INVESTIGATING THE INTERACTION OF CANONICAL SMAD AND NON-CANONICAL TGF- β INDUCED PATHWAYS IN THE FORMATION OF EMT AS A MODEL FOR CATARACTS IN THE VERTEBRATE LENS

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Cataracts are the leading cause of blindness worldwide. The surgical removal of cataracts is a common and mostly successful method of treatment, although there are still chances of postsurgical complications, such as secondary cataracts, also known as posterior capsular opacification (PCO). PCO occurs after surgery when the remaining lens epithelial cells (LECs) in the capsular bag are induced to undergo epithelial to mesenchymal transition (EMT), which results in a change in LEC morphology causing a proteinaceous opacity that leads to a loss of transparency. Previous research has shown that the induction of cataract formation in the lens is due to the signalling molecule, TGF- β , through multiple pathways, including TGF- β /Smad, β catenin/Wnt, and Rho/ROCK. The interactions between three key transcription factors involved in these pathways were examined using α -SMA as the molecular marker for EMT. Rat LEC explants were used as an *ex vivo* model and induced with TGF- β to undergo EMT then treated with either ICG-001 or SIS3 inhibitors. It was found that Smad3 inhibition prevented nuclear translocation of β -catenin and MRTF-A. However, the inhibition of β -catenin did not prevent nuclear translocation of MRTF-A, but resulted in an absence of EMT. In addition, β-catenin inhibition decreased the amount of phosphorylated Smad3. It is suggested that β -catenin may play a role in decreasing the expression of TBRI and TBRII, involved in the phosphorylation of Smad3, and that Smad3 may be a co-factor of MRTF-A. The findings illustrated in this thesis have implications for further research in therapeutics for the treatment of cataracts.

Oral Presentation Group A: Cell, Molecular, and Genetics

ACHAETE-SCUTE FAMILY BHLH TRANSCRIPTION FACTOR 2 REGULATES PROLIFERATION IN HUMAN TROPHOBLASTS

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The placenta is a transient organ that supports development and growth of the fetus. The parenchymal cells of the placenta are trophoblasts. Trophoblast progenitor cells, called cytotrophoblasts, differentiate into specialized trophoblast subtypes such as syncytiotrophoblasts and extravillous trophoblasts, which perform the majority of placental functions. The regulation of cytotrophoblast differentiation is not well known. Achaete-scute bHLH family transcription factor 2 (ASCL2) is a transcription factor known to play a role in maintaining stem cell traits in specific cell lineages, which we have shown to be expressed in human placenta. Thus, we hypothesize that ASCL2 maintains cytotrophoblast progenitor traits and prevents differentiation. In JEG3 cytotrophoblast cells, ASCL2 was shown to localize within the nucleus. Using gRT-PCR and western blotting, we found that ASCL2 mRNA and protein levels decreased upon cyclic adenosine monophosphate (cAMP) induced differentiation (n=3, p<0.05). We successfully transduced JEG3 trophoblast cells with pLX304 vectors encoding ASCL2, resulting in a 7-fold increase in ASCL2 expression compared to controls. ASCL2 overexpression did not affect expression of differentiation markers such as OVOL1, ERVFRD1, CGB, ERVV1, and ERVV2 upon cAMP treatment (n=3, p>0.05). However, cell growth rate increased by 90% in cells overexpressing ASCL2 at 72h (n=6, p<0.05). In conclusion, we demonstrate that ASCL2 does not play a role in trophoblast differentiation, however, it does regulate trophoblast proliferation. Future studies aim to determine ASCL2's role in proliferation via direct measures using bromodeoxyuridine assay and immunofluorescence staining for proliferation markers, such as Ki-67 and phospho-histone H3.

Oral presentation in Group A: Cell, Molecular & Genetics

DETERMINATION OF JAK3 AS A DRIVER GENE IN A ${\scriptstyle \bigtriangleup}$ SPIB/PU.1 MOUSE MODEL OF BALL

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Leukemia is a disease caused by driver mutations that confer survival and growth advantages to blood cells. Our research uses a mouse model of B cell acute lymphoblastic leukemia (B-ALL) initiated by deletion of genes encoding the related transcription factors SpiB and PU.1. These mice have a median survival of 18 weeks, suggesting that additional driver mutations are involved in B-ALL development. Whole exome sequencing on three Δ SpiB/PU.1 mice showed that two mice harbored missense mutations in Janus kinase 3 (Jak3): R633H, V670A, and T844M. Considering the recurrent identification of Jak3 mutations in human leukemia, it was hypothesized that Jak3^{R653H}, Jak3^{T844M}, and/or Jak3^{V670A} are driver mutations conferring growth advantages to pre-B cells in ΔSpiB/PU.1 mice. Sanger sequencing revealed that 26% (5/19) of ΔSpiB/PU.1 mice possessed mutations in the R653 codon, majority of which are R653H. Pre-B cells retrovirally expressing R653H, T844M, V670A, or T844M+V670A (double mutant, as found in a single mouse from whole exome sequencing) exhibited a growth advantage in low interleukin-7 (IL-7) concentration (0.5%) when competing with wild type pre-B cells (p<0.05). Furthermore, pre-B cells expressing each Jak3 mutation, except the double mutant, demonstrated superior viability in low IL-7 concentration compared to cells expressing an empty vector (p<0.05). In summary, a large fraction of analyzed Δ SpiB/PU.1 mice harbour R653 mutations while pre-B cells expressing mutant Jak3 demonstrated superior growth and viability. These results strongly suggest that specific mutations within Jak3 are driver mutations that cooperate with loss of PU.1 and Spi-B to drive leukemogenesis.

Oral presentation Group A: Cell, Molecular, & Genetics

Elucidating Signaling Pathways Involved in Syncytiotrophoblast Development

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The placenta is a transient organ that forms during pregnancy. The trophoblast cell layer within the placenta is essential for fetal growth and healthy development. Trophoblast progenitor cells, known as cytotrophoblasts, proliferate and differentiate via fusion with the overlying syncytiotrophoblasts. This fusion event is unique to the placenta, and mechanisms regulating this differentiation process are poorly understood. Disruption to the regulation of differentiation can lead to in utero complications such as preeclampsia and intrauterine growth restriction. OVO-like 1 (OVOL1) is a transcription factor involved in driving differentiation into syncytiotrophoblasts by supressing factors that promote the progenitor cell state of cytotrophoblasts. Cyclic AMP activation induces differentiation, and therefore OVOL1 expression. Cyclic AMP is implicated in many pathways, including the Protein Kinase A, β -Catenin, and Arylhydrocarbon Receptor pathways, indicating possible upstream regulators of OVOL1. Inhibition of these pathways in Jeg3 cells negatively affected the expression of differentiation markers including Syncytin-2, ERVV1, ERVV2, and HSD11B2, however, these pathways do not appear to be involved in the regulation of OVOL1 at the RNA level (N=3, n=2, p<0.05). Ectopic expression of OVOL1 in Jeg3 cells via lentivirus showed significant increase in OVOL1 at both the RNA and protein level following treatment of cAMP. This suggests that cAMP inducible signalling pathways involved in syncitialization may induce posttranscriptional mechanisms of OVOL1 regulation. By using inhibitors of cAMP inducible signalling pathways, as well as inhibitors of proteases and deacetylases, we investigate the pathways involved in the regulation of the OVOL1 transcription factor.

