undergraduate
RESEARCH AWARDS
symposium

EXPERIENCE. KNOWLEDGE.

Faculty of Science 2022 Undergraduate Research Awards

Olivia BOVEN, Biology (Dr. Michael Russello) NSERC-USRA

Sex Ratios of American Pika (*Ochotona princeps*) in Myra-Bellevue Provincial Park

American pikas (*Ochotona princeps*) are considered by some to be an indicator species for the effects of climate change, so monitoring populations is of great importance. This study is the first study to quantify sex ratios at Myra-Bellevue Provincial Park, Kelowna, BC, Canada. Typically, pikas settle close to their natal range and mate with their nearest neighbour. Previous studies have found a roughly even sex ratio. To determine sex ratios at Myra-Bellevue, fecal pellets were collected at 100 latrines across 11 sites. A PCR (polymerase chain reaction) was performed for each sample using primers for the microsatellite Ocp10 (autosomal) and the SRY (Y-specific) fragment. Products were then run on an agarose gel and samples with an SRY band were counted as male. In total, there were 27 females, 55 males, and 18 samples that could not be sexed. The results show that the sex ratio park wide was 2.03 males per females, with sex ratios within each site ranging from entirely male to 0.5 males per females. It is possible that more than one latrine may belong to an individual, however, so the sex ratios obtained here from fecal samples may not accurately align with the true populations. Moving forward, these same samples will be analyzed using a Genotyping-in-thousands by sequencing (GT-seq) panel under development, which will enable more accurate sexing and will be capable of assigning multiple latrines to a single individual. Additionally, it will provide broader genomic data, allowing us to study population connectivity between sites at Myra-Bellevue.

Liam BYSTROM, Chemistry (Dr. Kirsten Wolthers) NSERC-USRA

Investigating the Novel Complex Formation of B12-dependent Enzyme 5,6-LAM and Proposed Chaperone KamBC

The development of periodontal disease is closely associated with the formation of microbial layers, termed biofilm, in the oral cavity. Biofilm formation relies on the association (coaggregation) of various bacteria species. *Fusobacterium nucleatum*, a Gram-negative bacterium, has been linked with the coaggregation process and subsequent periodontal disease. Specifically, *F. nucleatum* ferments and removes lysine, a metabolite that can disrupt coaggregation. This project aimed to further characterize the lysine fermentation pathway of *F. nucleatum* to gain a better understanding of oral metabolites and periodontal disease. We focused on one enzyme from the fermentation pathway: 5,6-lysine aminomutase (5,6-LAM).

5,6-LAM uses a B_{12} cofactor for catalysis, which is occasionally inactivated. We hypothesized that two unknown gene products, KamB and KamC, form a dimeric complex (KamBC) and act as a B_{12} chaperone, restoring the B_{12} cofactor and enzyme activity. To test this hypothesis, we recombinantly expressed and purified KamB, KamC, and 5,6-LAM. We investigated binding using Native-PAGE, size exclusion chromatography, and isothermal titration calorimetry. We detected complex formation between KamB and KamC. However, experiments with 5,6-LAM and KamBC failed to show binding. This research has helped to provide further information about two unknown gene products in *F. nucleatum's* lysine fermentation pathway. The data we obtained here warrants further investigation into the exact role KamBC plays in lysine fermentation and periodontal disease.

Helen CHIANG, Biology (Dr. Deanna Gibson), URA

Can brain chemicals impact your gut? The investigation of kynurenine and serotonin on colitis and anxiety/depression

Inflammatory bowel disease (IBD) is characterized by digestive problems and gut inflammation. Approximately 30% of IBD patients also suffer with anxiety and/or depression as a result of their disease. An essential amino acid, Tryptophan breaks down to kynurenine (KYN) and serotonin (5-HT) which are implicated in inflammation and anxiety/depression. However, few studies have examined the relationship between depressive behaviors and serum, brain, and colon KYN and 5-HT levels in an IBD model. To fill this knowledge gap, we compared a mouse model ($Muc2^{-/-}$) which mimics ulcerative colitis; a subset of IBD, to a wild-type mouse. To assess anxiety/depression, the mice went through two maze tests - the open field (OF) and light/dark (L/D) test during the night and day time. To assess the biological response, tissues were assessed using enzyme-linked immunosorbent assay (ELISA) comparing serum, brain, and colon 5-HT and KYN metabolite levels. OF and L/D maze tests revealed that $Muc2^{-/-}$ and wild-type mice differ in behavior, but the behavioral differences in OF disappear at night time. Female $Muc2^{-/-}$ mice behaved differently from wild-type mice in the night-time L/D maze test. ELISA results revealed that serum KYN was significantly different between Muc2-/- and wild-type mice. 5-HT were not different across tissues at night, possibly explaining the night-time disappearance of behavioral differences seen in OF test. Daytime data is currently being analyzed. This project provides insights into IBD and its relationship between anxiety/ depression and potential avenues for new therapies.

