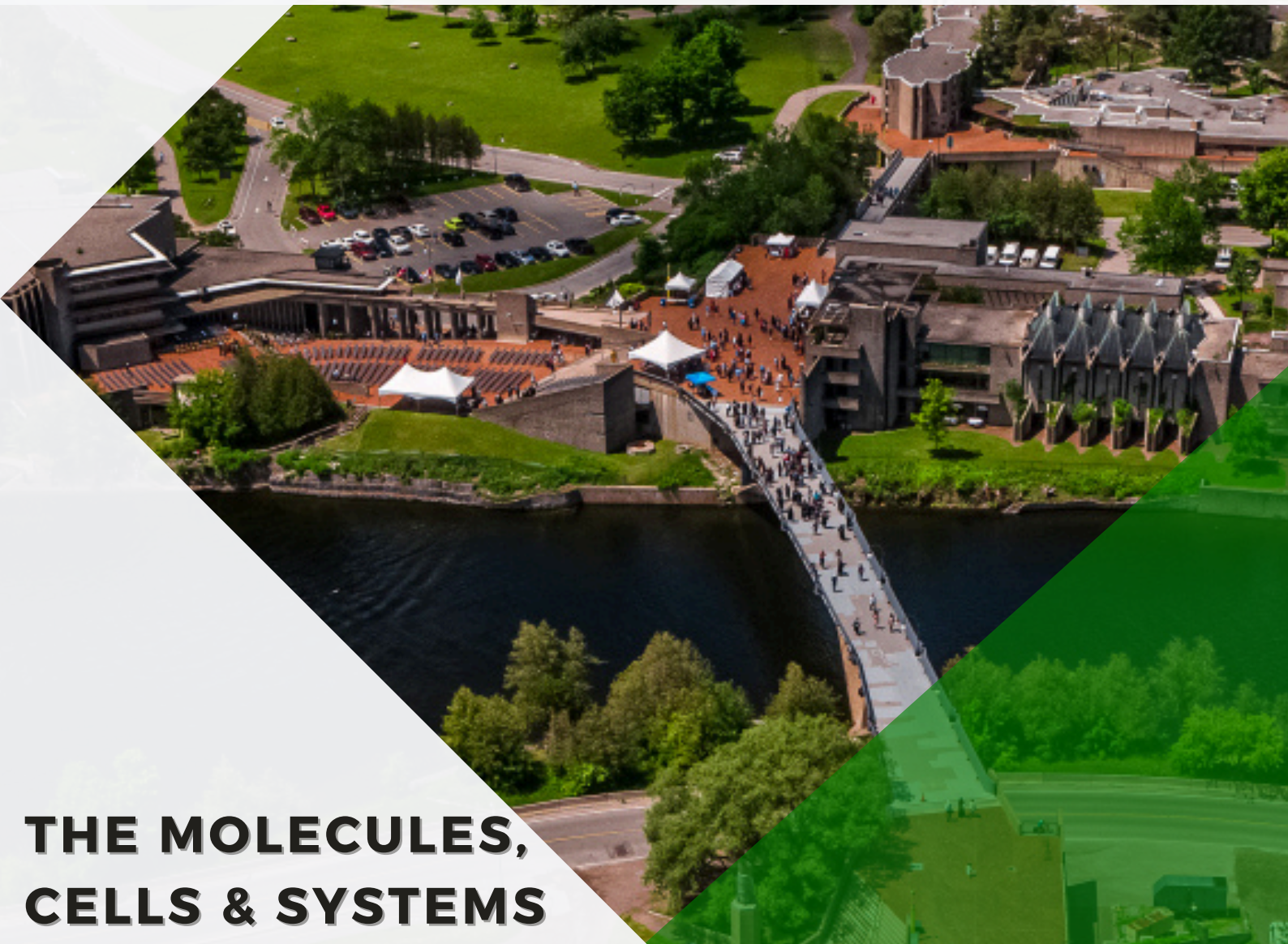
Molecules Cells & Systems

TRENT UNIVERSITY



THE MOLECULES, CELLS & SYSTEMS RESEARCH CENTRE PRESENTS:

3rd Annual Research Symposium

PROGRAM

APRIL • 24 • 2025

TRENT UNIVERSITY
STUDENT CENTRE, TSC 1.20
PETERBOROUGH, ON

MCS OVERVIEW

THE MOLECULES TO SYSTEMS APPROACH

The life sciences are a broad grouping of disciplines that study organisms across the tree of life. All organisms, whether unicellular (e.g., bacteria) or multicellular (e.g., plants, animals; including humans), require fundamental molecular building blocks. These building blocks facilitate the formation of a cell, which associates with other cells to form systems within a multicellular organism. Through evolution, organisms have become more complex, facilitated by the formation of specialized interdependent molecular and cellular systems. The Molecules to Systems Approach seeks to understand the mechanisms underlying each stage of this biological hierarchy.

OUR VISION

Trent University has a wealth of expertise in various disciplines across the life sciences including molecular biology, cell biology, biochemistry, microbiology, developmental biology, physiology, kinesiology, psychology, and neuroscience. To exploit this competitive advantage, the Molecules, Cells & Systems Research Centre brings together researchers from the Departments of Biology, Chemistry, Forensic Science, Kinesiology, and Psychology to collaborate and apply the Molecules to Systems Approach in their research.