Oral Presentation: Group A

THE NON-SPECIFICITY OF TRANSCRIPTION FACTORS IN SACCHAROMYCES CEREVISIAE

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Transcription initiation is regulated by transcription factors (TFs). Traditionally, individual TFs are thought to regulate a restricted set of genes, giving rise to a unique phenotype. Recent studies in *Drosophila melanogaster* suggest that TFs are much more non-specific than the traditional view as multiple TFs regulate the expression of genes required to give rise to a phenotype. To test whether TFs are specific or non-specific in *Saccharomyces cerevisiae*, three TFs (*GAL4, PHO2, NDT80*) will be knocked out and substituted with a randomly selected array of ten TFs. If TFs are indeed specific, there will be no rescue of the phenotypes by any of the ten TFs expressed from the three loci except for the original TF. However, rescue of any of the three phenotypes by a TF not normally encoded by the locus indicates non-specificity. Mutant strains of yeast containing either a knockout, reinsertion or substitution at the *PHO2* locus have been made and are being tested for acid phosphatase activity. *NDT80* locus plasmids have been cloned in bacteria and are being transformed into yeast in preparation for subsequent mating, diploid selecting and sporulation. Technical problems with the vector used to knockout the *GAL4* did not allow plasmid constructs for transformation of yeast to be established.

Oral Presentation – A: Cell, Molecular, and Genetics

ANALYSIS OF COPY NUMBER VARIANTS IN ASSOCIATION WITH LOCALIZED REGIONS OF HETEROZYGOSITY IN OUTBRED MICE

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

Oral presentation; Group A: Cell, Molecular & Genetics

Title: UNDERSTANDING THE EFFECTS OF SERINE AND GLYCINE METABOLISM ON P53-REGULATED CALTHRIN-MEDIATED ENDOCYTOSIS.

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Cancer cells require receipt of chemical signals and uptake of nutrients from the tumor microenvironment, processes that depend on myriad cell surface receptors and transporters. These cell surface proteins are controlled by clathrin-mediated endocytosis (CME), indicating that regulation of clathrin may in turn control aspects of tumor progression. How CME may be altered in cancer remains poorly understood. Recently, serine metabolism has emerged as a unique requirement for certain cancer cells. Serine and glycine are important precursors for the synthesis of lipids, nucleic acids, and proteins. The first committed step in the biosynthesis of serine and glycine requires phosphoglycerate dehydrogenase (PHGDH) and the glycolytic intermediate 3-phosphoglycerate (3PG). Importantly, deficiencies in serine metabolism lead to activation of p53, which is known to interact with clathrin. To understand how serine metabolism controls CME, we have used inhibition of PHDGH with the inhibitors NCT 503, CBR-5884, and PKUMDL-WQ-2101, and monitored the membrane traffic of transferrin receptor, a marker of CME. Our results to date indicate that treatment with NCT-503 reduces cell surface levels of transferrin receptor, indicating that perturbations of serine metabolism impact endomembrane traffic. Additionally. CBR-5884 has been shown to alter cell morphology. Future directions include determining the mechanism of action for inhibition. Due to the importance of serine and glycine in macromolecule production, the accumulation of these amino acids if advantageous for the survival and proliferation of cancer cells. Understanding the effects of serine and glycine metabolism on cellular processes such as CME can provide a new route for the development of drugs to target certain types of cancer.

ORAL PRESENTATION

Group: Cell, Molecular, and Genetics

REGULATION OF CANCER CELL MIGRATION BY CALCIUM

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Cell migration serves a critical role in physiological events such as tissue formation and maintenance. During such processes, individual cells sense and integrate numerous biochemical and mechanical cues from their microenvironment and migrate to specific locations according to those cues. Cancer cells, for instance, sample the rigidity of their surroundings to migrate to stiffer areas and thereby metastasize. Focal adhesions (FAs) are the main site of cell-extracellular matrix (ECM) contact. Cells exert traction forces at FAs and tug at the ECM to sample its rigidity. However, the exact mechanism through which stiffness signals are transduced into the cell through FAs is unknown. Recent reports show that stretch-activated cation channels localized in FAs may be involved in this process. This brought me to hypothesize that tugging on the ECM opens stretchactivated calcium channels in FAs and causes calcium entry as an indicator of stiffness. In the past year I have tested this hypothesis experimentally and showed that the local concentration of calcium at FAs indeed undergoes fast fluctuations over time. Such calcium influx at FAs decreases when we perturbed the ability of FAs to tug at their environment. In addition, interfering with calcium entry through calcium channels at FAs prolongs FA lifetime, which may slow down migration. Collectively, my findings will further our understanding of how cells sense the stiffness of their environment. It also reveals calcium and calcium-activated signaling pathways as prominent pharmacological targets to inhibit pathological processes dependent on cell migration towards stiffer tissue, such as cancer metastasis.

Oral and poster presentation, group A: Cell, Molecular & Genetics

THE EFFECTS OF BLOCKING WNT LIGAND SECRETION ON THE DIFFERENTIATION OF

F9 TERATOCARCINOMA CELLS

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What signaling plays a critical role in the establishment of extraembryonic endoderm in the early embryo, where activation of the canonical and non-canonical pathways induces primitive endoderm (PrE) and parietal endoderm (PE), respectively. Previous studies have shown that retinoic acid (RA) induces mouse F9 teratocarcinoma cells to differentiate into PrE: where subsequent treatment with dibutvrvl-cAMP (db-cAMP) further differentiates cells into PE. Porcupine (PORCN), a membrane-bound resident of the endoplasmic reticulum, is responsible for the post-translational palmitoylation of Wnt ligands required for secretion. This study focuses on the effects of the PORCN inhibitors, LGK-974 and C59, on the RA-induced differentiation of F9 cells and investigates whether PORCN inhibition blocks the secretion of Wnt(s) required to induce PrE and PE formation. It is hypothesized that PORCN function, and thus proper posttranslational modification of Wnt ligands, is required for F9 cell differentiation towards an ExE lineage. Relative attenuation of differentiation marker cytokeratin 8/18 was observed compared to the RA positive control when RA treated cells were co-treated with 10nM of C59 daily for 4 days (n=3). Preliminary data suggests that PORCN-inhibitor C59 may potentially block canonical signaling without major cell cytotoxicity, however additional experiments must be conducted to verify statistical significance of results.

Oral presentation Cell, Molecular, & Genetics

A BALANCING ACT: THE CYCLIN=LIKE PROTEIN PROMOTES CANCER FORMATION IN THE LIVER

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Hepatocellular carcinoma (HCC) is among the most aggressive and prevalent forms of liver cancer. With an increasing incidence in Canada and a five-year survival rate of 19%, a more comprehensive understanding of this disease is imperative in order to consider treatment options. Numerous lifestyle factors may underlie HCC, including chronic alcoholism, non-alcoholic fatty liver disease (NAFLD) and non-alcoholic steatohepatitis (NASH). These factors are known to elicit liver injury, inflammatory responses, and oxidative damage thereby imparting a regenerative response in this visceral organ. Regeneration is manifested as either fibrosis, in order to maintain structural integrity of the organ, or through hepatocyte proliferation, in order to restore functional mass. Progression through the cell cycle by the normally quiescent hepatocytes is thought to contribute to the proliferative response. Spy1 is a cyclin-like protein, which binds and activates CDKs at the G1-S and G2-M checkpoints, leading to cell cycle progression independent of cyclinbased regulation. A serendipitous discovery in the transgenic MMTV-Spy1 mouse demonstrates that Spy1 significantly increases the incidence of fatty liver disease and HCC. Combined with a methionine-choline deficient (MCD) diet in order to elicit liver injury, the MMTV-Spy1 murine livers response to these factors through inflammatory, proliferative and fibrotic responses is analyzed. Preliminary evidence demonstrates Spy1's role in favouring hepatic regeneration through a proliferative response rather than a fibrotic one, demonstrating a potential mechanism in the development of HCC. Results may shed light on Spy1's role in HCC, potentially allowing for determination of a diagnostic markers and pathways of the apeutic importance.