Dylan DASILVA, Biology (Dr. Andis Klegeris) URA

Histone-Induced Neuroinflammation as a Novel Mechanism for Alzheimer's Disease

Alzheimer's disease (AD) is one of the leading causes of mortality worldwide. The lack of effective treatment makes research to determine the mechanisms of AD critical to prevent future suffering of patients. AD is characterized by neuroinflammation resulting in the death of neurons, and ultimately, impaired cognitive function. Microglia, the immune cells of the brain,

play a critical role in the progression of AD and can adopt an activated, pro-inflammatory state that contributes to neuronal death. However, factors that promote pro-inflammatory behavior of microglia are not fully understood and improved knowledge of this process could uncover novel therapeutic targets. Histones are proteins that exist inside of cells and are normally involved in organization of DNA. Previous studies have demonstrated that when released from dying cells into the extracellular space, histones induce the production of excess pro-inflammatory mediators by various immune cells. Our study investigated the role of extracellular histones in modulating the immune responses of both human and murine microglial cell lines, and their toxicity towards neuronal cells. We observed that, along with established immune stimulants, histone subtypes, H1 and H3, induced the secretion of pro-inflammatory molecules and increased the cytotoxicity of microglial cells. Additionally, we showed that the toll-like receptor 4 (TLR4) inhibitor, TAK-242, eliminated the histone-mediated increase in pro-inflammatory molecules, indicating that the mechanism of histone signalling is TLR4-dependent. To the best of our knowledge this is the first study to establish histones as mediators of neuroinflammation. These novel data contribute to our understanding of AD disease progression.

Amaury DE BURGOS, Mathematics (Dr. Wayne Broughton), URA

Packings & Transversals In Tripartite Graphs

The field of graph theory studies abstract drawings consisting of points and lines, called *vertices* and *edges* respectively. If within a drawing, referred to as a *graph*, the set of all vertices (points) can be partitioned into three subsets such that any two vertices in the same partition have no edge (line) between them, we call the graph *tripartite*. If within a graph, three vertices are all connected to each other, the vertices (along with the edges that connect them) form a *triangle*. If a set of triangles in a graph is such that no two triangles share an edge, the set is called a *packing*. If a set of edges in a graph touches all triangles in the graph, the set is called a *T-transversal*. The question our research aims to answer is *"When does the size of the biggest packing equal the size of the smallest T-transversal in a tripartite graph?"*. Knowing that in 2012, Lakshmanan et al. proved the size of the biggest packing equals the size of the smallest T-transversal in a *complete tripartite graph* (there is an edge between every pair of vertices from different partite sets), we considered a tripartite graph with only two complete sides (since a complete tripartite graph has three complete sides). In these graphs, dubbed *nearly-complete tripartite graphs*, we proved the size of the biggest packing equals the size of the smallest T-transversal.

Krista FULTON, Chemistry (Dr. Wesley Zandberg), URA

Evaluation of O-Glycan Extraction Methods for Gastrointestinal Mucin Characterization

The gastrointestinal (GI) tract is protected by one or two layers of mucus, protein that is decorated with thousands of glycans (glycan = polymer of 2 - 20 simple carbohydrates (Figure

1). It has been shown that even small changes in the structures of these mucus-borne glycans trigger irritable bowel disease and inflammation that may lead to colon cancer [1]. To study these changes *O*-glycans need to be cleaved from the protein backbone for characterization of the gut *O*-glycome. A few methods have been reported that supposedly extract the *O*-glycans without causing degradation but none of the products have been evaluated despite these extracted glycans already being used in high impact studies [2,3]. Three such methods were tested: the current standard protocol which uses ammonia carbonate [4], one that uses the strong base 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) [5], and one that uses bleach (NaClO) [6]. The products yielded by each method were assessed using capillary electrophoreses (CE) which separates the glycans by size and charge. The CE profiles of each extraction method's products showed that all are producing something different which suggests that some or all of the methods are causing degradation of the glycan. In future projects the products of these extraction methods will be further evaluated using high-pressure liquid chromatography (HPLC) to separate the glycan products for characterization using mass spectrometry (MS), fluorescence detection (FLD), or charged aerosol detection (CAD). All of these methods will provide more detailed information about the structure of the glycan products and help determine which extraction method is producing whole *O*-glycans.