ORGANIZING COMMITTEE

Dr. Robert Huber, Dr. Stephanie Tobin, Josephine Esposto, Sean Condie

ACKNOWLEDGEMENTS

Session Moderators:

Dr. Leslie Kerr, Dr. Neil Fournier, Dr. Cayleih Robertson, Dr. Jenifer Hendel

Presentation Evaluators:

Dr. Sanela Martic, Mark Seegobin, Samantha Logan

Volunteers:

Thomas Burnside, Blythe Ferguson

FINANCIAL SUPPORT

SCHEDULE

TIME	EVENT
8:30 AM – 9:00 AM	**Registration**
9:00 AM - 9:05 AM	**Opening Remarks** ***Dr. Robert Huber and Dr. Stephanie Tobin***
9:05 AM – 10:00 AM	**Session 1: Trainee Presentations** ***Moderator: Dr. Leslie Kerr*** 1. **William Kim** - *Mutations in CLN5 disease impact lysosomal function and alter protein trafficking* 2. **Stephanie Sissons** - *Reactivating memory engrams to reverse impaired fear memory recall after chronic seizures* 3. **Melanie Marlow** - *Investigating the DNA-binding properties of the TATA-binding protein in* Giardia intestinalis 4. **Hannah Kavanagh** - *Adiposity in cardiac cachexia: Monocrotaline and its influence on the beiging of adipose tissue*
10:00 AM – 10:30 AM	**KEYNOTE SPEAKER** **Jeanette Wilkins,** Genetic Counsellor, Peterborough Regional Health Centre *Genetics in Healthcare: From Rare Insights to Everyday Impact*
10:30 AM – 10:45 AM	*Coffee/Tea Break*
10:45 AM – 11:45 AM	**Session 2: Trainee Presentations** ***Moderator: Dr. Neil Fournier*** 1. **Galair Prevost** - *The efficacy and timing of cytokinin-induced inhibition of frog virus 3 (FV3) replication* 2. **Gillian Ekins** - *Examining the mechanism of action for weight gain in a female rodent model of epilepsy* 3. **Isabelle Decorso** - *Oxidative stress and* Giardia *flavohemoglobins: A new perspective* 4. **Joel-Anthony Seaton** - *Isolation of soybean agglutinin for applications in glycan synthesis*
11:45 AM – 12:45 PM	*Lunch Break*

SCHEDULE

TIME	EVENT
12:45 PM – 1:15 PM	**KEYNOTE SPEAKER** **Dr. Jean-Paul Desaulniers,** Professor in Chemistry, Ontario Tech University *Photoswitchable siRNAs: A Gene-Silencing Symphony Light Show*
1:15 PM – 2:15 PM	**Session 3: Trainee Presentations** ***Moderator: Dr. Cayleih Robertson*** 1. **Linh Tran** - *Exploring the relationship between cytokinin and the encystation process in* Giardia intestinalis 2. **Pakin Pongpaiboon** - *Analysing sex-dependent effects of monocrotaline (MCT) through glycogen storage* 3. **Sean Condie** - *Exploring the function of the protein-sorting receptor sortilin in* Dictyostelium discoideum 4. **Lexi Thivierge** - *Role of the medial prefrontal cortex in mediating relief learning*
2:15 PM – 2:30 PM	*Coffee/Tea Break*
2:30 PM – 3:00 PM	**KEYNOTE SPEAKER** **Dr. Janice Strap,** Associate Professor in Biology, Ontario Tech University *Regulating the Matrix: Environmental and Molecular Control of Bacterial Cellulose in* Komagataeibacter *spp.*
3:00 PM – 3:45 PM	**Session 4: Trainee Presentations** ***Moderator: Dr. Jenifer Hendel*** 1. **Samer Owiar** - *Investigating the role of Cln5 in S-palmitoylation pathway in* Dictyostelium discoideum 2. **Jordan Webb** - *Evidence of allocentric spatial learning in male rats with large lesions of the hippocampus* 3. **Noah Fiorucci** - *Sex-specific responses of monocrotaline-induced cardiac cachexia on skeletal muscle inflammation, myofiber cross-sectional area, and intramuscular fat*
4:00 PM	**Presentation Awards & Closing Remarks** ***Awards Presented by Dr. Neil Emery***

ABSTRACTS

Keynote Speaker
Jeanette Wilkins, M.Sc., CCGC
Genetic Counsellor, PRHC Genetics Program, Peterborough Regional Health Center

Genetics in Healthcare: From Rare Insights to Everyday Impact

This lecture provides a bird's-eye view of clinical genetic services and explores how genetic knowledge and testing impact health outcomes. We will trace the evolution of genetics from a rare, specialized field to an essential tool integrated across all areas of healthcare. Through case examples and ethical discussions, you'll gain insight into the role of genetics in patient care and medical genetic decision-making. The specific objectives of this talk are to (1) explore clinical genetics through engaging case studies, (2) understand how genetic testing and knowledge improve health outcomes and patient care, and (3) learn about the roles of Genetic Counselors and Clinical Geneticists in healthcare.

Jeanette Wilkins is a Canadian Board-Certified Genetic Counsellor at Peterborough Regional Health Centre. She has played a pivotal role in expanding genetic services, leading the transformation of an outreach program into a comprehensive regional program covering cancer, prenatal, and general genetic services. Jeanette earned her Bachelor of Science (Honours) in Molecular Biology and Genetics from the University of Guelph, followed by a Master of Science in Genetic Counselling from the University of British Columbia. With expertise in both patient care and program development, she continues to advance the role of genetics in healthcare.