Oral Presentation Group A: Cell, Molecular and Genetics

EFFECTS OF NUTRIENT DEPLETION IN CELL PROLIFERATION AND GROWTH

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Tuberous Sclerosis Complex (TSC) is a rare genetic disorder affecting 1 in every 6000 births work wide. The disease is characterized by the formation benign tumors in the kidneys, brain, heart, lungs, skin, and eyes. TS is also related to other medical conditions and can progress to malignant cancers. TSC is caused by mutations in either the TSC1 or TSC2 tumor suppressor genes, also known as Hamartin and Tuberin respectively. The loss of Tuberin expression has been linked with most severe clinical outcomes and is found in several cancers including those of the breast, lung, ovaries, liver, brain, and urothelia. Our lab has shown that Tuberin binds the cell cycle protein, CyclinB1 at the G2/M checkpoint of the cell cycle. This interaction retains Cyclin B1 in the cytoplasm thereby slowing the onset of mitosis. Under low intracellular ATP conditions an upstream regulator of Tuberin, AMP-activated serine/threonine protein kinase (AMPK), phosphorylates Tuberin creating a downstream signal cascade inhibiting protein synthesis. We hypothesize that lack of nutrients may disrupt the Tuberin-Cyclin B1 complex to support enhanced cell division, thereby allowing tumours to form. For this work I have altered the TSC2 genome to mimic AMPK alterations in the Tuberin protein. I have tested the effects of alteration on binding to Cyclin B1 and on cell growth as compared to the wild-type protein. This work is dissecting the key mechanisms regulating cell growth and division and may reveal ways that tumour cells continue to grow under adverse, unfavourable conditions.

Oral Presentation A: Cell, Molecular, & Genetics

THE EFFECTS ON CELL MORPHOLOGY, CELL PROLIFERATION AND CELL VIABILITY AFTER REPEATED EXPOSURE TO NICKEL AGAINST MCF-7 BREAST CANCER CELLS

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Vale is the world's second largest producer of one of the most versatile metals found on earth: Nickel. Nickel is a known neurotoxin, immunotoxin, reproductive toxin, pulmonary toxin and carcinogenic agent. The Sudbury nickel refinery produces about 65 000 metric tons of nickel ore every year and this high consumption has inevitably lead to elevated levels of environmental pollution, which is generated throughout all its stages of production, recycling and disposal. The long term effects of cellular morphology, cell proliferation and cell viability are examined in vitro against the MCF-7 adenocarcinoma cell line after chronic exposure to both nickel chloride and nickel sulphate. There has been very limited research with respect to the epithelial origin MCF-7 breast cancer line and specific metals such as Nickel. Five varying concentrations of nickel (concentrations that can be found in the environment i.e. water, soil, air) were repeatedly exposed to subsequent generations of MCF-7 cancer cells 24 hours after being sub cultured and then left until a confluency of 80-90% as been met. The proposed research furthers the understanding of how repeated exposure to toxic metal such as nickel has on biological systems.

ORAL PRESENATION, GROUP A: Cell, Molecular and Genetics

THE ROLE OF THE SWI/SNF CHROMATIN REMODELLING COMPLEX IN THE NEURAL REMODELLING OF THE DEVELOPING *DROSOPHILA MELANOGASTER* MUSHROOM BODY.

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Components of the SWI/SNF chromatin remodelling complex have been implicated in the onset of intellectual disabilities. The effects of SWI/SNF can be explored by studying the highly plastic mushroom body structure in Drosophila melanogaster. The mushroom body (MB) of Drosophila undergoes extensive neural remodelling throughout development. During MB development, the y neurons initially extend both in the lateral and medial directions, prune back during the pupal stage, and re-extend in the adults, in the medial direction only. Recent research has shown that knockdown of SWI/SNF genes results in extra y neuron projections in the adults. The mechanism of these extra projections is unknown. To explore this phenomenon, I bred flies to induce knockdown of SWI/SNF proteins. I imaged flies at the larval and pupal stages of development to view the MB structure. I hypothesized these extra projections arise from inappropriate re-extension after puparium formation. I expected to see lateral and medial extensions in mushroom body at the larval stage of development in both the control and the knockdown conditions. However, at early pupal stage, I expected to see the neurons pruned back for each condition, indicating inappropriate re-extension. I have found no significant difference in MB morphology at the larval stage of development. However, at the pupal stage, there is a significant difference in knockdown morphology compared to the control. The neurons are pruned back in the control, as expected. However, the neurons in the knockdown flies failed to prune, indicating that the projections do not arise from inappropriate re-extension.

Oral Presentation

A: Cell, Molecular, & Genetics

Optimization of qPCR to analyze age, sex and isolation as factors affecting the expression of Neuroligin 3 in Drosophila Melanogaster

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Social behaviour is influenced by both genetic and environemntal factors. A family of postsynaptic adhesion proteins, Neuroligin, has emerged as potential candidate genes that may influence certain social behaviour. In previous studies exploring the honey bee, environmental influences has been observed to modulate expression of the *Neuroligin* gene family. In *Drosophila melanogaster*, the function of *Neuroligin 3* in social behaviour has yet to be discovered. Previous findings in the Simon lab exploring *D. melanogaster Neuroligin 3* mutants has suggested that Neuroligin 3 may play a role in determining social space of an individual fly. The aim of this study is to optimize a qPCR protocol specifically for Neuroligin 3 in *D. melanogaster.* By manipulating the environmental experience of the flies, the expression of Neuroligin in each experimental group can be quantified and measured. Using this data in combination with previous phenotypic findings in the lab a correlation between social spacing phenotypes and *Neuroligin 3* expression can be explored.

Abstract is for an Oral Presentation Group A (Cell, Molecular and Genetics)

OCTOPAMINERGIC NEURONS REGULATE FEMALE SEXUAL RECEPTIVITY IN DROSOPHILA MELANOGASTER

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In Drosophila melanogaster, females are the choosier sex. Females evaluate the quality of a mate using different neural networks that regulate sexual receptivity to male courtship. Neural networks controlling female receptivity could be modulated by extrinsic mushroom body neurons (EMBNs), namely by octopamine neurons. Octopaminergic neural regulation is conserved in most arthropods and is homologous to norepinephrine in mammals, a neurotransmitter responsible for promoting arousal, alertness, and memory retrieval. Two octopamine neurons called OA-VPM3 and OA-VPM4, innervate brain regions responsible for neural processing and are potential regulators of female sexual receptivity. If OA-VPM3 and OA-VPM4 regulate female sexual receptivity, then silencing or hyperactivating these neurons in female flies will lead to a change in the proportion of females that copulate in a no choice mating assay. Indeed, silencing OA-VPM4 individually is statistically significant, reducing the proportion of females that copulate in experimental genotypes in comparison to control genotypes. Silencing OA-VPM3 and OA-VPM4 concurrently is also statistically significant, reducing the proportion of females that copulate between experimental and control genotypes. OA-VPM3 and OA-VPM4 seem to be important in regulating female sexual receptivity and male rejection. Further research should test female receptivity when hyperactivating or silencing neurons downstream of OA-VPM3 and OA-VPM4, constructing a potential neural network responsible for regulating female sexual receptivity. Namely, gamma lobe dopamine neurons of the mushroom body represent possible future neurons to test.