Olivia IRELAND, Biology (Dr. Julien Gibon), NSERC-USRA

Investigating the role of ProBDNF on phosphatidylinositol 4,5-bisphosphate levels and caspase-3 cleavage in cortical neurons and PC12 cells

Persistent firing - a specific type of neuronal activity - is critical for working memory processing. This activity allows neurons to sustain their activity over a long period of time and it is regulated by secreted factors called neurotrophins. The precursor neurotrophin brain derived factor (ProBDNF) regulates persistent firing and synaptic activity in the entorhinal cortex. Upon maturation, ProBDNF is cut into two fragments: the prodomain peptide (pBDNF) and the mature neurotrophic factor (mBDNF). While it has been well established that mBDNF is an important regulator of synaptic growth and survival, little is known about the biological functions of ProBDNF and pBDNF. Previous research has shown that ProBDNF is capable of activating the p75 neurotrophin receptor (p75NTR) and consequently inducing synapse depletion and cell damage. On the other hand, recent evidence from the Gibon lab show that a mutated form of pBDNF inhibits persistent firing in a p75NTR-dependent manner. This project aims to understand how ProBDNF causes cell damage and how pBDNF inhibits persistent firing through their interactions with p75NTR. To measure the extent of cell damage, western blot analysis was used to quantify levels of cleaved caspase-3 (apoptotic markers) in PC12 cell models after incubation with ProBDNF. In addition, epifluorescence microscopy was used after acute application of pBDNF to measure the content of the membrane-bound phosphatidylinositol 4,5 - bisphosphate (PIP2) - a hypothesized inhibitor of persistent firing. The knowledge gathered from these experiments will help us understand how ProBDNF and pBDNF alter neuronal activity in the brain.

Jordan KATCHEN, Biology (Dr. Jason Pither), URA

Mycorrhizal Fungi and Plant Diversity within BC Forests

In forest ecosystems, most plants form symbioses with mycorrhizal fungi. These fungi form their symbioses by colonizing the plant's roots either intracellularly, as is the case with arbuscular mycorrhizal fungi (AMF), or extracellularly as with ectomycorrhizal fungi (EMF). These symbioses can be beneficial to plants, and through plant-soil feedbacks (PSFs) have the potential to influence the structure and functioning of forests. Using vegetation survey data from thousands of forest plots across British Columbia, we evaluate two competing hypotheses: (i) The EMF dominance hypothesis posits that local plant diversity decreases with increasing dominance of EMF symbioses, because these symbioses predominantly promote positive PSFs, thus favouring recruitment of the same host species over recruitment of different host species; (ii) The mycorrhizal dominance hypothesis posits that local plant diversity decreases with increasing dominance of either AMF or EMF symbioses, because both types of symbioses tend to promote positive PSFs. Using mixed effects models that account for the potential influences of other biotic and abiotic factors (e.g. canopy cover, mean annual temperature), we found greater support for the mycorrhizal dominance hypothesis. However, these findings remain to be evaluated against an appropriate null model, and their sensitivity to analytical choices (e.g. data exclusion criteria) remain to be evaluated. These findings are consistent with the hypothesis that AMF and EMF symbioses each tend to promote positive plant-soil feedbacks, and when either type of symbiosis dominates a forest stand, it tends to depress local plant species richness by promoting recruitment of the same species over different species.

