster
059	Paishegwon, R.D.	Laurentian	Talk
098	Pansuriya, S.	McMaster	Talk
062	Paquette, A.J.	Toronto	Talk
078	Park, S-B	Carleton	Poster
048	Pathmarajah, M.	McMaster	Talk
095	Péon, H.E.	Wilfrid Laurier	Talk
004	Pichette, J.	Laurentian	Talk
116	Pomero, B.	Wilfrid Laurier	Poster
074	Power, S.	Windsor	Talk
097	Rajic, M.	Toronto	Talk
056	Rancourt, K.	Laurentian	Talk
081	Renwick, M.G.	UWO	Talk
021	Rethi, S.	UWO	Talk
061	Rose, C.D.	UWO	Talk
136	Rossi, M.	Wilfrid Laurier	Poster
121	Sabourin, S.	Ottawa	Poster
047	Samani, A.M.V.	UWO	Talk
032	Sameem, M.	Windsor	Talk
010	Santangelo, J.	Toronto	Talk
135	Satti, S.H.	York	Poster
012	Scott, K.	Windsor	Talk
122	Seaver, K.	Wilfrid Laurier	Poster
031	Serré, J.	Laurentian	Talk
125	Siracusa, L.	Wilfrid Laurier	Poster
069	Smith, R.	UWO	Talk
106	Sreetharan, S.	McMaster	Talk
007	Sullivan, B.	Carleton	Poster
107	Szponarski, K.	Wilfrid Laurier	Talk
082	Tahir, Z.	Toronto	Talk
057	Thayaparan, D.	McMaster	Talk
002	Thrasher, C.	Windsor	Talk
109	Thuswaldner, J.	Carleton	Talk
065	Tieu, B.	UWO	Talk
029	Varela, B.	Laurentian	Talk
055	Vaz, B.	McMaster	Talk
118	Veras, M.A.	UWO	Poster
026	Violin, A.	Laurentian	Talk
131	Wilcox, H.	Wilfrid Laurier	Poster
044	Wilkes, S.	Laurentian	Talk
068	Williamson, C.	Laurentian	Talk

091	Willoughby, P.M.	UWO	Talk
015	Yabut, J.	McMaster	Poster
127	Yan, R.	McMaster	Poster
110	Yaworski, R.	Carleton	Talk
067	Zubic, A.	Windsor	Talk
033	Zygowska, A.	Windsor	Talk