Oral Presentation

A: Cell, Molecular and Genetics

DROSOPHILA MELANOGASTER WITH ATYPICAL SPERMATHECAL DEVELOPMENT: GENETIC BASIS AND REPRODUCTIVE EFFICIENCY

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Sperm storage plays a key role in the reproductive success of sexually-reproducing organisms, such as *Drosophila melanogaster*. *Drosophila* contain two types of sperm storage organs used for short-term and long-term sperm storage, the latter known as the spermathecae. Dipterans are thought to have the ancestral state of three spermathecae, while two spermathecae are most commonly observed within *Drosophila*. Molecular mechanisms controlling spermathecal development are not entirely known. Here, I examine a gene disruption line of *D. melanogaster* where ~50% of females produce an atypical three-spermathecae phenotype. Inverse PCR for the insertion site identified a candidate gene for spermathecal supernumeracy. Additional disruptions within the candidate gene were scored for spermatheca number. A disruption in the 3' untranslated region induced three spermathecae in ~30% of females, while a disruption in the spermathecae, females with three spermathecae did not produce a greater number of offspring after a single mating, but a greater proportion of these females are still producing some offspring after 15 days.

Oral presentation

A: Cell, Molecular, & Genetics

SEXUALLY DIMORPHIC SOCIAL SPACING IN *DROSOPHILA MELANOGASTER*: DETERMINING THE NEURAL CIRCUITRY INVOLVED

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The place an animal decides to stand relative to another is dependent on their ability to recognize others and respond appropriately. These responses have been observed to be sexually dimorphic and influenced by previous social experiences of individuals. I have studied social space, the distance between individuals in a group, to measure these responses. Two genes, *doublesex* and *fruitless* have been implicated in sex-specific traits in *Drosophila melanogaster*, including certain behavioural responses. Neurons with *doublesex* and *fruitless* are hyper-activated or silenced using drivers of these genes crossed with UAS-temperature dependent constructs. Distance between flies with altered neuronal activity will be measured to quantify social space. These results will be compared to the social space of wildtype flies who have been isolated to determine if social experience is processed through sex-specific neurons. These manipulations allow for better understanding of the neural circuitries involved in social responses.

Abstract is for an oral presentation in group A (Cell, Molecular and Genetics)

OPTOGENETIC CONTROL OF NOTCH-MEDIATED LATERAL INHIBITION IN *DROSOPHILA* NEUROBLASTS THROUGH CRY2-CIBN CLUSTERING

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Abstract

The process by which pluripotent cells in a developing embryo make decisions with respect to cell fate is critical for the formation of functionally distinct tissues from an equipotential precursor population. Communication between cells is achieved through many different molecular signaling pathways which regulate differential gene expression, allowing otherwise identical cells to adopt different fates. Notch, a highly conserved transmembrane receptor protein, is responsible for the activation and repression of proneural genes in a defined pattern within the neuroepithelium, specifying a subset of cells to adopt a neural fate in a process called lateral inhibition. Previous studies have shown that Notch-mutant Drosophila embryos adopt a lethal neurogenic phenotype, resulting in over-proliferation of neural precursors at the expense of epithelial tissue. However, the mechanisms by which lateral inhibition dynamically regulates neural precursor populations is not well understood. To determine whether developing Drosophila embryos can adapt to disruptions of Notch-mediated lateral inhibition, I propose an optogenetic strategy to selectively inhibit Notch signaling in the developing neuroepithelium. By introducing a synthetic Cryptochrome 2 (CRY2)::mScarlet fusion protein, used in combination with a transgenic Drosophila line with the Notch ligand Delta fused to an N-terminally truncated cryptochrome-interacting basic-helix-loop-helix 1 (CIBN) and a fluorescently tagged Notch receptor (Notch-YFP;Δ-CIBN), it will be possible to exogenously control the process of lateral inhibition during embryonic neurogenesis using light as a molecular switch. This project could potentially provide insights as to the ability of the developing Drosophila nervous system to dynamically respond to changes in the pattern of cell-fate determination.

For oral and/or poster presentation

Group A: Cell, Molecular, & Genetics

DEVELOPMENT OF SELF-REPLICATING NON-INTEGRATING LENTIVIRAL VECTORS FOR SAFE MOLECULAR-GENETIC IMAGING OF TRANSPLANTED CELLS IN VIVO

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Cell-based therapy is an emerging therapeutic for many diseases and cellular imaging with reporter genes (RGs) can allow one to precisely monitor cellular viability, location and numbers *in vivo* to help predict therapeutic effectiveness and patient outcome. However, how we label cells with RGs needs significant consideration, as many integrating vectors pose a risk of unwanted genotoxicity. Here we explore the use of non-integrating lentiviral vectors (NILV) containing the scaffold-matrix attachment region (S/MAR) motif for stable RG expression, as S/MAR-NILVs can enable stable transgene expression without integration (Verghese *et al.* 2014). We first produced NILVs expressing fluorescence and bioluminescence RGs by mutating the catalytic domain of integrase gene necessary for integration and confirmed expected loss of RG expression over time. We recently confirmed correct engineering of S/MAR-NILV transfer plasmids. Next steps include S/MAR-NILV production, and evaluation of RG expression over time in cancer cells both *in vitro* and *in vivo*. If successful, we believe this new gene vector can be broadly used to label cells with RGs, enabling long-term and safe imaging of cells *in vivo*.

Type: Oral Presentation

Category: A- Cell, Molecular, and Genetics

RNA INTERFERENCE AS A GENE SILENCING TOOL TO CONTROL THE TWO-SPOTTED SPIDER MITE *TETRANYCHUS URTICAE*

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The two-spotted spider mite, Tetranychus urticae, is one of the most polyphagous arthropods feeding over 1000 plant species with a global distribution. Despite effort to control the twospotted spider mite using acaricides, this pest remains a serious problem for agriculture as populations grow quickly and develop resistance to pesticides in only a few years. Recently, RNA interference (RNAi) technology was developed as an alternative for pest control. RNAi is an evolutionarily-conserved cellular process, during which short RNA molecules can interfere with complementary messenger RNA transcripts and degrade them. This research is focused on testing the efficiency of double stranded RNA (dsRNA) molecules to reduce mite fitness by targeting genes essential for mite biology including: Srp54, Rop, SNAP-a, Rpt3, Rpn-7, Shibire, Pcf11, Cactus, Pp1α-96A, Pp1α-96A(2), Hsc-70 and, Gawky. Application of Srp54, Rop, SNAPα, Rpt3, Rpn-7, Shibire, Pcf11, Hsc-70 and, Gawky dsRNA fragments were found to significantly reduce survivorship of newly molted adult females when compared to controls. In addition, mite fecundity was reduced by 91.2%, 85.0%, 52.6%, 45.3% and 34.1% using dsRNA against Rop, SNAP-a, Rpt3, Rpn-7 and Srp54 respectively. This research has demonstrated the potential of using dsRNA as an alternative to pesticide to control mites and should be further investigated to elucidate a practical application in an agricultural setting.