Samantha KOLE, Physics (Dr. Alex Hill), URA

Faraday tomography: features in simulated space

Faraday tomography is a technique used in radio astronomy to help us map out magnetic fields and other related quantities in outer space. Improving the methods, equipment, and analysis used in this process can aid future investigations. In my research, I used models of the interstellar medium (ISM) – the thin, ionized, magnetized gas in space - to simulate Faraday tomography. Using graphical analysis and equations from literature, I aimed to find the best practical wavelength range for doing Faraday tomography on the ISM. I also investigated the limitations of using a model ISM for Faraday tomography. While I did not find the best wavelength range, I showed that the effectiveness of the process is affected by both the minimum and maximum wavelengths used. I found the model ISM limiting because of its resolution, but found practical ways to fill in data and make it usable. As new observations make the Faraday tomography technique feasible, this work builds our understanding of what these observations tell us about the real ISM.

Melanie LEMAIRE, Biology (Dr. Mark Rheault), URA

The effects of long term dietary TEA exposure on fruit fly morphology

All organisms must eliminate toxic compounds from their bodies. These toxic compounds can be formed inside the body of the organism or found in the environment. Fruit flies have Malpighian tubules (MTs) that are functionally similar to the renal tissues found in vertebrates. MTs are responsible for the formation of primary urine and the detoxification and excretion of xenobiotics such as pesticides, drugs, and plant defensive compounds. In this project I was interested to see if long term exposure (>200 generations) to a xenobiotic such as tetraethylammonium (TEA) would result in a decrease in morphometric measurements in adult fruit flies. I recorded body mass, external body structure measurements, MT mass, and MT measurements. Results showed that long term exposure to dietary TEA resulted in a decrease in body mass, a decrease in size in a number of anatomical structures, and a decrease in mass and overall size of MTs compared to control flies. Additionally, the results suggest that TEA may illicit sex specific effects in adult fruit flies in chronically exposed TEA diet populations compared to controls. This study could inform further beneficial research in pesticide detoxification and resistance which can have both agricultural and economic effects. It also provides foundational knowledge for new studies exploring sex specific functional differences in xenobiotic detoxification.

Clara LETEF, Biology (Dr. Sanjoy Ghosh), NSERC-USRA

Investigating factors controlling the impact of oxidized linoleic acid metabolites in cardiac fibroblasts

The Canadian diet has become rich in omega-6 polyunsaturated fatty acids (ω-6 PUFAs) such as linoleic acid (LA) found in corn oil. LA gets oxidized by 15-lipoxygenase, then glutathione peroxidase-4 (GPx4) to produce 13-HODE, an oxidized linoleic acid metabolite (OXLAM). Literature proposes that OXLAMs are harmful to cells, yet minimal studies investigate whether OXLAMs, or other factors present cause toxicity. This study used cardiac fibroblasts-collagen producing cells- with MitoSoX dye (measuring reactive oxygen generation in oxidative stress) for visualization, and resazurin assays for an approximation of cell viability. To determine the impact of OXLAMs in fibroblasts, we added LA with 15-lipoxygenase inhibitor, 9c(i472) - preventing OXLAM formation- and found OXLAMs essential at low doses. Hyperglycemic conditions were investigated by adding glucose to LA-treated cells, seeing an increase in death nd oxidative stress. Antioxidant supplementation with glutathione (GSH- coenzyme for GPx4) was studied by adding 2mM of GSH (healthy heart cell concentration) to cells with different concentrations of LA. When we inhibited OXLAM formation, cell viability decreased, concluding OXLAMs as essential for GSH function. Finally, the effects of metabolic substrates on OXLAMs were explored. Sodium pyruvate easily enters the mitochondria and is converted to energy (Krebs Cycle). When sodium pyruvate was added with LA and inhibitor, cell viability increased, showing the reversal of oxidative stress caused by excess OXLAMs. As the industrialization of the food industry continues, determining the impact these factors play on OXLAMs can lead to breakthroughs in the treatments of adverse health effects linked to high ω-6 PUFA diets.

Gustavo M. MARCHANI, Biology (Dr. Soheil Mahmoud), IURA

Life Bulb – Light production in cyanobacteria with fungal genes

Autobioluminescent organisms are considered to be the future of lighting as alternatives to electrical light production in an effort to reduce greenhouse gasses, energy consumption, and as a tool to deal with the climate crisis. Recently, the bioluminescent pathway of fungi has been elucidated which is very efficient thanks to its cyclical nature recycling its substrate. Nevertheless, this bioluminescent system has only been applied to plants which have a slow growth and limitations in scalability and placement. In this project, we will engineer the photosynthetic bacteria *Synechocystis sp.* PCC6803 to autobioluminesce using fungal genes to create an easily scalable, carbon-negative, and versatile light source alternative to help us reconcile with nature. We revised taxonomic variations in the genetic structure of the pathway of bioluminescent fungi coupled with observations of locals and researchers to select bioluminescent genes and to design our genetic circuit. These were then optimized and assembled using the Modular Cloning system for easier troubleshooting in cyanobacteria. We have successfully assembled most of the genetic constructs and established guidelines for the growth of our cyanobacteria. This proof-of-concept study will open the doors for a more versatile and efficient way of "growing " light.