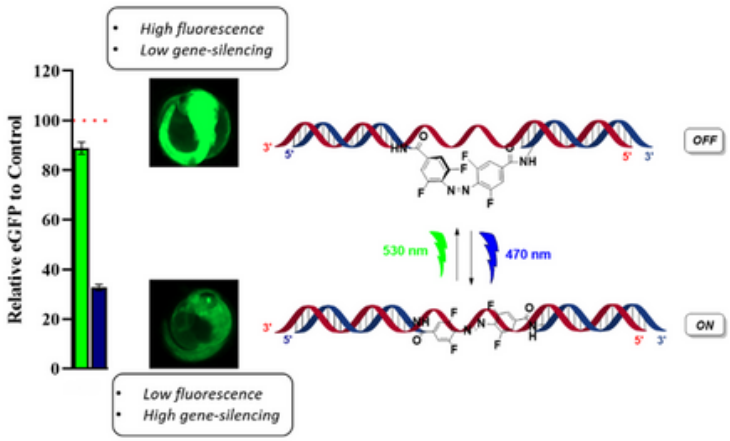
ABSTRACTS

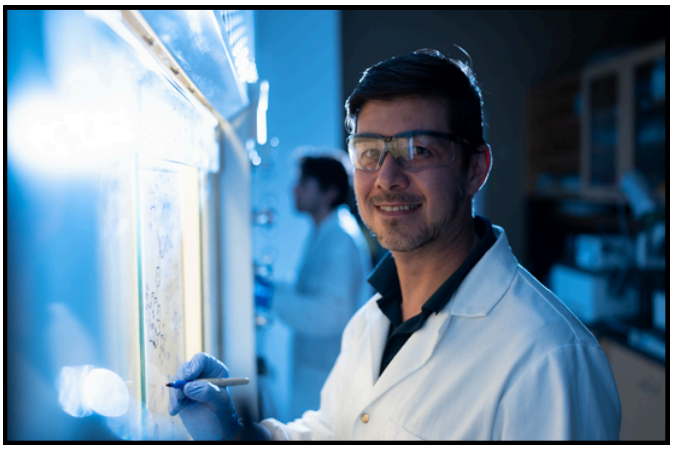
Keynote Speaker

Jean-Paul Desaulniers, Ph.D.
Professor, Faculty of Science: Chemistry, Ontario Tech University

Photoswitchable siRNAs: A Gene-Silencing Symphony Light Show

Short-interfering RNAs (siRNAs) are gene-silencing oligonucleotides that have an important role in biotechnology applications and oligonucleotide-based therapeutics. For ex-ample, siRNAs are important to elucidate gene function and to characterize complex biological pathways. One of the challenges in using siRNAs involves controlling its use once adminis-tered inside of the cell. One way to control an siRNA material is to use a photoswichable molecule called azobenzene. Azobenzene is characterized by two λmax areas, a π to π* transition in the UV area that has a large extinction coefficient, and a weak n to π* transition in the visible area. For azobenzene, UV light promotes a *trans* to *cis* isomerization via π to π* excitation, whereas visible light promotes a *cis* to *trans* isomerization via n to π* excitation. We have developed an siRNA material that contains an azobenzene embedded within its structure and this can be reversibly activated and inactivated within cells with visible and UV light, respectively. We have also developed *ortho*-functionalized tetra-chlorinated and tetra-fluorinated azobenzene derivatives that are capable of isomerizing between *cis* and *trans* with different colored lights. Our tetra-fluorinated azobenzene-containing siRNA system is highly robust, and gene silencing can be controlled *in vivo* in Japanese Medaka (*Oryzias latipes*).





Dr. Jean-Paul Desaulniers is a Professor of Chemistry at Ontario Tech University, and holds an Ontario Tech Research Excellence Chair in Chemical Biology where he leads cutting-edge research in the field of chemical biology. His expertise centers on nucleic acid chemistry and biology, with a focus on designing and synthesizing novel nucleic acid derivatives for therapeutic applications like RNAi.

A native of London, Ontario, Dr. Desaulniers completed his undergraduate degree in Chemistry and Biochemistry at Western University in 2000. He then completed his PhD at Wayne State University under the mentorship of Dr. Christine Chow in 2005, where his research focused on the organic synthesis of pseudouridine and its role within RNA. Following his doctoral studies, Dr. Desaulniers held a prestigious American Cancer Society postdoctoral fellowship at the University of Michigan, working in Dr. Anna Mapp's lab until 2008. There, he designed small-molecule inhibitors to target protein-protein interactions.

Since joining Ontario Tech in 2008, Dr. Desaulniers has built a research program focused on nucleic acid chemical biology. His work explores the synthesis of nucleic acid derivatives, such as photoswitchable siRNAs, which can be used to control gene silencing in biological systems. His group has achieved notable success in creating tetrafluorinated azobenzene-functionalized siRNAs, which can be optically activated using light to regulate gene expression in model organisms like Japanese Medaka fish embryos with collaborator Dr. Simmons from Ontario Tech. His lab was awarded a patent in 2020 for this work, and commercialization funds from NSERC I2I and Lab2Market to further develop this technology.

ABSTRACTS

Keynote Speaker

Janice Strap, Ph.D.
Associate *Professor, Faculty of Science: Biology, Ontario Tech University*

Regulating the Matrix: Environmental and Molecular Control of Bacterial Cellulose in *Komagataeibacter* spp.