Oral Presentation

A: Cell, Molecular, & Genetics

HOMOLOGY INDEPENDENT TARGETED INTEGRATION (HITI) CRISPR/CAS9 SYSTEMS FOR EFFICIENT REPORTER GENE KNOCK-IN IN CELLS

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With the ever-extending use of cell therapies in regenerative medicine, there lacks precise monitoring tools needed to distinguish responder from non-responders to cell based treatments. Non-invasive tracking of cells with imaging reporter genes can allow one to monitor the fates of implanted cells, enabling quantitative assessment of treatment response. CRISPR/Cas9 when coupled with the Homology Independent Targeted Integration (HITI) system allows safe and efficient genome editing of cells with reporter genes (RG) for non-invasive and longitudinal monitoring of transplanted cell fate. Here we compared the traditional and inefficient Homology Directed Repair (HDR) CRISPR/Cas9 system to the HITI system by co-transfecting HEK-293 cells with either Cas-9 guide or Cas-9 scrambled (control) expressing green fluorescence RG together with either a HDR RG system or a HITI RG system expressing red fluorescent RG. Following transfection, cells are FAC sorted for cells that exhibit both green and red fluorescence, followed by flow analysis for red fluorescing cells for the next 3 weeks. Genomic DNA is isolated in order to confirm integration of the RG system into the correct loci. Our preliminary results show that the HITI RG system results in a higher percentage of red cells compared to the traditional HDR system. This work lays the foundation for an effective and safe genome editing tool for RG tracking of cells in vivo.

Oral Presentation

Cell, Molecular & Genetics

DEVELOPING TARGETED RNAi KNOCKDOWNS OF *fruitless* SPLICE VARIANTS FOR *Drosophila melanogaster* AND *Drosophila simulans*

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Reproductive isolation reduces gene flow between populations and drives speciation. In *Drosophila melanogaster* and *Drosophila simulans*, the *fruitless* gene influences sexual development and female rejection of copulation attempts from heterospecific males. The gene has four potential first exons (P1 - P4), the first of which is sex-specifically spliced. The role of the P1 transcript has been studied extensively in males, yet very little is known about the functions of the other transcripts, or which transcripts influence female behaviour, despite the role female choosiness plays in reproductive compatibility. The effect of *fruitless* on female receptivity is challenging to study as pre-developmental knockout of most *fruitless* transcripts is lethal. Pairing the Gal4/UAS expression system with RNAi specific to the *melanogaster* and *simulans* sequence for each exon will allow for localized, temporally controlled silencing of three *fruitless* transcripts (*fru-Fem, fru-P3* and *fru-P4*) which circumvents developmental lethality. Selectively silencing expression of one species' alleles of these transcripts in *melanogasterl simulans* hybrids will allow for assessment of their role, if any, in female rejection behaviour.

Oral Presentation

Group A: Cell, molecular, and geneticsdsa

DEVELOPING IN VITRO REGENERATION AND TRANSFORMATION METHODS FOR APPLE PLANTS

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The apple industry is a large contributor to the Canadian economy, but commonly faces yield loss due to disease. Fire blight is a common bacterial disease, causing an estimated annual loss of \$4 million to the Canadian apple industry. Genetic resistance via genome editing is a potential method for developing resistance to bacterial diseases. Genome editing relies upon efficient regeneration of transformed tissue. Currently, genetic transformation of 'Fuji' apple species is still not routine, due to low cell regeneration efficiency. In this study, Murashige and Skoog (MS) and Woody Plant medium (WPM) were tested across various concentrations of plant growth regulator, thidiazuron (TDZ), to determine optimal regeneration methods for apple cultivar 'Fuji'. Murashige and Skoog medium, paired with 1 mg/L TDZ, was found to provide optimal regeneration conditions. This method was then used for *Agrobacterium* mediated transformation of 'Fuji' apple plants, testing both a reporter construct and a genome editing construct. Shoot regeneration efficiency on selection medium was determined to be 6.4% and 20.5% for tests with a reporter construct and a genome editing by this study will aid in future development of resistance to bacterial diseases via genome editing.

Oral Presentation

Group A: Cell, Molecular, & Genetics

IDENTIFICATION OF SPECIES – DIAGNOSTIC DNA MARKERS DIFFERENTIATING RED MAPLE RED MAPLE (*Acer rubrum*) AND SILVER MAPLE (*A. saccharinum*)

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Identifying DNA markers within a species is an important aspect in genetics. We focused on red maple (Acer rubrum), silver maple (Acer saccharinum), and freeman's maple (Acer × freemanii) - a hybrid between the two. This research will fill the gap of the DNA composition of the species by using Inter Simple Sequence Repeat technique. The objective of this research is to identify markers that uniquely represent each of the maple species. Therefore, we will search for DNA sequences that are different among the species, but present throughout the samples. A. rubrum and A. saccharinum should have distinct markers, and A. freemanii should contain the diagnostic markers of both parental species. DNA was extracted from A. rubrum, A. saccharinum, and A. freemanii. Once the DNA had been extracted, a PCR was conducted to amplify the DNA of each sample using different primers. Using gel electrophoresis, we could analyze the bands to determine if any are unique to the individual species. We found speciesspecific DNA markers that are unique to both species of maple to conclude their distinctive genes. These primers include ISSR 5, ISSR 8, ISSR 10, and ISSR 825. These markers were also present in the hybrid, which further supports the findings that these genotypes are speciesspecific. This research will enable further molecular studies of the investigated species, as well as, aid in the development of new molecular markers, and provide application in plant conservation projects. The results may also be useful for botany forensic studies involving such maple species.

Oral presentation

Judged by Group A

BIRD WINDOW COLLISIONS IN LIGHT OF BUILDING & HABITAT CHARACTERISTICS DURING THE FALL MIGRATORY PERIOD AT UNIVERSITY OF TORONTO MISSISSAUGA

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Rapid urbanization and anthropogenic infringement upon natural habitats have been associated with an increased quantity of bird window collisions in recent years. Avian field research indicates that a plethora of factors including modern building design and species characteristics are each, individually associated with this increase in collisions. Nevertheless, the collective impact of such factors has not been sufficiently covered in present literature. Particularly, this topic has not been extensively explored in an environment similar to University of Toronto Mississauga, which consists of a number of mid-rise buildings on developed land surrounded by a number of important bird habitats (mixed forest and cultivated grassland). The purpose of this study was to determine the collective impact of migratory status, weather conditions, habitat proximity and percent window coverage of building facades on bird window collisions at University of Toronto Mississauga. This study involved monitoring bird window collisions across 40 building facades (spanning over six buildings) on campus through daily site surveys during the 2017 fall migratory period. During the study, a great number of window collisions by migratory birds were recorded in sites with relatively low window coverage contrary to the majority of currently published research. As such, a multivariate analysis of the impact of specific building, environment and species characteristics on the number of collisions at each building facade was necessary. Ultimately, the results of this study may contribute to a future action plan outlining specifically tailored, tangible preventative measures necessary to reduce bird window collisions at University of Toronto Mississauga.

