CONFERENCE ABSTRACT BOOK OPEN ACCESS

Richard E. Peter 15th Annual Biology Conference (2024)

Lisa R. MacLeod, MSc Student [1]*, Carina L. Lopez, MSc Student [2],
Aduratomi Etuk, MSc Student [1], Prabashi Wickramasinghe, MSc Student [1]

[1] Department of Biological Sciences, University of Alberta, Edmonton, Alberta, Canada
[2] Department of Agricultural, Food & Nutritional Science, Faculty of Agriculture, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

*Corresponding Author: lmacleod@ualberta.ca

Note: Correction added after original version published on March 13, 2024. We regret any inconvenience caused.

Abstract:
The Richard E. Peter 15th Annual Biology Conference is organized by the Biology Graduate Student's Association at the University of Alberta. This event is a multi-day, interdisciplinary, student run conference that showcases research conducted by graduate and senior undergraduate students in the biological sciences. In alignment with the conference theme, "Sustainability," presentations explore innovative research that addresses environmental challenges and conservation, as well as fosters sustainable practices. This conference serves as a platform for students to present their findings in various fields, including Molecular Biology and Genetics, Paleontology, Microbiology, Plant Biology, Physiology and Development, Marine Biology, Immunology and Infection, Entomology, and Health Sciences. For more information, visit the conference website https://repeter2024.weebly.com/.

Keywords: biology; ecology and evolution; microbiology cells and systems; sustainability; genetics; health sciences; immunology and infection; plant biology; entomology; paleontology

Table of Contents

Conference Abstracts

Note: These abstracts have been reproduced directly from the material supplied by the authors, without editorial alteration by the staff of the URNCST Journal. Insufficiencies of preparation, grammar, spelling, style, syntax, and usage are the authors.

Oral Presentations in Ecology and Evolution

Apis mellifera Maternal Vaccination Against Israeli Acute Paralysis Virus (IAPV)

Chenoa Kaufman, Undergraduate Student [1], Prabashi Wickramasinghe, Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Pollinator health, especially of the honey bee (Apis mellifera) is a critical area of study that is directly implicated in the global food supply chain and economy. Israeli Acute Paralysis Virus (IAPV) is responsible for high honey bee mortality and the corresponding reduction in pollination and economic productivity suggests that prophylactic strategies to improve honey bee health are needed urgently. We currently lack any practical treatments for IAPV or any other honey bee virus, but transgenerational immune priming (TGIP), the process by which offspring are prepared to combat disease-causing agents the parent(s) experience in their immediate environment, is an interesting mechanism that can potentially be exploited to promote resistance to pathogens. The present study relied on honey bee queen vaccination with inactivated IAPV and testing offspring survival against active IAPV with the wider goal of evaluating the presence and efficacy of TGIP to defend against IAPV and the potential use of TGIP to combat other honey bee viruses. We hypothesized that honey bees can prime their offspring against IAPV and thus predicted that the offspring of queens exposed to thermally inactivated IAPV would exhibit higher survival of an acute IAPV infection than the offspring of unmanipulated control queens. Heat-inactivated IAPV was applied to the shaved thoraces of queens, and offspring were assigned into random experiment cages, exposed to an active IAPV inoculum, and subjected to a 12-day mortality assay. Statistical analysis was performed and revealed that exposure to IAPV results in a decrease in survival compared to control groups, but offspring from immunized queens did not exhibit significantly improved survival against acute IAPV infection. This result suggests a lack of TGIP against IAPV in honeybees and overall underscores the intricate nature of honeybee transgenerational immune response and the need for further exploration in developing effective strategies against viral threats.

Individual Variation in Diurnal Body Temperature and Foraging Activity in Over-Wintering Black-Capped Chickadees (Poecile atricapillus)

Deborah M. Hawkshaw, PhD Student [1], Jan J. Wijmenga, Kimberley J. Mathot
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Small birds in the winter can mitigate energetic shortfalls via increases in foraging rates and/or via reductions in metabolic rate and body temperature (torpor). The ability to both increase foraging rates and use torpor during the day could have profound implications for an individual's daily energy budget and, ultimately, their over-winter survival. Trade-offs between foraging efficiency and daytime torpor use may exist but have not been explicitly investigated. Here, we investigated the presence of within and among-individual correlations between daytime body temperature (Tb) and foraging rate in over-wintering black-capped chickadees (Poecile atricapillus). Using temperature-sensing passive integrated transponder (PIT) tags, we measured daytime body temperature and foraging rates in 20 free-living black-capped chickadees over 49 days in a single winter (January – February). Body temperature during visits to the feeder was, on average, 41.67 ℃ but ranged between 25.0 and 44.9 ℃. In general, chickadees exhibited colder daytime Tbs, shorter intervals between successive feeder visits (inter-visit interval, or IVI) and increased feeder visits as the ambient temperature decreased. However, within-individuals, there was only evidence of a weak positive correlation between visit Tb and IVI and no correlation between total daily feeder visits and daily mean Tb. We found that visit Tb, daily mean Tb and daily visits to the feeder were repeatable traits, while IVI was not. Sex did not explain a significant amount of variation in daily foraging rates or daytime Tb, nor was there evidence of among-individual correlations between daily mean Tb and daily feeder visits. Our results suggest that chickadees may independently regulate foraging rates and daytime Tb; however, more studies investigating the expression of these traits are needed. Overall, our study provides insights into how small birds in the winter use multiple traits to overcome energetic challenges.

Plant Social Networks: A Tool for Understanding Community Assembly and Coexistence

Emily M. Holden, PhD Student [1], James F. Cahill Jr.
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Diverse plant species can live together, though classical ecological thinking suggests competition should prevent such coexistence. Theory suggests a combination of abiotic resources and plant social interactions determine plant performance and abundance so that the resulting community is a complex product of the filters' interaction. Social context is expected to influence cooccurrence patterns as species are differentially excluded from or admitted into local communities as a result of their associations with other species. Ecology is moving in a new direction where efforts are made to quantify and harness the complexity of these interactions through network analyses, which map species and their associations, thus integrating species composition with interspecific interactions. Network descriptors have been linked to community resistance and stability, but their ability to explain community dynamics has not been explicitly tested. Here, we ask if basic co-occurrence metrics can be used to understand changes in biodiversity. We created cooccurrence networks for plants in a local grassland, then calculated the average weighted positive and negative cooccurrences of species interactions in communities. We then asked if these weighted co-occurrence values protect changes in biodiversity in communities using linear mixed models. Our results show that co-occurrence data predicts species gains and richness, suggesting biotic processes govern community invasibility. This work shows the importance of plants' social environment in assembly processes and highlights the role of biotic interactions in maintaining biodiversity.

Assessing the Intraspecific Osteological Variation of Notropis hudsonius (Cypriniformes: Leuciscidae)

Erika Jessen, MSc Student [1], Alison Murray, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The spot tail shiner (Notropis hudsonius) is a widely distributed Leuciscid species found across the northeastern USA and Canada. Traditionally considered a basal, plesiomorphic species, most research on the osteology of N.hudsonius has been in the context of wider phylogenetic studies or studies focusing on a specific osteological characteristic amongst other North American cyprinoids, like the caudal skeleton of pharyngeal teeth. However, no previous studies have specifically focused on the osteology of N.hudsonius, nor the intraspecific osteological variation of a single Notropis species. A part of my thesis project focused on describing the osteology of N.hudsonius and documenting all osteological variants observed. Analyzing cleared and stained specimens representing 15 populations from Alberta, the Northwest Territories, western Ontario and Manitoba, I noted several differences in the osteological characteristics of N.hudsonius. Variants were most prominent in the suspensorium, caudal skeleton, weberian apparatus, pectoral girdle, oral jaws, branchial apparatus and hyoid bar. These findings reveal what osteological traits are subject to variability in N.hudsonius and what osteological differences are specific to a certain region, population or individuals and provide important insight into the intraspecific variation and evolution of N.hudsonius and North American Leuciscids as a whole.

Analyzing Collective Thermoregulation in Fish Using a Novel Enclosure and Machine Learning-Assisted Multitracking

Ethan Proctor, MSc Student [1], Daniel Barreda
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Environmental stressors caused by anthropogenic climate change have significantly altered the biodiversity, physiology, and behaviour of fish species. Warming of aquatic habitats may disrupt collective behaviours in gregarious fish that are critical for foraging and predator avoidance. Recent studies analyzing environmental stress on collective behaviour are often limited by small group sizes tested in discreet conditions. Here, we present a freely-selective thermal enclosure that accommodates larger groups and facilitates social behaviour. Paired with machine-learning assisted multitracking, this methodological system allows automated analysis of collective behaviour under a continuous thermal gradient. As a proof of principle we observed goldfish populations of different sizes with our system and found that group cohesion increases with group size but not with temperature.

Effect of Rhizobium Strains on Pea Leaf Weevil (Coleoptera: Sitona lineatus) Herbivory and Development

Olajide I. Fatukasi, PhD Student [1], A Wijerathna, Malinda Thilakarathna, Maya Evenden
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Pea leaf weevil (Sitona lineatus L.) is a major pest of field peas and faba beans (Fabaceae). Economic damage to these legumes is caused by S. lineatus adults and larvae that feed on foliage and the rhizobia-containing root nodules, respectively. The host-specific rhizobia fix atmospheric nitrogen and receive carbon nutrients in exchange. The rhizobia-plant interaction may affect pea leaf weevil herbivory and development by influencing plant food quality and chemical defense. We tested the hypothesis that Rhizobium-field pea interactions influence S. lineatus foliar feeding and development. Field pea plants received one of four treatments in each of three experiments assessing foliar feeding and development: 1) inoculated with the wild-type Rhizobium leguminosarum strain, WT3841; 2) inoculated with a mutant Rhizobium leguminosarum strain, MT3940 that does not fix nitrogen; 3) treated with nitrogen; and 4) control plants that received only water. After 3 and 5 weeks, male (4) and female (4) S. lineatus were introduced into the cages containing the variously treated plants and were allowed to feed for 8 days. There was no treatment effect on reproductively active overwintered and spring adult S. lineatus herbivory, which could be due to a trade-off between nutritional food quality and chemical defense. Wild-type Rhizobium strain, however, significantly supported S. lineatus development from egg to adult stage compared to the mutant-type, and other treated plants in the development experiment. Lower development of S. lineatus on plants inoculated with the mutant-type, MT3940 could be due to smaller nodule size and a lack of fixed nitrogen compared to wild-type strain, WT3841 treatment.

Effects of Soil Texture and Light Availability on Caragana Arborescens Performance

Haley Lacza, Undergraduate Student [1], Martin Hinojosa, Viktoria Wagner
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Caragana is a non-native shrub introduced to the Canadian prairies in 1880 from Eurasia and has been widely planted to prevent soil erosion and act as a windbreak along crop fields. Since being introduced, it has escaped from cultivation and is now invading native ecosystems and national parks across Alberta, Saskatchewan, and Manitoba. However, not much is known about the environmental settings that allow Caragana to invade. Previous references suggest that this shrub prefers to grow in bright locations with sandy soil, yet this hypothesis has never been tested. To explicitly test the growth of Caragana under different environmental conditions, we chose to grow this shrub in a variety of soil types and light availabilities. We hypothesized that Caragana growth would be highest in a sandy soil treatment with no shade, and lowest in the deep shade and loamy soil treatment. Our initial results show that neither soil type nor the amount of light influence Caragana germination, however these variables interact to influence seedling survivability. Both the partial and deep shade loam treatments had the highest mortality amongst treatments, followed by the partial and deep shade sand-loam mix treatments. The bright treatments had very little mortality regardless of soil type. These results suggest that locations with little canopy cover, such as forest clearings or edges, may be the most at risk of Caragana invasion.

Seasonal and Social Influences on Caching Behaviour in Black-Capped Chickadees (Poecile atricapillus)

Ipshita Gayen, PhD Student [1], Deborah Hawkshaw, Megan LaRocque, Jan Wijmenga, Kimberley Mathot
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Many resident wintering parids in temperate regions engage in high-intensity caching in fall. Caching acts as an external energetic reserve and it is predicted that situations that reduce foraging success (predictability) or increase energetic needs will encourage individuals to hoard more food to increase the probability of overwinter survival. Multiple models have attempted to address how the optimal caching strategy might vary within a population as members differ in their energetic needs and access to resources. However, until the recent advancement in automated tracking technology (e.g., RFID) collecting enough high-resolution individual foraging data to test theoretical predictions was logistically challenging. This study investigates individual differences in the seasonal caching patterns from mid to late fall. We quantified how priority access to food resources within a flock influences an individual's choices for relative use of cacheable and non-cacheable food. The experiment was conducted in a marked wild population of black capped chickadees (Poecile atricapillus) in its natural habitat with supplemented food. The data has been collected but the analysis is still underway. We will evaluate support for a decrease in caching rate and increase in feeding rate as winter approaches, which is predicted from theory because of build up of caches and higher energetic needs at lower ambient temperatures, respectively. Additionally, we

predict that as black capped chickadees maintain social hierarchy in resource acquisition, dominant flock members will have lower caching tendency as compared to subordinate members.

Ecology of Fear: Ontogeny-Mediated Non-consumptive Effects

Lisa R. MacLeod, MSc Student [1], Lien T. Luong
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Non-consumptive effects (NCEs) arise in the presence of predators or parasites even when death or infection does not occur and can include changes to host behaviour, physiology, or morphology. A growing body of literature offers evidence of various NCEs in parasite-host interactions, but the focus has been primarily on a single developmental stage of the host. Using the Drosophila nigrospiracula – Macrocheles subbadius fly-mite system, we investigate the impact of parasite exposure (sans infection) during the pupal and adult pre-reproductive stage. First, we exposed fly pupae to ectoparasitic mites—either indirectly (caged mites) or directly (free roaming mites). We found that direct exposure significantly decreased the rate of successful eclosion by 20% (development from pupa to adult) compared to unexposed pupae; however, the duration of pupation was not significantly affected. The indirect exposure did not have a significant effect on either successful eclosion or duration of pupation. Second, we tested how exposing adult female flies to ectoparasitic mites prior to reproduction affected fecundity during the post-exposure, reproductive period. Females indirectly exposed to caged mites had a 35% reduction in the number of offspring produced compared to control flies, but only for the first few days post-exposure; i.e., the effect was reversible after mites were removed. Offspring production returned to levels comparable to the control flies, 4 – 5 days after the removal of mites. The exposure to mites had no significant effect on offspring weight compared to unexposed females. Investigating the diverse NCEs associated with parasite exposure at various life stages of the host is important in understanding the ecology of fear and its total impact on hosts throughout their entire lifespan, with consequences for host ontogeny and population growth.

Salmonella's Phopq Reveals Fundamental Insights into the Evolution of Two-Component Systems in Specialized Niches

Luke Barretto, PhD Student [1], Casey Fowler, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The acquisition of novel genes is the primary driver of bacterial evolution. However, acquiring ecological innovations from new genes must be tempered by a robust system for integrating these genes into existing regulatory networks. One of the most well studied such systems are two-component systems (TCSs), a versatile combination of two proteins—a sensor kinase and its response regulator—that govern the expression of a bacterial cell's genetic repertoire in response to diverse changes in its environment. This research seeks to advance our understanding of bacterial evolution through the lens of TCSs and their central role in accommodating the acquisition of evolutionary novelties. Through this analysis of TCSs, we hope to uncover the mechanisms which cause their divergence among bacteria, and how their differences have fueled bacterial evolution. We hypothesize that TCSs fine-tune their sensory capabilities and their regulation of newly acquired genes to fit the demands of diverse bacterial niches. To characterize TCS evolution, our research examined the mutational consequences of placing Escherichia coli's PhoPQ system (i.e. 'ecoli-cizing') in a genetically related bacterium, Salmonella enterica serovar Typhi. Using molecular cloning and allelic exchange, we constructed a library of PhoPQ mutants in reporter strains of S. Typhi and assessed their ability to regulate S. Typhi's genes through β-galactosidase assays. Our results demonstrate that the fully 'ecoli-cizing' S. Typhi severely diminishes its ability to regulate its own genes. Intriguingly, restoring S. Typhi's PhoP and PhoQ domains in the E. coli PhoPQ construct leads to a varied gradient of complementation, with certain domains restoring more gene expression than others. Ultimately, we hope to refine our research into the evolution of TCSs by using cell culture models to investigate the phenotypes of our constructs in infection settings as well as establishing the mechanistic contributions of the domains of the PhoPQ TCS.

Chickadees on the Move: Age, Sex, and Energetics Influence Space Use in Black-Capped Chickadees

Megan LaRocque, MSc Student [1], Jan J. Wijmenga, Kimberley J. Mathot
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Animal movement decisions are shaped by both extrinsic and intrinsic factors. Historically, spatial ecology studies have focused on average movement patterns within animal groups; however, recent studies highlight the value of considering movement decisions both within- and among-individuals. Using a marked population of black-capped chickadees (Poecile atricapillus), we used the number of unique feeders an individual visits within our study area as a proxy for space use to explore how: i) within-individual space use and foraging rates are affected by ambient temperature; ii) among-individual differences in space use and foraging rates are affected by sex and age; and iii) space use influences survival. We found that as temperature decreased, feeding rate increased; however, females increased their space use, while males did not. This may be due to sex-related differences in dominance, where males (i.e., dominants) have priority access to feeders, while females expand their foraging areas to meet higher energetic demand. We found that independent of temperature, juvenile males used more unique feeders than adult males. We suggest this may be due to age-specific benefits of male space use, where un-paired juvenile males may increase feeder exploration to gain information about potential mates. Finally, we found no effect of space use on annual survival. Overall, our results suggest that dominance hierarchies and individual energetics impact within- and among-individual variation in space use. We provide suggestions for future studies to enhance understanding of fitness-related consequences of within- and among-individual variation in space use.

Exploring Sources of (Co-)Variation in the Timing and Intensity of Foraging in a Wild Population of Black-Capped Chickadees (Poecile atricapillus)

Nathan Hobbs, MSc Student [1], Deborah Hawkshaw, Jan J. Wijmenga, Kimberley J. Mathot
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The timing and intensity of foraging behaviour in birds are shaped by both extrinsic (e.g., temperature, daylength) and intrinsic (e.g., sex, age) factors. Although many of the same factors have been shown to affect both traits, the covariation between these traits has not been explored. We observed a population of 143 individually marked black-capped chickadees (Poecile atricapillus) over a period of 90 days during the non-breeding season and recorded the time when each individual began and ended foraging, and their total number of feeding events in a given day. Within-individuals, later initiation of first feeds relative to sunrise and earlier termination of feeding relative to sunset, were both associated with lower daily intake rates. This demonstrates that plasticity in timing of activity can have important impacts on total daily food intake. We also found evidence for different chronotypes of birds. Individuals that consistently started feeding earlier relative to sunrise also ended feeding later relative to sunset and had higher total food intake. This suggests that extending the foraging period can enable birds to meet higher energetic needs. Taken together, our results show that timing and intensity of foraging are integrated traits that do not vary independently from one another.

Artificial Selection for High and Low Virulence of an RNA Virus in Apis Mellifera Workers

Prabashi Wickramasinghe, MSc Student [1], Chenoa Kaufman, Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The molecular mechanisms driving Israeli acute paralysis virus (IAPV) pathogenesis, especially in the context of global honey bee colony decline, remain poorly understood. Unraveling IAPV pathogenesis involves identifying key determinants of IAPV virulence. In our experimental evolution study, we hypothesized that IAPV virulence rapidly evolves in response to artificial selection. We predicted that subjecting IAPV to serial passages through honey bee workers, coupled with bi-directional artificial selection for opposing virulence, would yield two divergent IAPV strains - one highly virulent and the other benign - with noticeable genetic and phenotypic deviations from the original strain. A selection experiment was set up with 11 parallel lines for each direction. Mortality assays were performed for each of the lines and ten generations of the experiment, involving the sampling of deceased bees every 6 hours over a span of 5-7 days. IAPV titers were quantified from the isolated virus of the earliest and latest dying bees in each generation, and inoculum prepared to infect the subsequent generation. Kaplan-Meier survival analysis of the phenotypic data indicated substantial heterogeneity among lines with a significantly higher survival rate of bees subjected to “benign” lines relative to the “virulent” lines or the original IAPV strain. This promising result will be combined with high-throughput genome sequencing to identify the molecular changes underlying the phenotypic IAPV differentiation and further our understanding of virus pathogenesis.

The Effects of the Soil Microbiome Under Prior Resource Conditions on the Plant Community

Rebecca Innes, MSc Student [1], Emily Holden, James Cahill
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The availability of light and nutrients has a direct effect on the composition of plant communities and the soil microbiome. However, the impacts this availability has on the soil microbiome and how those changes affect the plant community are not well understood. Here, we aim to explore if prior resource availability changes the effect of the soil microbiome on the future plant community, and if these effects are contingent on the presence of current nutrients. To test this, we set up a mesocosm experiment using soil inocula samples taken from an ongoing field experiment that manipulated shade and nutrients in four distinct regimes. We grew six common grassland plant species in these different soil inocula, and after 5 months we collected above ground biomass to determine if there is a difference in biomass between plant communities grown in each soil treatment. Preliminary results suggest lower germination rates in sterile pots with no added field soil compared to those with a present microbiome. These results potentially suggest that the soil microbiome could be negatively affecting these plant communities by reducing germination. By understanding if resource availability is indirectly affecting the plant community through changes to the soil microbiome, we can begin to predict what changes will occur in plant dominated ecosystems, such as grasslands, under different environmental shifts.

Food Supplementation of Peregrine Falcons Improves Offspring Survival and Condition Without Changes in Parental Provisioning Behaviour

Rebekah Alice McKinnon, PhD Student [1], Erik Hedlin, Kevin Hawkshaw, Kimberley J. Mathot
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Parents are expected to exhibit intermediate levels of investment in parental care that reflect the trade-off between current versus anticipated future reproduction. Providing parents with supplemental food with which to provision young may allow for increased care to current brood (additive model), re-allocation of parental effort to other behaviours such as self-care (substitution model), or may provide parents with a buffer against provisioning shortfalls (insurance model). Here, we investigated the impact of parental food supplementation on provisioning behaviour and breeding success in Arctic-breeding Peregrine falcons (Falco peregrinus tundrius) over five successive breeding seasons (2013-2017). The results reveal that supplemental feeding significantly increased offspring survival probability, supporting both additive and insurance models. However, there was no evidence of changes in provisioning effort or fledgling body condition, challenging the predictions of these models. The study is consistent with parents adopting a hybrid of the additive and substitution models, where food supplementation enables increased investment in other forms of parental care, such as nest defence, without altering provisioning rates. The lack of observed effects on provisioning rates, coupled with increased survival, suggests a potential reallocation of parental effort. The findings contribute to understanding the nuanced responses of Peregrine falcons to food supplementation, highlighting the need for future studies to explore broader environmental contexts and potential long-term effects on parental survival and future reproduction.

The High Spatiotemporal Variability of Phytoplankton Communities: Implications for Managing Harmful Algal Blooms in Central Alberta Lakes

Renz Layugan, MSc Student [1], Rolf Vinebrooke
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Many stakeholders of recreational lakes in Alberta have a growing concern for cyanobacterial harmful algal blooms (cHABs). Thus, a comprehensive monitoring program is key in mitigating its harmful effects. This can be difficult as cHAB dynamics show high spatiotemporal variability. Traditional monitoring programs often summarize cHAB status by generating a proxy value to represent the entirety of the study lake at a certain sampling period, which do not fully capture the spatial variability of cHABs. As a part of a larger project in tracking spatiotemporal dynamics of cHABs, we generated data for taxonomically diagnostic phytoplankton pigments from multiple sampling points (20-30 sites) at multiple sampling periods (2-3 times) in six recreational lakes in central Alberta. Here, we applied multivariate analyses to (a) describe the spatiotemporal patterns of phytoplankton abundance and community both within and between the six recreational lakes, and (b) determine which environmental parameters dictate this spatiotemporal variability. As expected, our preliminary analyses show that phytoplankton abundance and communities highly vary throughout space and time, likely driven by local factors since spatiotemporal patterns are lake-specific. Our findings can provide evidence to pinpoint the origin of cHABs to improve management applications.

Evidence of Mutually Detrimental Interactions Between Urban Coyotes and People Experiencing Homelessness

Sage Raymond, PhD Student [1], Colleen Cassady St. Clair
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Cities across North America exhibit increasing rates of both human-coyote conflict and people experiencing homelessness, but potential relationships between these phenomena have received no peer-reviewed study to date. We synthesize personal observations from a long-term study of urban coyotes in Edmonton, Canada, with evidence from the literature to describe, and then make recommendations for reducing, three types of human-coyote conflict in the context of homelessness. First, people and coyotes seek similar habitat, often in dense vegetation, for encampments and denning, respectively. These sites minimize interactions with recreating people and their dogs but increase spatial overlap between coyotes and the encampments that house people. A second type of conflict, food conditioning, results from shared space and the opportunities it necessarily creates for habituation. Habituated coyotes are more likely to access human sources of food near encampments, which reduces wariness towards people in other contexts. Third, attraction to people caused by both spatial overlap and food conditioning may increase human exposure to zoonotic diseases carried by coyotes, especially a novel tapeworm to which people experiencing homelessness may be especially susceptible. These sources of conflict may be reduced with directed research about the three types of conflict we propose, direct support and targeted health care for people experiencing homelessness, and education for health care professionals and others who support people experiencing homelessness. Ideally, these actions would embrace a socio-ecological approach, recognize the social complexities and inequities that lead to homelessness, address other urban wildlife species, and encourage reconciliatory actions for marginalized groups.