Bacterial cellulose (BC) is not merely a structural polymer, it is the molecular matrix through which many microbes anchor themselves, withstand environmental stress, and mediate interkingdom interactions. In my lab, we use *Komagataeibacter* spp. as a model to understand how environmental and intracellular cues converge to control BC biosynthesis. These acetic acid bacteria produce highly crystalline cellulose that serves as the structural scaffold of their biofilm, and notably, this process is finely regulated by signals both from within and beyond the cell. This talk will highlight our work uncovering how plant-derived phytohormones modulate cellulose biosynthesis, bacterial morphology, and cellulose crystallinity in a concentration-dependent manner. Certain hormones, like indole 3-acetic acid, inhibit cellulose yield while stimulating growth, decoupling biomass accumulation from matrix synthesis. These findings provide insights into how plant-associated bacteria may synchronize their biofilm physiology with fruit ripening stages. Beyond exogenous hormones, our work demonstrates that *Komagataeibacter* also synthesizes its own phytohormones, positioning this genus as both a sensor and contributor to the plant hormone milieu. We've identified key regulatory nodes in this signaling network that integrates oxidative stress, quorum sensing, and redox homeostasis with cellulose biosynthesis. Together, these findings advance our understanding of the environmental and molecular regulation of BC in *Komagataeibacter* and underscore the sophisticated strategies these bacteria employ to thrive in plant-associated niches.

Dr. Janice Strap is a microbiologist and Associate Professor in the Faculty of Science at Ontario Tech University. She earned her Ph.D. in Microbiology and Biotechnology at the University of Alberta and completed post-doctoral research at the University of Idaho in the Department of Microbiology, Molecular Biology and Biochemistry, as well as the Environmental Biotechnology Institute (EBI). Dr. Strap's current research focuses on bacterial biofilms, environmental microbiology, and the molecular mechanisms underlying cellulose biosynthesis in acetic acid bacteria. A key focus of her work is understanding how environmental cues, including plant-derived molecules, influence bacterial physiology and behaviour, particularly in the context of host-microbe interactions. Her lab has made important contributions to the understanding of how bacterial cellulose enhances ecological fitness, and how its production can be modulated by both natural signaling molecules and synthetic inhibitors. By integrating molecular genetics, bioinformatics, and chemical biology, her interdisciplinary research has broad implications across biotechnology, agriculture, and materials science.

ABSTRACTS
Trainees

Mutations in CLN5 disease impact lysosomal function and alter protein trafficking

William D. Kim (1), Samer A. Owiar (1), Cassandra H. Pyne (2), Robert J. Huber (1,2)
1. Environmental and Life Sciences Graduate Program, Trent University, Peterborough, Ontario, Canada
2. Department of Biology, Trent University, Peterborough, Ontario, Canada

Batten disease is a devastating neurological disease that primarily affects children but occurs in all ages. The disease is caused by mutations in ceroid lipofuscinosis neuronal (CLN) genes (CLN1-CLN8, CLN10-CLN14), of which there are 13 total. Each gene is linked to a distinct subtype of Batten disease (e.g., mutations in CLN5 cause CLN5 disease). CLN5 is a soluble protein that localizes to the endoplasmic reticulum and lysosome, as well as extracellularly. CLN5 has been implicated in many cellular processes such as autophagy and protein degradation. However, its precise function within the cell is still under investigation. Previous work showed that mutations in residues that are N-glycosylated in CLN5 impact the structure and intracellular trafficking of the protein, as well as its function within lysosomes. While some of these mutations have been identified in patients with CLN5 disease, the effects of other more common mutations have not been previously studied. Here, we used the social amoeba *Dictyostelium* to study the molecular effects of several CLN5 disease-causing mutations. We observed that mutations in CLN5 increase the number of acidic vesicles, indicating potential lysosomal dysregulation. The mutations also alter intracellular and extracellular lysosomal enzyme activity, which correlates to effects on protein ubiquitination and proteasome-mediated degradation. This is supported by our findings that CLN5 disease-causing mutations alter the intracellular and extracellular amounts of various lysosomal enzymes. As described above, CLN5 is detected extracellularly, and we observed that CLN5 disease-causing mutations significantly reduce the secretion of Cln5. Altogether, this work reveals the effects of CLN5 disease-causing mutations on lysosomal function, protein degradation, and intracellular trafficking.

Reactivating memory engrams to reverse impaired fear memory recall after chronic seizures

Stephanie Sissons (1), Hugo Lehmann (1), Neil M. Fournier (1)
1. Department of Psychology, Trent University, Peterborough Ontario, Canada

Epilepsy is a chronic neurological disorder characterized by recurrent seizures and cognitive impairments, including memory deficits. Up to 50% of individuals with epilepsy experience persistent memory deficits, highlighting the necessity for targeted intervention. Memory formation relies on the activity of patterns of neuronal populations engaged at the time of a learning experience. Reactivation of these cells at the time of retrieval is believed to be necessary for successful memory recall. Pathological activity has been shown to disrupt engram circuit connectivity which could lead to their defective reactivation during memory retrieval. However, whether disruption in engram cell reactivation contributes to cognitive deficits in epilepsy remains largely unknown. To better understand the mechanisms underlying seizure-related memory impairment, we employed an activity-dependent neuronal tagging and chemogenetic viral strategy to selectively visualize and manipulate memory engrams. To achieve this, we “tagged” CA1 hippocampal memory engrams formed during context fear learning in male rats. After successful memory encoding, the rats underwent either repeated convulsive seizures induced by pentylenetetrazole (PTZ) or received vehicle injections over a two-week period. Preliminary results show that context fear memory recall was significantly impaired following PTZ seizures. However, pharmacogenetic reactivation of the original engram ensemble was sufficient to partially reverse the memory deficits associated with PTZ seizures. These results demonstrate that at least part of the original memory engram remains intact despite extensive pathological circuit changes induced by repeated seizure activity. Furthermore, our findings also suggest that reactivating this circuit can drive memory recall indicating that memories are not permanently lost with chronic seizures but can be potentially recoverable.