Simona MASTROIANNI, Chemistry (Dr. Susan Murch), URA

Analysis of Kratom

Mitragyna speciosa (Korth.) Havil., or Kratom is an evergreen tree native to Southeast Asia. Kratom is traditionally consumed as a stimulant by labourers and can also be used as an opium substitute. Kratom has multiple varieties that are said to result in different effects when consumed. There are over 30 monoterpene indole alkaloids (MIAs) present in Kratom. The main active opioids are mitragynine and 7-hydroxymitragynine. The overall objective of this research was to analyze three varieties of Kratom: (1) Indonesia; (2) Borneo; and (3) Rifat (Red Vein Thai Kratom) by Liquid Chromatography-Mass Spectrometry (LC-MS) and analyze the chemical composition of each variety. Using untargeted metabolomics, the alkaloids mitragynine and 7-hydroxymitragynine were putatively identified in all three varieties. Rifat contained the highest level of MIAs compared to Indonesia and Borneo. Anthocyanins were putatively identified all three strains. Rifat contained the highest levels of anthocyanins, which is hypothesized to contribute to the strain’s distinguishable red colour.

Emily MELLORS, Physics (Dr. Rebecca Feldman), URA

Design and Assessment of a Low-Field Tabletop MRI System

Magnetic Resonant Imaging (MRI) systems consist of three powerful magnets. The magnetic fields produced by the magnets create an environment where signal can be elicited from a sample by exciting the MRI-visible protons with a signal oscillating at the resonant radio

frequency (RF). Often, there are local variations in the MRI fields which lead to image blurring, geometric distortions during image acquisition, and signal void artifacts. Low-field MRI systems cannot provide the resolution of high-field systems, but are of interest for applications requiring portable, less expensive devices. In this project, a tabletop spoke-and-hub permanent magnet array was constructed and assessed for homogeneity. Spatial encoding in x and y directions were achieved by tilting one hub relative to the other to generate a linearly increasing field in the tilt direction. An RF coil for emitting the stimulating signal was designed and optimized for low field strengths, where the signal-to-noise ratio is limited by the weak signal from the sample. The optimized coil was tuned and matched to the resonant frequency of the spoke-and-hub magnet. Finally, a transmit/receive switch was designed to allow stimulation of the sample and collection of the emitted signal with a single coil. This design demonstrates a potential low-cost method of generating MRI images without the siting and expense constraints of conventional high-field MRI.

Nasser MOHAMMED, Physics (Dr. Alex Hill), NSERC-USRA

Calibration of the CHIME (Canadian Hydrogen Intensity Mapping Experiment) Telescope's all-sky radioastronomical polarization angle.

Radio sources tend to be polarized. Strong magnetic fields in the galactic medium accelerate electrons, emitting cyclotron radiation in the radio frequency rage. The CHIME telescope is a stationary radio telescope in the Okanagan Valley, and much of the polarized radiation data it collects is from this type of emission. This project aimed to investigate the accuracy of the polarization angle of the data collected by CHIME, and to implement techniques to correct any angle offset. We implemented two methods to calibrate the raw data: utilizing trusted data in finding the correct gain terms, and approximating the gain differences between orthogonal channels to be very small. Having accurate polarization data of the galactic plane is key for continued research into mapping the galactic interstellar medium's magnetic field. The CHIME data shows promise to produce the first accurate all-sky polarization maps covering the 400-800 MHz frequency range.