Patterns and Temporal Dynamics of Canine Breakage and Intraspecific Injuries in Western Hudson Bay Polar Bears (Ursus maritimus)

Simonne S. Tremblay, Undergraduate Student [1], David McGeachy, Nicholas J. Lunn, Evan Richardson, Andrew E. Derocher
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Canine teeth are essential components of the anatomies and ecologies of carnivores as they are utilized in prey apprehension and behavioural displays; however, this tooth type has high incidences of injury which may reduce survival. Polar bears (Ursus maritimus) typically have polygynous mating systems where males compete for access to females. Injuries, especially to the canines, are frequent in these encounters and can be used as proxies for mating system dynamics. The western Hudson Bay (WHB) subpopulation has been experiencing rapid decline in recent decades which may impact traditional characteristics of their mating system. We examine the overall patterns of canine breakages and its temporal dynamics within the WHB using field data from 1981-2023 (n=3493) through non-parametric statistical analyzes and linear mixed effect models. We found differing rates of mean breakage and coinciding intraspecific injury between males and females; proportions of maximum breakage are similar between the sexes until approximately ten years of age, where males then begin accruing more serious damage to their canines. The interactive effect between the age of an individual and the year of observation was a significant predictor in mean breakage throughout the study period suggesting that breakage is beginning to increase over time, potentially in response to shifting population structures. The comparative results of canine breakage occurrences and related injuries between the sexes reveal WHB males to be under more intensive intraspecific pressures than females within the subpopulation, which aligns with previous research regarding canine breakage and its use in understanding mating systems of polar bears. The presence of a relationship between year and canine breakage in both sexes suggests that there may be increased intraspecific competition occurring within the subpopulation, however continued investigation into the temporal dynamics is needed as stronger causal relationships are likely to arise as decline continues in the subpopulation.

Effects of Artificial Light on Bat Activity Around Urban Ponds of Edmonton

Victoria Berg, Undergraduate Student [1], Camila Hurtado, Tyler Cobb, Carol Frost
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Water bodies in heavily urbanized areas are valued for their ecosystem services, including their ability to support biodiversity. However, lamp posts are often installed around urban ponds to increase accessibility and safety for people, but the influence of these artificial light sources on habitat use and behaviour of nocturnal species like bats is not well understood. Using bioacoustic recording equipment, we examined bat echolocation calls near 27 urban ponds of varying light conditions to determine the effect of artificial light from lamp posts on bat occurrence and behaviour in Edmonton.

Throughout the summer of 2023, bat echolocation calls were recorded using Wildlife Acoustics' Song Meter Mini Ultrasonic Recorders. At each sampling location, two acoustic recorders were installed, one on the water's edge and the other 50 m away from the water's edge, for a period of 30 minutes before sunset until 30 minutes after sunrise. Artificial light datasets were collected using aerial imagery to identify individual lamp posts within 50 m of each acoustic recorder, yielding a range of 0-7 lamp posts within a given buffer. Preliminary results suggest that low light conditions within 50 m from each recorder yield optimal artificial light conditions for bat activity. Results from this study can be used to determine the best solutions to decrease habitat disturbance resulting from artificial light and support biodiversity around Edmonton's urban ponds.

After the Gold Rush: The Impact of Gold Mining on Wolverines, Lynx, and Marten

Zachary Fogel, MSc Student [1], Karlie Knight, Piia Kukka, Paul Boyce, Alice McCulley, Thomas Jung, Fiona Schmiegelow
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Nätra/Wolverines, Ninju/Lynx, and Tsŭk/Marten are threatened by human disturbance across their range. In central Yukon, Tr'ondëk Hwëch'in First Nation's (THFN) stewardship of the land and its inhabitants has been interrupted by industrial mining. These furbearers are bioculturally significant for THFN, who have identified mining as a threat to furbearers and other wildlife in their Traditional Territory. Academic research indicates that furbearers are affected by industrial activity, but little research has focused on mining. In response to its high biodiversity and current changing landscape, Yukon South Beringia, which encompasses the THFN Traditional Territory, has been identified as a Priority Place. The Priority Place Initiative is a commitment to shift toward collaborative management that explicitly includes Indigenous Peoples and Traditional Knowledge. To meet this commitment, we are partnering to quantify the relationship between landscape condition and furbearer abundance and distribution. We have deployed trail cameras and autonomous sound recording units (ARUs) across a gradient of disturbances. The trail cameras will photograph animals that pass by the cameras and the ARUs will record ambient sound. We will estimate wolverine abundance using camera data, quantify industrial activity using ARU data and aerial imagery, and evaluate the relationship between wolverine abundance and industrial activity. We expect that abundance will be lower in areas of higher disturbance. Our results will inform support THFN's stewardship responsibilities, regional land use planning and environmental assessments, and inform wider conservation efforts for furbearers.

Oral Presentations in Marine Biology

Nutritional Values of Small Pelagic Species Vary More by Taxa and Size Than by Region

Alana M. Krug-MacLeod, MSc Student [1], Elan J. Portner, Miram R. Gleiber, C. Anela Choy, Elizabeth A. Daly, Natasha A. Hardy, Larry B. Crowder, Stephanie J. Green
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Nutritional content can vary significantly both within and among marine species, in conjunction with organisms' life history traits and environmental conditions. Monitoring the value and circulation of energy and nutrition can aid in explaining and predicting shifts in species distributions and abundance—supporting fisheries and conservation management as environmental conditions change. We used proximate composition and morphometrics to compare nutritional values and traits of five small pelagic species that are prey for albacore. These characteristics were also used to assess whether there were differences between northern and southern regions of the California Current. We used simple linear regressions and Bayesian analytical tools to assess the relationships among nutritional metrics, regions, and traits. The nutritional patterns in the selected SPS were conserved between the northern and southern regions of the California Current in this study period and the relative nutritional value of the species remained constant. Overall, we found that size (length and mass) was a greater predictor of nutritional value than region. Further, we found that size better predicted percent protein than percent lipid in squids and that protein contributed more to total energy in squids than did lipid. The reverse was true in fishes: size better predicted percent lipid than percent protein and lipid contributed the most to total energy. Practically, our findings suggest that size can be used to predict nutritional content of small pelagic species, which would simplify indirect calculation of nutritional value for researchers, saving time and resources. We found that the strength and direction of size-to-nutritional value relationships differed among species, but we also identified broad trends that may be specific to vertebrate and invertebrate species across a wider spectrum. If this proves true, it will help predict foraging behaviour in changing environments, facilitating fisheries management and conservation.

Oral Presentations in Plant Biology

Enhancing Resilience to Drought Stress: The Role of Legumes in Forage Mixtures

Chathuranga De Silva, PhD Student [1], Malinda Thilakarathna
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Drought stress significantly impacts forage production, underscoring the need to deepen our understanding of how forage plants respond to such conditions. The primary aim of this study was to assess the response of legume-grass mixed stands to drought stress in comparison with grass monoculture. A greenhouse pot experiment was conducted by subjecting red clover (Trifolium pratense L.)—timothy grass (Phleum pratense L.) mixed stand and a timothy monoculture stand to severe drought (20% field capacity—FC), moderate drought (40% FC), and well-watered (80% FC) conditions for four weeks, followed by a four-week recovery period during which moisture levels were restored to 80% FC. Moderate and severe drought significantly decreased the shoot biomass of the mixed stand, while no significant difference was observed in the timothy monoculture. However, plants subjected to moderate drought recovered shoot growth during the recovery phase. Comparatively, the total biomass of the legume-grass mix stand was notably higher than grass monoculture in well-watered control, moderate, and severe drought treatments during both drought and recovery phases. Drought stress reduced shoot biomass and nitrogen fixation capacity in red clover. Grass in the mixed stand exhibited a significantly lower C:N ratio and higher leaf chlorophyll content compared to monoculture. These findings highlight the inclusion of legumes in forage mixtures as an effective strategy to enhance resilience against moderate drought stress.

The Effects of Volatile Tree Emissions on Detection of Host Lodgepole Pine by Mountain Pine Beetle

Madelyn O'Hara, MSc Student [1], Antonia Musso, Colleen Fortier, Marion Mayerhofer, Maya Evenden, Janice Cooke
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Recent outbreaks of Dendroctonus ponderosae (mountain pine beetle, MPB) in the western Canadian provinces have wreaked havoc on forest dynamics, particularly in lodgepole pine trees, the historic host of MPB. However, even when faced with intense outbreaks, some trees survive on the landscape. Research from the Cooke lab has shown that a distinct genetic difference exists between the progeny of trees that were killed during a MPB attack and those that survived. We hypothesize that survivors exhibit traits that render them more resilient to MPB. I hypothesize that MPB can detect differences between progeny of MPB-killed lodgepole pine and progeny of survivors during host selection, and that progeny of survivors are less attractive to MPB. To understand how MPB interpret these trees, I have three main objectives: 1) determine if MPB can sense a difference in the volatile profiles emitted by the two progeny types through the use of olfactometer assays in which beetles are exposed to both classes of progeny, 2) quantify the volatile profiles of the two progeny classes, and 3) identify molecular and/or gene expression differences in the two progeny types that may influence whether MPB choose to attack a particular tree. If MPB find one class of progeny more suitable for attack and subsequent colonization, it is expected that the beetles will spend more time in its proximity, and this will be associated with distinct differences in the profiles of these trees. I have completed olfactometer assays, in which I collected video data to document adult beetle (n=78) choice between seedling progeny of MPB-killed and MPB survivor lodgepole pines. Analyses of these video data are in progress. The outcomes of this research will reveal whether lodgepole pine traits that influence MPB host selection contribute to genetic resilience of lodgepole pines against this devastating forest insect pest.

Low-Cost Forage Management Impacts on Soil Biological Health Indicators of Old Grassland: Legacy Effects

Megan MacElheren, MSc Student [1], Emmanuel Mapfumo, Vern S. Baron
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Increasing costs of fertilizer application and low legume content are contributing factors to the loss of grasslands in Canada. Sod-seeding legumes may improve grassland productivity and soil health. The current study was established in 2022 at Lacombe Research Centre and utilizes 8 pens previously managed under either annual cereals or perennial grasses since 1993. Biological soil health indicators were assessed to determine legacy effects of management practices preceding the application of new treatments (sod-seeded legumes). Indicators assessed were permanganate oxidizable carbon (POXC), autoclave extractable (ACE) soil protein, and bacteria composition and diversity. The analysis of variance (ANOVA) General Linear Model procedure in SPSS was used to test for differences in soil health indicators under annuals/perennials.

Regression analysis was conducted on POXC data's relationship to soil depth. Bioinformatic analyses at phylum level for 16S rRNA marker gene sequenced data using Microbiome Analyst included alpha diversity and interactive heat map with cluster analysis using the Ward algorithm. POXC and ACE soil protein were significantly higher under perennials than annuals. POXC decreased with soil depth (curvilinear)($R^2>0.85$). Bioinformatic analyses indicated higher microbial diversity (Shannon index) at OTU level under perennials compared to annuals. The actual abundance of bacteria phyla was 19% higher in perennials than annuals. Relative abundances of Acidobacteriota and Firmicutes were higher under annuals compared to perennials, and Verrucomicrobiota and Actinobacteriota were lower under annuals compared to perennials. Sampling and assessment of biological health indicators will continue until 2026 to assess effects of sod-seeding of legumes on productivity and soil health.

Effects of Introduced Cicer Milkvetch on Plant and Soil Ecology in Dry Mixedgrass Prairie

Michele Tran, MSc Student [1], Rhea Amor Lumactud, Batbaatar Amgaa, Cameron Carlyle, Malinda Thilakarathna
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Cicer milkvetch, an introduced perennial forage legume in Canada, has been previously found to increase forage biomass while reducing soil carbon (C) and vegetation diversity in the Canadian dry mixedgrass prairie region. We sought to understand these ecological changes by examining the effect of cicer milkvetch on vegetation, soil carbon and nitrogen, and microbial composition. A field trial was conducted at the University of Alberta Mattheis research ranch, located in Southern Alberta, in the dry mixedgrass prairie region where cicer milkvetch had been introduced historically. Ten plots of cicer milkvetch were randomly selected, each with an adjacent plot of cicer-free grassland and a second grass plot fertilized with nitrogen at a rate similar to the nitrogen provided by cicer as a legume. Plant species cover, plant biomass, and soil cores were collected from each plot in 2022 and 2023 growing seasons. First-year (2022) results showed that total above-ground biomass was significantly higher in cicer milkvetch plots, with lower plant species richness compared to grass and fertilizer treatments. Available soil nitrogen, measured through Plant Root Simulator (PRS) soil probes, was higher under cicer milkvetch plots and nitrogen-fertilized grass plots compared to the grass plots without nitrogen fertilizer. Soil carbon concentration and C:N ratio were not significantly different between cicer milkvetch and grass plots. Soil microbiome data showed no significant differences in alpha and beta diversity at the genus level among all three treatments. Plant and soil sampling was conducted in the second year (2023), and data is currently being processed to understand the effect of cicer milkvetch on the above parameters.

Genetic Analysis of Clubroot Resistance in the C Genome of Brassica napus and Identification of Molecular Markers

Sonia Navvuru, MSc Student [1], Habibur Rahman
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Brassica napus canola is the major Brassica species cultivated worldwide for its oil. Due to its widespread cultivation, it has become a venerable crop affected by several biotic and abiotic stresses. Among these, clubroot disease, caused by Plasmodiophora brassicae, is one of the most devastating stresses causing a yield loss of 29-90%. To date, clubroot resistance of the A genome of B. rapa has been used in the breeding of clubroot-resistant canola cultivars; nevertheless, the major gene resistance of the A genome has been reported to become ineffective after growing a cultivar only for a few years. On the other hand, the C genome of B. oleracea houses several quantitative trait loci (QTL) and exhibits a broad spectrum of resistance; yet, this resistance has not been exploited to date. The purpose of my research is to develop different segregating populations, such as F2, B1, B2, F3, B1F2, B2F2, by crossing a clubroot-resistant B. napus line, carrying resistance in the C genome, to a clubroot-susceptible canola line, and phenotype the populations for resistance to P. brassicae pathotypes 3A and 3H in order to understand the genetic control of this resistance. Further, these populations will be genotyped by molecular markers and linkage association of the genotypic and phenotypic data will be carried out to identify the polymorphic markers associated with the resistance. The results from this research will facilitate combining the resistances of the A- and C genomes to develop canola cultivars with higher stability of clubroot resistance.

Effects of Humic-Based Soil Amendment on Canola (Brassica napus L.) Growth Under Different Moisture Conditions

Tianyi Zhao, MSc Student [1], Pramod Rathor, Linda Gorim, Malinda Thilakarathna
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Canola is a dominant revenue-generating crop in Western Canada, yet the limited water availability often restricts its production. Humic-based soil amendments have shown promise in enhancing soil health and plant growth. Humalite is a naturally occurring humate material rich in humic acids deposited in southern Alberta. In this study, we evaluated the effects of humalite on canola growth, photosynthesis, and seed yield, under varying moisture conditions. Canola was grown in pots under greenhouse conditions with four different humalite rates (0, 400, 800, and 1600 kg ha-1) and maintained at 80% and 30% field capacities (well-watered and drought-stressed conditions, respectively). Results indicated that drought stress prominently reduced canola performance on tested parameters such as photosynthesis, transpiration, stomatal conductance, biomass, and seed yield. Humalite-supplied plants at 400 kg ha-1 demonstrated significantly improved photosynthesis (13%; 53%), seed yield (19%; 18%), and oil content (24%; 26%) under well-watered and drought stress conditions, respectively. The increases in shoot (14%) and root (31%) biomass were significant only under well-watered conditions. The research findings highlight the potential humalite as a soil amendment for improving canola growth and yields, particularly under moisture-deficit conditions, and provide insights into sustainable agricultural practices.

Oral Presentations in Entomology

Insecticide Susceptibility and Biological Control of Agriculturally-Important Insect Pests on the Canadian Prairies

Alexis Blanchette-Arnold, MSc Student [1], Héctor Cárcamo, Boyd A. Mori
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Insecticide resistance is an increasing concern in agricultural systems globally due to the widespread use of chemical insecticides for insect pest management. Currently, a limited number of insect species have demonstrated resistance to pyrethroid insecticides on the Canadian Prairies; however, susceptibility/resistance levels of many species have not been tested. This is especially concerning given that some invasive species present on the Canadian Prairies are known to have evolved resistance in their native region. For example, in the native range of cabbage seedpod weevil, Ceutorhynchus obstrictus, in Europe, populations in oilseed rape (Brasica napus) fields have developed resistance pyrethroids. Ceutorhynchus obstrictus is an invasive species in Canada, and is a significant pest of canola (Brassica napus) grown across the southern Canadian Prairies. The pea leaf weevil, Sitona lineatus, is another invasive species on the Canadian Prairies, and is a major pest of pulse crops. Here, we will investigate the susceptibility of C. obstrictus, and S. lineatus populations from across the Prairies to pyrethroids. Alternatives to foliar applied pyrethroid insecticides can include other chemical (e.g. insecticide seed treatments) and non-chemical (e.g. biological control) management strategies. To examine these potential alternatives, we will also investigate insecticide seed treatments for S. lineatus management, as well as current parasitism levels of C. obstrictus. Overall, this project will provide a baseline of current pyrethroid resistance, optimal seed treatments, and parasitism levels which can inform management practices.

Effects of Water Chemistry on Dominant Aquatic Invertebrates in Urban Stormwater Ponds

Emily Armfelt, Undergraduate Student [1], Tyler P. Cobb., Olivia DeBourcier, Carol Frost
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Urban stormwater ponds (SWPs) are designed to reduce localized flooding and erosion, but there is a growing interest in their potential additional benefits for biodiversity. Similar to aquatic ecosystems outside the urban setting, water chemistry of SWPs is strongly influenced by factors surrounding the catchment basin. As a result, research is needed to better understand biodiversity responses to water chemistry in city environments. Ecosystem function is often similar within orders of aquatic invertebrates, and dominant taxa contribute the most to ecosystem function. Therefore, by studying water chemistry and its effect on dominant orders of aquatic invertebrates, we can gain insight into how different water chemistry may impact urban SWP systems via selecting for aquatic taxa with different functions. This study investigates relationships between the five most dominant aquatic invertebrate orders of each SWP (taxonomic identity, number of individuals, and proportion of total number of individuals made up by each of these orders), as well as several water chemistry parameters (conductivity, pH, phosphate load, and heavy metal content) in 353 SWPs in the city of Edmonton. At each SWP, a dip-net sample will be collected and then sorted, identified to the order level, and enumerated. We will also collect water samples for chemical analysis. By assessing the effects of differing water chemistry conditions on dominant orders of aquatic invertebrates in SWPs, our results will support residents, the City of Edmonton, and utilities companies (e.g., EPCOR) in making informed decisions about the management and future development of these urban ecosystems.

Contarinia nasturtii-Arabidopsis thaliana: A Model System to Study Gall Insect-Plant Interactions

Carina L. Lopez, MSc Student [1], Boyd A. Mori
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Invasive species are a threat to biodiversity and ecosystem function. In Canada, the invasive swede midge, Contarinia nasturtii (Diptera: Cecidomyiidae), threatens agricultural production of Brassicaceae plants. Midge larvae manipulate their host plant to create galls—abnormal plant tissue deformations produced in response to salivary secretions—which can cause yield losses up to 85% in Brassicaceae crops. A model system to understand the mechanisms involved in host plant selection and manipulation by this insect pest would enhance our knowledge of this pest and possible management strategies. This project aims to develop a model system with C. nasturtii and Arabidopsis thaliana wherein insect-plant interactions can be thoroughly studied. This unique system will explore the plant signaling pathways and defense responses that are manipulated by the midge. Specifically, the objectives of this project are 1) To investigate the life cycle and biology of C. nasturtii on A. thaliana, and 2) To investigate the manipulation of host plant pathways using defense-related mutant lines of A. thaliana relevant to insect-plant interactions. These objectives will be achieved by using a series of no-choice tests with A. thaliana at three distinct growth stages, and infesting plants at each growth stage separately with five densities of adult female midges. Due to initial experiments which found an extremely short life span of C. nasturtii adults, an experiment was conducted to evaluate the effects of water access on the longevity and survival rate of individual male and female C. nasturtii adults. Results of this experiment will provide important information as the model system is refined. Ultimately, this research will begin to elucidate the mechanisms of midge manipulation of the plant and the role of plant defense compounds in response to C. nasturtii.

Would You Still Infect Me if I Was a Worm? Ecology of Fear on Larval Drosophila

Caroline Liang, MSc Student [1], Lien Luong
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Parasites can indirectly impact hosts through non-consumptive effects (NCEs) via changes in behaviour, morphology, and/or physiology. Organisms respond to parasite cues in order to avoid infection, and avoidance is time and energy intensive. These responses can be understood in terms of the ecology of fear framework. Parasite infections can be chronic, resulting in strong cumulative responses to parasites. The study system involves a cactophilic fruit fly (Drosophila nigrospiracula) and a naturally occurring parasitic mite (Macrocheles subbadius). When flies are exposed to mites chronically, their fecundity and longevity is reduced, although that effect has only been studied for adult flies. Larvae cannot be infected by mites although previous work has shown that mite presence can reduce pupation success, and larvae preferentially pupate away from mite cues. We aimed to assess whether M. subbadius exert non-consumptive effects on D. nigrospiracula larvae that persist through development to adulthood. We measured fecundity and longevity in control (no history of mite exposure) and

exposed (exposed to mites as larvae) adult female flies. We did not detect any downstream effects on fecundity or longevity. Control and exposed flies produced similar numbers of offspring and they had similar lifespans. Certain behaviours may respond to immediate shifts in parasite risk, and since flies no longer encountered parasite cues post-emergence, there was no long-lasting effects of mite exposure. Our study highlights the importance of the ecology of fear and how variable environments can modulate the effects of fear.

Flea Beetle and Pea Leaf Weevil Insect Impact on Pea-Canola Intercrop and its Sole Crops

Dominic Donkor, PhD Student [1], Maya Evenden, PhD, Boyd Mori, PhD, Viktoria Wagner, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Intercropping can be defined as the growing of two or more crops together in the same space and harvestable together either at the same or different times. Benefits of intercropping include increased crop productivity, reduced reliance on chemical resources and lower insect pest risk. This study aims to investigate the impact of specialist flea beetles and pea leaf weevils on pea-canola intercrop, and canola or pea monocultures, as well as the influence of climatic and environmental variables on insect pests within a pea-canola intercrop or its sole crop system. Comprehensive commercial field and plot to field site trials will be conducted at different locations throughout central Alberta, in collaboration with the Alberta Pulse and Canola Growers. Laboratory experiments, including olfactometer studies, will also be conducted. Insect pests will be sampled. Treatments will include peaola intercrop, pea monoculture, and canola monoculture with varying fertilization regimes in the intercrop treatment. This study addresses a gap in the current research by examining the mechanisms behind intercropping practices involving peas and canola in Alberta Province. The findings will contribute to a better understanding of the potential benefits and challenges associated with intercropping and provide insights into insect pest management strategies within intercrop and monoculture systems.

Precocious Foraging Behavior in Honey Bees (Apis mellifera) Occurs Independently of Chemical Stress Tolerance

Gursimran Toor, MSc Student [1], Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The western honey bee faces numerous stressors when foraging. Among these stressors, honey bees will inadvertently encounter some form of chemical stress, either from plant-produced phytochemicals or harmful compounds utilised in crop protection, which might lead to the forager's mortality or the accumulation of the compound once they return to the colony. The increased loss of foragers has long-term implications; notably, young, less developed honey bees are recruited as foragers – referred to as precocious foragers. These foragers are often less capable of performing successful foraging flights than normal foragers. One cause of precocious foraging is stress. In this study, we tested the hypothesis that chemical stress tolerance is linked to precocious foraging. We tested the prediction that workers from colonies that are particularly susceptible to the neonicotinoid insecticide imidacloprid also forage more precociously than workers from colonies that are less susceptible. We measured survival of caged bees from 26 colonies of three different stocks from two locations in Alberta over 48 hours when exposed chronically to imidacloprid. Precocious foraging of workers from the same colony cohort sources was studied in three single-cohort-colonies by daily observation of the hive entrances. Honey bees were marked using acrylic paint to identify their origin colonies, and foragers were marked additionally with black paint. We found significant differences between colonies for both chemical stress tolerance and age of first foraging. However, we found that the age at which our honey bees transitioned to foragers was not significantly correlated to the chemical stress tolerance across colonies. Our results suggest the onset of precocious foraging in honey bees likely occurs independently of their susceptibility to chemical stressors. However, our findings also suggest the source colony is a significant factor in determining the age at which they transition to foragers, indicating the presence of an unclear mechanism.

Investigating Virus Presence in Three Different Genetic Stocks of Honey Bee Queens (Apis mellifera)

Heather Anderson, Undergraduate Student [1], Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Honey bees (Apis mellifera) provide important pollination services for crops in North America. Alberta in particular relies on honey bee pollination for canola production. High rates of colony loss continue to affect the beekeeping industry due to various factors, such as disease, parasites, pesticides, and overall environmental change. Apis mellifera are eusocial and reproduction is performed only by the queen and male drones. Therefore, queen bees and their health are highly important for colony success. Beekeepers are increasingly purchasing queen bees from various international and national sources to help increase colony numbers after winter mortality. Honey bee viruses are found in hives worldwide, likely including queens being shipped to beekeepers. This could facilitate the long-range dispersal of viruses. We investigated the presence of six common honey bee viruses in queens collected from three different breeders, one local, one national, and one international. We analyzed the queen tissue using RNA extraction and qPCR analysis to quantify the viral concentrations in the incoming queens. Upon analysis, we expect to find viruses present in the queens we have collected. Variation among sources can affect management decisions and the virus prevalence and abundance in queens directly determine their capacity to serve as long-range vectors, further threatening the sustainability of the beekeeping industry. We conclude that more awareness of queen virus impacts on colony health is needed.