Investigating the DNA-binding properties of the TATA-binding protein in *Giardia Intestinalis*

Melanie Marlow (1), Kieran Freitag (1), Steven Rafferty (2), and Janet Yee (3)
1. Environmental and Life Sciences Graduate Program, Trent University, Peterborough, Ontario, Canada
2. Department of Chemistry, Trent University, Peterborough, Ontario, Canada
3. Department of Biology, Trent University, Peterborough, Ontario, Canada

Giardia intestinalis is a protozoan parasite responsible for the disease Giardiasis. As a highly divergent eukaryote, *Giardia* lacks several characteristic organelles, including peroxisomes, stacked Golgi, and has a reduced form of mitochondria called mitosomes. The TATA-binding protein is a transcription factor that helps to regulate the genes transcribed by three RNA polymerases: I, II, and III. The TATA-box binding protein (TBP) homolog from *Giardia intestinalis* (gTBP) is highly divergent as it lacks key phenylalanines required for binding and unwinding double-stranded DNA. Additionally, structural modelling shows that the gTBP DNA-binding pocket is narrower than other eukaryotic TBPs. Our lab has been investigating the DNA-binding properties of gTBP using electromobility shift assays (EMSA). We have determined that gTBP preferentially binds to single-stranded DNA (ssDNA) over double-stranded DNA (dsDNA) and that gTBP binds DNA in two distinct modes we have named A mode and B mode. For the A mode, gTBP binds to ssDNA that contain four or more consecutive guanine bases. For the B mode, gTBP binds in a manner dependent on DNA structure rather than sequence. We used base stacking energy potentials between adjacent dinucleotides as a simple proxy for per-nucleotide flexibility and found that gTBP binds highly flexible regions of DNA rich in cytosine and thymine nucleotides. Overall, we present new insights into transcription in *Giardia* by investigating gTBP DNA-binding specificity. Future work will include characterizing gTBP binding in vivo using techniques such as co-immunoprecipitation to determine protein-binding partners and chromatin-immunoprecipitation to confirm DNA-binding preferences.

Adiposity in cardiac cachexia: Monocrotaline and its influence on the beiging of adipose tissue

Hannah Kavanagh (1), Stephanie Tobin (1), Holly Bates (1)
1. Department of Biology, Trent University, Peterborough, Ontario, Canada

Cardiac cachexia, a rapid and severe weight loss associated with chronic heart failure, resulting from dramatic changes in metabolic processes. While lean body mass loss is noted in cardiac cachexia models using monocrotaline (MCT) injections, the impact on adipose tissue, particularly inguinal (subcutaneous) white adipose tissue (iWAT), remains unclear. This study investigates whether beiging occurs in the iWAT depot in response to low-dose MCT. We quantified the expression of key genes involved in adipose tissue function (Ucp1, Prdm16, and Zfp516) and inflammatory cytokines (Il16 and Tnf alpha) using qPCR. Statistical significance was assessed using t-tests. We hypothesized that male MCT-injected mince would show increased expression of these genes compared to female MCT-injected mice, and male and female saline-injected mice. Previous findings in the Tobin lab demonstrated that male MCT-injected mice had a significant reduction of body mass. Analysis revealed that Zfp516 expression was elevated in the male and female MCT-injected mice. Similarly, Il16 and Tnf alpha showed increased expression, with Tnf alpha exhibiting a significant elevation compared to the male saline-injected mice. However, the expression of Ucp1 and Prdm16 were inconclusive. These findings contribute to understanding the role of adipose tissue beiging in cardiac cachexia, offering insights into the disease's progression and potential therapeutic strategies to combat cardiac cachexia-related weight loss.

The efficacy and timing of cytokinin-induced inhibition of frog virus 3 (FV3) replication

Galair Prevost (1,2), RJ Neil Emery (1,2), and Craig Brunetti (1,2)
1. Environmental and Life Sciences Graduate Program, Trent University, Peterborough, Ontario, Canada
2. Department of Biology, Trent University, Peterborough, Ontario, Canada

Cytokinins (CKs) are signaling molecules that are present in all kingdoms of life. These N6-adenine derivative molecules are vital in the development and differentiation of cells. In vertebrate systems, there are still gaps of knowledge in our understanding of what these molecules can do and the mechanisms behind their documented activity. Recently, we observed that specific CKs can significantly inhibit viral replication. To examine these effects we employed frog virus 3 (FV3), a double-stranded virus that primarily affects ectothermic vertebrates. Expanding on previous work and are testing the antiviral activity of two aromatic CKs, kinetin and kinetin riboside (KR). We investigated the timing of antiviral CK effects and determined their optimal concentrations for activity. Our results show that, at 15 μM of kinetin and KR, there is a significant decrease in FV3 replication. We then compared a 1-hour pre-treatment to a concurrent treatment of the CKs and our results show that both timing treatments inhibited FV3 replication, although concurrent treatments showed a greater magnitude of decreased viral activity ($p<0.05$). These results confirm the ability of CK to inhibit viral replication and further demonstrate that concurrent application with infection give the clearest effects. This will help future work that will delve further into the mechanisms behind CK inhibition of viral infection.