Oral Presentation (Group: Ecology & Evolution)

Discerning Cyclic Patterns and Habitat Variation in Underwater Soundscapes in the Laurentian Great Lakes

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Passive acoustic monitoring (PAM) is a non-invasive method that can be used to assess the underwater soundscape, and through it the health of an ecosystem, by providing researchers with a non-biased comprehensive ecosystem visualization. While PAM has a rich history in marine systems, it is less commonly used in freshwater, limiting our understanding of sound levels in freshwater habitats. Here we measure timing and location-based variation in sound in the Laurentian Great Lakes (LGL) to estimate the likelihood of adverse effects on native fish. Sound levels varied in areas with recreational boating on a diurnal cycle, with levels up to 140 dB re 1 μ Pa/Hz in the morning and evening and quieter periods midday. Areas without boats had no clear patterns and were generally quieter, where peak sound energy was concentrated around 100 Hz and below. Areas with increased boat traffic had significant sound energy at 800-1000 Hz, a bandwidth that overlaps with fish species possessing hearing specializations. While still preliminary, these results identify the potential for habitat-specific noise effects in the LGL and allows us to test potential for anthropogenic effects in local fish species of interest.

This is for an Oral presentation. Group B: Ecology & Evolution.

DECLINES IN WINTERING SNOW BUNTING POPULATIONS AS A FUNCTION OF CLIMATE CHANGE AND AGRICULTURAL INTENSIFICATION IN NORTH AMERICA

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Populations of many bird species are in decline around the world, a situation which has been attributed to many factors such as habitat loss and climate change. The snow bunting (Plectrophenax nivalis) is no exception to this calamity, as its North American population has declined over 60% in the last 50 years. Despite being studied extensively in Europe, the dietary habits, habitat preferences, and population dynamics of snow buntings during the winter months are poorly understood in their North American range. This species is known to inhabit farmland and feed on grain left behind after harvest, though the extent of their reliance on cropland is unknown. We seek to understand relationships that could exist between snow bunting numbers, climate change, and changes in agricultural practices in Canada and the United States, using long-term records (over 100 years) from Audubon's Christmas Bird Count (CBC), climate and weather data from the Canadian government and NOAA databases, and crop yields and farmland area reports from Statistics Canada and the USDA. We also aim to examine possible relationships between chemical applications to farms and the decline in snow bunting populations. We hypothesize that declines in snow bunting numbers are related to decreasing farmland habitat and increased crop yield over the last century. Results for the effect of climate and chemical application on snow bunting populations are forthcoming. This study highlights the need for a more thorough inquiry into the environmental impacts of human activities on birds.

Oral Presentation

Group B: Ecology & Evolution

EFFECTS OF ANTHROPOGENIC WASTE ON THE RACCOON, *PROCYON LOTOR* (CARNIVORA: PROCYONIDAE) IN SOUTHERN ONTARIO

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Historically, food waste produced by humans has been a food source for many animals globally; particularly scavengers, which have moved into urban areas to live alongside humans. The rise in unhealthy Canadians may have something to do with the products we consume, products we also throw away as waste. This study aims to analyze the effects of anthropogenic food waste on animals consuming it; in particular, the raccoon (Procyon lotor). Raccoons were live trapped across southern Ontario in the fall of 2017 as a part of the Ministry of Natural Resource's rabies surveillance where their weights were recorded. Using ArcGIS® and ArcMap ™ software by Esri, the weight (kg) of raccoons can be tested against the number of households, grocers, apartments and restaurants in the area. Using this data, I can look for a relationship between the amount of waste available for consumption and the weight of the animal. If raccoons are feeding predominately on anthropogenic waste, we predict that raccoons living in close proximity to larger amounts of available food waste will be relatively larger than those in low availability areas. Here, we observed that sex of the animal plays a role in weight variation, but that the number of houses, restaurants, grocery stores and apartments in an individual's home range is not significant in determining weight. This information is useful in furthering our knowledge of the health effects experienced by wildlife living in proximity to humans.

Keywords: raccoon, Procyon lotor, live trapping, weight, body condition, waste production

Supervisor: Albrecht Schulte-Hostedde, Department of Biology, Laurentian University, Sudbury.

Oral Presentation

Group B: Ecology and Evolution

USE OF GUT MICROBIOMES IN CREEK CHUB (*SEMOTILUS ATROMACULATUS*) TO ASSESS THE ENVIROBMENTAL EFFECTS OF MINE EFFLUENTS IN SUDBURY STREAMS

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Sudbury, ON has a history of mining derived environmental damage in its watersheds. In recent years, enormous investments have been conducted to reduce the release of metals from waste storage areas. Water quality has greatly improved in many streams and some fish, such as the Creek chub (*Semotilus Atromaculatus*) have recolonized, but we still have little means of assessing the health of these Cyprinidae. This project focuses on the use of the gut microbiome as a possible environmental health indicator. We sampled Creek Chub from Junction Creek, a 20km stream that goes directly across the city, receiving drainage from five tributaries that flow from some of the biggest mining and waste storage areas. We sampled the reference stream LSR-07 and three streams from impacted tributaries on Junction Creek; Garson, Maley and Nolin. Fish were dissected with sterile equipment and their hindgut and its contents were extracted. DNA extractions were performed on each sample and sent out for illumina MiSEQ of the 16s rRNA gene. The samples analyzed from the polluted tributaries are expected to differ in bacterial community structure compared to those from the reference site. The results of this project will guide future studies exploring microbial-host associations in compromised aquatic environments of Sudbury.

Abstract is for Oral presentation

Judged in group B: Ecology and Evolution

PERSONALITY: HOW DO SQUIRRELS HANDLE STRESS?

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The stress response is used by all organisms to react to new and potentially dangerous circumstances. Individuals generally have an undifferentiated behavioural response over a wide range of situations; this is known as their personality. Currently it is unknown how personality is related to an individual animal's background stress levels. The goal of this study is to observe how individuals react to handling and how it is connected to their short-term and long-term naturally caused stress. Background research shows that animals that have a reduced behavioural reaction to stressful stimuli generally produce more glucocorticoids. Therefore I hypothesized that both short and long-term stress mediums would show higher levels of cortisol in animals that were more docile. Red squirrels (*Tamiasciurus hudsonicus*) were trapped at various protected sites in Ontario. Docility was measured using a handling bag test. Fecal and hair samples were obtained from the squirrels and sent to the Toronto Zoo for cortisol extraction and enzyme immunoassay. Data did not fit parametric assumptions so a Spearman's Rank Correlation test was run. The fecal cortisol levels of male squirrels were negatively correlated with inactive time significantly (p = 0.03849). Females, however, showed the opposite trend while approaching significance (p = 0.06479). Hair cortisol appeared to have no relationship with docility in either sex. This research will help us to understand the differences between conspecifics and how they vary in their behavioural response to stressful stimuli.

I would like to give an oral presentation judged under group B: Ecology and Evolution (OR Physiology & Toxicology)

COMMON TERN AGGRESSION AND FLEDGING SUCCESS AT BREEDING SITES SHARED WITH CASPIAN TERNS

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Reproductive success (RS) is central in determining the productivity and interpreting population dynamics of colonial avian species. RS is usually difficult to measure and thus fledging success (FS) is commonly used as a proxy for colonial birds. Common terns (COTE; Sterna hirundo) are a well-studied colonial species; however, there is limited information on the effect of non-predatory neighbouring species on COTE RS. Recent literature on RS has focused on the effects of abiotic, human, and predatory disturbances, overlooking the potentially large role interspecies interactions play in the productivity of a particular species. In this study, the interactions between COTE and Caspian terns (CATE; Hydroprogne caspia) are examined through behavioural observations over the fledging period on islands with and without the presence of CATEs. One island containing only COTEs had significantly lower levels of aggression and less aggressive interactions overall, these results may be due to predation or extensive vegetation growth on the island. No significant difference in the number of aggressive interactions or level of aggression was seen on the remaining islands, and FS was not significantly different between groups with and without CATEs. Statistical power was low because of issues resulting from visibility, which can underestimate FS. We tested a methodology using a Radio-Frequency Identification (RFID) system to monitor COTE FS remotely. However, due to software glitches and loss of Passive Integrated Transponder (PIT) tags during the experiment, no conclusive results were obtained.