The Effects of Sleep Deprivation on Susceptibility to Parasitic Infection

Holly Tang, Undergraduate Student [1], Lien Luong, Jacob Berry
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Sleep is an important biological function in many species, and a lack of sleep has been shown to have adverse effects on an individual's metabolism, immune system, homeostasis, and circadian rhythm. In this study, I aim to determine the effects of sleep deprivation on susceptibility to parasitic infection. Parasites make up a large proportion of earth's diversity of life, yet, the interaction between the ectoparasitic mite, Macrocheles subbadius, and the fly, Drosophila nigrospiracula, are one of the only two known parasitic interactions for Drosophila. Therefore, I wish to gather a better understanding of factors that influence parasitic infections between the species of interest. Grooming is a behavioural defensive mechanism seen in D. nigrospiracula against ectoparasitic infections by M. subbadius. However, grooming is energetically costly, and previous studies have suggested that grooming increases a host's susceptibility to future infections. Therefore, I hypothesize that if sleep deprivation increases the flies' susceptibility to infection, there will be higher infectivity rates among the sleep deprived (SD) flies than the non-sleep deprived (NSD), because they will have lower endurance and be less likely to successfully defend themselves against mites. I predict the infectivity rate will be higher among the sleep deprived SD flies compared to NSD. I tested susceptibility to infection of 14 day old and 21 day old flies by placing either 14 or 21 day old SD and NSD flies into a jar with mite filled media for 2 hours, and then checked for attachment of mites on flies. As of now, I have found no statistically significant difference between SD and NSD flies. Through this experiment, we can better understand the effects of sleep deprivation on parasite resistance.

The Weak Worker Hypothesis: A New Framework for Understanding Division of Labor in Social Insects

Jacob J. Herman, PhD Student [1], Alexander Walton, Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Individual variation is important for group function in all social species. Many highly integrated social insect colonies rely on division of labor among colony members and emergent properties of their collective behavior and physiology. Response threshold models are a prominent, proximate explanation of division of labor in many contexts, but how different response thresholds arise is largely unexplored. We propose the "Weak Worker Hypothesis", suggesting that specific tasks are preferentially performed, or at least initiated by the individuals that are most susceptible to the underlying stressor. Here, the response threshold is an internal evaluation of a task-specific stimulus that is influenced by the severity of the physiological perturbation of the individual, which simultaneously determines the susceptibility of this individual to succumb to the external disturbance. Under this premise, varying individual stress susceptibilities thus generate division of labor and group stability. The Weak Worker Hypothesis provides a functional explanation for individual-level responses to environmental deviations from optimal conditions, which equally represent stimuli and stressors. We discuss how the known relation between the nutritional status of individuals and their foraging behavior fits the Weak Worker Hypothesis framework. Moreover, we present a new dataset in support of the hypothesis on the relation between individual heat tolerance and

fanning behavior in honey bees (Apis mellifera L). We conclude that the Weak Worker Hypothesis could provide a useful extension of response threshold models for understanding the division of labor in social groups and might have repercussions for applied social insect science, such as selective breeding and eradication efforts.

Oribatid Diversity, Distribution, and Dispersal Along Alberta's North Saskatchewan River

Jessica Lario, MSc Student [1], Lisa Lumley, Heather Proctor
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The North Saskatchewan River (NSR) is a complex network connecting large portions of land. This network provides a mechanism for long-range dispersal that could be used by many organisms to travel. Limited research has focused on terrestrial organisms using these systems for passive dispersal, such as oribatid mites. These terrestrial arthropods are typically found in soil environments and may be important indicators of soil health. Several species have been found only in the NSR valley, within and downstream of Edmonton. This pattern could be due to limited research, and they may be an endemic species to central Alberta, or it could be an indication of introduction through urban run-off from Edmonton. This project aims to determine the diversity of species found within the NSR across the province of Alberta and determine if this pattern suggests oribatid mites could be using the river for dispersal. Soil samples are collected upstream, downstream, and within Edmonton, and invertebrates are extracted using Tullgren funnels prior to using microscopy to sort and identify oribatid mites to species level. The data collected in this study can be applied to other terrestrial organisms that may be using this long-distance mode of transportation to new habitats.

Buzzing With Potential: Prediction of Honey Bee Queen Success by Stock, Physical Attributes, and Behavioral Traits

Kayla deJong, Undergraduate Student [1], Olav Rueppell
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

The management of honey bees in Alberta is important both for pollination and economic purposes. However, Albertan beekeepers face many challenges – overwintering colony loss rates approaching 50% being one of the most important issues in recent years. One of the most frequently cited reasons for overwintering failure by beekeepers is poor queen quality. The queen may be only one individual among thousands in a given colony, but as the mother of all other bees in the hive she has potential to influence colony-level behaviours and success, both through her own behaviour and the genetic attributes she passes on to her offspring. Beekeepers face trade-offs in selecting genetic stock when purchasing new queens, and evaluating a queen's quality before placing her in a colony is very difficult. Here, we aim to both evaluate differences between queens from local and non-local lineages, and to test the efficacy of a variety of queen-specific physical measurements and behavioural tests on predicting eventual colony success. 45 queens from three different stocks (Alberta, Quebec, Hawaii) were evaluated by physical and behavioral traits in a central location prior to being distributed to three apiaries (Northern, Central, and Southern Alberta) and placed in colonies for a period of two years. During this period, colony-level attributes, such as temperature variation and disease level, were recorded. High honey production and overwintering success are desired by beekeepers, thus queen and colony level traits were evaluated as predictors of these parameters. Our results indicate that queen traits, both physical and behavioural, are poor indicators of colony success. However, disease level does appear to influence overwintering success, indicating that environmental conditions may predominate. Additionally, overwintering success, honey production, temperature variance, and queen longevity vary by stock. Despite the general assumption that locally-adapted queens outperform queens shipped from other locations, we found queens of Albertan stock to underperform, and queens from the Quebec breeder to perform the best.

Population Structure and the Evolutionary Significance of Elytral Colour Patterns in the Big Sand Tiger Beetle (Cicindelinae: Cicindela formosa)

Wei Han Lau, MSc Student [1], Felix Sperling
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The Big Sand Tiger beetle (Cicindela formosa) is a large, charismatic beetle that has become a focus of conservation efforts and evolutionary studies. Due to its narrow ecological tolerance, C. formosa is at risk of being driven to extinction in the western regions of its range, especially within Alberta (COSEWIC 2012). C. formosa exhibits highly variable elytra (hardened outer wings) colour patterns. These elytral colour patterns form the basis of subspecies delimitation in C. formosa. However, these patterns may be a highly adaptive trait, and thus unsuitable for delimiting evolutionary significant units (ESUs) (Schultz and Hadley 1987; Nayuta and Teiji 2020). Further, discordance between genetic relatedness and elytral patterns is found within this species, and broadly among tiger beetles as well (French et al. 2020, Laroche et al. 2023). As such, and despite the species' significance for evolutionary studies and the threat of extinction this species faces, ESUs within this species remain unresolved. Are there only three subspecies in Canada? Should some populations be given conservation priority over others? This project will answer these questions by using genome-wide DNA sequence data to test prior taxonomic hypotheses. Further, as part of this project, I will identify the genomic basis for elytral patterns by coupling genomic, phenotypic, and climatic data. In summary, this project aims to reassess the ESUs of an imperiled species complex while contributing to our understanding of the selective pressures driving colour pattern variability in Cicindela.

Oral Presentations in Molecular Biology and Genetics

Characterizing a Novel Memory Suppressor Gene, SEC22

Akhila Eswaran, PhD Student [1], Anna Phan
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The identification and characterization of genes that are required for normal memory processes, has been the focus of research over the past few decades. However, recent studies have shown that genes functioning to suppress memory also exist. Through a targeted small scale RNAi memory screen conducted using Drosophila melanogaster, sec22 was identified to constrain memory as a novel memory suppressor. sec22 is ubiquitously expressed and plays a vital role in the transport of vesicles between the endoplasmic reticulum and Golgi apparatus. However, sec22's role in learning and memory is unclear. Using the aversive olfactory behavioral assay, we found that knockdown (KD) of sec22 in all neurons, dopamine neurons and mushroom body neurons (MBNs), significantly improved memory due to a specific enhancement of learning. We also found using a temporal gene expression system (GAL80ts) that this effect is due to sec22's function during development as well as in adulthood. sec22 is part of the Synaptobrevin family of genes consisting of the vesicle fusion and secretion associated genes, ykt6, vamp7, syb and nsyb. Interestingly, all these genes were found to affect memory acquisition as ykt6 KD in MBNs enhanced learning while KD of syb, vamp7 and nsyb, impaired it. To elucidate sec22's mechanism of memory suppression, the effects of sec22 KD on live neuronal function, neuronal anatomy and neurotransmission will be investigated using immunohistochemistry, in-vivo imaging and confocal imaging methods. It is postulated that characterizing sec22 as a memory suppressor will reveal key insights into cellular mechanisms of learning and memory. Moreover, studying memory suppressors may help reveal therapeutic targets for neurological disorders associated with memory loss.

Identification of Photoreceptors as Rods vs Cones in Early Vertebrates

Aprajita Gupta, Undergraduate Student [1], Gavin Neil, Ted Allison
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Ciliary photoreceptors allow for vision in all vertebrates, but their evolutionary origins remain obscure. Rods allow for vision in poorly lit conditions, whereas cones allow for coloured vision and greater visual acuity. Early vertebrates only contained cone-like photoreceptors. During evolution, rods began to dominate the mammalian retina, as a result of the transcription factor, neural retina leucine zipper (Nrl). Nrl specifies rod-cell fate in mammals and is capable of inducing this fate even in cone-fated precursors when expressed transgenically, as shown in our previous studies. My study tests whether Nrl homologs from different species also specify rod cell fate in zebrafish through transgenic engineering of zebrafish. Studying Nrl function in photoreceptors will allow us to determine whether agnathan photoreceptors exhibit similarities to either rods or cones. Using transmission electron microscopy and immunohistochemistry, we aim to determine whether these cells exhibit morphologically similar phenotypes to that of normal rod photoreceptors. We have found that Nrl homologs from various

species are able to specify rod cell fate, using the rod specific antibody 4C12. We are currently working on obtaining data depicting morphological features of these rods. If we confirm hagfish homologs of Nrl are able to exhibit similar features of that of rod photoreceptors, then this will help us develop a better understanding of the retina present in the last common vertebrate ancestor. This will allow us to gain a deeper understanding of the evolutionary origins of the photoreceptors.

Spermatogenesis to Oogenesis: How Does the Regulation of the Switch Differ Between Caenorhabditis tropicalis And C. elegans

Chloe Cettiga, Undergraduate Student [1], David Pilgrim, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The Caenorhabditis genus is made up of nematodes that have binary male/female fates, except for three species that have independently evolved as male/hermaphrodite: C. elegans, C. briggsae, and C. tropicalis. The regulation of spermatogenesis and oogenesis in C. elegans is thought to be controlled by the CUL2/FEM complex, which acts as an E3 ubiquitin ligase to repress TRA-1 from turning on oogenesis-related genes, thereby promoting continued spermatogenesis. One of the FEM proteins, FEM-1, is thought to act as the substrate recognition subunit (SRS) of the CUL2/FEM complex and recognizes TRA-1 for degradation. A specific motif called the VHL box indicates whether a protein acts as the SRS in an E3 ligase complex. The consensus sequence for this motif was found in the C. elegans FEM-1 amino acid sequence. Because of this finding and FEM-1's interaction with TRA-1, it's been determined that FEM-1 acts as the recognition component for this regulatory complex, with TRA-1 as its substrate. However, after employing bioinformatic analysis to compare the C. tropicalis FEM-1 sequence to the VHL box consensus sequence, it was found that this motif is not conserved. The question we now ask is how did the regulation of the C. tropicalis spermatogenesis-to-oogenesis switch evolve differently compared to C. elegans, and how did this contribute to the independent evolution of C. tropicalis self-fertile hermaphroditism? For this project, the interaction between FEM-1 and TRA-1 is being assayed using the RTA yeast two hybrid system. As well, to further analyze the conservation of the VHL box motif, a bioinformatic comparison has been conducted, and compares the VHL box sequence of the male/female sister species to those of the hermaphrodite/male species.

Deciphering Distinct Signaling Networks That Regulate Dopamine-Mediated Learned and Innate Behaviours

Dana Guhle, PhD Student [1], Jacob Berry, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

A critical function of the brain is to elicit optimal and adaptive behavioural responses needed to survive. An animal's response must adapt based on past events (i.e., memory) or internal state (i.e., starvation-altered innate behaviour). Interestingly, in Drosophila melanogaster, dopaminergic (DA) signalling through a single receptor, Dop1R1, regulates both learned and internal state driven behavioural plasticity. Dop1R1is highly expressed in the memory center and is critical for encoding memories through adenylyl cyclase (ADCY1 ortholog) driven synaptic depression. DA to Dop1R1 signaling through a separate adenylyl cyclase (ADCY2 ortholog) also drives state dependent changes to innate odour preference. It remains unclear how these Dop1R1 pathways bi-directionally regulate memory synapses. To advance our understanding of Dop1R1 signaling environments, we utilized TurboID proximity labelling proteomics and RNAi screening to identify interactors that help regulate learning and innate behaviour. Proximity labelling in active neurons in vivo using a CRISPR knock-in of a Dop1R1-Turbo-V5 construct identified candidate proteins significantly enriched around the Dop1R1 receptor compared to another memory regulator, Dop1R2. Disruption of candidate proteins in memory circuits lead to significantly altered behavioural plasticity in both starved odour preference or associative learning contexts. Interestingly, candidate interactors affect either internal state or learned behavoural plasticity but not both, indicating distinct pathways. In vivo functional imaging experiments will be conducted to reveal if these candidates alter Dop1R1 receptor mediated synaptic depression or potentiation, and downstream cAMP signalling. Altogether, our study will identify and characterize novel pathways regulating dopaminergic signalling to illicit two distinct behaviours and illuminate how the brain genetically fine-tunes these behaviours through one receptor.

Pharmacological Inhibition of ROMO1 Attenuates Ovarian Cancer Cell Proliferation

Dania Al-Rimawi, Undergraduate Student [1], Jason Dyck
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

In Canada, it is estimated that 6 women die of ovarian cancer every day, suggesting a need for improved targeted anticancer therapies. Previous studies have demonstrated that a protein called Reactive Oxygen Species Modulator 1 (ROMO1) is expressed at very high levels in the mitochondria of ovarian cancer cells and is associated with rapid cancer cell growth. Our lab used in silico screening of an FDA-approved drug library and has identified a drug with an off-target effect of inhibiting the protein ROMO1. Based on this, we hypothesized that pharmacological inhibition of ROMO1 with this drug will prevent cell cycle progression, impairing cancer cell growth and survival. Methods: To verify that high ROMO1 levels are associated with more severe outcomes, Kaplan-Meier (KM) curves were generated using ovarian cancer patient data based on low or high ROMO1 expression (kmplotter.com). Progression-free survival (PFS) and overall survival (OS) were assessed as endpoints. To determine if this drug reduces ovarian cancer cell growth, human ovarian cancer cells were treated with either vehicle or ROMO1 inhibitor. After 24 hours, cellular metabolic activity, which is positively correlated with proliferation, was quantified using colorimetric MTT assays. Cells were also lysed and subjected to western blot analysis for expression levels of p27, a tumour suppressor protein which regulates cell cycle progression. Results: Ovarian cancers with high ROMO1 expression had greatly reduced progression-free survival (PFS) and overall survival (OS) compared to those with low ROMO1 expression. Furthermore, ovarian cancer cells treated with the ROMO1 inhibitor exhibited a 56.1% (n=7) decrease in proliferation compared to vehicle-treated control cells. Lastly, cells treated with the ROMO1 inhibitor exhibited a significant increase in p27 expression, indicative of cell cycle arrest. Significance: This work may ultimately lead to the development of an effective anticancer therapy to improve clinical outcomes of women with ovarian cancer.

Dmd Exons 3-9 Skipping by Antisense Oligonucleotides

Eric Yan, Undergraduate Student [1], Harry Wilton-Clark, Rohini Roshmi, Toshifumi Yokota
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Duchenne muscular dystrophy (DMD) is a rare disease often caused by large deletions in DMD, leading to reading frame shifting and lack of the product, dystrophin. Progressive muscle loss occurs, followed by premature death. Recent research has attempted to treat the disease with splice-switching antisense oligonucleotides (SSOs), which modify splicing so exon(s) adjacent to the deletion are excluded from the transcript and the reading frame is restored. The resulting product is truncated but largely functional. There exist 4 such FDA-approved drugs, though they function by skipping single exons near the C-terminus, leaving N-terminal mutations underserved. Our project objective is to simultaneously skip exons 3-9 of DMD. Natural history studies have recorded exons 3-9 deletions producing a practically healthy phenotype, showing more restorative potential than smaller in-frame deletions. Further, the wide range of skipping increases applicability; exons 3-9 skipping could be used to treat the majority of patients with N-terminal mutations. Project workflow began with differentiating a DMD patient cell line into myotubes to represent skeletal muscle. At this point, we treated with individual SSOs or SSO combinations and extracted RNA from cells for RT-PCR. This identifies the proportion of DMD mRNA with the desired exon(s) skipped, showing SSO effectiveness at the transcript level. We extracted protein from cells for Western blots to identify dystrophin restoration and SSO effectiveness at the protein level. At the cellular level, we are currently attempting immunocytochemistry to identify dystrophin localization and validate other results. These assays will be repeated in patient-derived cardiomyocytes to represent cardiac muscle, a tissue SSOs are known to be poorly delivered into. We expect our results to reveal the restorative potential of exons 3-9 skipping for Duchenne muscular dystrophy. This may inform future SSO drug design, potentially laying a foundation for a new treatment option for patients.

The Effects of Chronic Social Isolation on Drosophila Sleep

Heba Mahmood, Undergraduate Student [1], Anna Phan
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Adverse effects brought on by social isolation during the COVID-19 pandemic have become increasingly evident. In humans, social isolation has been reported to disturb sleep. Fruit flies (Drosophila melanogaster) are a social species that demonstrate cooperative behavior and a preference for social stimuli over non-social stimuli. Interestingly, social isolation of Drosophila has also been shown to result in sleep loss. Due to 65% of human disease-causing genes found in fly genomes, findings have implications for human research. My research focuses on characterizing behaviors of socially isolated flies. Objective one was to test sleep disruption using the DAM system, which records locomotor behavior. Data was analyzed with MATLAB

and results showed that isolated males have fragmented sleep at night compared to grouped housed males, and isolated females sleep more during the day compared to grouped housed females. Objective two was to determine whether the effects of social isolation on sleep are intergenerational. Results revealed that female progeny of isolated flies are less active when awake at night. Objective three was to differentiate the effects of isolation on sleep behavior between flies isolated during development versus flies isolated in adulthood. We hypothesize results will indicate that developmental and adult isolation both lead to similar sleep disruption in flies. This research is significant because determining the behavioral effects of social isolation lays the foundation for future research that will examine how isolation alters the brain to cause these behaviors. Investigating these cellular mechanisms is critical for attenuating the negative consequences of social isolation. This project will also aid in further understanding implications of social isolation on mental health.

Unveiling the Molecular Tapestry: Downstream Transcriptional Targets That Mediate Stromalin's Effects on Synaptic Vesicle Pool Size

Illia Pimenov, MSc Student [1], Courtney M. MacMullen, Ronald L. Davis, Anna Phan
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Learning and memory are two of the fundamental cognitive processes. While a lot is studied about genes that promote learning, little is known about genes that limit memory formation and why such functions exist. Recently, *stromalin* has been shown to be a learning suppressor that functioned by restricting the synaptic vesicle (SV) pool size in dopamine neurons (DANs) in Drosophila melanogaster. While SV regulation, function, and numbers are critical for learning and memory, the transcriptional regulation of SV numbers remains unexplained. To advance our understanding of this enigma, a DAN-specific RNA-Seq with silenced stromalin was performed to identify the genes downstream of it that mediate its effects on SV numbers. The identified significantly differentially expressed candidates then underwent a primary aversive olfactory memory screen and a secondary transgenically expressed SV marker (Syt:eGFP) screen to further narrow down the list of potential targets. After performing validation experiments and using Nanostring's nCounter to test mRNA levels of genes with memory effects consistent with stromalin's transcriptional and behavioral effects using whole brain control and pan-neuronal stromalin knockdown, two genes were outlined as primary candidates: *nep1* and *su(z)12*. Further nCounter experiments tested whether *su(z)12*, a transcriptional regulator, may regulate *nep1* itself. Based on the results, we are now evaluating the hypothesis that *nep1* mediates stromalin's memory suppression effects via limiting SV numbers. We are doing this by testing whether overexpression of *nep1* can rescue stromalin knockdown effects on memory. Additionally, using advanced in vivo functional imaging, we intent to look at the dopamine release from DANs and mushroom body neurons with silenced stromalin and/or *nep1* to further test the hypothesis that *nep1* is indeed a downstream transcriptional mediator of stromalin's SV pool size effects in neurons. In the future, we aim to unravel the mechanism of how *nep1* alters neuronal SV pool size.

Genetic Landscape of Epilepsy: Diagnostic Yield of Clinical Exome Sequencing

Jaya Bansal, Undergraduate Student [1], Saadet Andrews
[1] Department of Medical Genetics, Faculty of Medicine & Dentistry, University of Alberta, Edmonton, Alberta, Canada

Seizures are due to unusual electric brain activity and result in uncontrolled body movements. When seizures occur more than once, they are called epilepsy, a common neurological disorder. A vast majority of patients with epilepsy to date have an unknown diagnosis. Currently, the approach is to conduct multiple investigations to formulate a diagnosis. It not only prolongs the time that a patient must wait but also hinders the timely receipt of an appropriate treatment, complicating the process of identifying an optimal course of action. Therefore, achieving a prompt and accurate diagnosis is crucial as it will be the key to delivering disease-specific treatments. Previously. Dr. Andrews reported the diagnostic yield of genetic investigations in childhood epilepsy - about one-third had a genetic diagnosis, and one-third had a specific treatment implication due to their genetic diagnoses in those studies. Since then, she has provided genetic diagnostics to about 200 individuals with epilepsy. Our study intends to investigate the diagnostic yield of molecular genomic investigations in adult and pediatric patients, bridging the age cohort. We hypothesize that there are specific phenotypes, electrophysiological abnormalities, and neuroimaging features to suggest an underlying genetic disease in individuals with epilepsy. To test our hypothesis, electronic charts of approximately 150 individuals with epilepsy seen in the Epilepsy Genetics Clinic will be reviewed for phenotypes, electrophysiological investigations, neuroimaging, and genetic investigation results. We will divide the study cohort into two groups: 1) individuals with genetic epilepsies and 2) individuals with no genetic epilepsies. We will develop a diagnostic algorithm based on our study results. We expect that one-third of individuals with epilepsy have an

underlying genetic disease. The main goal our algorithm sets to achieve is to shorten the diagnostic odyssey for individuals with epilepsy.

Regulatory and Downstream Elements of the Hippo Pathway in Zebrafish During Hindbrain Development

Jorge Hernandez, Undergraduate Student [1], Renée Dicipulo, Anna Desharnais, Andrew J. Waskiewicz
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The hindbrain is a major developmental region of the nervous system that gives rise to most cranial nerves and CNS organs, controlling several autonomic functions and voluntary motor movements. Because hindbrain and brain ventricle system development are highly coupled, any defective ventricle formation will cause severe neurological abnormalities. Consequently, research is focused on elucidating multiple pathways that control hindbrain maturation and patterning. One currently explored is the Hippo signalling pathway, as it remains unclear how it affects hindbrain ventricle development. The Hippo pathway has as a central protein the WW domain containing transcription regulator 1 (also known as Taz), that is regulated by phosphorylation and activate different transcription factors that control neurogenesis and segmentation of the hindbrain in units known as rhombomeres. This research focuses on investigating the regulatory and downstream elements of Taz. Previous research showed that zebrafish taz-/- mutants exhibit abnormal ventricle formation, demonstrating the critical role of Taz. Additionally, taz-/- mutants showed downregulation in rac3b (GTPase) and rasgef1ba (Guanine exchange factor) genes. Therefore, to determine the role of rac3b and rasgef1ba activity driving the aberrant phenotype in Taz mutants, we plan to generate CRISPRANTs in wildtype zebrafish to downregulate rasgef1ba and rac3b, and compare the obtained phenotype with the aberrant phenotype, confirming that these genes play a role in ventricle development. Moreover, we have shown that in taz-/- mutants, mpz and dnajc9 genes were upregulated in the hindbrain. To confirm the role of mpz and dnajc9 driving ventricle development, we will overexpress mRNA of these genes in both wildtype and taz-/- mutants. Testing whether overexpression exacerbate the aberrant phenotype or result in a wildtype phenotype. Overall, this work will contribute to understanding the regulation and downstream effects of Taz during hindbrain and ventricle development.