Jordan SAWCHUK, Physics (Dr. John Hopkinson), URA

Cation ordering on a lattice of corner-sharing tetrahedra: Investigation of a simple Ising-like model

The onset of order in physical systems as temperature decreases is a nearly universal phenomenon. However, in frustrated magnetic materials, geometric constraints imposed by their crystal structure suppresses ordering. Unexpected properties in these materials - and the inability of established models to accurately describe them - has spurred a vigorous research effort. A vital task in this field is the discovery and synthesis of new frustrated materials. The spin-ice pyrochlore $Dy_2Ti_2O_7$ is a prototypical example of a frustrated magnet in which magnetic cations form a sublattice of corner-sharing tetrahedra (CST). A CST sublattice is also seen in

spinel $Na_4Ir_3O_8$, in which a 3:1 ratio of magnetic-to-non-magnetic cations share the CST sites and order to form a magnetic sublattice called a hyperkagome lattice. Theoretical studies indicate that the emergence of a hyperkagome lattice in a ¼-doped pyrochlore would create a new form of spin ice. Though pyrochlores are amenable to chemical substitution, no experimental evidence of hyperkagome cation order has been seen in doped pyrochlores. In this study, we investigate cation ordering of a 1/4-doped CST lattice under a simple model of cation repulsion. We analytically derive a ground state phase diagram for the smallest systems, finding three ground states: the hyperkagome, kagome and clustered states. Using geometric arguments about the ground state energy landscape supported by classical Monte Carlo simulation results, we propose that the general features of this phase diagram are invariant of system size. We also elucidate interesting open questions for future work.

Megan SCHROEDER, Chemistry (Dr. Kirsten Wolthers), URA

Functional Characterization of Heme binding Flavodoxins in *F. nucleatum*

Fusobacterium nucleatum is an opportunistic pathogen associated with periodontal disease and colorectal cancer. Flavodoxins are electron-transfer proteins that are essential for bacteria but are absent in animal cells, making them excellent antibiotic targets for treating such diseases. *F. nucleatum* utilizes seven flavodoxins, about which there is limited knowledge. This study provides new insights into the functional roles of two unique flavodoxins in *F. nucleatum* that have both flavin mononucleotide (FMN) and heme cofactors. The first goal of the study was to investigate the redox activity of the flavodoxins' heme and FMN cofactors. The second goal was to determine whether the heme cofactor can bind to diatomic gases like O_2, carbon monoxide (CO) and nitric oxide (NO). Through purifying and manipulating these flavodoxins, it was discovered that the heme cofactors in both flavodoxins are redox active, that they bind to CO and NO, but are readily oxidized upon exposure to O_2. These findings are important because a similar gas-binding heme protein called soluble guanylyl cyclase (sGC) has been used in biotechnology as an optical oxygen sensor and an MRI contrast agent; these applications may also apply to *F. nucleatum*'s heme-bound flavodoxins. Furthermore, advancing knowledge about each flavodoxin's role in *F. nucleatum* contributes to the future development of antibiotics that combat colorectal cancer.

Kristyn TAMELIN, Chemistry (Dr. Wesley Zandberg), NSERC-USRA

The Preparation and Purification of Mucins For Bacterial Utilization

Glycans are sugar molecules that compose the protective mucus lining of the gut. The relationship between glycans and colon bacteria is vital to colonizing the microbiome in babies and maintaining a healthy gut in adults. Glycan research is vital to understanding bacterial entry to immune tissues and developing preventative measures against infection. The protective glycan layer prevents bacteria from accessing underlying immune tissue. Microbes can degrade glycans at specific sites, which causes the glycans to be cut from the protein of the glycoprotein

and puts immune tissues at risk of infection. In this study, we developed and compared several methods for splitting glycans from a mucus glycoprotein. We fed the prepared glycans to bacteria and measured bacterial growth based on optical density: the sample's ability to absorb light when it passes through the sample. While it is common to use the compound ammonia to split glycans, this procedure is not feasible on a large scale because it is explosive. Therefore, we investigated a novel treatment containing bleach as a possible alternative for releasing glycans. We demonstrated that these preparation methods are unique and do not produce identical results. We found that colon bacteria do not grow when fed ammonia or bleach prepared glycans and therefore do not utilize or break down these types of prepared glycans. This knowledge will contribute towards establishing rational modifications to glycan purification procedures and growth assays. Future studies will investigate the composition of the glycan in detail and determine how its structure influences bacteria growth.