Oral presentation: B

EFFECT OF GUT MICROBIOME ON MODULATING SOCIAL SPACING IN DROSOPHILA MELANOGASTER

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Drosophila melanogaster social behavior is mediated by both genetics and environment, including the gut microbiome. The gut microbiome has been shown to modulate *D. melanogaster* mating and egg-laying site preference. I have investigated the specific relationship between the gut microbiome and one rarely investigated aspect of social behavior: social spacing. Social space is defined as the distance between an individual fly and its closest neighbour. The aim of my research is to characterize the extent to which the gut microbiome affects social space of various *D. melanogaster* strains by comparing between control and microbe-free flies. Previous studies conducted in the Simon lab suggest a sex effect of the gut microbiome on social spacing in the relatively recently wild-caught *D. melanogaster* BJS strain, where axenic females are closer at proximal distances compared to control counterparts, while males showed no difference in social space. Gut microbiome-mediated social space of the lab strain Canton-S as well as another relatively recently wild-caught strain called Elwood were also investigated.

Abstract is for an Oral Presentation Group B (Ecology & Evolution)

NETWORK ANALYSIS OF LIVING SOCIAL POPULATIONS: LESSONS FROM FRUIT FLIES

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Network analysis can help model different aspects of social life, from the social structure of animal population to the communicable spread of contagion. Despite its utility, however, network analysis has not been fully integrated into the study of animal behaviour, perhaps because it can be difficult to track and score large numbers of social interactions in real time. *Drosophila* is a highly tractable model organism that may lend itself well to network analysis. Using a video-to-software analysis pipeline that I helped to develop, I am testing how the structure and function of insect social networks vary as a function of genotype. A neuroligin 3 gene broadly implicated in social behavior appears to promote fly interactions, as evidenced by more connections between nodes and shorter path lengths relative to the networks of wild type flies. I interpret genotypic variation in fly networks within a statistical and sociobiological framework.

Oral Presentation

Ecology and Evolution

SINGLE-SONG REPERTOIRES CHANGE THROUGHOUT A LIFETIME IN SAVANNAH SPARROWS

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Many species across the animal kingdom are able to communicate with each other through many different modalities. Bird song is an auditory method of communication that has a dual purpose: mate attraction and territory defence. As a bird ages, the importance of attracting a mate and defending a territory is expected to change, suggesting its song may change in parallel. Previous studies have considered changes in song structure with age in species that have multiple song types, yet most species of birds have just a single song type, and variation with age in these species has been largely overlooked. In this study, we explore Savannah Sparrows (*Passerculus sandwichensis*), a single-song type species, to determine whether or not Savannah Sparrow song changes with age. We recorded and measured the fine structures of 21 males' songs from a population on Kent Island, New Brunswick. We found that as birds age, their songs remain similar in structure but become increasingly shorter with time. Our results provide evidence in support of the hypothesis that bird song changes with age in a species with a single-song repertoire. Therefore, male song length may be an age cue that other males can use to assess potential rivals, and females can use to assess potential mates.

Oral presentation

Ecology and Evolution

THE DIET OF GREAT HORNED OWLS (*Bubo virginianus*) FROM HIGH BLUFF ISLAND, SOUTHERN ONTARIO

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High Bluff Island, Presqu'ile, Ontario consist of two major woodlands on the western and eastern side of the island separated by fields and thickets. The unique landscape on the island is characterized by the age of the trees, uncommon associations between species, and the rarity of the mature forest on Lake Ontario islands. Many species including the Great Horned owl seek refuge in dense or mature foliage on High Bluff Island. The objective of this study is to obtain dietary information for Great Horned Owls (Bubo virginianus) in a region where little to no previous data had been collected. Remains from owl pellet material on High Bluff Island were collected during the summer season of 2017. A total number of 78 prey items were collected from pellet samples or remains around 3 roost sites on the island. All prey items were identified as Meadow Voles (*Microtus pennsylvanicus*). This diet data revealed that Great Horned Owls on High Bluff Island feed on strictly voles from the island. The increased consumption of voles and negligible number of other remains in the diet indicates that island habitat is sustainable enough for Great Horned Owls and other raptor species. Ecosystem interactions are crucial in understanding prey biodiversity, which can limit populations of target conservation species, including birds of prey and the Great Horned Owl.

Oral Presentation

B: Ecology & Evolution

REPRODUCTIVE EFFORT OF *BRACHYHYPOPOMUS OCCIDENTALIS* AND ELECTRIC SIGNAL VARIATION

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Life history strategies create trade-offs between growth, survival and reproduction allowing species to become specialized in their environment thus affecting reproductive effort. Brachyhypopomus occidentalis, a pulse-type electric fish with an annual spawning season, was studied at eight different sites in Panama. Few studies exist on the Brachyhypopomus genus. fewer still have information on the reproductive cycle of specific species such as *B. occidentalis*. We aimed to add to the general understanding of this fish's reproductive cycle in relation to its morphology, particularly gonad size as well as to determine if a correlation between gonad size and electric signaling, predator richness and reproductive strategy existed. Electrodes were used to locate individuals who were then caught by dip net. Three electric signals were recorded for each fish who was then euthanized by thermic shock and transported to the Smithsonian. They were then photographed and analysed to determine body length, mass, presence of predatory injuries (rank), sex and gonad mass. Preliminary data does not suggest a relationship between energy invested in gonad development in the fish and their predator's prevalence (rank). Total rank is not significantly different by site and gonad weight is not dependent on rank, but on total body size. The importance of this research is to gain information on this species and on their reproduction to better understand reproductive strategies in relation to predation.

Oral presentation

I would like to be judged under group B.

CHEMOTOPIC ORGANIZATION OF ODORANT RESPONSES IN THE SEA LAMPREY (*Petromyzon marinus*) OLFACTORY EPITHELIUM

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In sensory systems such as audition, vision and proprioception, specialized receptors are topographically organized to enhance detection sensitivity. However, it is unclear how the olfactory system is spatially organized in a way that aids in odour detection. In sea lamprey (Petromyzon marinus), odorants stimulate the olfactory sensory neurons within the olfactory epithelium lining. It is unclear how patterns of odorant responses are distributed throughout this structure. This thesis investigated different regions of the olfactory epithelium in the larval sea lamprey through the odorant responses to a pheromone (3kPZS) and an amino acid (L-arginine) odorant likely a feeding cue. I used a calcium imaging technique to study the chemotopic organization of odorant responses in the olfactory epithelium and found that the olfactory sensory neurons responding to the odorants are broadly distributed in the olfactory epithelium. However, results suggest odorant responses appear to be more concentrated on the right side of the olfactory epithelium for 3kPZS and the left side for L-arginine. The broad zonal patterning for odorant responses may serve as an initial organizing step in olfactory sensory information coding that ensures a response to an odorant that stimulates any region of the olfactory epithelium. This is advantageous to the animal as the detection of odors is used for a variety of behaviours such as feeding, migration, spawning and survival of the animal, which serves an evolutionary purpose for the survival of these species.