Examining the mechanism of action for weight gain in a female rodent model of epilepsy

Gillian Ekins (1), Neil Fournier (1), Holly Bates (2)
1. Department of Psychology, Trent University, Peterborough, Ontario, Canada
2. Department of Biology, Trent University, Peterborough, Ontario, Canada

Epilepsy is a prevalent disorder which impacts several different biological systems, which can result in behavioural, affective, cognitive, and physiological changes. Rodent models, such as electrical kindling, simulate neurological changes associated with epilepsy by repeatedly stimulating a brain region over several weeks. In addition to memory impairments and increased rates of anxiety and panic disorders, obesity is also a common comorbid diagnosis in individuals with drug-resistant epilepsy. Rodent models of epilepsy tend to show a pattern of increased weight in kindled rodents compared to controls. To date, the relationship between epilepsy and weight is not clear. The present study examines female Long-Evans rats (n=20) kindled via the basolateral amygdala, which is a location prone to epileptic changes, to 75 stimulations and how their weight, blood glucose and food consumption differs from control rodents. Preliminary results suggest that kindled rats gain weight significantly faster ($p>0.001$) and consume more food compared to control rats ($p=0.055$). Additionally, kindled rats demonstrate higher fasting blood glucose levels as well as a reduced blood glucose response to intraperitoneal injections of dextrose compared to control rats. This study aims to explore the relationship between epilepsy and weight gain by examining glucose metabolism and food consumption in kindled rats. Understanding these connections may provide insights into the physiological impact of drug-resistant epilepsy in patients who live with this disorder.

Oxidative stress and *Giardia* flavohemoglobins: A new perspective

Isabelle Decorso (1), Steven Rafferty (2)
1. Environmental and Life Sciences, Trent University, Peterborough, Ontario, Canada
2. Department of Chemistry, Trent University, Peterborough, Ontario, Canada

Giardia flavohemoglobin (gFlHb) is the sole heme enzyme in *Giardia intestinalis*, a microaerotolerant protist that causes giardiasis, the most common parasitic intestinal infection worldwide. gFlHb is primarily known for protecting the parasite against nitrosative stress in mammalian small intestines. However, my research suggests a potential role for gFlHb in oxidative stress. Specifically, hydrogen peroxide stimulates gFlHb's NADH oxidase activity nearly threefold and induces its expression approximately twofold, suggesting a regulatory response to oxidative conditions. Additionally, the active site of flavohemoglobins resemble those of type I and II peroxidases, enzymes that typically break down peroxides. In *E. coli*, flavohemoglobin acts as an alkyl hydroperoxide reductase under anaerobic conditions, breaking down peroxide. When similar experiments were conducted aerobically with gFlHb, the enzyme unexpectedly generated hydrogen peroxide rather than degrading it. This was demonstrated both indirectly using a horseradish peroxidase/o-dianisidine assay and directly using a free radical analyzer equipped with a hydrogen peroxide sensing electrode. Furthermore, the accumulation of hydrogen peroxide led to heme degradation, suggesting a possible self-regulatory mechanism. These findings support the idea that gFlHb may participate in a feedback loop in which excessive hydrogen peroxide generation triggers heme degradation, ultimately downregulating gFlHb expression to conserve cellular resources.

Isolation of soybean agglutinin for applications in glycan synthesis

Joel-Anthony Seaton (1), Jenifer Hendel (2)
1. Department of Biology, Trent University, Peterborough, Ontario, Canada
2. Department of Chemistry, Trent University, Peterborough, Ontario, Canada

Glycans, complex carbohydrate polymers, are vital macromolecules in all living organisms, contributing to energy metabolism, cell recognition, adhesion, and cellular structural integrity. The field of glycoscience has grown in importance due to the extensive biological roles of glycans. However, research in this area faces a significant challenge: the limited accessibility and availability of complex glycans. Traditional production methods, such as chemical or chemoenzymatic synthesis, are difficult to scale because of their synthetic complexity or the high cost of enzymes. An alternative option is isolating glycans from natural sources, but existing methods are designed for small-scale isolation and have not been adapted for largescale production. This study aims to optimize methods for isolating milligram quantities of the high mannose glycan M9, which is biologically significant due to its presence on viral shields and its overexpression in certain cancers. M9 is found in soybeans, specifically as the sole glycoform on the soybean agglutinin (SBA) glycoprotein. I will present our research that is focused on comparing methods of isolating and purifying SBA with the goal of optimization. In this workflow, the glycans are cleaved from SBA using NaOCl and analyzed using MALDI-MS. Future work will focus on optimizing glycan purification processes such as hydrophobic interaction chromatography with cotton. The outcome of this study and future work, will be scalable methods for isolating complex N-glycans from natural sources. These materials will lead to a better understanding of the biological roles of N-glycans and new therapeutic applications.