Greta TODTMANN, Chemistry (Dr. Gino DiLabio), IURA

Computational Study of the PINOylation Mechanism at Benzylic C-H Bonds

Benzylic alcohols and aldehydes, whose wide range of applications include pharmaceuticals, are easily synthesized in nature. A new method of this synthesis, which is difficult in vitro, has been developed by the Stahl group. It involves using the PINO radical to abstract a hydrogen radical from the benzylic position and then to trap the resulting benzylic radical in a process called PINOylation. This reaction yields a C-O bonded product, and unexpectedly, a ring expanded C-N bonded product. Varied reaction conditions have no influence over the ratio of these products; however, the product ratio varies significantly between different substrates. This project aimed to identify the reaction mechanism which yields the C-N bonded product and to understand the influence of different substrates on it. Reaction mechanism structures and energetics were calculated using quantum chemical modelling methods. Thorough examination of the energetics of different possible reaction mechanisms leading to the C-N-bonded product confirmed that the most plausible mechanism is the attack of the benzylic radical at the N-position of PINO, and subsequent ring expansion. Preliminary calculations with the different substrates studied by the Stahl group seem to support this mechanism as well. The substrates which show little to no C-N bonded product influence the reaction mechanism to have a free energy reaction barrier above zero, making the process of C-N bonded product formation energetically unfavourable. These results support the initial reaction mechanism proposed by the Stahl group, deepening the understanding of the PINOylation reaction, and allowing for greater control of future synthetic applications.

Carolina TOMIYAMA, Chemistry (Dr. Wesley Zandberg), IURA

Characterization of Novel Milk Oligosaccharides: A Qualitative Analysis

Oligosaccharides are short sugars naturally present in fruits, vegetables, and milk. Few are digested in the small intestine; therefore, they primarily act as prebiotics. It is well known that

their intake greatly impacts immunity and development. This study focuses on milk oligosaccharides and aims to better understand their specific biological activity. Furthermore, it investigates oligosaccharides with glucuronic acid (GlcA) that were recently reported. To do so, we analyzed milk samples from eight species: human, cow, camel, goat, sheep, elephant, pig, and horse. Human milk samples were segregated according to genetics (secretors and non-secretors). All the data was acquired through high-resolution mass spectrometry using LC-MS (Liquid Chromatography-Mass Spectrometry). We were able to characterize 25 glucuronic-acid-containing oligosaccharides based on their presence in different species' milk and their abundance in humans with different genetic conditions. Although the results were satisfactory, the novel milk oligosaccharide's exact structures remain a mystery that should be further investigated.

Lilian TON NU, Chemistry (Dr. Isaac Li), URA

Using a DNA-based probe to quantify cell adhesion at the molecular level

Cell Adhesion is one of the diverse behaviors mediated by mechanical signaling where piconewton levels of force are exchanged between the extracellular matrix (ECM) and adjacent cells. Despite its importance, the mechanism behind mechanical signaling is not well understood. To quantify and characterize these mechanical cellular events, our study uses an irreversible DNA molecular force probe called the Tension Gauge Tether (TGT) that fluoresces upon application of force. This fluorescence pattern allows us to visualize and map the force history of the cell during the adhesion process. As mechanical cues orchestrate multiple cell behaviors, a better understanding of the mechanical signaling process can be extended to stem cell or cancer cell research.

Spencer URSEL, Biology (Dr. Kirk Bergstrom), URA

Combating intestinal diseases with mucus producing organoids

The health of the gut microbiota has large effects on the health of the gut. Intestinal diseases such as inflammatory bowel disease (IBD) and colon cancer are connected to the health of the microbiota. The gut microbiota can be modulated to help combat chronic intestinal diseases such IBD and colon cancer. One method of promoting gut health is supplementing the gut microbiota with complex carbohydrates known as O-glycans. O-glycans are found in the protective mucus barrier present in the colon and have been found to promote gut microbiota to release anti-inflammatory metabolites. Colon epithelial cells can be cultured to create colon organoids capable of mucus secretion. The purpose of this research is to create a colon organoid system able to release MUC2, the glycoprotein that makes up mucus present in the colon. This mucus could then be collected and analyzed for its ability to promote growth and anti-inflammatory functions of the gut microbes. To create a system capable of secreting MUC2 novel techniques were implemented to create an air-liquid interface, which allowed mucus

secretion and collection. The presence of mucus was confirmed via end-point histological processing.