Looking into the Evolution and Early Development of Photoreceptors: Determining the Specificity of Nrl for Cell-Fated Cone Conversion to Rod-Like Morphologies

Kaitlyn H Kluttig, Undergraduate Student [1], Gavin J, Neil, Ted W. Allison
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

NRL is a rod-determining transcription factor where loss of function leads to an absence of rods and an excess of S-cones (short wavelength cones, or UV cones). In addition, NRL driven with an sws1 opsin promoter in nrl mutant zebrafish transgenics has shown the ability for cell-fated S-cones to develop rod-like morphologies (Oel et al. 2020). These discoveries led to questions of photoreceptor evolution and processes in early development. These speculations address the generally accepted notion that cones developed first, with rods developing from these cells (Shichida and Matsuyama 2009), where perhaps these photoreceptors evolve and develop closer together than was previously believed. This study investigates the genetic relationship between photoreceptors in relation to NRL's specificity on effects towards cone cell-fate. Transgenic plasmid constructs using the Tol2 system are injected at the single-celled stage to drive zebrafish NRL (zfnrl) with different cone-opsin promoters to determine if NRL has the potential to change a broad selection of cone cell-fate to rod-like development, or if this manipulation is specific for S-cones. Through varying zebrafish genotypes and cone-opsin promoter utilization, early results have implicated towards S-cone specificity in this cell-fated switch. This suggests that S-cones and rods are more closely related than other cones in evolution, with potentially being closer in early development as well. Continuing research is focusing on increasing sample sizes, quantifying cells, using statistical analysis, and looking into more promoters to strengthen these findings. Future uses of this research can lead to clinical applications in treating rod degenerative diseases with therapeutic approaches using photoreceptor generation with aims to regain the loss of rod cells.

Disentangling the Link Between Neuroinflammation and Seizures Post-traumatic Brain Injury: Implications for Tauopathy and Dementia Prevention

Leith AlKhatib, Undergraduate Student [1], Ted W. Allison
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Each year in Canada, over 20,000 people are hospitalized due to Traumatic Brain Injury (TBI), which is an independent risk factor for developing prominent, devastating dementias. Our lab has shown that mitigating post-TBI seizures can reduce accumulation of a dementia-linked protein, called tau. Increased inflammation has also been linked to TBI. Our goal is to disentangle which mechanisms are dominant (and most accessible for prophylactic intervention) after TBI. We will approach this by knocking down the inflammatory response through Toll-like receptor 4 (TLR4), a key component of inflammation. Then, we will determine if anti-inflammatory mitigation of tauopathy is reversed when seizures are re-kindled in the presence of convulsant drugs such as kainate. Utilizing a Zebrafish model with a CRISPR/Cas9-induced loss-of-function mutation in TLR4, we seek to validate the model's diminished inflammatory response following TBI through quantification of inflammatory biomarkers via qPCR. Specifically, we anticipate an upregulation of pro-inflammatory cytokines, such as interleukin 1-β, interleukin-6, and Tumor Necrosis Factor α, in wild-type (WT) fish compared to our TLR4 knockout (KO) model. Subsequently, we will investigate the impact of reduced inflammation on post-TBI seizure characteristics and tau protein levels, aiming to discern whether anti-inflammatory strategies can mitigate tauopathy. Preliminary experiments have confirmed successful CRISPR-induced deletion mutations in injected fish, enabling our establishment of a stable transgenic line. Moving forward, our qPCR analyses will provide insights into the inflammatory profile post-TBI in our TLR4 KO model. By elucidating the interplay between inflammation, seizures, and tau pathology, our study aims to pave the way for innovative therapeutic interventions aimed at preventing dementia onset following TBI.

Environmental Nucleic Acid Applications for Freshwater Fish Biomonitoring

Melissa D. Misutka, MSc Student [1], Chris N. Glover, Micah Brush, Greg G. Goss, Heather D. Veilleux
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Environmental nucleic acids (eDNA & eRNA) are a non-invasive tool for the biomonitoring of aquatic species. To detect a species of interest, water samples are collected and filtered, and nucleic acids are extracted and analyzed in downstream applications such as qPCR and Next-Generation Sequencing. While eDNA barcoding is a relatively well-established approach, representing a highly sensitive and specific method for determining species distribution, spawning patterns, or migration patterns, the utility of eRNA is less well known. We designed and optimized an eDNA and eRNA detection assay for a species of special concern in Alberta, Arctic grayling. This freshwater fish is an iconic species, believed to be in decline, but our understanding of its distribution and factors affecting it, is poor. Improved tools for monitoring current and future species distribution are required. To this end, we developed assays, tested their specificity and sensitivity, and determined best practices for achieving maximum yield from field samples. To gain an understanding of the spatiotemporal acuity of both eDNA and eRNA, we determined decay and shedding rates and modelled detection probabilities in laboratory settings under various flow rates. Environmental RNA had lower shedding rates, but similar decay rates, to eDNA. Both molecules are predicted to be detectable in a pool of volume < 32,500m3 and flow rate < 0.5m3/s-1. Our assays were successfully applied to 2 watersheds across Alberta: Athabasca and Peace/Slave River Basins. This assay will help establish a baseline for Arctic grayling distributions, and how they will change over time under anthropogenic pressures such as climate change. Our eRNA assay is under further development with the goal of understanding fish population health. Additionally, this work will provide insight into the applications and limitations of environmental nucleic acids as a tool for aquatic biomonitoring.

What Can Worms Teach Us? Evolutionary Novelty in the Sex Determination Pathway of Caenorhabditis nematodes

Montana Bobinski, PhD Student [1], David Pilgrim
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

From an evolutionary perspective, it is well-understood why the forelimbs of mammals are similar in structure: they are born of descent from a common ancestor. However, the genes which regulate limb formation in mammals have diverged drastically over time to produce a variety of limbs in extant species. This process by which pre-existing factors change to generate new structures is known as evolutionary novelty. And on this topic, one question remains unanswered: to what degree can related species change on a developmental and genetic level to adopt novel traits? Two of the three hermaphrodite/male nematode species in the Caenorhabditis genus, C. elegans and C. briggsae, are robust for the study of the function and the evolution of genetic pathways that regulate developmental decisions. Under the assumption that sex

determination pathway genes found in C. elegans and C. briggsae will allow for the discovery of similar sex-determining genes in other nematode species, this project has entailed the use of the third male/hermaphrodite Caenorhabditis species, C. tropicalis, which is not closely related to either C. briggsae or C. elegans. Currently, the Pilgrim lab has had much success in using targeted mutagenesis techniques, such as CRISPR/Cas9, to knockout sex-determining homologues in C. tropicalis to observe their effects on germline cell production, and I am excited to report a number of sex cell-related phenotypes which implicate the C. tropicalis sex determination strategy as similar to C. elegans in some instances while apart from C. elegans and C. briggsae in others. As a poorly understood genetic system, C. tropicalis will provide the opportunity to not just continue the development of the Caenorhabditis genus as a principal model for developmental biology, but will also allow us to explore how signal transduction pathways regulating developmental decisions in related species can diverge in the genes recruited over long stretches of evolutionary time.

Investigating the Role of UNC119 in Left-Right Patterning in the Zebrafish

Olive Lo, MSc Student [1], Francesca Jean, Dave Pilgrim
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Cilia can be categorized into primary cilia and motile cilia. Regardless of the motility, the fundamental structures of both types of cilia are the same – both are composed of microtubules. During ciliogenesis, microtubule subunits are transported from the ciliary base and are added to the tip by intraflagellar transport (IFT) complexes. Malfunctioning of IFT proteins results in disrupted cilium biogenesis and function, eventually leading to ciliopathic diseases such as kidney diseases and retinal degeneration. Unc119 proteins regulate ciliogenesis via lipidated intragflagellar targeting (LIFT), and loss of function mutations may result in disorganized microtubule structures and sensory defects. My project tries to understand how unc119 regulates ciliogenesis in the Kupffer's vesicle (KV) of Zebrafish embryos, and aims to provide insights on whether unc119 is involved in Left-Right (L-R) patterning in mammals. The KV is a fluid-filled organ lined with both primary and motile cilia and plays a critical role in establishing the L-R axis of the fish. My hypothesis is that unc119 mutants will have disturbed microtubule organization in both types of cilia in the KV, leading to ciliary defects and abnormalities in L-R patterning. I will test my hypothesis by generating mutants using CRISPR/Cas9 and screen for fishes that exhibit abnormal L-R axis. I will then examine the structure, distribution, and function of both primary and motile cilia in the KV of the mutants. As the KV is equivalent to the organizer of asymmetry in mammalian species, this study can provide understanding on how unc119 may be involvled in the establishment of L-R asymmetry in mammals including human, in which abnormal L-R patterning results in situs inversus that can lead to cardiac problems.

Characterizing Learning-Induced Sleep in Drosophila melanogaster

Rebecca Dust, Undergraduate Student [1], Jacob Berry
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

It is well-established across the animal kingdom that sleep regulates memory and the neurotransmitter dopamine (DA) is critical for both memory and sleep. However, it is much less clear how learning in turn regulates sleep and how neural circuits that store memories communicate sleep needs. While sleep has been proven to support consolidation of long-term memory (LTM), a recent study indicates that, at least for courtship memory, post-learning sleep is driven by the learning itself, and it requires DA receptor Dop1R1 and output from the memory encoding circuit MBON-g2a'1. While courtship memory is complex and multisensory, we hypothesized that a simpler unimodal learning could also drive sleep and may use similar or even distinct molecular and circuit mechanisms. We utilized a series of classic olfactory learning paradigms to train flies to associate odours with a punishing electric shock and then assay their sleep and activity across 24 hours post-learning. Consistent with the prior study, we found brief training does not alter sleep, but spaced training, a paradigm that creates consolidated LTM, also drives robust increases in sleep during the first 3 hours compared to untrained animals. We are currently testing if this post-learning sleep requires Dop1R1 and synaptic output from MBON-g2a'1. Thus, we offer evidence that the formation of long-lasting simple unimodal memories drives sleep that presumably aids consolidation. By using this simple well-studied spaced olfactory assay, we expect this project will add significantly to uncovering molecular, synaptic, and circuit mechanisms that connect memory storage to sleep needs.

Prophylactic Treatment with Anticonvulsants in a Zebrafish Model of Post Traumatic Seizures, Demonstrates Seizure and Tau Pathology Reduction

Sam Tan, MSc Student [1], Laszlo F. Locskai, Lance Doucette, Melissa Kinley, Zixuan Li, Eva Dhupia, Richard Kanyo, Simonetta Sipione, Ted W. Allison
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Traumatic brain injury (TBI) is a common affliction found from mild to severe levels in millions of individuals worldwide. It is also a known risk factor for tauopathies, dementia, and other neurodegenerative diseases. Tauopathies in particular describe a family of neurodegenerative diseases characterized by the abnormal formation of tau aggregates like chronic traumatic encephalopathy (CTE) and Alzheimer's. Seizures have also been shown to be a mechanistic link between the prion like spreading of tau and cell death surrounding neurodegeneration. We have utilized a novel TBI method, replicating blast induced TBI and a tauopathy reporter in zebrafish, able to effectively report the presence of human tau aggregates. With these tools, we have determined that anticonvulsant treatment of zebrafish with post traumatic seizures after TBI, reduces tau aggregates, cell death, and seizure activity. A goal of this project, is to test further anti-epileptic drugs (AEDs) and their synergistic effects with other AEDs, where we will examine reduction on neurodegenerative pathologies of tau and seizures. Calcium imaging of brain activity (CaMPARI) is another tool we are continuing to optimize for imaging brain activity during seizures and after anticonvulsant treatment. Future work includes removal of the receptors that bind to these AEDs with CRISPR gene editing to further elucidate the drugs' specificity and receptor necessity for the post-traumatic seizure phenotype in zebrafish. Treatments with the AEDs removal and TBI will demonstrate receptor importance in antiepileptic treatments. This investigation will allow us to build on ideas to further refine clinical treatment aspects of post traumatic seizures. In particular, this contributes to work seeking better clinical outcomes concerning Alzheimer's and other related neurodegenerative diseases.

Exploring the Effects of Traumatic Brain Injury on Zebrafish Cognition and its Potential Reduction Through Anti-Epileptic Drugs

Shayleen Ghassemi, Undergraduate Student [1], Trevor Hamilton, Ted Allison
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Traumatic brain injury (TBI) is a leading cause of global mortality and disability; but beyond its immediate impact, TBI emerges as a substantial risk factor for dementia, encompassing conditions like chronic traumatic encephalopathy (CTE) and Alzheimer's disease (AD). This study explores the realm of TBI and its potential cognitive consequences in zebrafish. It discovers whether these defects can be observed with behavioural assays such startle response tests, anxiety tests (novel object approach), and memory tests (y-mazes, and novel object recognition), and can validate larval zebrafish as a reliable model for TBI-induced cognitive decline akin to mice and human patients. The research employs a diverse battery of behavioural assays on both larval and adult zebrafish to assess cognitive functions following TBI. Furthermore, the study explores the impact of post-TBI seizures and investigates whether a brief application of antiepileptic drugs (AEDs), specifically Levetiracetam, can mitigate the observed cognitive decline. The results of this study show a decrease in the level of startle response following TBI and a subsequent rescue of this reduction in movement through the application of Lev. The adult tests also show a reduction in anxiety levels following TBI. These results could potentially reshape therapeutic interventions, not only regarding post-TBI cognitive decline, but also regarding the potential application of antiepileptic drugs in dementias such as Alzheimer's disease and chronic traumatic encephalopathy. Overall, this research not only advances our understanding of TBI-induced cognitive deficits but also holds implications for broader neurodegenerative research and potential therapeutic strategies.

Estimation of Genetic Variability of Digestibility in Pigs

Sonja Allen, PhD Student [1], Hélène Gilbert, Céline Carillier-Jacquin
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Digestive efficiency is an important factor in pig production, influencing nutrient absorption and addressing feed cost challenges while promoting environmental sustainability. However, measuring digestive efficiency is expensive as it requires housing animals in individual digestive cages and heavy chemical analyses. Following an earlier proof-of-concept in the French Large-White breed, this study examined a simplified Near Infrared Spectrometry (NIRS) methodology applicable to fecal samples collected on farms. The aims of the study were to quantify the genetic variability of digestive efficiency in

French Landrace and French Piétrain breeds and to test the relevance of this methodology to capture digestive efficiency differences in two production systems, namely conventional and organic. Fecal digestibility coefficients (DCs) of energy, nitrogen, and organic matter were predicted from samples collected at 21 weeks of age with a total of 758 samples for the breed comparison and 278 samples for the production system comparison. Additionally, measurements of feed intake and efficiency, growth rate, and carcass traits were recorded for the three breeds within each production system. The findings indicate that there exist genetic variability for digestive efficiency traits in the examined breeds, with heritability estimates ranging from 0.17 ± 0.13 to 0.20 ± 0.15 for the Landrace breed and 0.31 ± 0.14 to 0.34 ± 0.14 for the Piétrain breed. Furthermore, differences in digestive efficiency were observed between conventional and organic production systems, with conventional farm animals exhibiting significantly higher DCs for energy, nitrogen, and organic matter (8.07%, 10.13%, and 7.56%, respectively, $p < 0.001$) compared to organic farm animals, due to dietary differences. Implications: The use of this cost-effective and simplified NIRS protocol could ensure accurate ranking of pigs based on their digestive efficiency. This could then serve as a potential solution to mitigate high feed costs and reduce the overall environmental footprint associated with pig production.

Oral Presentations in Microbiology

Bacteriophages May Be the Key to Mitigating Ammonia-Oxidizing Bacteria-Driven Pollution

Aaron Turner, MSc Student [1], Dominic Sauvageau, Lisa Y. Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Ammonia-oxidizing bacteria (AOB) play a pivotal role in nitrification – oxidizing ammonia to nitrite – and contribute to nitrous oxide production. Widely used synthetic fertilizers exacerbate this process, leading to increased nitrous oxide emissions, creating an imbalance in the delicate nitrogen cycle. This causes greenhouse gas emissions, ozone depletion, and nitrate pollution. A proposed solution to this problem is using bacteriophages to target AOB. However, little is known about these phages. This project aims to discover and understand the complex relationship between AOB and phages. One way bacteria persist in the environment is by integration of phages into their genomes (prophages), potentially conferring benefits including biofilm formation, gene transfer, and environmental adaptation. To understand how prophages may interact with their hosts, we first investigated the diversity and characteristics of AOB prophages and their virions. This involved predicting where phages may be hiding within AOB genomes. These viral regions were annotated and their diversity was observed by comparison against known phages. Genomes containing intact prophage were induced by causing DNA damage, signaling to prophages to 'abort ship'. By doing this, we can extract the virions from integrated viruses in host genomes. The next steps are to fundamentally understand how these virions differ from their genomic counterparts, and how they interact with hosts on an evolutionary, functional, and genetic basis. Furthermore, to harvest phages for nitrification control, we must screen for lytic (virulent) phages capable of targeting AOB. With our current findings, and future approaches, we aim to understand how phages may confer benefits to hosts, and alternatively, may be used to combat AOB to halt their pollutive effects.

Characterization of Arac Negative Regulator (Anr) Family-Like Protein in MP1 E. coli Phage-Inducible Chromosomal Island (Pici) ECCIMP1

Caroline MacDonald, Undergraduate Student [1], Valeria Tsviklist, Kat Pick, Tracy Raivio
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Escherichia coli, a gram-negative bacterium prevalent in numerous ecosystems, is known for its ability to colonize the mammalian gastrointestinal (GI) tract. This interaction between bacteria and host may be pathogenic or commensal, depending on the strain and host. Bacteria often have their genes controlled by proteins encoded on mobile genetic elements such as plasmids, prophages, and pathogenicity islands. Among these, Phage-Inducible Chromosomal Islands (PICIs), such as EcCIMP1, have emerged as major contributors to horizontal gene transfer. PICIs are a type of phage satellite that will hijack certain phage elements needed for their lifecycle. They replicate and disseminate throughout their surroundings, integrating into other bacteria. EcCIMP1 was originally found in E. coli strain MP1, and contains an uncharacterized protein known as EcCIMP_025. EcCIMP_025 has been previously shown to repress Locus of Enterocyte Effacement (LEE) pathogenicity expression in Citrobacter rodentium, a murine model for enteric infections. This EcCIMP_025 shows homology with the AraC Negative Regulator (ANR) family of transcriptional regulators. Often associated with virulence regulation, the existence of this unknown protein in a non-pathogenic strain of E. coli poses a unique scenario. What is its purpose if not to regulate virulence? This study selected E. coli MP13 and E. coli MP7, as well as mouse pathogen C.

rodentium DBS100 to investigate the effects of the unknown EcCIMP_025. Deletion mutant strains were constructed, followed by luciferase assays using reporter plasmids to assess expression of genes related to responses to nutrient stress, envelope stress, and DNA damage. Bacterial growth was assayed to monitor growth dynamics between groups. It has been found that the deletion of EcCIMP_025 leads to significant repressor effects on two of the five reporters tested in wildtype C. rodentium DBS100; recA and cpxP. These results provide an exciting segue into future research regarding impacts of EcCIMP_025 on E. coli gene expression.

Elucidating the Molecular Biogenesis of Outer Membrane Vesicles in Methylomicrobium Album BG8

Elizabeth Arenas, PhD Student [1], Dominic Sauvageau
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Methylomicrobium album BG8 is a methanotroph with a large biotechnology potential. One feature that makes this bacterium attractive to study is its unique production of outer membrane vesicles (OMVs) which could carry valuable cargo like isoprenoids. Outer Membrane Vesicles (OMVs) are buds that are produced and released by the spontaneous bledding of the outer membrane of Gram-negative bacteria. OMVs are utilized to carry outside the cell a variety of cargos for different physiological processes such as quorum sensing, horizontal gene transfer, interbacterial killing, toxin delivery and nutrient hydrolysis. Due to its unique properties to export outside the cell different materials, there is a growing attention in the study of OMVs to develop them as a controllable delivery platform of specific cargos. My research will probe how OMVs are regulated using genetics and proteomics of specific secretion pathways. Specifically, the comparison of the genomes of a natural OMV producer and a non-OMV producer to elucidate the molecular biogenesis of these nanostructures. If our hypothesis that M. album BG8 can be used as a delivery platform is valid, we will examine the ability of the OMVs to package and deliver valuable commercial products by increasing the yield of vesicle production by promoting the proteins related to OMV production identified in this research.

A Novel Salmonella-Specific Lipoprotein that Modulates the Bacterial Response to Antimicrobial Peptides

Paris Brown, MSc Student [1], Casey Fowler, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Bacteria deploy many mechanisms to sense changes to their external environment and respond accordingly. One such mechanism is via two-component systems, which are highly prevalent signalling pathways found throughout the bacterial kingdom. They are commonly composed of an environment-sensing kinase that activates a DNA-binding regulatory protein. Within Enterobacteriaceae, PhoPQ is an established member of the two-component protein family. PhoQ responds to low extracellular concentrations of cations, low pH, and cationic antimicrobial peptides (CAMPs) by phosphorylating PhoP. This allows for PhoP to regulate genes involved in virulence, resistance, outer membrane remodelling, and nutrient starvation.
Sty1874 is an uncharacterized gene that is unique to the Salmonella enterica species. We have identified that this gene is a member of the PhoPQ signalling pathway and aids in activating virulence and antimicrobial resistance factors. The involvement of small proteins in two-component system regulation has been a factor of interest for many research groups, in part due to their complicity in bacterial virulence. This project characterizes Sty1874 as a small, putatively inner membrane lipoprotein. We demonstrate that this lipoprotein facilitates PhoPQ activation strongly in response to the presence of CAMPs in vitro. Intriguingly, Sty1874's ability to activate PhoPQ does not translate functionality within the closely related bacterial species Escherichia coli (E. coli), providing insight into the diversification of two-component system regulation in response to varying environmental challenges.
Sty1874 represents an additional modicum of gene regulation that has not yet been characterized in the Enterobacteriaceae family. This project will inform on how the well-studied PhoPQ system is able to respond to a variety of different signals across different species. Ultimately, this research will contribute to the growing body of knowledge in two-component system evolution and regulation.

Macrophage Cell Biosensor System to Evaluate Immunotoxicity of Ospw and Implications on Cell Activation and Signaling

Kendra M. Woodall, MSc Student [1], James L. Stafford
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Alberta's expansive oil sands deposits undergo oil sands extraction and processing which uses substantial amounts of water. This water is referred to as oil sands process-affected water (OSPW) and during oil sands processing, it amasses toxicants like naphthenic acids, which are recognised as a major source of OSPW-mediated toxicity. Thus, OSPW must be treated and remediated before ultimately returning to the environment. Treatments such as advanced oxidation processes (AOPs) degrade naphthenic acids, and these treatments must be evaluated to determine efficacy. Our lab proposes an immune cell-based in vitro biosensor system to detect immunotoxicity in untreated and treated OSPW. Macrophages, a type of innate immune cell, detect environmental stimuli using cell surface receptors and rapidly respond with secreted proinflammatory proteins. OSPW has been found to induce immunotoxic effects on macrophages and AOPs are effective in reducing immunotoxicity. We have found that OSPW induces comparable pro-inflammatory responses to endotoxins such as lipopolysaccharides (LPS) and appear to activate similar intracellular signaling proteins such as IRAK1/4 and NF-κB. Toll-like receptor 4 (TLR4), which is known for detecting LPS, is implicated in OSPW detection by macrophages. As TLRs and TLR-activated signaling proteins are implicated in both OSPW and LPS-mediated signaling, this suggests that the same cell receptor and signaling pathway may potentially play a role in detecting both. The similarities between OSPW and LPS detection and cell signaling enhances our understanding of OSPW immunotoxicity and provides insight on how contaminated waters affect mammalian cells. This research offers a sensitive and reliable method to assess contaminated waters and treatments, and provides further understanding of the immunotoxic effects of OSPW.

Optimizing Methane Bioconversion: Harnessing Methylomicrobium Album BG8 for Sustainable Biotechnological Applications

Fabián G. Rondón, PhD Student [1], Dominic Sauvageau, Lisa Y. Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Amid escalating climate crisis concerns, the scientific community has proposed methanotrophs, renowned for their ability to consume methane, as a possible dual-purpose solution: mitigate methane's environmental impact while converting it into commercially significant bioproducts like biodegradable plastics, biofuels, vital acids, and food supplements. However, the path to industrial scalability is fraught with challenges, notably the methanotrophs' low metabolic activity and the energetically intensive nature of methane biocatalysis. Methylomicrobium album BG8, distinguished by its exceptional metabolic versatility in methane utilization, stands out as a particularly promising candidate for this endeavor. Our project seeks to navigate these obstacles by strategically employing metabolic engineering and co-culture techniques to enhance the cultivation of M. album, thereby unlocking new, more viable avenues for industrial exploitation. In the realm of bioengineering methylotrophs, metabolic engineering emerges as a pivotal approach, offering a detailed comprehension of an organism's metabolic network by harnessing genome-scale metabolic modeling (GSM). These GSMs, rooted in genomic information, provide a detailed mathematical outline of an organism's metabolism, aiding in pinpointing metabolic targets and unraveling physiological complexities, making them cost-effective for diverse bioengineering strategies. This research leverages the GSM model for M. Album BG8, aiming to refine its predictive power through rigorous validation (multiomics) and meticulous debugging. The primary goal is to enhance cultivation metabolic engineering techniques to boost yields and capitalize on M. Album BG8's methane-to-metabolite conversion. By applying Flux Balance Analysis (FBA) and exploring symbiotic co-cultures with photosynthetic or methylotrophic partners, the study addresses issues like methanol inhibition and oxygen transfer limitations, presenting a viable economic approach for methane biocatalysis.