Exploring the relationship between cytokinin and the encystation process in *Giardia intestinalis*

Linh Tran (1), Neil Emery (1), and Janet Yee (1)
1. Department of Biology, Trent University, Peterborough, Ontario, Canada

Giardia intestinalis is a protist that causes diarrheal disease in humans. The infection initiates when a host ingests infectious cysts in contaminated water. Once inside the host, the cyst hatches in the stomach to release the trophozoite form, which swims down and proliferates within the intestinal tract. In the small lower intestine, some trophozoites develop back into cysts, which are released in the feces where they become a source of new infections. The conversion of the trophozoite into the infectious cyst form is called encystation and this can be induced in laboratory cultures. Cytokinins (CKs) are signaling molecules that are well-studied in plants for their important roles in the growth and developmental processes. Recently, CK metabolites have been found to be present in all kingdoms of life. Preliminary work done suggested that *Giardia* can metabolize CKs during encystation, and N6-benzyladenosine (BAR), a synthetic form of CK, may have inhibitory effect on the *Giardia* encystation process. My research is to investigate the roles BAR has on encystation. Efficiency of encystation of the *Giardia* cultures was monitored by the appearance and localization of a cyst wall protein (CWP1) by using immunofluorescence microscopy, and by quantifying the increase of this protein by using immunoblotting. Those experiments were performed in parallel with endogenous measurements of relevant metabolites inside *Giardia* cells during encystation using liquid chromatography-high resolution mass spectrometry. Lastly, flow cytometry was used to analyze and monitor the G2 phase of the cell cycle that is the entry point to the encystation pathway.

Analysing sex-dependent effects of monocrotaline (MCT) through glycogen storage

Pakin Pongpaiboon (1), Stephanie Tobin (1)
1. Department of Biology, Trent University, Peterborough, Ontario, Canada

The monocrotaline (MCT) model is a pharmaceutical approach to studying cardiac cachexia, a multiorgan syndrome that results in extreme lean body mass and skeletal muscle loss in heart failure patients. Although the MCT model of cardiac cachexia is prevalent, the physiological basis for sex-dependent effects is not well-studied. Previously, our lab has detected results that suggest that male mice are more adversely affected by MCT-induced cachexia as the whole-body weight of male but not female mice was reduced. It was found that the cross-sectional area of skeletal muscle was reduced in male and female samples while fat was reduced in male samples. As skeletal muscles are a site for glycogen storage and many skeletal wasting conditions are associated with reduced glycogen, we suspected that glycogen could be different in MCT-treated mice, particularly in males with reduced body and adipose mass. We hypothesized that skeletal muscle glycogen storage of males will be more adversely affected than females treated with MCT. Greater glycogen storage in females will provide support that skeletal muscle and body weight are lost in a sex-dependent fashion through glycogen changes in the MCT model of cardiac cachexia. Periodic Acid Schiff (PAS) stain was applied to paraffin-embedded skeletal muscle sections of the tibialis anterior and gastrocnemius from the previous study to quantify glycogen. The results and analysis, which are in progress, will provide a physiological basis for skeletal muscle loss in MCT-induced cachexia.

Exploring the function of the protein-sorting receptor sortilin in *Dictyostelium discoideum*

Sean Condie (1), Robert Huber (2)
1. Environmental and Life Sciences Graduate Program, Trent University, Peterborough, Ontario, Canada
2. Department of Biology, Trent University, Peterborough, Ontario, Canada

Intracellular protein sorting is essential for supplying resources needed by the organelles of a cell. For example, lysosomes require enzymes that break down bulk material like food sources and defective cellular components. How these enzymes get to the lysosome is well studied in mammals and begins with sorting receptors in the Golgi apparatus. Sorting receptors bind to an enzyme in the Golgi apparatus and allow the enzyme-receptor conjugate to be packaged into a vesicle and sent into the cytosol. One of these sorting receptors is sortilin (SORT1), which also participates in the trafficking of other non-enzymatic proteins. In our work, we use the social amoeba *Dictyostelium discoideum* to study sortilin-mediated protein sorting. *D. discoideum* has been used for close to a century to better understand conserved cellular and developmental processes and encodes a homolog of sortilin (Sort1). Unlike mammalian cells, *D. discoideum* has a unique life cycle comprised of both unicellular and multicellular phases. Cells lacking sort1 display reduced proliferation in liquid culture but grow normally on solid agar containing their bacterial food source. Immunofluorescence imaging revealed a cytokinesis defect, smaller and less distinct secretory vesicles, and poor actin recruitment to the cell membrane. Western blots revealed reduced intracellular autocrine proliferation repressing factors and reduced intracellular density sensing factors. We also generated a custom antibody against Sort1, enabling a significant advantage in future assay designs. Together, this research lays the foundation for establishing *D. discoideum* as a model organism for studying sortilin-mediated protein trafficking.

Role of the medial prefrontal cortex in mediating relief learning

Lexi Thivierge (1), Neil Fournier (1)
1. Department of Psychology, Trent University, Peterborough, Ontario, Canada

Mistakenly associating danger with a harmless stimulus can lead to maladaptive fear responses. Most research has focused on fear extinction, where a once-threatening cue is repeatedly shown without consequences, reducing fear. However, animals can also learn to associate cues with safety in other ways. Previous studies have shown that stimuli encountered after an aversive event can signal relief, also known as relief learning. In this study, rats undergo either threat conditioning, where a tone (5000 Hz, 12s) precedes a foot shock (1.0mA, 1.5s), or relief learning, where a shock precedes a tone. This pairing is presented 6 times, each one minute apart. Twenty-four hours later, a recall test is conducted by presenting the tones in the training environment. Results show that the relief learning rats exhibit significantly less freezing during the tone presentation compared to the threat-conditioned rats, indicating that they associate the tone with relief from the aversive stimulus. Human neuroimaging and animal studies indicate that ending an aversive stimulus activates brain regions like the medial prefrontal cortex (mPFC), which regulate defensive responses. Using c-fos as a marker for neuronal activation, this study revealed significant activation of the infralimbic cortex in relief learning animals. Future experiments will investigate activity in projections to the mPFC in relief-conditioned rats using a viral green fluorescence protein (GFP)-conjugated retrograde tracer. This research aims to enhance our understanding of the neural circuitry involved in relief learning and how the mPFC regulates behaviour in response to threats.