Oral Presentation, Judged on the group of C: Physiology & Toxicology

DISTRIBUTION OF SOLITARY CHEMOSENSORY CELLS FOUND ON THE BRANCHIAL

PORES OF NEWLY TRANSFORMED SEA LAMPREY, Petromyzon marinus.

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The agnathan vertebrate, the sea lamprey, *Petromyzon marinus* uses a diffuse chemosensory system with microvillous solitary chemosensory cells (SCCs) innervated by single nerve fibers located on the branchial pores. Previous research has shown small gill papillae containing SCCs in lampreys newly transformed from larvae into adults, when water flow for breathing has changed from mouth to gill, to tidal gill ventilation. This study explores the SCC morphology and distribution along the posterior surface of the seven external gill slits in newly-transformed sea lamprey, along with corresponding innervation. The microvilli of the SCCs were labeled with phalloidin while the anti-acetylated α -tubulin immunolabelling labeled the innervating neural fibers. The SCCs were grouped together at specific ventral and dorsal regions of the gill pores. These specific regions indicate association with early development of the gill papillae seen in the juvenile and adult stages. Specific variations in the morphology of the microvilli may indicate SCC development. This study suggests that the SCC chemosensory system is functioning in the newly transformed stage of lamprey development. Neurophysiological studies are needed to investigate the chemosensory properties of these cells.

Oral Presentation

Group C: Physiology and Toxicology.

EFFECTS OF GALLIUM AND INDIUM MIXTURES ON THE AQUATIC INVERTEBRATE DAPHNIA MAGNA

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There is currently an increasing demand for trace elements such as gallium, and indium in new and emerging technologies. These are considered Technology Critical Elements (TCE's) and are expected to be increasingly exploited which may result in their release into the environment and subsequent exposure. There is little information on the effect these elements in the environment, and their potential hazards to organisms that are exposed to them. Since they are often found associated with the same elements, such as zinc, they are likely to be introduced into the environment simultaneously during the extraction of the desired element, leading to mixture formation. To test if mixtures of these elements had an impact on aquatic organisms, the individual effects were first determined. EC₅₀ tests (48h) were conducted for both elements on the aquatic invertebrate Daphnia magna. This test procedure followed the Environment and Climate Change Canada standard method for EC₅₀ tests performed on *Daphnia magna*. Once the individual effects were determined, tests were conducted with two-element mixtures on D. magna using a toxic unit approach following the same standard test method. Results from mixtures tests showed synergistic interactions between gallium and indium. Individual toxicity tests indicated gallium was the more toxic element, but indium was found to play a greater role in the toxicity of the mixtures based on secondary mixtures tests. These findings are important as gallium and indium were found to have a greater aquatic toxicity when they are present in mixtures than present as individual elements.

Abstract for oral presentation

Judged for Physiology and Toxicology

CORRELATION BETWEEN PANGENOME SIZES & RATES OF HORIZONTAL GENE TRANSER IN BACTERIAL GENOMES

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Horizontal gene transfer (HGT) is a critical component of bacterial evolution. The concept of a pangenome is also gaining prevalence within the field of comparative genomics. It is the all-inclusive, cumulative set of genes which can be present within any strain of a selected species. It is generally assumed that higher rates of HGT correlate to a higher pangenome size. For example, large pangenome sizes would provide for a larger sample of available genes allowing for greater possibilities of genes to be transferred. Higher HGT rates should also correlate to a higher chance of introducing unique or rare genes to an existing genome. Despite this intuitive assumption there is no evidence to explicitly support it and, there remains many confounding variables which must be considered (e.g. phylogenetic distance, environment). Through a bioinformatics approach, this study seeks to demonstrate the correlation between HGT rates and pangenome size within ten different bacterial species, while accounting for phylogenetic distance as one confounding variable.

Category - A: Genetics

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ALTERNATIVE LAMPRICIDE TREATMENTS: PHYSIOLOGICAL EFFECTS ON JUVENILE LAKE STURGEON (*ACIPENSER FULVESCENS*).

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The piscicide 3-trifluoromethyl-4-nitrophenol (TFM) is used to control the invasive sea lamprey populations in the Great Lakes. Applied to tributaries at the minimum lethal concentration (MLC; meant to kill 99.9% of the larval lampreys over 9h), TFM selectively targets the sea lamprey. Lake sturgeon juveniles (<200 mm), often found in lamprey streams, have an increased sensitivity to TFM at this life stage. We hypothesize that this increased sensitivity to the lampricide occurs because the sturgeon's detoxification capacity is overwhelmed during regular treatments. Here, we tested the effectiveness of an alternative TFM treatment, where fish were exposed to the 24h MLC of the lamprey over 24 h (lower dose over a longer time), to test whether it is reduces sturgeon mortality. Fish (1+ year old, 116±1mm on average) were either exposed to the 9h or 24h MLC. Mortality was reduced from 55.0% at the 9h MLC to 8.8% at the 24h MLC. Blood, plasma, bile and muscle were collected and analyzed for TFM, its metabolites and physiological disturbances. Plasma lactate levels significantly increased in sturgeon exposed to the 9hMLC, indicating a switch to anaerobic glycolysis, but glucose remained unaffected. Hematocrit and mean corpuscular hemoglobin concentration decreased in fish exposed to the lamprey 24h MLC, but no mortality was observed. Analysis of TFM and its metabolites led to the identification of TFM-glucuronide and TFM-sulfate as major metabolites of TFM in lake sturgeon. The "long-and-low" TFM treatment appears to be an effective method to reduce sturgeon mortality without affecting treatment efficacy.

Abstract for Oral Presentation: to be judged in group C: Physiology & Toxicology.

FISH BEHAVIOUR AND PHYSIOLOGY ACROSS TWO WASTEWATER EFFLUENT GRADIENTS IN HAMILTON HARBOUR

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Chemical inputs from wastewater treatment plants (WWTPs) pose serious environmental stressors for urban watersheds. Hamilton Harbour is one of 43 locations around the Great Lakes identified as an Area of Concern due to environmental damage and poor ecosystem health. While habitat remediation has reversed some ecosystem degradation, treated wastewater continues to enter the harbour, hindering restoration efforts. In this study, we examined the effects of WWTP effluent on multiple levels of biological organization in wild fish. We studied two native species - green sunfish (Lepomis cyanellus) and bluegill sunfish (Lepomis macrochirus) – and the invasive round goby (Neogobius melanostomus) along an effluent gradient near two WWTPs entering Hamilton Harbour. We used field-based behavioural tests and scored activity levels, boldness, and predator responses. We also assessed morphological and physiological markers: body mass, Fulton's body condition, gonadosomatic index (GSI), liver investment, haematocrit (Hct), and critical thermal tolerance (CTmax). We found that activity, boldness, and predator responses were not affected by proximity to WWTP effluent. Body mass, body condition, liver investment and Hct all tended to be higher in fish found closer to WWTPs, suggesting that fish near effluent outflow may be in better condition. While CT_{max} did not vary with proximity to WWTPs, the three species showed strong differences in thermal tolerance: green sunfish had the highest CT_{max} and round goby had the lowest. Understanding the ecological thresholds for various fish species and how environmental stressors like wastewater effluent affect fish behaviour and physiology is vital for shaping resource management decisions and conservation policy.

Oral Presentation

C. Physiology and Toxicology