Adapting Clinical Antimicrobial Assessments for Detecting Oil Sands Processed Water Toxicity

Kareem F. Moghrabi, MSc Student [1], Isaac J. Sánchez-Montes, Mohamed Gamal El-Din, James L. Stafford
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Current approaches for petroleum extraction in Northern Alberta oil sands use hot water to separate bitumen from oil sand deposits. These techniques use large quantities of water, leading to the accumulation of 1.44 billion m3 of oil sands process waters (OSPW) as of 2022. In general, OSPW contains several inorganic and organic components that contribute to acute and chronic toxicity. These waters are currently held under a zero-discharge policy, but government and industry have recently initiated plans for treatment and release of OSPW. These treatments will target organic toxicants in OSPW such as

naphthenic acids (NAs) and will require intensive toxicity monitoring to inform water quality status. As these waters are complex mixtures, the tools used to assess their toxicity and treatment efficacy must be equally varied and sensitive. While in-vivo assays provide biologically relevant data for affected organisms, in-vitro approaches allow for rapid, high-throughput screening of water samples to inform treatment decisions and further toxicity testing. Standardized bacteria-based water toxicity screening approaches have been applied for detecting OSPW toxicity, but are susceptible to photometric interference. In clinical settings, broth microdilution minimum inhibitory concentration (MIC) assays are a reliable, standardized method to determine effective concentrations of antimicrobial compounds against indicator organisms. These methods allow for longer exposure times, less sample modification, and direct assessment of cell viability. Through a modified MIC, we demonstrate significant, dose-dependent loss of Staphylococcus warneri viability upon exposure to both NAs and whole OSPW. Additionally, treatment of these samples with advanced oxidation processes reduce NAs and recover bacterial growth. This photometry independent method correlates with chemical analysis of these waters and standardized toxicity assays, serving as a valuable supplementary approach for screening water toxicity.

Mitigating Agricultural Environmental Impact: Evaluating Plant-Derived Biological Nitrification Inhibitors on Methanotrophic Bacteria

Mehnaz Bushra Hamid, MSc Student [1], Lisa Y. Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Agricultural practices relying on ammonia-based fertilizers contribute significantly to environmental issues such as greenhouse gas (GHG) emissions and water eutrophication. In the past 40 years, Human-induced global emissions, primarily driven by nitrogen additions to agricultural lands, have increased by over 30%. Ammonia-oxidizing microbes (AOM) play a crucial role in the rhizosphere as they convert fertilizers into the potent greenhouse gas, nitrous oxide. One potential solution to mitigate this environmental impact is using Biological Nitrification Inhibitors (BNIs), derived from root exudates of plants. BNIs are able to suppress key enzymes in AOM. However, BNIs come with a limitation as they may also inhibit similar enzymes in methane-oxidizing bacteria, which could reduce the biological methane sink, leading to net methane emissions. This study investigates the efficacy and mode of action of BNIs on methane-oxidizing bacteria. We will determine the EC50 of BNIs from commercial sources and wheat root exudates on several methanotroph species and determine which enzymes in methanotrophs are affected using substrate-dependent oxygen consumption assays and proteomics analysis. Our hypothesis is that BNIs fall into two classes: one that selectively inhibits ammonia oxidizers but not methane oxidizers, and one that inhibits both ammonia- and methane oxidizers. This research contributes to a comprehensive understanding of BNIs, ensuring their safety and promoting sustainable agricultural practices by reducing emissions of GHGs and promoting nitrogen use efficiency by crops.

Engineering Methylomicrobium Album BG8 for Gene Analysis

Nina Deschner, Undergraduate Student [1], Brittany Green, Dominic Sauvageau, Marina Lazic, Lisa Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Methanotrophs are obligate aerobic bacteria capable of utilizing methane and/or methanol (C1 compounds) as their sole carbon source. The availability and cost of C1 feedstocks makes methanotrophs an interesting area for research from a scientific and industrial perspective. Regardless of obligate aerobic requirements, Methylomonas denitrificans FJG1 is the exception that can thrive under oxygen depletion. A homologue of eukaryotic hemerythrin, called Bacteriohemerythrin (bhr), is a protein associated with oxygen binding. A methanotroph-specific Bacteriohemerythrin gene “bhr-00” has been observed to be upregulated under hypoxia in M. denitrificans FJG1. We hypothesize that the bhr protein in this bacterium enables it to survive in hypoxic ecosystems. To validate the hypothesized function of the bhr gene, we sought to express it in the related gammaproteobacteria methanotroph Methylomicrobium album BG8 that does not contain bhr. To achieve this, we obtained a synthesized version of the bhr-00 gene and utilized Gibson Assembly to clone the gene into a broad-host-range IncP plasmid. Once successful cloning was confirmed, the plasmid was transformed into the E. coli S17-1 λpir conjugative donor strain. To ensure proper transfer of the bhr-containing vector into M. album BG8, conjugation-mediated plasmid transfer between donor and recipient was performed. Once successful plasmid uptake by M. album BG8 is confirmed, the strain will be cultured in hypoxic conditions with methane as a carbon source. Growth, as well as methane and oxygen consumption will be analyzed via GC-TCD. If the hypothesized function of bhr in methanotrophs as an oxygen scavenger is correct, M. album BG8 will be able to grow in extreme hypoxia, expressing the characteristic pink colour of the protein in the cells. From an industrial perspective, this study presents an opportunity to overcome a major limitation in industrial fermentation of

methanotrophic organisms due to microaerophilic fermenter conditions. Furthermore, this study provides applications for the genetic engineering of non-model microorganisms.

Adaptive Evolution of Methylomicrobium Album BG8 to Low pH Conditions

Rachael Rieberger, Undergraduate Student [1], Marina Lazic, Dominic Sauvageau, Lisa Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Within the context of escalating impacts of methane emissions, innovative microbial solutions are imperative for mitigation efforts. Methanotrophs, a diverse cohort of microorganisms capable of utilizing methane as their sole carbon source, offer potential solutions. While consuming methane, methanotrophs synthesize certain metabolic products that can be considered value-added compounds. However, industrial scale-up for these compounds can be troublesome, with temperature fluctuations, pH variation and synthesis of inhibitors impeding methanotrophic growth and production. Addressing these challenges, Methylomicrobium album BG8 emerges as a compelling candidate for industrialization due to its rapid growth rate, high growth yields, and adaptability under various nutrient conditions. Consequently, this project endeavors to address one of the key scale-up issues (acidification) by employing adaptive evolution strategies in M. album BG8. To achieve this goal, we aim to cultivate a strain adapted to low pH conditions developed via serial passages starting from pH 6.6, two units' lower than parental, with the goal of reaching the lowest pH still capable of growth, around pH 4.0 – 4.5. Adaptive evolution, a targeted trait selection method, is used to produce advantageous alterations in rapidly reproducing microbes. Sequential passage of M. album BG8 at progressively lower pH levels is expected to yield mutations that provide survival advantages to the strain. Observations reveal that as the pH of the medium decreases, M. album BG8 exhibits a tendency to alkalize its surroundings, potentially through the secretion of an unknown metabolite. To determine the strains adaptation mechanisms, the parental strain and select isolates will be subjected to the whole-genome sequencing. Comparative analysis between isolates and the parental strain will reveal specific mutations and provide valuable insight into potential gene functions. This study has potential to address crucial hurdles for industrialization of methanotrophs and offers a sustainable solution to help mitigate methane emissions.

DLP6 is a Stenotrophomonas maltophilia Phage that Uses Cira as its Receptor

Umut Oktay, MSc Student [1], Jonathan Dennis
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

With the increase in antibiotic resistance, new therapeutics in the fight against bacterial pathogens are desperately needed. One alternative therapy uses bacteriophages (phages) as a means of overcoming bacterial infections. Stenotrophomonas maltophilia is an opportunistic Gram-negative pathogen that threatens immunocompromised individuals and those with cystic fibrosis. The drug-resistance characteristics of S. maltophilia make this bacterium an excellent candidate for the application of phage therapy. However, one limitation is that most of the bacteriophages in our library against S. maltophilia use type IV pilus as their receptor for infection. This limitation may be combated by a novel approach that involves creating phage cocktails (combinations of phages) to improve the effectiveness of combating infections and resistance to singular phages, which can emerge easily in bacteria. Nevertheless, the potential for combining these phages is limited, as even minor mutations in bacteria could confer resistance to both phages. DLP6, on the other hand, is a broad host range phage that can infect pilus-defective S. maltophilia. Our preliminary results displayed that DLP6 might use a CirA orthologue which is a TonB-dependent iron transporter. To confirm the DLP6 receptor, we created a clean cirA knockout strain that showed that DLP6 requires CirA for infection since the clean deletion of cirA abolishes phage infection. Our current work is exploring phenotypic differences between DLP6-resistant S. maltophilia isolates and wild-type strains to investigate if resistance to DLP6 has any negative consequences for these bacteria. We are also exploring DLP6 infection profiles in various situations including low iron conditions as well as the combination of DLP6 with our pilus-binding phages for investigating phage-phage synergy against S.maltophilia. The outcomes of our study will demonstrate the therapeutic efficacy of DLP6, whether used individually or in combination with other phages, in addressing S.maltophilia infections.

Oral Presentations in Physiology and Development

Sodium Uptake Recovery During Low-pH Exposure in Zebrafish

Anthony Kovac, MSc Student [1], Greg Goss
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Sodium homeostasis in fish gill cells is integral for maintaining proper cellular and bodily function. However, there are several confounding factors that make maintaining sodium homeostasis difficult in certain environments. The low sodium concentration in freshwater in tandem with low pH in some environments severely hinders the uptake of sodium through electroneutral/passive transporters. However, many fish species live in low pH/low sodium freshwater which has caused debate in the literature about what ion transporters act in these fishes in these environments. Our study focuses on a novel model of sodium uptake in zebrafish (Danio rerio) that uses a yet-to-be-determined potassium-driven sodium transporter that circumvents these thermodynamic constraints. This potassium-driven sodium transporter has been shown to allow sodium uptake to recover in zebrafish after long exposure to low-pH environments. Using radiotracer-based ion flux technologies, we measured sodium uptake in zebrafish after either 2 h or 8 h of exposure to pH 4.0. I will use differential gene expression analyses to identify possible candidate genes that act to restore sodium transport capacity. Preliminary findings show reductions and recoveries of sodium uptake in zebrafish in response to low pH and sensitivity to mesylate (KB-R7943), a known inhibitor of sodium-calcium exchangers. The differential gene expression analysis remains to be elucidated.

Exploring Larval Carcinus Maenas' Potential as a Marine Toxicological Model

Dustin Doty, MSc Student [1], Tamzin Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The effects environmental pollution has on ecosystems are assessed through a battery of standardized toxicity experiments on model organisms which inform policy and set environmental limits for chemical contamination. Many of these tests use models from freshwater environments that may not be reflective of the habitat in question but have quick developmental times and are easy to culture (i.e. zebrafish and Daphnia). Even fewer model organisms exist in the marine environment, leaving an opening for understanding marine toxicology and the potential effects of environmental chemical release on the development of seawater organisms. C. maenas is an invasive crustacean found throughout all inhabited continents which have a sensitive planktonic larval stage; thus, they are globally applicable and can be used for developmental assays. This investigation seeks to delineate the feasibility of using larval C. maenas as toxicology models utilizing two metal contaminates, copper and cadmium. They were chosen as they are representative essential and non-essential metals respectively, their toxicity is extensively described, and are both prevalent throughout the marine environment. Larvae were exposed to a concentration series of a given metal to determine the median concentration that results in the death of 50% of the test population (LC50). The 48-hour LC50 of copper was 261 µg/L (95 % CI: 209 – 344 µg/L), and 653 µg/L (95% CI: 520 – 849 µg/L) for cadmium. Sublethal immobilization effects were also monitored resulting in 48-hour EC50 values of 123 µg/L (95% CI: 105 – 146 µg/L) and 414 µg/L (95% CI: 348 - 501 µg/L) for copper and cadmium respectively. These concentrations encroach on environmental relevance supporting their usefulness as a model which is furthered by their relative ease to culture. Establishing this model will provide consistency and replicability across laboratories, provide valuable data for regulators, and aid in the management efforts of this invasive species.

Investigating the Potential Toxicity of Copper Nanoparticles from Agricultural Run-off on Daphnia magna

Fleur Issac, MSc Student [1], K Phillips, A Poulin, Jeffrey M. Farner, Tamzin A. Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Agricultural pesticides can potentially leach into the environment, causing detrimental effects on surrounding aquatic biota. Nanopesticides (e.g., copper nanoparticles (CuNPs)) are cost-effective, have improved dispersibility, and provide a greater surface area for application ensuring a more targeted pesticide delivery. However, nanoparticles (NPs) tend to aggregate and fall out of suspension; they can dissolve over time, raising concerns about the long-term effects on aquatic organisms and environmental deposition. While useful for reducing pests, nanoformulations exhibit novel properties with unknown effects compared to conventional counterparts. We aimed to compare and contrast the established impacts of acute and chronic toxicity of dissolved copper (Cu) to that of CuNPs in agricultural pesticides, as its toxicity is unclear. The 48-hour lethal median concentration (LC50) of CuNPs was tested using the freshwater crustacean Daphnia magna, as they represent a critical species in many food webs. The observed acute LC50 concentration for ionic Cu was 38.5 µg/L (95% CI 32.48-

46.68), while only partial mortality was observed during acute CuNP exposures, suggesting CuNPs are less acutely toxic than ionic Cu. However, the toxicity effects of CuNPs change as the suspensions age. LC50's on aged (~2 weeks) CuNPs showed no clear pattern of an increase or decrease in toxicity (i.e., Day 3 LC50 = 248.0 mg/L (95% CI 65.9 - -105 mg/L) vs Day 15 LC50 = 65.8 mg/L (95% CI 54.4 - 96 mg/L). Further experiments need to determine the potential risk of aged CuNPs. Understanding the toxicity of nanopesticides used in agriculture will help us understand the potential risks associated with NPs and allow for better regulatory limits on pesticides, protecting sensitive species. This project is funded by the Alberta Conservation Association.

The Effect of Movement on the Development of the Peripheral Nervous System in Zebrafish (Danio rerio)

Gabriel Dillenburg, PhD Student [1], Zach Hall, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

During development, the nervous systems grow and starts to gather sensory inputs that can stimulate neurogenesis. For example, the sensory feedback generated by movement has been shown to stimulate zebrafish larvae brain development, with larvae that swim more generating more new neurons in the forebrain than those swimming less. Dorsal root ganglia (DRG) are a type of peripheral sensory cells in the zebrafish larvae body that detect movement during swimming and convey the sensory inputs to the brain. Cell populations of DRG grow during postembryonic development through neurogenesis like the brain, so it's possible that movement directly impacts the generation of DRG. I hypothesize that movement during postembryonic development affects the production of DRG. To test this, I used three different approaches: (1) I raised fish under restraint using smaller wells that reduce larval swimming; (2) I raised the fish in 6% methylcellulose, a thick media that reduces swimming; and (3) I used chrna1 fish, mutants that can't move due to the lack of synaptic transmission at the neuromuscular junction. For all approaches, I used transgenic zebrafish (Isl2b:gfp) that produce a green fluorescent protein in their DRG allowing for in vivo visualization and resampling over development to count DRG cells. All treatments reduced larval swimming compared to controls and consequently reduced the number of DRG generated. Therefore, I conclude that the amount of postembryonic movement that a zebrafish larvae experience affects DRG growth.

Dietary Trimethylamine N-Oxide Supplementation Improves Insulin Sensitivity in High-Fat Diet Fed Mice

Hailey Van Allen, MSc Student [1], Bergen Vetsch, Boshra Mandour, Bryan Lum, Kelly-Ann Leonard, Randal Nelson, Gia V. Shelp, Clara E. Cho, Jessica Yue, René Jacobs
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Trimethylamine N-oxide (TMAO), a choline-derived metabolite, is a product of gut microbiome metabolism that is associated with conflicting positive and negative impacts on glucose metabolism and insulin resistance. The objective our study is to determine whether dietary TMAO supplementation affects glucose metabolism in high-fat diet (HFD) fed C57BL/6 mice. 11-week-old male and female C57BL/6 mice were fed a HFD (42% fat) with or without 0.2% TMAO supplementation (TS) for 8 weeks. At weeks 5 and 6, select mice were housed in metabolic cages for 48 hours, and at week 8, select mice underwent a glucose tolerance test (GTT) or a euglycemic hyperinsulinemic clamp (EGHIC). Plasma, liver, skeletal muscle, and white adipose tissue were collected following a 16-hr fast. Plasma TMAO levels were increased in TS male and female mice. Weight gain was reduced in TS female mice, likely due to increased whole-body oxygen consumption. Neither body weight or energy expenditure were altered in males. There were no significant differences in fasting hepatic or plasma lipid levels. Both male and female TS mice exhibited improved glucose tolerance. EGHIC indicated that TS increased whole-body insulin sensitivity; interestingly, male mice displayed reduced insulin-stimulated glucose production, while TS female mice had increased glucose utilization in peripheral tissues. Dietary TMAO supplementation may be beneficial for attenuating weight gain (females) and improving glucose tolerance and insulin sensitivity (both sexes) in mice fed a HFD. Further research is warranted to determine the mechanism by which TMAO impacts adiposity, glucose metabolism, and insulin sensitivity HFD-fed mice.

Assessing the Multigenerational Effects of Daphnia Magna to Naphthenic Acids Sourced From Oil Sands Process-Affected Water

Mathias Segura Castillo, Undergraduate Student [1], Jenelle McCuaig, Tamzin Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The continuous increase of oil sands production, particularly in Northern Alberta, has also led to an increase in their hazardous by-products. Oil sands process-affected water (OSPW) is a common by-product of the bitumen extraction process, which is required to be stored in artificial reservoirs referred to as tailings ponds. Any leakage or seepage of OSPW from these tailings ponds has the potential to contaminate nearby freshwater systems. While OSPW contains various toxic components, naphthenic acids (NA) are generally considered the primary source of OSPW toxicity. Due to their slow biodegradation in aquatic ecosystems, NAs are capable of persisting in these ecosystems and affecting a wide variety of biota across multiple generations. Although research efforts have shifted towards studying NAs, the majority of this research has revolved around the acute effects of NAs, whereas the chronic effects remain understudied. Taking this into account, 4 generations of Daphnia magna, a prevalent bioindicator species, will undergo continuous 21-day exposures to the following NA concentrations: 0 (control), 10, 20, or 40 mg/L. Various growth and reproduction endpoints will be used to determine the chronic effects of NAs on D. magna as well as their potential to recover from NA toxicity across generations. Understanding the chronic toxicity of naphthenic acids is crucial for informing policy decisions regarding oil sands production and the management of OSPW-contaminated sites.

The Effects of Muscle Dysfunction on Developmental Brain Cell Proliferation in Larval Zebrafish

Micaiah Achtymichuk, MSc Student [1], Zachary J. Hall, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Development is neither predetermined nor a unidirectional flow of information. Development is a multifaceted process, where the interplay of genetics, environment, and additional factors, such as organismal behavior, intertwine to achieve the genesis of an organism. Unlike most developmental studies that focus on environment or genetics solely or in combination, the basis of this experiment is the effect of genetics on behavior, and behavior on development. This project elaborates on previous findings which demonstrated that the development of the larval zebrafish brain is profoundly influenced by organismal behavior, finding that forebrain neurogenesis (the creation/integration of new brain cells) is highly impacted by the magnitude of the swimming behavior of a zebrafish larva (Hall & Tropepe., 2018). Muscle disorders (myopathies) are conditions that ultimately affect the ability of an animal to move, including early in development. Accordingly, I hypothesize that myopathies may also mediate developmental neurogenesis via swimming behavior. Thus, my work aims to discover the interplay between muscle dysfunction, swimming behavior, and developmental neurogenesis early in the life and genesis of zebrafish. I will achieve this through targeted mutagenesis of some genes implicated in myopathies (dmd, neb, ryr1b) to interfere with the function of the target genes, and thus the zebrafish muscular system. The resulting mutant larvae with impaired muscle function will be assessed for behavioral differences and for any associated neurogenic changes compared to non-mutant larvae throughout the embryonic and the start of post-embryonic stages of development. My study will show if developmental movement, something common to all vertebrates, is a vital factor in brain development.

Out of Body Heart Preservation: Is More Blood Better?

Parham Hassanzadeh, Undergraduate Student [1], Mitchell Wagner, Guilherme Mainardi, Darren Freed
[1] Department of Physiology, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Heart failure is a leading contributor to mortality worldwide, claiming more lives than any other condition. While heart transplantation is the gold standard for treatment, transplant volume is currently limited by conventional static cold storage (SCS), which limits out of body preservation of the heart to a six hour window. This limits transplant volumes by preventing long distance transport. Ex-situ heart preservation (ESHP) is a technology that extends the preservation of the organ by continuously providing oxygen and essential nutrients. While ESHP demonstrates superior preservation of heart functionality compared to SCS, function still declines throughout the procedure. Hemoglobin (Hb) is a key oxygen delivery molecule in the blood, which composes the perfusion solution for ESHP. A widely accepted notion of the field of ESHP is that more blood is better as it increases oxygen delivery, however it is also possible that more blood can increase the delivery of free heme, a toxic byproduct, when red blood cells break (hemolysis). Therefore we tested the hypothesis that increasing Hb levels could lead to decreased functional preservation by increasing hemolysis. The hearts of juvenile Yorkshire pigs were perfused on an ESHP apparatus for 11 hours. Four groups were formed varying in levels of hemoglobin. Cardiac function

and ion concentrations were assessed bi-hourly throughout the run. Hearts perfused with high hemoglobin (more blood) demonstrated worse preservation of several cardiac function parameters over the 11 hour perfusion when compared with the other groups. In addition, high Hb groups demonstrated a significant increase in potassium levels (a marker of hemolysis) versus the other groups. These findings demonstrate that hemolysis is detrimental to the preservation of the heart. By understanding the effects of Hb, hearts procured for transplant can be preserved with enhanced longevity, which will aid in expanding the pool of hearts deemed suitable for transplantation.

Reasons for My Back Pain in the Summer of 2023: Understanding the Differences in Lab Reared & Wild Caught Daphnia pulex Responses to Ultraviolet Filter Exposure

Sidney Martin, Undergraduate Student [1], Aaron Boyd, Connor B. Stewart, Jenelle D. McCuaig, Alexandra R. Legge, Dustin Doty, Tamzin Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Lab-reared organisms are important for researchers to understand aspects of our natural environment, under controlled conditions. These findings contribute to policy development, risk assessment frameworks, and discovery-based knowledge on environmental systems. However, an important question for those in environmental research is whether lab reared organisms are a representative model for their wild counterparts? Using a comparative approach, both lab-reared and wild-caught Daphnia pulex were raised and housed in their respective waters (lab OECD or wild lake water) and exposed to ultraviolet filters (UVFs) to understand the effect of life-history on environmental exposures. Daphnids were exposed to avobenzone (AVO), oxybenzone (OXY), and octocrylene (OCT), in acute and chronic experiments. EC50's & LC50's assessed acute responses, where across several concentrations a median concentration that produced an effect in 50% of daphnids was found. To observe the differences between the daphnids (lake and lab) after chronic exposures to UVFs, a 21 day exposure was implemented. In this exposure, daphnids were treated with one of three UVF concentrations (nominally, 10 µg/L, 50 µg/L, and 100 µg/L) of either AVO, OXY, or OCT. Immobilization (EC50) and mortality (LC50) were measured after acute UVF exposures while reproductive endpoints, body length, and metabolic rate were measured after chronic exposures. Lethality after acute exposures appear relatively unchanged between the daphnids (i.e. Lab Daphnia, OCT LC50: 2.67 mg/L (CI 2.13-3.55 mg/L), Lake Daphnia, OCT LC50: 4.65 mg/L (CI 3.09-7.40 mg/L)). The effects of chronic exposure included a 53% reduction of total reproduction of the wild daphnids compared to lab daphnids after exposure to the high concentration of OCT. Our experiment highlights a clear difference in the ability of lab and wild organisms to handle a toxicant exposure, calling attention to the need for more environmentally relevant experimental methods to provide greater insight into what happens in natural environments.

Does Sociality Influence Toxicology?

Sienna L. Overduin, PhD Student [1], Kayla Lottin, Sidney Martin, Tamzin Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Many animals live in groups due to greater foraging opportunities, reduced likelihood of predation, and social buffering (i.e., reduced stress concurrent with exposure to members of the same species). Fluctuating environmental parameters due to climate change and increases in anthropogenic outflows are damaging aquatic ecosystems, causing additional stress on fish populations and their social structures. Current risk assessment practices for environmental protection rely on standardized social groupings regardless of species, ignoring natural species assemblage. To understand the importance of sociality in the sensitivity of fish to contaminants, we performed a 96-hour copper (Cu) median lethal concentration (LC50) assessment in marine threespine stickleback (Gasterosteus aculeatus) whilst manipulating the social context. Specifically, we sought to understand whether the social stress of isolation would increase the fish's ventilation rate leading to greater brachial accumulation of Cu, resulting in a larger proportion of isolated fish experiencing lethality than fish exposed in groups to the same concentration of Cu. Wild caught stickleback were exposed to one of eight Cu concentrations (nominally; 0 (control), 50, 100, 200, 400, 600, 1000, 1500 µg/L of Cu) for 96-hours in either isolation (1 fish), pairs (2 fish), or groups (6 fish). Further assessments included tissue-specific Cu distribution and burden in gill, intestine, liver, and whole-body samples, and gill lamellae samples for histological analysis to determine if any differences occurred at the sub-lethal level. Cu's 96-hour LC50 did not change across the different social contexts tested with values (and 95% confidence intervals) of 560 [454, 639] µg/L, 596 [491, 751] µg/L, and 537 [453, 605] µg/L calculated for isolated, paired, and grouped fish, respectively. The importance of understanding whether fish sociality can alter the response to a given toxicant is rarely if ever investigated. This assessment will help to inform the importance of sociality in the field of toxicology.