Investigating the role of Cln5 in S-palmitoylation pathway in *Dictyostelium discoideum*

Samer Owiar (1), William D. Kim (1), and Robert J. Huber (1,2)
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S-palmitoylation is a post-translational modification that adds palmitic acid onto a protein to regulate its trafficking and function. S-palmitoylation is a subcategory of lipidation and what makes it unique is that it is the only form of lipid modification that is reversible. Previous work suggested that the lysosomal enzyme CLN5 may function as a depalmitoylase, which is a protein that removes palmitate from palmitoylated proteins. Mutations in CLN5 cause a rare form of neurodegeneration called CLN5 disease, which is a subtype of neuronal ceroid lipofuscinosis. In my research, I am investigating the potential depalmitoylase activity of Cln5 in the eukaryotic microbe Dictyostelium discoideum, which is an established model system for studying CLN5 disease. There are two main objectives of my research. First, I optimized the use of acyl-Resin Assisted Capture (acyl-RAC) in D. discoideum to capture palmitoylated proteins from wild-type (WT) and cln5- cells. I then screened several candidate proteins and identified proteins that were differentially palmitoylated in the two cell lines, which aligned with the predicted depalmitoylase activity of CLN5. In my second objective, I am assessing the effect of a palmitoylation inhibitor on the growth and multicellular development of D. discoideum, which will lead to a better understanding of the role of palmitoylation in regulating conserved cellular and developmental processes. Overall, this work will provide insight into the molecular function of CLN5 and the importance of palmitoylation during the D. discoideum life cycle.

Evidence of allocentric spatial learning in male rats with large lesions of the hippocampus

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The hippocampus (HPC) plays a crucial role in spatial learning and navigation. It helps to create cognitive maps of the environment, which include two main types of information: allocentric (viewpoint invariant), and egocentric (viewpoint dependent). The HPC supports allocentric spatial learning, but is not necessary for egocentric. In the Morris water task, rats with HPC lesions often rely on egocentric strategies, whereas control rats use allocentric strategies for more efficient navigation. Evidence suggests that other types of memory deficits depend on the extent of HPC lesion, however, it is unclear if this relationship applies to spatial memory. My research aimed to investigate whether allocentric and egocentric strategy use could be predicted by the extent of HPC damage. Analyses provided remarkable evidence of consistent allocentric strategy use in a subgroup of lesioned rats. Contrary to the literature, the current evidence suggests that allocentric learning is not solely dependent on the HPC.

Sex-specific responses of monocrotaline-induced cardiac cachexia on skeletal muscle inflammation, myofiber cross-sectional area, and intramuscular fat

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Cardiac cachexia (CC) is a severe comorbidity of chronic heart failure characterized by muscle wasting, inflammation, and metabolic dysregulation. Although sex differences influence disease progression, their role in CC remains understudied. This study investigates sex-dependent effects of monocrotaline (MCT)-induced CC on skeletal muscle inflammation, myofiber cross-sectional area (CSA), and intramuscular fat mobilization. We predicted that as CC progresses, inflammation will be increased while myofiber CSA and intramuscular fat deposition will be reduced. C57BL/6N male and female mice (n=4) received subcutaneous injections of 200 mg/kg MCT or saline/DMSO for eight weeks. Tibialis anterior muscles were analyzed for inflammation via CD45 and F4/80 immunohistochemistry, intramuscular fat content via Oil Red O (ORO) staining, and myofiber CSA using laminin and ORO staining. Statistical analysis was performed using two-way ANOVA. MCT treatment significantly reduced CSA in males ($p < 0.01$) across both cohorts, while no statistical difference was seen in females. CD45+ and F4/80+ cell staining revealed no significant difference in response to MCT in either sex. This suggests a potential delay in the inflammatory response, insufficient potency of MCT, or the involvement of alternative mechanisms in muscle wasting. Additionally, intramuscular fat content remained unchanged across all experimental conditions, likely due to the young age and dietary conditions of the mice. These findings suggest a sex-dependent susceptibility to MCT-induced CC, with males exhibiting greater muscle atrophy than females. Further research is needed to elucidate the underlying pathways driving these sex differences.

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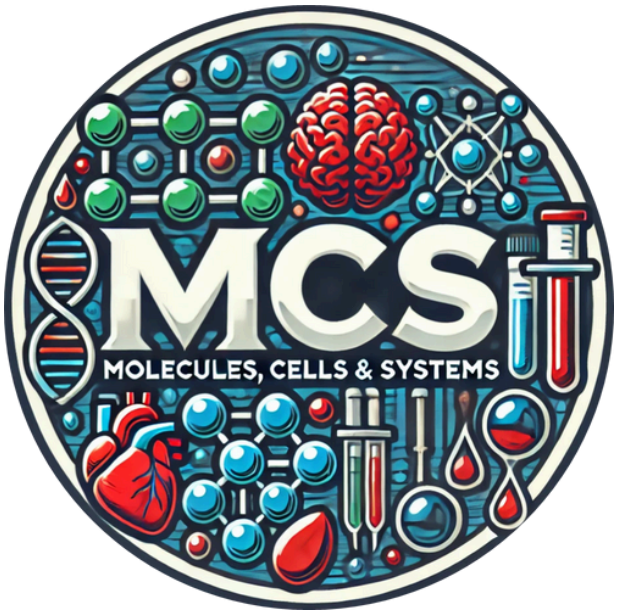

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