Kalli VANSTONE, Computer Science (Dr. Ifeoma Adaji), NSERC-USRA (12 to 2 pm)

Sqilxwcawt calendar mobile application design

This research details the design methodology used to create a syilx wellness-based calendar application and immersive user experience. The design process, user research and user experience are considered from a syilx perspective of relationships to place and time, the self and the collective, and seeking wellness through balancing those relationships. This design brings together the enowkinwix nested inquiry model representing the self, family, community, land and all life, and the oppositional dynamics dialogic model, represented by the elder-youth and tradition-innovation relationship dynamics to craft a syilx understanding of wellness that is informed by the First Nations Health Authority's Determinants of Well-being, creating distinctly syilx space and set of tools is crafted to facilitate reflection, learning, data collection, decision making, and transforming plans into actions. These combined methodologies adapt to suit the needs of syilx people across different spectrums, which is missing within most design approaches which typically consider the individual and are reminiscent of police profiling of indigenous users based upon their where-abouts and associations. This research involves defining the methods, showing how they are applied in the design, and illustrating the efficacy of these methods through the process of designing components intended to support menstruators. The proposed application will be shaped by the user's real-world relationships and experiences, and the user's relationships and experiences will be shaped by the application, a form of the Task-Artifact Cycle. These real-world impacts resulting from this cycle weave syilx knowledge into reality and the digital world, offering a means of expressing spatio-temporal soverignty.

Sofia VENTOSA, Chemistry (Dr. Thuy Dang), IURA

Characterization of Alcohol Dehydrogenases from *Rauwolfia serpentina*

Rauwolfia serpentina (Indian snakeroot) is a plant traditionally used in Southeast Asia and China for its medicinal properties, which are linked to its diverse metabolite classes, such as alkaloids. *R. serpentina* is especially known for its production of yohimbines and heteroyohimbine (HY) alkaloids. The low production of these compounds in slow-growing and endangered species, however, make its natural extraction unsustainable. Simultaneously, these metabolites' complex structure makes chemical synthesis impossible or expensive. Using a heterologous expression system in a microbial host would be an innovative biotechnology solution for society's needs for these bioproducts. The HY and yohimbines biosynthesis pathways, however, have not been elucidated in *R. serpentina*. In this project, we aimed to discover new alcohol dehydrogenases (ADHs) involved in the production of HY and yohimbines in this plant. Previously cloned ADH candidate genes were expressed in *Escherichia coli*, then purified using

his-tagged cobalt beads. Successfully purified ADH candidates were assayed with their substrate strictosidine aglycone and product formation was monitored by liquid-chromatography mass-spectrometry (LC-MS). Strictosidine aglycone was reduced to the HYs ajmalicine or tetrahydroalstonine; which were identified based on comparison to HY standards. These initial results indicate that one of the ADH candidates works in similar ways to the tetrahydroalstonine synthase enzyme from *Catharanthus roseus*, another plant that produces HYs. Further purification of ADH candidates is necessary for obtaining enough product for NMR analysis, so these results can be confirmed. Future work should focus on purification and LC-MS methods optimization to ensure proper protein purification and product analysis.

Paula WONG-CHUNG, Computer Science (Dr. Patricia Lasserre), IURA

Deep Learning-Based Digitization of Plains Cree Documents

The digitization of historical documents plays a significant role in the preservation and revitalization of indigenous languages in Canada. This study focuses on the digitization of a set of Plains Cree biblical newsletters, the *Kicitwaw Miteh* (Sacred Heart) corpus, loaned to us by the University nuhelot'ĵne thaiyots'ĵ nistameyimâkanak Blue Quills for this purpose. Digitization is achieved through Optical Character Recognition (OCR). The OCR process generally consists of two steps: the detection of text in a document during the segmentation stage and the transcription of the located text during the recognition stage. While there exists previous work on Plains Cree Standard Roman Orthography (SRO), to the best of our knowledge, no recognition work has yet to be performed on Plains Cree (nêhiyawêwin) syllabics. Furthermore, attempts are met with several challenges such as the lack of benchmark datasets, insufficient data, and language complexity. We trained a recognition model based on our manually created dataset using Convolutional Neural Networks (CNNs), a deep-learning method appropriate for low resource datasets, and a benchmark Character Error Rate (CER) of 0.27% was achieved. Such a high rate of success will not only eliminate the need to transcribe the documents by hand and significantly reduce the manual effort needed to correct the machine-produced results, but also pave the way for future OCR work on Plains Cree syllabics.