Oral Presentations in Immunology and Infection

Examination of Zebrafish (Danio rerio) Leukocyte Immune-Type Receptor-Mediated Crosstalk Regulation of Phagocytosis

Najia Ul Huda, MSc Student [1], James L. Stafford
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Immunoregulatory receptors play a crucial role in coordinating immune responses against microbial invaders by modulating intracellular signaling events. Leukocyte immune-type receptors (LITRs) are a diverse group of proteins found in fish that share similarities with the mammalian immunoglobulin superfamily (IgSF) members. Traditionally, immune-type receptors have been categorized as either stimulatory or inhibitory based on the presence of specific motifs within their CYT regions, known as immunoreceptor tyrosine-based activation motifs (ITAMs) or immunoreceptor tyrosine-based inhibitory motifs (ITIMs), respectively. Overall, LITRs regulate innate immune functions through their cytoplasmic tail (CYT) regions, inducing various intracellular signaling networks. When multiple immunoregulatory receptors interact with common ligands and recruit signaling molecules, it fine-tunes the overall response and this phenomenon is known as receptor crosstalk. Previous research in our lab involving the catfish stimulatory receptor IpLITR 2.6b and putative inhibitory receptor IpLITR 1.1b has shown that IpLITR 1.1b reduces the phagocytic capacity of IpLITR 2.6b. This reduction occurs through distinct cytoplasmic tail regions coordinating different aspects of inhibition. The focus of my research project is to examine receptor crosstalk between two unique zebrafish LITR-types using a flow cytometric-based phagocytic assay approach developed in our lab. I have developed a stably co-expressing cell line in AD-293 HEK cells through successful cellular transfection. This cell line expresses a putative stimulatory construct, the N-terminal FLAG tagged DrLITR 1.2wt which consists of an ITAM and ITIM within the same CYT, while the CYT of the N-terminal HA-tagged DrLITR 15.1wt contains two ITIM motifs and an immunoreceptor tyrosine-based switch motif (ITSM). Ongoing work is focused on optimizing conditions for co-engagement of αHA mAb and αFLAG mAb with co-opsonized beads to examine phagocytosis by conducting meticulous comparisons of monoclonal antibody concentration on opsonized bead targets and bead-cell incubation times. This research underscores the significance of employing imaging flow cytometry as a valuable platform for exploring the dynamic signaling potential of DrLITR-types in regulating immune cell effector responses.

Examination of the Immunomodulatory Properties of Oil Sands Process-Affected Water and Commercial Naphthenic Acids Using Macrophage Cells

Sunanda Paul, PhD Student [1], Isaac Sanchez-Montes, Hadi Mokarizadeh, Mohamed Gamal El-Din, James L. Stafford
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Alberta oil sands bitumen mining generates large volumes of oil sands process-affected water (OSPW), which contains variable quantities of organic and inorganic components exhibiting acute and chronic toxicity. Previous studies showed that OSPW organic components are predominantly responsible for causing toxicity. Recent studies have used advanced oxidation processes (AOPs) to treat OSPW, targeting toxic organic compounds like naphthenic acids (NAs). However, there is no established biological system to assess the OSPW treatment samples. Recently, we have developed immune cell-based bioassays to examine the exposure effects of OSPW before and after treatment. In the present study, we investigated the immunomodulatory effects of OSPW samples from different industrial sources which were treated by AOPs. For this study, human macrophage-like cells (THP-1) were exposed to untreated and treated OSPW samples, and cytokine multiplex assays were performed to measure the cytokine secretion levels following 24hr exposure. We also performed synchronous fluorescence spectroscopy (SFS) analysis to measure the organic compounds present in OSPW samples before and after treatment procedures. As NAs are reported to be the most potent organic toxicant present in OSPW, we have also examined the effects of commercial NAs on macrophage cells. Our results show that untreated OSPW has higher secretion levels of pro-inflammatory cytokines which is significantly different than the AOP-treatment samples. Our results also indicate that OSPW samples from different sources have distinct inflammatory properties that correlate with the organic components (e.g. NAs) present in individual OSPW. However, the reduced inflammatory properties of the AOP-treated samples suggest that the removal of specific organic components (i.e. NAs) is responsible for abrogating the inflammatory activity in macrophage cells. Moreover, commercial NAs induced pro-inflammatory cytokine secretion, confirming their inflammatory nature. Overall, our study indicates that macrophage cells can serve as sensitive bioindicators for examining OSPW inflammatory components and remediation strategy evaluation.

Oral Presentations in Health Sciences

Studies on a Novel Cross-Protective Lipopeptide-Based Intranasal Vaccine for Sars-Cov-2 Infections

Adrien Lam, Undergraduate Student [1], Mohammad R. Vakili, Afsaneh Lavasanifar, Raj S. Patel, Babita Agrawal
[1] Department of Surgery, Faculty of Medicine and Dentistry, University of Alberta, Edmonton, Alberta, Canada

The SARS-CoV-2 virus, responsible for the COVID-19 pandemic, has infected over 750 million individuals globally. Despite control measures, emerging new variants over the past two years threaten the efficacy of existing vaccines, highlighting the need for a cross-protective vaccine capable of combating these varied strains. Lipopeptides (LP), derived from the highly conserved epitopes from Spike (S) protein of SARS-CoV-2, demonstrated the induction of protective B-cell and T-cell responses upon intranasal immunization in mice. The incorporation of these LPs in nanoparticle (NP) carriers based on poly-(lactic-co-glycolic acid) (PLGA) can enhance vaccine stability, its ability to effectively cross the epithelial barrier, and immunogenicity to facilitate a robust immune response. Thus, the objective was to develop a NP formulation of LP- based epitopes (LP1 and LP2) from highly conserved regions of the S antigen of SARS-CoV-2 as a broadly protective intranasal COVID-19 vaccine. We hypothesize that incorporating LP1 and LP2 into PLGA NPs will enhance their immunogenicity, leading to long-lasting robust mucosal immune responses and an efficient mucosal vaccine candidate. Experiments with varying PLGA concentrations will be used to maximize LP loading in NPs. The size and aggregation of the NPs will be determined by Zetasizer software while LP loading will be examined through Liquid Chromatography/Mass Spectrometry. Initial formulation attempts resulted in large aggregated NPs. Several experiments were conducted to modify the protocol, resulting in NPs with a Z-average (size) of 6288.0 (SD = 6.1) d.nm, and a volume distribution of 94.4 (SD = 11.3). As the Z-average is still high, further modification of the protocol and loading efficiency of LPs into NPs remains to be determined. Future studies will evaluate the NP-based vaccine efficacy using animal models. Subsequent studies will encompass the inclusion of other LPs derived from the M and N proteins of SARS-CoV-2.

Detection and Treatment of Legionella Pneumophila in Public Building Water

Brittyne Hlavay, MSc Student [1], Norman Neumann, Mathew Diggle, Patrick Hanington
School of Public Health, University of Alberta, Edmonton, Alberta, Canada

Legionella pneumophila is an opportunistic water-borne pathogen that causes respiratory illnesses in humans, known as Pontiac fever and the more severe Legionnaire's disease. This bacterium inhabits a variety of aquatic environments, including both natural and man-made (i.e., premise plumbing in buildings). Transmission occurs through the aerosolization and subsequent inhalation of water droplets, which makes cooling towers, hot tubs, and decorative fountains common sources of L. pneumophila outbreak. This research aims to validate methods of Legionella detection, enhance surveillance efforts, update existing water treatment guidelines, and inform public health responses to mitigate future outbreaks. To better understand the growth of Legionella in premise plumbing we have collected water from the bathroom taps and drinking fountains of 10 public buildings. Through qPCR, Legionella spp. was detected in 7 of these buildings, and the pathogenic L. pneumophila was also detected in one of these buildings using a species-specific qPCR assay. Viable and culturable L. pneumophila was consistently recovered from this same building over a 4-month period. Consequently, hot water (60oC) was used to sanitize the building plumbing systems, and bacterial levels were monitored by both molecular and culture-based methods before and after. This heat-treatment was shown to be successful at reducing culturable L. pneumophila, however, L. pneumophila DNA remained detectable in most sites sampled, suggesting persistence of L. pneumophila in the plumbing. While qPCR analysis of Legionella was shown to be an efficient method to detect the presence of the bacteria in building water, the standard culture methods used by most jurisdictions are essential for evaluating risk to human health. Future work will focus on a comparative genomics assessment of these environmentally - derived isolates with clinical isolates from patients with Legionnaire's disease, and in order to further assess the public health risk associated with L. pneumophila in this building.

Investigating the Innervation and Muscularization of the Mammalian Diaphragm in a Mouse Model of Diaphragmatic Hernia

Jaida F.P. Albrechtsen, Undergraduate Student [1], Nicole H.M. Applin, Juan F. Garcia Rivas, Robin D. Clugston
Faculty of Kinesiology, Sport, and Recreation, University of Alberta, Edmonton, Alberta, Canada

Introduction: Congenital Diaphragmatic Hernia (CDH) is a significant congenital anomaly that occurs in 1/3000 live births and is marked by the incomplete development of the diaphragm leading to abdominal herniation and pulmonary hypoplasia. The cause of CDH has not been determined, although some studies suggest that irregular retinoid signaling during the early stages of diaphragm development plays a role in the etiology of CDH and maternal retinoic acid (RA) supplementation can at least partially rescue CDH. Diaphragm organogenesis is a highly coordinated process in which the phrenic nerve develops with the mesenchyme of the primordial diaphragm in parallel with muscularization. To understand these processes in the context of CDH, we aim to visualize the phrenic nerve and musculature of the diaphragm. Methods: Transgenic C57BL/6 mice, which carry the dominant negative RA receptor (Rardn) were crossed with Prrx1-Cre mice, resulting in double transgenic fetuses and littermate controls. Three experimental groups will be used for immunostaining: control, CDH, and RA rescued diaphragms. Fetuses will be collected at E16.5, dissected via light microscopy and whole mount immunostaining will be used to visualize the phrenic nerve and diaphragm muscularization. Results: In preliminary studies, we have visualized 3 control diaphragms using whole mount immunostaining for the phrenic nerve and are in the process of staining for diaphragm muscularization. We expect to see a smaller phrenic nerve ipsilateral to the CDH defect due to atrophy of the nerve. We plan to visualize muscularization as well as phrenic nerve preservation due to RA rescue of CDH. Conclusion: This study will enable us to visualize the impact of RA signaling and supplementation on the diaphragm muscle and phrenic nerve of our double transgenic mouse model, providing novel insight to the innervation and muscularization of the Prrx1-Cre:Rardn fetal diaphragms.

Poster Presentations in Ecology and Evolution

Leave It, Burn It, or Cut It Down? Best Management Practices for Birds in Post-Mountain Pine Beetle-Attacked Forests

Emily Swerdfager, MSc Student [1], Erin Bayne
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The Canadian Rocky Mountains and Foothills of Alberta are exhibiting increasingly frequent and severe forest disturbances, as a result of climate change, including an unprecedented mountain pine beetle (MPB) hyperepidemic, subsequent wildfires, and salvage-logging practices. These disturbances alter the vegetation structure and composition of the forest, which presumably results in changes to avian communities. The aim of this research is to evaluate how bird communities vary between standing MPB-attacked forests, MPB-attacked forests that have burned, and salvage-logged MPB-attacked forests. Our goal is to provide guidance on best management practices for birds in post-MPB-attacked forests by evaluating how community metrics like species richness, community composition and functional diversity change over time. Using a time-for-space substitution, we have deployed 160 Autonomous Recording Units in targeted areas in Alberta in 2023 and we will return to the field in 2024 to balance out sample sizes across treatments, in order to measure changes in avian communities over time.

Geographic Song Variation in Passerines with Changes in Seasonal Photoperiods

Emma Cowley, Undergraduate Student [1], Lionel Leston
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The vocal behaviour of birds serves a number of important purposes in avian sexual selection, communication, and gives insight into avian phenology. Birds are known to alter their singing behaviour depending on environmental factors and social circumstances, depending on the purpose of their singing. Birds have an endogenous circannual clock that alters various life history traits to maximize their fitness with the changing seasons. Photoperiods serve to calibrate circannual clocks and can vary as a function of season and latitude. How photoperiods may affect bird singing activity as latitude increases is poorly understood, thus, our project decided to test just that. The goal of the study is to use acoustic data to conduct site-specific surveys of Tennessee Warbler and White-Throated Sparrow, to improve our understanding of how increasing latitudes affects bird singing behaviour with the variance of seasonal daylight. With the selection of six sites (3 northern and 3 southern), we listened to 1-minute segments recorded in intervals of 40-minutes over a 24-hour period spanning an average of 5 days.

Whenever a song from either of the target species was detected, it was tagged. The findings of this study will give insight into the function of bird singing and potential drivers in the process of speciation. The research may also serve to improve the understanding and implications of using acoustic recording units to inform projects on ecological monitoring.

Variation in the Relative Size of Nucleus Basalis in Songbirds Related to Seed-Husking Behaviour

Julia A. Bowen, Undergraduate Student [1], Cristián Gutiérrez-Ibáñez, Douglas R. Wylie
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Seed-husking, the ability to remove the husk of hard seeds, is an essential feeding mechanism of birds that has evolved independently multiple times within the superfamily Passeroidaea. In this group of birds, seed-husking is largely accomplished by precise manipulation of the seeds with the beak and tongue. Precise manipulation of object requires increased sensory feedback and neural processing. Previous research has shown that some seed-husking birds, like finches of the family Fringillidae and Estrildidae, have a large amount of touch receptors in the orofacial region, similar to the increased amount of touch receptors in the tip of the finger of mammals that use their hands to manipulate objects. In birds, somatosensation to the orofacial region is conveyed by several cranial nerves to the principal trigeminal nucleus (PrV) in the brainstem which then sends direct projections to the nucleus basalis (NB) in the forebrain, a pathway analogous to the somatosensory cortex of mammals. Given the increase in manipulation with the beak and tongue, and amount of touch receptors in some seed-husking birds, it is possible that NB is enlarged in the groups that have developed seed-husking. In this study we measure the volume of NB in songbirds, including several different families of seed-husking birds within the family Passeroidea. Our preliminary results suggest that NB is enlarged in seed-husking species.

Effects of Winter Severity on Hunter Success of White-Tailed and Mule Deer in Alberta

Kathryn Vaughan, MSc Student [1], Mark Boyce
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

White-tailed deer (Odocoileus virginianus) and mule deer (O. hemionus) play crucial roles in Alberta's ecosystems and economy, impacting vegetation, predator-prey dynamics, hunting practices, and overall biodiversity. However, no studies have addressed the combined effects of winter severity and hunting leading to unpredictable deer harvest fluctuations across Alberta. We aim to fill this knowledge gap by comprehensively documenting factors influencing deer harvest fluctuations, especially during periods of low hunter success, to inform effective management strategies. To achieve this, we will develop a Winter Severity Index (WSI) to predict winter severity in various regions across the province, identify drivers of deer hunter success (e.g., winter severity, Normalized Difference Vegetation Indices, hunting), and explore a potential Hydra Effect—a phenomenon wherein harvests foster populations stabilization through density dependence and harvest interaction. Leveraging data from the Government of Alberta, Forestry and Parks, historical WSIs, and NASA Daymet V4 satellite weather data, we will assess hunter success responses to WSIs and analyze the impact of varying hunting pressure on harvest. This research significantly contributes to wildlife management and conservation in Alberta, bridging knowledge gaps and offering insights into hunter success and harvest fluctuations. The study benefits hunters, researchers, and conservation authorities alike, emphasizing the positive role of hunting in conservation efforts. By developing a predictive Winter Severity Index and understanding factors shaping deer populations, we aim to ensure the health and stability of white-tailed and mule deer populations amid Alberta's evolving environmental and economic landscape.

Bottom-up Factors Influencing Activity and Diet of the Ronald Lake Bison Herd During Their Neonatal Period

Sebastian Buitrago Gutierrez, MSc Student [1], Mark A. Edwards, Scott E. Nielsen
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Optimal Foraging Theory predicts that places with the highest quality forage will be preferred to maximize energy intake, although forage quantity can also play an important role for some species. The Ronald Lake wood bison (Bison bison athabascae) herd is a small herd (~200 animals) of conservation and cultural importance. The herd exhibits an annual migration (~ 28-km) that is outside of their core range to a meadow complex and surrounding forests at the base of the Birch Mountains. Reasons for this migration are unknown, however, it corresponds to the neonatal period when the selection of habitats that provide the highest energy intake is important due to the high nutritional demand of parturition and maternal care. Here we examine characteristics of this seasonal range in terms of forage quantity, quality, and foraging intensity. Specifically, we collected vegetation and foraging data in the herd's summer range using transects deployed inside 250-m grid cells that were stratified and sampled based on their location with respect to the meadow (meadow, near, far). Bison foraging signs were tallied and vegetative samples clipped for biomass and macronutrient content inside quadrats along transects. Using Generalized Linear Models, we found that biomass, macronutrients (i.e. metabolizable energy), and forage intensity decreased around 3-6% per 100-m distance from the meadow. Our results suggest that forage quantity and quality may be playing an important role in the herd's decision to migrate to the meadow complex during their neonatal period, although other factors, such as predation avoidance, could also be important.

Prey Preferences Under Thermal Stress in the Invasive Green Crab (Carcinus maenas)

Sofia Panzetta, MSc Student [1], Tamzin Blewett, PhD, Stephanie Green, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

In ecosystems across the globe, the expansion of invasive species has emerged as a critical challenge, with their ranges increasing dramatically by over 70% in the past 50 years. Recognized as a prominent risk to biodiversity, understanding the impact of these invaders has become increasingly vital for the implementation of effective management strategies. At the forefront of this global biological invasion is the European Green Crab (Carcinus maenas), colonizing every continent except Antarctica. Despite its invasive success, a severe knowledge gap persists, particularly on the West Coast of North America, hindering effective management strategies. In response to escalating environmental challenges such as rising ocean temperatures and acidification, C. maenas has demonstrated an ability to out-compete native species, posing threats to commercially and ecologically important species. This project addresses these gaps by investigating prey consumption rates of C. maenas, particularly under thermal stress. Through feeding experiments, we explore the relationship between body size, temperature, and prey consumption rates, shedding light on how environmental variables shape the foraging strategies of the green crab. Preliminary findings suggest a positive correlation between temperature and prey consumption, with smaller crabs exhibiting higher consumption rates than larger crabs. Through experimental trials and analyses, this research aims to fill critical knowledge gaps surrounding the ecological impacts of the European Green Crab invasion, providing insights essential for the development of informed conservation and management strategies. Preliminary findings underscore the significance of understanding the interplay between environmental variables and body size in predicting the ecological impact of this invader.

Changes in Black Bear Habitat Use in Response to the Construction and Operation of a Wind Power Plant

Tawnee Dupuis, MSc Student [1], Jaclyn Comeau, Katherina Gieder, Mark S. Boyce
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The American black bear (Ursus americanus) is the most widely distributed bear species in North America. Historically, black bears occupied the majority of forested areas on the continent (Hall 1981). However, with the growth of the human footprint, forested habitats available to bears have decreased substantially in quality and quantity (Foley et al. 2005). To prevent human-bear conflicts while maintaining a sustainable black bear population, preserving high-quality habitats is essential. One emerging threat to habitat quality is wind power development (Diffendorfer et al. 2019). While the impacts of wind energy projects on bats, migratory birds, and raptors are documented, there is no research on the potential effects that these developments have on terrestrial mammals, such as bears. The objective of this project is to quantify changes in black bear habitat use in response to the construction and operation of a wind power plant. Between 2011 and 2020, 40 black bears were collared and equipped with GPS transmitters in southwestern Vermont, USA, where the first industrial-sized wind

project within a National Forest was built in 2017. This construction footprint overlaps with areas of previously intact black bear habitat, namely stands of American beech trees (Fagus grandifolia). These mature trees produce beech nuts which are an important source of fat and protein for bears in the fall as they prepare for hibernation. Geospatial satellite-derived data from collared bears was collected during all three phases of the wind development: before, during, and after the construction of the power facility. We contrasted bear location data between the different construction phases using a latent selection difference (LSD) function. We aim to quantify the magnitude and extent to which habitat use is impacted by wind development. These findings will direct future wind energy development plans to mitigate impacts on black bears and other terrestrial mammals.

Terrain and Snow Beds Structure Evolutionary Strategies Across Plant Life-Forms in an Alpine Environment
Zach Madsen, MSc Student [1], Scott Nielsen
[1] Department of Renewable Resources Sciences, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Grimes CSR theory represents the evolutionary trade-offs between competition (C), stress-tolerant (S), and ruderal strategies (R), and can be calculated using individual functional leaf traits. The CSR analysis is a useful tool for analyzing community assembly across environmental constraints, especially one that is highly heterogenous, such as alpine ecosystems, which are characterized by environmental "extremes" within and across local areas. Our goal was to test environmental constraints against CSR strategies across broad life-form, using four environmental gradients: snow persistence (SP), solar severity index (SSI), bare rock cover (BRC), and max soil depth (MSD), which were proxies for the four major constraints in alpine ecosystems (growing season length, temperature, habitable space, and resource availability). Our results showed species and community CSR scores weighed heavily toward S strategy, with individuals showing some trend toward R strategies, and only one individual with a noticeable trend toward C strategy. We also showed that SP & SSI affected forb and shrub abundance, but the results conflicted across individual life-history traits. These results support our initial hypotheses that (1) S was the more dominant strategy, with a gradient between R score and little trend towards high C score; (2) Individuals within life-forms would demonstrate unique life-history strategies across different environmental constraints. Warming temperatures are increasing during early growing season, causing accelerating melt rate of snow patches, and causing an upward migration of shrub and tree species. Using environmental gradients to test evolutionary trade-offs within life-forms like shrubs, and their unique individual's life-history strategies, we lay the ground work for future predictions of shrub and tree expansion threats on community assembly at large scales.

Poster Presentations in Paleontology

Description of Enchodus Specimens from the Albertan Bearpaw Formation
Luke Nelson, MSc Student [1], Alison Murray, Don Brinkman
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Enchodus is an extinct genus of teleost fish from the order Aulopiformes. The genus lived in Cretaceous marine formations from around the globe, and it contains approximately 24 species that exhibit a wide range of morphologies. The large number of species and a lack of strong characters have prevented the resolution of phylogenetic relationships within the genus. Several contrasting phylogenies have been proposed for Family Enchodontidae, and while some larger clades have been consistently recovered, the genus Enchodus is only united by homoplastic characters. My research is focused on the description of several specimens of Enchodus from Southern Alberta. These specimens are the first articulated Enchodus material from the Bearpaw Formation, which was deposited along the Western coast of the Western Interior Seaway (WIS). Five species of North American Enchodus are considered valid, and some lived contemporaneously with the Alberta specimens on the Eastern WIS. Several of the specimens are highly complete and well-preserved, with articulated cranial and postcranial material, in addition to a variety of less complete specimens. The specimens show affinities with Enchodus petrosus. My research is focused on the description of these specimens, along with a phylogenetic analysis of the genus Enchodus, using discrete morphological characters.

Tooth Development and Resorption in Squamates - Methods in Comparing Living Teiidae and Fossil Mosasauridae
Fatima Iftikhar, MSc Student [1], Michael W. Caldwell, Aaron R. H. LeBlanc
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Teeth are a defining characteristic that can be used to construct evolutionary relationships. Despite its diagnostic power, this feature is trivial within squamate phylogeny as squamates have a wide variety of tooth attachment types. Most notably, hard tissue histology and computerized tomographic (CT) scans have revealed that fossil marine lizards within Mosasauridae differ from their primitive ancestors by having a deep, socketed geometry of attachment instead of a shallow, asymmetrical one. Considering this morphological variation, internal investigation through soft tissue histology is necessary to search for relatedness between living and extinct squamates. This reasoning has been used to discover that extant lizards, particularly Tupinambis (Teiidae), share a socketed mode of tooth attachment, symmetrical root structure, and three-layered attachment tissue system with mosasaurs. Because of these similarities, further comparative investigation is necessary to fully understand the geometry and mechanisms underlying tooth structure and resorption within Mosasauridae. Thin sectioning and histological staining of tooth tissues in extant Tupinambis will reveal the complex and unique attachment and resorption of teeth in teiid lizards, that will in turn help us understand the dynamics of mosasaur soft tissues at different stages of tooth development. In collaboration with the King's College London, various histological staining on extant Tupinambis (Teiidae) jaws and hard tissue histology on fossils within each sub family of Mosasauridae is to be performed. Overall, this data will be used to understand the complex and unique attachment and resorption of teeth in teiid lizards, and to help understand the dynamics of mosasaur soft tissues at different stages of tooth development. These studies will lead to various research outputs: a description of teiid tooth resorption, a comparison of mosasaur and teiid attachment geometry and tissues, as well as a description of mosasaur teeth that encompasses both tissue analysis and tooth reabsorption mechanisms.

Poster Presentations in Plant Biology

Sod-Seeding Perennial Legumes into Beef Cattle Pastures in Central Alberta
Georga Boffen Yordanov, MSc Student [1], Bismark Asante-Badu, Erick Santos, Cameron Carlyle, Edward Bork
[1] Department of Agricultural, Food and Nutritional Science, Faculty of Agricultural, Life and Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Agricultural outputs make up a significant portion of global greenhouse gas emissions. An increase in productivity of already-established pastures while also decreasing greenhouse gas emissions from these pastures is possible when sod-seeding perennial legumes into established forage stands. In this project, we are examining the recruitment dynamics of legumes sod-seeded into pastures as a method of rejuvenation (and the following changes in forage mass, nutritive value, and quantity post-seeding) and investigating legume contributions to greenhouse gas mitigation. Our study examines three sites in central Alberta that sod-seeded perennial legumes into existing pastureland in mid-to-late June 2023. Each site has eight plots in a randomised complete block design (four control plots and four sod-seeded plots), with each plot at least 5 m wide by 100 m long and containing four subplots where subsampling occurs. Per subplot, there were two shallow soil cores taken (at 0-30 cm depth, and will be used to quantify soil organic matter, pH, electrical conductivity, and total carbon and nitrogen), grazed and non-grazed biomass has been clipped (will be tested for neutral detergent fibre, acid detergent fibre, and protein, will be repeating clipping in 2024), and greenhouse gas emissions sampled (thrice in 2023 and monthly in 2024 growing period, measuring emitted carbon dioxide, methane, and nitrous oxide). This study will produce quantitative information on the effectiveness of sod-seeding for pasture rejuvenation, a cost-benefit analysis of the agronomic benefits of sod-seeding, and insight into how legume sod-seeding alters ecosystem services associated with climate change mitigation.

Effects of Humic-Based Soil Amendment on Legume-Rhizobia Symbiosis
Oshadhi Athukorala Arachchige, MSc Student [1], Malinda S. Thilakarathna
[1] Department of Agriculture, Food & Nutritional Sciences, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

"Legumes play a vital role in establishing sustainable agricultural practices. Through their distinct capability to form symbiotic associations with Rhizobium bacteria and fix atmospheric nitrogen, they serve as integral components in ecologically friendly forage systems, enabling high yields with minimal resource usage and reducing the dependency on nitrogen fertilizer. Establishing a fully functional symbiosis involves completing multiple steps, starting with the exchange of recognition signals between plants and bacteria and progressing to the formation and function of root nodules. Additionally, the process of symbiotic nitrogen

fixation (SNF) in legumes is intricate and highly influenced by various environmental and soil-related factors. Efforts towards enhancing long-term sustainability and productivity in agriculture have seen ongoing exploration of various soil amendments. Among these, humic-based products (HPs) stand out as organic amendments rich in humic acids (HA), serving as vital plant biostimulants. They have been documented to stimulate microbial growth and modulate cellular metabolism regulation. However, the efficacy of HPs is intricately linked to the concentration of HAs, necessitating the identification of the optimal application concentration to maximize benefits. While research on the effect of HAs on legumes exists, Humalite, a rich source of HA found in southern Alberta, Canada, remains understudied. This proposed study will investigate the effects of different concentrations of Humalite (ranging from 0.025% to 0.4% v/v) on alfalfa and red clover nodulation, root traits, plant growth, and nitrogen fixation compared to the untreated control. Plants will be grown under controlled environmental conditions using a hydroponic system. Plants will be sampled after eight weeks of growth, and data will be collected on nodule number, nodule dry weight, root length, surface area, volume, plant dry weight, shoot nitrogen assimilation, and SNF. This study aims to elucidate the potential benefits of Humalite application on legume growth and nitrogen fixation.

Comparing the Effectiveness of Different Methodologies for Detecting Escaping Caragana Arborescens

Jacob Mamchur, Undergraduate Student [1], Martin Hinojosa, James F. Cahill, Viktoria Wagner
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The purpose of this study was to evaluate the extent of Caragana arborescens invasion into natural habitats in Alberta and Saskatchewan and to compare the effectiveness of two different protocols for detecting escaping Caragana populations. Caragana arborescens, an introduced shrub widely planted as a shelterbelt species, has been reported to escape into natural habitats. However, the full extent of its escape and a protocol for detecting these populations are currently lacking. Our first approach utilized a database provided by the Prairie Farm Rehabilitation Administration (PFRA), containing geographic information on distributed Caragana saplings. We contacted 165 localities across Alberta and Saskatchewan, identified by the PFRA, of which 40 reported a positive presence of Caragana bordering natural habitats, thereby indicating potential for invasion. In contrast, our second method, termed "drive-by" sampling, identified 104 populations of Caragana bordering natural habitats. Cross-referencing these findings with the PFRA database showed that 89% of these localities were not listed by the PFRA, suggesting a significant underreporting in the official data. Our findings indicate that reliance on PFRA data alone is inadequate for accurately identifying escaping Caragana populations. The drive-by sampling approach, supplemented by collaboration with local communities, appears to be a more effective and reliable method for locating populations that have escaped cultivation. This study underscores the need for improved monitoring protocols to better manage the spread of Caragana arborescens and protect natural habitats from invasion.

Determining Protein-Protein Interactions Using Split-Luciferase Assays

Yang Yi Zhong, Undergraduate Student [1], Qiaomu Li, Glen R. Uhrig, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Post-translational modification (PTM) is an essential process in plants and other organisms. PTMs require protein-protein interactions between the modifiers and their substrates. Ubiquitin ligases and kinases are the two main enzyme classes that catalyze PTMs through ubiquitination and phosphorylation. These chemical modifications alter protein structures, influence protein activities and functions, and consequently influence various physiological processes, including growth and stress response. Previous Arabidopsis thaliana interactomic results of our lab identified numerous putative protein-protein interactions. These include interactions between SNF1-related kinases 2 (SnRK2s) and E3 ligase-encoding gene Histone Monoubiquitination 2 (HUB2), and between splicing kinase (SK1) and Serine and arginine-rich (SR) proteins. This project aimed to establish a reliable workflow to verify these interactions using split-luciferase complementation (Split-LUC) assays. Split-LUC assays rely on two complementary fragments of the firefly luciferase (LUC) gene, inserted separately into two putative protein interactors using recombinant cloning and expressed in Nicotiana benthamiana leaves. Should the two proteins interact, the proximity of two complementary LUC fragments would allow for the reconstitution of a functional LUC protein and quantification through luminescence measurements. Here, we observed significantly elevated bioluminescence signals representative of LUC reconstitution, demonstrating protein-protein interactions between HUB2 and SnRK2s. Our findings reaffirmed the previous hypothesis of HUB2 phosphorylation by SnRK2s and further resolved the signaling pathways in plants. In the future, we will employ this split-LUC workflow to verify the protein-protein interactions between SK1 and SR proteins.

Enhancing Carbon Sequestration and Microbial Activity Through Native Plant Restoration in Saskatchewan Grasslands

Dauren Kaliaskar, PhD Student [1], Cameron N. Carlyle
[1] Dept of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Grasslands are endangered ecosystems because of conversion to cropland, which contributes to climate change through increased greenhouse gas emissions and reductions in biodiversity. Restoration of cropland back to grassland can help restore biodiversity through the planting of native vegetation and reduce greenhouse gases through the establishment of perennial plants. The research I am developing will examine the effects of grassland restoration on forage production, soil carbon storage and indicators of soil health related to nutrient cycling and soil microbial communities. This will be done using two approaches. First, I will use a chronosequence to examine the long-term effects of restoration on plants and soils, and will take measurements at locations where the restoration history is well known. These sites range in age from a few years to decades, which will enable estimation of carbon sequestration and development of the soil microbial community over time. Chronosequences are a solution to long-term studies but have faults, so in a second approach, I will examine short-term (3 years) change in vegetation, soil properties and microbial communities at newly established restoration sites where I can measure baseline conditions prior to restoration. This comprehensive analysis will elucidate the relationship between reintroduced plant species, microbial activity, and the carbon sequestration capability of restored grasslands. Based on previous studies, we anticipate that changes in plant communities will lead to shifts in microbial communities, subsequently resulting in an increased carbon turnover rate, which is expected to enhance the soil carbon storage pool. This research will aid producers in optimizing land use for their production purposes by supporting the conversion of marginal cropland back to native vegetation, help the agricultural sector reduce its greenhouse gas (GHG) footprint, and contribute to understanding of succession of microbial communities and their function in grasslands.

Characterization of Alternative Splicing Regulation in Arabidopsis

Antonio Sebastian Hernandez Rodriguez, Undergraduate Student [1], Glen R. Uhrig, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Alternative splicing (AS) is a vital cellular mechanism in eukaryotic cells that can lead to the generation of diverse protein isoforms from a gene. AS is carried out by a protein complex known as the spliceosome, which is regulated, in part, by protein phosphorylation. The mechanisms and timing of how protein phosphorylation regulates the spliceosome, and the impact of phosphorylation activity has on plant growth and development, is just now being explored. Recent research has provided compelling evidence that AS impacts plant abiotic stress responses. For example, when exposed to high temperatures, AS-induced changes in protein localization activate the transcription of other genes responsible for protein folding and degradation, enhancing the plant's ability to cope; a critical trait for agricultural sustainability. To further understand the relationship between protein phosphorylation and AS we propose to characterize the molecular machinery suggested to reversibly phosphorylate the plant spliceosome. To do this, we characterize a compendium of Arabidopsis thaliana mutants lines using diverse functional genomic techniques to define AS control. Correspondingly, we will grow these plants under variable environmental conditions to explore the impacts of reversible phosphorylation on AS. Ultimately, the results from these experiments will offer valuable information for future targeted characterization of newly resolved AS regulators.

Exploring Growth-Defense Tradeoffs in Progeny of Lodgepole Pine

Lucas Iwamoto, MSc Student [1], Marion Mayerhofer, Jeffrey Kiely, Aryn Laxton, Colleen Fortier, Janice Cooke
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Lodgepole pine (Pinus contorta var. latifolia) has co-evolved with mountain pine beetle (MPB, Dendroctonus ponderosae). In recent years, a massive outbreak caused by the MPB, has impacted over 27mil hectares of forests throughout western North America. To protect against such pests, trees have evolved protection mechanisms, such as constitutive defences, independent of recent contact with a specific threat, or induced defences, specific responses to particular threats. However, to produce and maintain these mechanisms the plant has to allocate energy, which might be affected by environmental and genetic aspects. The goal of this project is to discover whether there are growth-defence trade-offs that can be detected or not between young lodgepole pine trees that were grown from seed collected from mature lodgepole pine trees that were either killed during the MPB epidemic (MPB-susceptible) or survived the epidemic (MPB-resilient). For this purpose, plant productivity measurements and wood core samples were collected from a total of 458 11-year-old MPB-susceptible and MPB-resilient trees that were planted in two research forests located in British Columbia. We hypothesize that MPB-resilient trees grow more slowly than MPB-susceptible trees because of the greater allocation of resources to defence strategies. And so MPB-resilient trees are expected to have reduced productivity, thinner phloem, smaller annual growth rings and earlier transition to latewood compared to MPB-susceptible trees. We would also expect MPB-resilient trees to have a greater number of resin ducts, reflecting a prioritization of resources to defence mechanisms. To test this hypothesis and these predictions, I am conducting statistical analyses of the tree height and diameter data that were collected, and I will carry out dendroecological analyses of the tree cores. This summer, I plan to collect additional cores for more detailed anatomical and histochemical analyses.

Advancing Wheat Genomics: A Comparative Analysis of Direct vs Indirect Gene-Editing

Sandhya Gautam, PhD Student [1], Fengying Jiang, Lam Tran, Gavin Chen, Andre Laroche, John Laurie
[1] Department of Agricultural, Food & Nutritional Science, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Wheat (Triticum aestivum) is a complex polyploid crop with a large genome. Genetic modification in bread wheat is challenging due to the redundancy present across the three ancestral genomes. This study addresses this complexity by conducting a comparative analysis of two innovative gene editing methods; “direct editing” and “indirect editing” via the haploid method. Direct gene editing via CRISPR-Cas9 begins with wheat transformation and leads to the development of a transgenic line. This method effectively modifies the plant genome; however, the key challenge is to develop a gene-edited line free from transgene. Addressing this, indirect editing via the haploid method comes forefront. The haploid method initiates with the transformation of maize and develops transgenic maize lines. Subsequently, this transgenic maize line is utilized to pollinate wheat lines to produce edited, haploid, and transgene-free wheat lines. Comparing the two approaches, direct editing has higher editing efficiency (42-44%) than the haploid method (4.29%). While the haploid method has lower efficiency, it offsets the shorter time required to develop a fixed-edited line. Thus, this study offers valuable insights into the two gene modification methods and serves as a significant resource for future wheat genome research.

Characterizing Genes That Influence Host-Quality of Lodgepole Pine to Mountain Pine Beetle

Raymond Kwok, Undergraduate Student [1], Colleen Fortier, Samuel Beck, Marion Mayerhofer, Mohana Talasila, Samson Osadolor, Glen Uhrig, PhD, Janice Cooke
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Lodgepole pine has co-evolved with mountain pine beetle (MPB), a bark beetle that has destroyed millions of hectares of forests in Canada. Even during severe MPB outbreaks, however, some lodgepole pines within a forest evade or survive MPB attacks. We have determined that progeny of survivor trees can be genetically distinguished from progeny of MPB-killed trees. We hypothesize that a subset of the pine genes making up this genetic fingerprint for MPB resilience assists in evasion or survival of MPB attack through establishing undesirable host characteristics for MPB selection and colonization. To test this hypothesis, we have begun characterizing the functions and expression patterns of these genes to determine their potential for contributing to an MPB-resilient phenotype. As the first step, we have cloned lodgepole pine cDNAs corresponding to these genes, verifying their sequences for functional characterization. Using BLAST with these DNA sequences, I have determined putative functions of these genes through sequence similarity with annotated genes from other plant species. Gene expression profiling will be completed quantitative RT-PCR. Using samples harvested from MPB-attacked mature lodgepole pine or lodgepole pine trees inoculated with the MPB-vectored fungal symbiont, Grosmannia

clavigera, that acts as a plant pathogen, we will investigate whether expression of these genes change in response to MPB attack or fungal inoculation. Because we cannot make transgenic lodgepole pines to test gene function, we will carry out complementation assays using the model plant Arabidopsis thaliana. The cloned cDNA of the pine genes will be transformed into an A. thaliana mutant of the corresponding ortholog to assess if the pine DNA restores a wildtype phenotype, suggesting that the pine gene carries out similar biochemical functions to the A. thaliana orthologs. Overall, the characterization of these genes will contribute to understanding the traits that contribute to lodgepole pine resilience to MPB.

Poster Presentations in Entomology

Assessment of the Efficacy and Non-Target Effects of Bacillus thuringiensis israelensis (BTI) for Mosquito Control in Edmonton

Ang Thompson [1], Ronald Batallas Huacon, Maya Evenden
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Mosquitos (Diptera: Culicidae) are well known as a nuisance pest species, but are also some of the world's most effective vectors of diseases such as malaria, dengue, Zika virus and West Nile virus. One of the most effective ways of controlling mosquito populations is via the bioinsecticide Bacillus thuringiensis isarealensis (Bti), which kills only mosquitos and their close relatives. In Edmonton, Bti is applied by hand to temporary ponds and ditches to control flood water mosquitoes, such as Aedes vexans. Aerial application of Bti to ephemeral water bodies outside of Edmonton was canceled in 2022 due to concerns about its effect on nontarget organisms, particularly members of the family Chironomidae. Chironomids are a family of non-biting midges closely related to mosquitos that are a common food source for a wide variety of insectivorous animals. As a result, the need for clarification of Bti's environmental impacts, as well as its impact on local mosquito dynamics is needed to better inform Edmonton's mosquito control procedures. This project aims to a) assess the local mosquito population and community dynamics and b) provide a comprehensive review of the effects of the bacterial insecticide Bacillus thuringiensis israelensis (Bti) on nontarget non-biting midges. In collaboration with the City of Edmonton, we will model the effects of certain environmental variables on mosquito populations (temperature, precipitation, seasonality), analyze mosquito community dynamics in response to local environmental conditions, perform a landscape analysis to determine the spatial and temporal effects on different local mosquito species, and compare larval abundance pre- and post-Bti treatment. A meta-analysis will be done to assess the literature's current understanding on the effects of Bti on non-biting midges.

Improving Management Tools for Low-Density Mountain Pine Beetle Populations in Novel Habitats: Investigating Anti-attraction Properties of Fungal Volatile Compounds

Leah Crandall, MSc Student [1], Rashaduz Zaman, Nadir Erbilgin
[1] Department of Renewable Resources, Faculty of Agricultural, Life & Environmental Sciences, University of Alberta, Edmonton, Alberta, Canada

Mountain pine beetles (MPB) overcome host tree defenses with the aid of symbiotic ophiostomatoid fungi, most commonly Grosmannia clavigera and Ophiostoma montium. During beetle colonization, antagonistic, saprophytic fungi such as Aspergillus and Trichoderma sp. can also colonize attacked trees. These fungi are commonly found in MPB galleries and are known to compete with MPB and their symbiotic fungi, thereby impacting beetle survival and reproduction. Recent work by Zaman et al. (2023) found that MPB are strongly attracted to fungal volatile organic compounds (FVOCs) emitted by symbiotic fungi. However, little attention has been given to the FVOCs emitted by antagonistic fungal species. Given that MPB attraction is known to be influenced by FVOCs emitted by symbiotic fungi, it is plausible that a similar phenomenon may occur with FVOCs from antagonistic species. Therefore, we will be investigating whether FVOCs emitted from antagonistic fungi may play a role in the inhibition of MPB attraction. We will characterize the volatile profiles of Aspergillus and Trichoderma species to identify possible anti-attractant compounds. We will then conduct choice assays in order to determine the impact of those individual FVOCs on MPB attraction. In the presence of antagonistic fungi, we expect to observe reduced MPB feeding and attraction to their symbiont FVOCs. Once we identify individual compounds with anti-attraction properties, we will conduct field experiments using beetle pheromones. This work will allow us to better understand interactions between MPB and FVOCs during host selection and will expand MPB monitoring tools.

Susceptibility to Temperature Stressors at the Individual and Colony Level

Kaitlin Kinahan, Undergraduate Student [1], Jacob J. Herman, Olav Rueppell
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

In social insect colonies individuals depend on each other to contribute to colony success. They exhibit division of labor within the caste system with individuals of different traits undertaking a variety of roles. At the colony level, we heated to a temperature within tolerance to initiate fanning behavior. This behavior is done to improve circulation and bring down the internal temp of the hive. Bees were divided into two categories, fanners and non-fanners, according to the observation of this trait at the colony level. These bees were then individually temperature tested determining their susceptibility to temperature changes. As measured by survival when heated to a critical temperature, we hypothesized the bees initiating fanning in the hive to also be more susceptible to temperature at the individual level. The results are discussed in the context of the previously proposed weak worker hypothesis. In the future, we expect similar results on cooling experiments regarding the initiation of clustering.

Susceptibility of Non-sclerotized Invertebrates to Ectoparasite Infection
Sean Jay Yuan Chua, Undergraduate Student [1], Colin Macleod, Lien Luong
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Invertebrate exoskeletons have been shown to provide defense against pathogens, parasites, and parasitoids due to their tough covering. Behavioral defenses also play a significant role in infection avoidance and parasite removal. Previous observations in our lab have shown that newly eclosed flies had higher levels of infection than fully sclerotized flies. From this study, my study seeks to uncover the mechanism that increases susceptibility to infection in newly enclosed flies, focusing on behavioral and physical defense. Using the cactophilic fly, *Drosophila nigrospiracula*, and its ectoparasitic mite Macrocheles subbadius to investigate the relative importance of parasite resistance. I test the hypothesis that newly eclosed flies are more susceptible to mites because the cuticle is not yet fully sclerotized and vulnerable to mite attachment. Alternatively, newly eclosed flies may be more susceptible because they are less active and have nonfunctional wings, a behavioral defense that is important to mite resistance. I experimentally removed the wings from flies to determine the role of behavioral resistance by clipping the wings from both groups. If behavioral resistance is important, then I expect to see higher infections compared to flies with intact wings, but no difference based on level of sclerotization. However, if sclerotization plays a bigger role, I expect to still see differences between newly eclosed flies and fully sclerotized flies, even without wings. Additionally, we tested the endurance of sclerotized and newly eclosed flies; previous studies have demonstrated that endurance is correlated with the level of resistance. I hypothesize that newly eclosed flies are energetically spent following metamorphosis, thus predicting lower endurance in newly eclosed flies. Investigating the underlying mechanisms that increase susceptibility to mite infection reveals traits applicable to other invertebrates' defense against ectoparasites, and evaluate how crucial these defenses are to resist infection, especially during the vulnerable period immediately after molting.

Poster Presentations in Molecular Biology and Genetics

Fruit Flies 'Seize' to Exist: Examining the Effects of Stromalin on Seizure Behavior in Drosophila
Amoolya Sai Dwijesha, MSc Student [1], Celina Phan, Anna Phan
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The cohesin complex, an evolutionarily conserved structure, is composed of 4 core subunits: Stromalin (STAG1/2), Rad21, Smc1 and Smc3, and accompanying proteins that help the complex with its roles in chromosome segregation and transcriptional regulation. Perturbations in the subunits and associated proteins lead to a collection of rare, developmental disorders engulfing various organ systems, known as cohesinopathies that manifest in behavioral, neurological, and physiological symptoms. A few years ago, the Drosophila homolog of the STAG 1/2 subunit known as Stromalin was found to function as a memory suppressor gene by restricting synaptic vesicle (SV) pool sizes in dopamine neurons. Prior work on in-vivo and in-vitro models of cohesinopathies have suggested interferences in cohesin complex's role in gene expression and not chromosome segregation as a possible causal mechanism for the disease. This led us to hypothesize that Stromalin as a member of the cohesin complex may regulate SV numbers transcriptionally, and thus induce neurological symptoms like increased seizure frequencies in cohesinopathies. We observed that, like cohesinopathy patients, pan-neuronal reduction of Stromalin resulted in an enhancement in seizure frequencies. We then identified possible downstream mediators of Stromalin's seizure effects by testing the 5 candidates uncovered by our lab via RNA-Sequencing: nep1, CG17698, cox7c, ttm2 and su(z)12. Excluding cox7c, prior studies on the mammalian homologues of the candidates provided evidence for either altered expression or mutation of the candidates in certain human disorders characterized by seizures or in rodent models of status epilepticus. We found CG17698 and cox7c to be potential mediators of Stromalin's seizure inducing effects

as pan-neuronal reduction of the two emulated the seizure phenotype produced by Stromalin. At present, we are investigating whether depleting SV numbers at presynaptic terminals by blocking their transport in the whole brain can reverse the seizure phenotype observed in our cohesinopathy model.

Aberrations in Antioxidant Expression in the Liver, Spleen, and Heart Occur 4 Hours Post-sepsis

Avery Noppers, Undergraduate Student [1], Ibrahim Khodabocus, Rohini Roy Roshmi, Si Ning Liu, Jad-Julian Rachid, Claudia Holody, Hélène Lemieux, Stephane Bourque
[1] Department of Biochemistry, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Sepsis is a life-threatening condition caused by a dysregulated host response to infection. Sepsis is associated with mitochondrial dysfunction and perturbations in metabolism. This damage is likely due to the overaccumulation of reactive oxygen species (ROS), which can lead to pathophysiology. Antioxidants neutralize excessive ROS pools under physiological conditions, conferring homeostasis. Here, we attempted to explore the immediate impact of fecal slurry-induced peritonitis (FIP) on antioxidant expression in metabolically important organs such as liver, spleen, and heart to gain further insight as to the impact of FIP on antioxidant expression in adult mice. Additionally, we hoped to gain an understanding in the differences in responses between sexes. FIP was induced in male and female mice by injecting fecal slurry into the abdominal cavity, while controls were given vehicle. After 4 hours, all mice were euthanized, and the tissues were collected for RT-qPCR experiments. We chose to look at the expression of catalase, glutathione peroxidase 4 (GPX4), superoxide dismutase 1 (SOD1), and superoxide dismutase 2 (SOD2) in all tissues. We found a reduction in the expression of catalase in male and female liver and spleen, as well as an increase in female heart samples. We found a reduction in the expression of GPX4 in male and female liver, as well as female spleen, but no differences in heart samples. We found a reduction in the expression of SOD1 in male and female liver, as well as female spleen, but found no differences in heart samples. We found a reduction in the expression of SOD2 in male and female liver and spleen, but no differences in heart samples. The findings of this research may highlight key tissues where the greatest perturbations from homeostasis lie, creating the foundation for future pre-clinical experiments aimed at generating bespoke medical interventions based on sex.

Identification of Neurons Activated by Social Experience

Brian Kong, Undergraduate Student [1], Illia Pimenov, Anna Phan
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Social isolation is a growing issue among Canadians which can significantly impact human mental and physical health, and has been linked to adverse outcomes such as cardiovascular disease and depression. With social isolation being a phenomenon that affects Canadians of all walks of life, research that focuses on the biological causes and consequences of social isolation can have a widespread impact. To address this issue, our project aims to determine which neurons are involved in the experience of social isolation in Drosophila melanogaster. Our lab has previously found that social isolation leads to a profound reduction of neural synapses, reduces social interactions, disrupts sleep, and impairs learning and memory in Drosophila. Notably, these behaviors are similar to how humans and rodent models respond to social isolation as well. We plan to use novel genetic tools that sum neural activity over long periods of time (such as the Transcriptional Reporter of Intracellular Calcium (TRIC) and Calcium-dependent nuclear import of LexA (CaLexA) tools) to study neuronal activity in socially experienced and isolated flies. We will use these tools to identify the neurons that are disproportionately affected by social isolation, which will allow us to focus our research efforts on deciphering the mechanisms behind social isolation behaviors. Ultimately, understanding the neural mechanisms behind the experience and consequences of social isolation will aid the development of effective intervention strategies and treatments, and may help give us insight into the long-lasting effects of social isolation on humans.

Investigating the Effect of Sars-Cov-2 ORF10 in C. elegans

Darren Lepp, Undergraduate Student [1], Martin Srayko
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

SARS-CoV-2 continues to threaten human respiratory health. One CoV-2 protein, ORF10, has been shown to interact with Zyg11B, a component of the Cullin-2 E3 ubiquitin ligase complex in human cells. Interestingly, this interaction causes overactivity of the Cullin-2 E3 Ubiquitin Ligase Complex, ultimately causing degradation of a protein found in cilia (IFT46). Loss of IFT46 could be responsible for some SARs-related symptoms by impairing cilia biogenesis/maintenance. The Srayko lab showed that ZYG-11/CUL-2 is required for degrading the MEMI proteins in C. elegans during oocyte maturation. MEMI proteins are required for oocytes to undergo meiotic cell divisions but must then be removed prior to the first mitotic cell divisions in the embryo. If MEMI is removed prematurely, female meiosis II does not occur, but if MEMI is not degraded at the end of meiosis, the embryos remain in meiosis II and fail to transition into mitosis properly. My research is focused on determining whether or not ORF10 has an effect on C. elegans ZYG-11/CUL-2. To study this, an ORF10-6xHis sequence was cloned into an expression plasmid. Conditions for protein expression are currently being optimized. Protein purification will be done via nickel-affinity-column chromatography. Once purified, the protein will be injected into C. elegans hermaphrodites and the resulting offspring will be observed. If ORF10 activates the ZYG-11/CUL-2 complex in worms, then we would expect to see phenotypes consistent with degradation of downstream components. Because MEMIs are potential substrates of ZYG-11/CUL-2, the ORF10 injection might cause phenotypes that resemble MEMI loss. This work will be informative on the evolution of host-virus interactions and the molecular basis for viral proteins pathology, which could help direct the development of therapeutics in the future.

Exploration of Biomarkers in Chronic Complex Diseases

Devanshi Pandit, Undergraduate Student [1], Zoe Wang, Gane Wong, David Wishart
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Complex multifactorial diseases are influenced by many factors including genetic, environmental and lifestyle factors that play into risk factors and presentation of the disease. They have a variety of consequences that can be assessed by the presence of biomarkers. Biomarkers can be protein, genetic, metabolic, imaging and more which serve as biological cues. They play a role in prognosis, diagnosis, therapy, and screening. Knowledge of biomarkers in chronic diseases can help us characterize these diseases better as well as aid in developing new treatments. It can be challenging to find and assess the various scientific studies that exist in literature. There is a need to determine a set value that can be tested, how this can be used in testing of individuals. This study explores the multifaceted role of biomarkers in complex multifactorial diseases specifically focusing on cardiovascular disease and Type 2 Diabetes.

Fecal Slurry-Induced Peritonitis Causes Sex-Specific Alterations in Mitochondrial Function and Reactive Oxygen Species Generation in the Liver and Kidney

Ibrahim Khodabocus, PhD Student [1], Jennie Vu, Rohini Roshmi, Avery Noppers, Si Ning Liu, Jad Rachid, Claudia Holody, Helene Lemieux, Stephane Bourque
[1] Department of Pediatrics, Faculty of Medicine and Dentistry, University of Alberta, Edmonton, Alberta, Canada

Introduction: Sepsis is estimated to underlie 20% of all global deaths. Males appear to be more susceptible to sepsis-induced organ injury than females, although the mechanisms underlying these sex-differences are unclear. Here, we studied the impact of fecal slurry-induced peritonitis (FIP) on mitochondrial function in male and female mice to gain insight into the sex-specific metabolic consequences of sepsis. Methods: C57BL/6 mice were injected with fecal slurry (FS, 0.55 mg/g) or vehicle. Buprenorphine (0.5 mg/kg, at 4h), Ringer's Lactate (15 mL/kg, at 12h) and Imipenem (25 mg/kg, at 12h) were administered post FIP-induction. At 4h, 12h, and 24h post-FIP, mice were euthanized, and tissues were collected; liver and kidney homogenates were assessed for mitochondrial function by high resolution respirometry. Biochemical assays were used to assess mitochondrial content and oxidative stress (8-oxo-dG). Results: FIP caused: (1) no changes in renal mitochondrial content, but reduced content in the liver of males (P=0.012), but not females (P=0.095) by 24h; (2) reduced respiration through complex(C)II in the renal medulla of males (P=0.002), but not females (P=0.75), as early as 4h post-FIP; (3) increased respiration through CI in kidneys of both males (P=0.03) and females (P=0.006) by 12h post-FIP, but reduced respiration through CI in the liver of males (P=0.031) and females (P=0.0005) by 12h post-FIP; (4) increased liver 8-oxo-dG in females (P=0.01), but not males (P=0.35) by 12h post-FIP. Significance: This work may provide insights into the sex differences in susceptibility to sepsis-induced organ dysfunction. Funding: Project funding was provided by a CIHR Project Grant (PS178007) held by SB.

Scalloped-Complex Regulation of Cell Identity

James Stewart, Undergraduate Student [1], Julie Haskins, Kazuki Ueda, Karen He, Andy Cheng, Andrew Simmonds
[1] Department of Cell Biology, Faculty of Medicine and Dentistry, University of Alberta, Edmonton, Alberta, Canada

TEAD proteins are an evolutionarily conserved family of DNA binding proteins. These require 'cofactor' proteins with transactivation domains to act as transcription factors. TEAD cofactors include Vestigial-like proteins (VGLLs) and Yes associated protein or Transcriptional coactivator with PDZ domain (YAP, TAZ). In Drosophila melanogaster, the paralogs are Scalloped (Sd) for TEAD; Vestigial (Vg) and Tondu-domain-containing Growth Inhibitor (Tgi) for VGLLs; and Yorkie (Yki) for YAP/TAZ. Multiple cofactors may be present simultaneously in a cell, and it is hypothesized they compete to bind Sd to drive separate cellular processes. Thus far, the interactions of Yki and Tgi with TEAD have been the primary focus of previous studies, mostly without consideration for the role of Vg. Notably, one major regulatory event in VGLL, YAP/TAZ, and TEAD transcription factor activity is the sequestering of one or more protein components in or out of the nucleus. A second regulatory event is phosphorylation of TEAD cofactors which alters cellular localization and protein stability. Utilizing immunofluorescent imaging in Schneider 2 cells (derived from Drosophila embryos), I demonstrated that overexpression of transgenic Vg affects cellular localisation of co-overexpressed Sd and its other cofactors. In addition, post-translational phosphorylation of serine 215 in Vg has been previously reported to affect Vg functionality in tissue. I showed that overexpression of phosphomimetic or non-phosphorylatable VgS215 mutations also has a significant effect in terms of localization in or out of the nucleus, especially when Sd or other cofactors are co-overexpressed. Finally, I demonstrate that Sd's cofactors can colocalize with one another even in the apparent absence of Sd, both in and out of the nucleus.

Measuring Voltage-Gated Potassium Channel Expression Using Alfa Tags in a Fluorescence-Based Assay

Mark Wong, Undergraduate Student [1], Shawn Lamothe, Harley Kurata
[1] Department of Pharmacology, University of Alberta, Edmonton, Alberta, Canada

Background: The voltage-gated potassium channel Kv1.2 plays a crucial role in neuronal excitability. Kv channels exert their effects by controlling ion movement across the plasma membrane. Mutations, regulatory proteins, and drugs can affect ion channel localization, leading to disease. Mutations in Kv1.2 in the brain are associated with a spectrum of seizure disorders and neurodevelopmental delay. Electrophysiology techniques such as patch clamping allow for measuring real-time ion channel activity. Still, methods to assess the effects of drugs, regulatory proteins, and mutations on Kv channel expression are underdeveloped. Objectives: This study implements a fluorescent assay using the small peptide ALFA tags recognized with high specificity by an ALFA nanobody (NbALFA) to assess Kv1.2 channel expression. We hypothesize that using the ALFA tag will not affect normal functions of the Kv1.2 channel and can be used to quantify channel expression at the cell surface. Methods: We engineered an ALFA tag into one of the extracellular loops of Kv1.2 and then fluorescently labeled it using an NbALFA. A glycosylation-deficient mutation construct of the channel (ALFA-Kv1.2 [N207Q]) was created to test if glycosylation affects ALFA-tag binding. Patch clamp recordings and Western blotting were used to test if the ALFA tag affects Kv1.2 expression and response to known regulatory proteins. Fluorescently-labeled channels were detected using a plate reader and visualized using microscopy. Results: No difference was seen in expression between untagged Kv1.2 and ALFA-Kv1.2. Also, ALFA-Kv1.2 and ALFA-Kv1.2 [N207Q] are sensitive to known regulatory proteins of the channel. Using a fluorometric plate reader was sub-optimal because high background fluorescence persisted in the plate reader detection. Discussion: The ALFA tags detect Kv1.2 channel localization at the plasma membrane without affecting normal channel activity and are unaffected by channel glycosylation. The effect of known regulatory proteins of Kv1.2 can be visualized; however, quantifying fluorescence using a plate reader is not optimal due to high background fluorescence.

Using a Macrophage Bioassay to Examine the Effects of Nickel at Oil Sands Process-Affected Water Relevant Concentrations

Nishita Shukla, Undergraduate Student [1], Nora Hussain, James Stafford
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Oil sands process-affected waters (OSPW) is a by-product of crude oil extraction, and contains environmentally hazardous concentrations of cytotoxic organic and inorganic contaminants. Previously, our lab has shown that inducible nitric oxide synthase (iNOS) in macrophages is stimulated by organic contaminants and secretes nitric oxide (NO) in response, allowing them to function as a water quality biosensor. Although the organic fraction —particularly naphthenic acids— has been extensively studied, the inorganic fraction remains mostly unexplored. Metals which reside in the inorganic fraction are an under-investigated component of OSPW, but can be extremely toxic and are difficult to break down or remove. This project will examine the effect of nickel (Ni2+) on murine macrophages at OSPW-relevant concentrations. While Ni2+ is known to activate proinflammatory responses in human immune cells, it has been shown to interfere with NO production in mouse macrophages. This project exposed cells to various concentrations of Ni2+ in combination with heat-killed Escherichia coli (HKEc), to examine if the addition of Ni2+ affected the NO production induced by HKEc. The effect of nickel on immunomodulation will be measured by comparing macrophage cells that are pre-exposed to Ni2+ at OSPW relevant concentrations before adding HKEc, to cells exposed to Ni2+ and HKEc simultaneously. A better understanding of the immunomodulatory impact of specific heavy metal concentrations in OSPW will be useful in interpreting biological impacts in the context of developing future remediation methods.

The BMP Ligand gdf6a Regulates Zebrafish Craniofacial Development

Sabrina Fox, PhD Student [1], Sarah Hay, Andrew Waskiewicz
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The vertebrate craniofacial skeleton is crucial for communication, feeding, and breathing. Improper craniofacial development leads to congenital abnormalities that severely impact the quality of life of affected individuals. Currently, the factors that regulate the morphogenesis of the craniofacial skeleton during development remain incompletely understood. Here, we demonstrate that the Bone Morphogenetic Protein (BMP) ligand Growth and Differentiation Factor 6a (gdf6a) is a regulator of craniofacial development in zebrafish. gdf6a is expressed in the zebrafish pharyngeal arches from 48-72 hpf; more specifically, gdf6a is expressed along the ventral midline of the developing zebrafish craniofacial skeleton. Accordingly, gdf6a mutant larvae have malformed craniofacial cartilage elements. Consistent with the expression pattern of gdf6a, these malformations primarily affect the midline of the cartilage elements in mutant larvae. gdf6a mutants also display aberrant muscle fiber localization with no effect on tendon morphology, suggesting that gdf6a regulates muscle fiber attachment to the skeleton independently of tendon formation. Analysis of BMP signaling using phosphorylated Smad (pSmad) immunofluorescence and BMP-regulated GFP expression indicates that BMP signaling is active along the ventral midline of the zebrafish craniofacial skeleton from 48-72 hpf and corresponds directly with gdf6a mRNA expression, suggesting that gdf6a induces BMP signaling in these structures. Consistent with this, inhibition of BMP signaling from 48-72 hpf phenocopies gdf6a mutants, and readouts of BMP signaling are reduced in gdf6a mutants, indicating that gdf6a regulates craniofacial development via BMP signaling. Future studies will focus on identifying of the downstream transcriptional targets of gdf6a-mediated BMP signaling in the craniofacial skeleton.

TGT-B Signaling Regulates the Expression of BMP Inhibitors During Vertebrate Jaw Development

Sarah Hay, Undergraduate Student [1], Sabrina Fox, Andrew Waskiewicz
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Vertebrates rely heavily on their craniofacial skeleton for communication, breathing, and feeding; abnormal formation of the jaw during embryonic development can negatively impact these behaviors, thereby lowering the quality of life of affected individuals. Several families of signaling proteins are involved in development of the jaw, including members of the bone morphogenetic protein (BMP) and transforming growth factor beta (TGT-B) superfamilies. The components of these signaling pathways are involved in a wide array of processes including bone formation, cartilage maintenance, and homeostasis of the skeleton in adult organisms. We have shown that BMP and TGT-B signaling are active in distinct domains of the developing zebrafish jaw and that they have opposite roles during jaw development. We have also shown that these pathways antagonize one another, with TGT-B restricting the domain of BMP signaling in the jaw. However, the mechanism by which TGT-B restricts BMP signaling in this context is not fully understood. Our lab has found that TGT-B signaling restricts BMP signaling by promoting the expression of nog3, a BMP antagonist. Here, we further investigate the relationship between BMP and TGT-B signaling by conducting in-situ hybridization to characterize the spatial expression of the BMP inhibitors. We find that the spatial expression of these genes is consistent with a role in inhibiting BMP signaling during jaw development. Additionally, manipulating TGT-B signaling with pharmacological agents alters the spatial expression of these BMP inhibitors, indicating that the TGT-B signaling spatially regulates BMP signaling in jaw development by inducing the expression of genes that encode BMP inhibitors. Future studies will focus on characterizing the expression of these genes in other conditions to better understand which TGT-B family members regulate the expression of these BMP inhibitors. Taken together, this data contributes to our understanding of the signaling networks that regulate vertebrate jaw development.

The Development of a Liver Organ-On-Chip for Studying Liver Metabolism and Toxicity Assessments

Suyenna Huang, Undergraduate Student [1], Upasana Singh, PhD, David Wishart, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Liver organ-on-chips have emerged as a novel, alternative platform for studying liver metabolism and compound toxicity assessments, compared to the conventional two-dimensional (2D) cell culture of primary hepatocytes. 3D culture addresses the limitations of 2D culture by growing cells in a more physiologically relevant environment with the addition of extracellular matrix components and allowing for complex spatial interactions. To study liver metabolism that is more reflective of in vivo conditions, two issues need to be addressed: (i) the maintenance of non-parenchymal cells that support hepatocyte function through cytokine secretion, and (ii) the creation of the sinusoid, the liver capillaries where metabolites and oxygen can be exchanged with the hepatocytes. The use of organ-on-chips (OoCs), a type of 3D culture, addresses both issues by co-culturing multiple cell types within a microfluidic chip and controlling the fluid flow over the cells, mimicking the flow of the bloodstream within the sinusoid and interactions with the hepatocytes. The long-term goal of this project is to develop a liver organ-on-chip, which is specifically modeled after the hepatocyte-sinusoid interface by using 3 different cell lines: induced pluripotent stem cell (iPSC) derived hepatocytes, human microvascular endothelial cells (HMEC-1), and THP-1 which are monocytes. The HMEC-1 and THP-1 represent the endothelial cells of the sinusoid and the Kupffer macrophages respectively. The current objective is the optimization of all 3 cell lines growth and the differentiation of the iPSCs. iPSCs are differentiated into hepatocyte-like cells using a laminin matrix and various media over the course of 21 days. The genetic stability of the iPSCs are confirmed by karyotyping. The differentiated iPSC-derived hepatocytes are validated by morphological analyses and albumin secretion testing. Finally, all 3 cell lines are seeded into 2 bordering channels within a microfluidic chip, therefore mimicking the in vivo environment between the sinusoid and the hepatocytes.

Poster Presentations in Microbiology

Urec Diversity and the Potential for Microbially Induced Calcite Precipitate in Permafrost

Jarod McLeay, Undergraduate Student [1], Camilla Nesbø, Scott Cocker, Brian Lanoil, Duane Forese
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Permafrost, covering ~25% of the northern hemisphere land area, is the only component of the cryosphere that people live upon. As ground temperatures warm, much of this area is compromised by the loss of ground ice and the associated integrity of the soil. Thawing permafrost thus has the potential to impact critical infrastructure along with northern ecosystems. There are few options to mediate these changes outside of costly engineering interventions. This project aims to investigate the potential for microbially induced calcite precipitation (MICP), a mitigation approach used to improve soil strength and increase soil stability, though not yet investigated in permafrost areas. In MICP, microbial activity raises the pH within the soil, which, along with an injection of calcium salts, leads to the precipitation of calcite (CaCO3), stabilizing the soil and increasing its shear strength. There are several challenges to applying MICP to existing microsystems in situ, including stimulating microbial processes that raise pH, such as urea hydrolysis by the urease enzyme encoded by the ureC gene. As a first step toward evaluating the potential for MICP for use in permafrost environments, we explored ureC biodiversity in permafrost metagenomes in public databases, comparing these to metagenomes from non-permafrost regions. This analysis suggests that ureC biodiversity and composition from permafrost and non-permafrost areas are similar, indicating methods to enrich urease-degrading bacteria in thawed permafrost may be similar for different biomes. Using ureC primers, we amplified ureC genes from permafrost samples taken at several depths in our focus region of central Yukon. We will sequence these genes and add them to our metagenomic tree to examine the diversity of ureC genes from this region. Future research will also involve tracking ureC gene enrichment under other MICP conditions, correlating microbial communities to soil stabilization, and optimizing MICP treatments of thawed permafrost soils.

Enterobacteriaceae Two-Component System Crosstalk

Kayla Surgenor, Undergraduate Student [1], Ceara Cabana, Casey Fowler, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Bacterial two-component systems (TCS) are a common form of signal transducers that allow bacteria to respond to their dynamic environment. TCS consist of a membrane bound histidine kinase (HK) that senses environment stimuli and interacts with a cytosolic response regulator (RR) to translate the stimulus into gene expression. Bacteria encode for several TCS; however, these systems are normally isolated from one another to prevent cross-system interactions. Despite TCS highly favouring insulation and specificity, interactions between systems are occasionally observed through a process termed crosstalk. QseBC is a TCS that regulates flagella and motility and is documented to undergo a crosstalk interaction with the TCS PmrAB. While this interaction is well documented, the purpose of this crosstalk is poorly understood. Potential hypotheses state that crosstalk may be advantageous and resulted from coevolution between these systems or this interaction evolved from a gene duplication event and this interaction will be lost over time. The objective of this project is to determine how prevalent this crosstalk interaction is within the Enterobacteriaceae family to better understand the evolution and purpose of this unique interaction. Chimeric plasmids were created to express different combinations of a RR and HK from QseBC and PmrAB. The prevalence and strength of these system interactions will be tested using RR promoter:LacZ reporter fusion and β-galactosidase assays. The prevalence and strength of the interaction between proteins of different species will reveal how conserved crosstalk is within the Enterobacteriaceae family to determine if this is an advantageous interaction and provide a better overall understanding of crosstalk evolution.

Towards the Industrialization of Methylorubrum extorquens AM1: Adaptive Evolution in Response to Acid Stress

Maryssa Iacobelli, MSc Student [1], Dominic Sauvageau, Lisa Y. Stein, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Bioprocessing is a sustainable method for producing various commercial products such as bioplastics and enzymes from living organisms. One bacterium, Methylorubrum extorquens AM1 has shown potential for bioprocessing, as it uses methanol as a feedstock, which can be derived from methane – a potent greenhouse gas. However, translation into industry is challenging due to harsh industrial conditions like extreme temperature, salinity, and pH. Thus, the aim of this research is to address one of these stressors by adapting M. extorquens AM1 to low-pH conditions using adaptive evolution. Adaptive evolution is a selection method for generation of beneficial mutations in rapidly growing microbes. By culturing the strain

over many generations and gradually decreasing the pH, it is expected that various mutations allowing for the strain's persistence will emerge. As the strain adapts, it is expected that metabolite production will improve. To evaluate this, gas chromatography will be used to follow the production of polyhydroxy butyrate, a precursor for bioplastics and known metabolite of M. extorquens AM1. To uncover how the strain adapts to acid stress, some select isolates as well as the parental strain will be sent for whole-genome sequencing, RNA seq analysis and metabolomic profiling. This analysis will identify different mutations that have been selected for and suggest how the genes are functioning. Since proton pumps, amino acid-dependent tolerance systems and adapted membranes are all ways in which bacteria may overcome acid stress, mutations related to these systems may be observed. As global trends shift towards a bioeconomy, efficient systems to compete with the current industrial standards are imperative. This study will address acid stress and provide insight on how to overcome it using selection from laboratory adaptation.

Induction and Characterization of Bacteriophages from the Clinical and Environmental Pathogen Burkholderia gladioli

Angelle P. Britton, PhD Student [1], Brittany S. I. Supina, Philip Lauman, Jonathan J. Dennis, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

By 2050, a predicted 40% of hospital acquired bacterial infections will resist standard drug therapies. Concerningly, discovery of novel antibiotics is not keeping pace with developing antibiotic resistance. Burkholderia gladioli is an emerging, broad-host-range pathogen that causes serious respiratory infections in individuals with cystic fibrosis and chronic granulomatous disease, and infects several plant species including onions, carnations, gladiolus, and rice. B. gladioli is highly resistant to antibiotics, urgently necessitating novel treatment strategies for clinical and agricultural infections. Bacteriophage (phage) therapy is a treatment option that offers strong pathogen specificity without significant side effects. Combined with the high environmental availability of phages, these advantages make phage therapy an attractive alternative to antibiotics. While many phages active against members of the closely related Burkholderia cepacia complex (Bcc) have been characterized, few B. gladioli-targeted phages have been isolated, thus the need for further investigation is required. Most known Bcc phages are lysogenic, meaning that they can integrate into their host genome upon infection. In this dormant form, these integrated phages (prophages) may make the host bacteria resistant to lysis and secondary infection. My work aims to harness prophage-containing strains of B. gladioli as a source for phage isolation to increase our arsenal of phages specific to B. gladioli, which may target environmental or clinical pathogens. Nine B. gladioli strains were treated with Mitomycin C, a DNA cross-linking agent known to induce stress, and six prophages (AB1-AB6) were isolated. Through whole genome sequencing, AB1, AB2, and AB5 were identified as novel phages containing phagemid characteristics (AB1 and AB2), or transposable elements (AB5). Transmission Electron Microscopy classified AB1 as a myovirus. Future work will continue characterizing these and other B. gladioli phages, while exploring their unique genetic features.

Poster Presentations in Physiology and Development

Environmental Impact of Organic Ultraviolet Filters on Aquatic Organisms: Insights from Lab and Lake Water Exposures

Ally Legge, Undergraduate Student [1], Aaron Boyd, Sidney Martin, Connor Stewart, Jenelle McCuaig, Dustin Doty, Tamzin Blewett, PhD
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Organic ultraviolet filters (UVFs) are compounds that protect against UV radiation and are commonly found in a variety of cosmetic products, such as sunscreens. During recreational activities, these chemicals can leach into aquatic ecosystems and cause environmental contamination. UVF toxicity to aquatic organisms has been observed, with Daphnia often being a model test species. Previous UVF toxicity experiments have focused on lab-raised aquatic organisms in lab-prepared water samples. This study investigated UVF toxicity in lab-reared Daphnia pulex in both lab water and lake water in order to observe how environmentally-relevant water samples impact the toxicity of these chemicals. The toxicity of three UVFs, avobenzone, oxybenzone, and octocrylene, was compared. 48 hour LC50 tests and 21-day exposures were conducted for each UVF and several endpoints were measured, including reproductive effort, metabolic rate, and body length. LC50s for all UVFs were higher in the lake water treatments (i.e. avobenzone LC50 26.4 µg/L 95% CI (20.6-35.9)) compared to lab water treatments (avobenzone LC50 5.30 µg/L 95% CI (3.67-8.04)), suggesting lowered toxicity. However, lake water treatments exhibited greater mortality to UVFs over the long-term exposures. High levels of DOC in the lake water is likely the cause of this difference, as DOC binds to the UVFs and are then subsequently eaten by the daphnids over the course of the exposure. Body

length measurements and reproductive effort decreased in lake water treatments due to the faster rates of mortality observed. No significant differences in metabolic rate between the water types was observed, which is consistent with results from previous studies on UVF toxicity in Daphnia. With aquatic ecosystems around the world facing the threat of human activity, it is vital to understand how environmental contaminants, such as UVFs, impact the biota.

24-Hour Exposure to Cb_1R Agonist Acpa in Developing Zebrafish and Subsequent Primary Motor Neuron Morphology at 5 Days Post Fertilization

Hudson Bovee, Undergraduate Student [1], Declan W. Ali
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The endocannabinoid system (ECS) is a whole-body signalling network that contributes to organismal homeostasis. In its coordination with the nervous system, the ECS has been implicated in neuronal proliferation, myelination, connectivity, and morphology; synaptic functioning and plasticity; and neurogenesis. Endogenously produced cannabinoids, associated enzymes, proteins, signalling targets, and cannabinoid receptors make up the endocannabinoid system. Among these constituents is the G-protein coupled CB_1 cannabinoid receptor, which is abundantly expressed in the central nervous system and has been found to influence neuronal outgrowth. CB_1 receptors are also found in skeletal muscle and are thought to mediate muscle contraction. Here, we aim to elucidate the role of the CB_1 receptor in the early morphological development of primary motor neurons in zebrafish. Zebrafish will be exposed to either a water control, a vehicle control, or the CB_1 agonist arachidonylcyclopropylamide (ACPA) for 24 hours beginning during gastrulation. Exposure at this time coincides with the beginning and early stages of neurodevelopment in zebrafish. Following exposure, they will be left to develop under standard conditions until five days post fertilization (dpf). At 5 dpf, zebrafish will be euthanized. Then, immunohistochemistry will be performed on the euthanized larvae using a fluorescently tagged antibody to label the surface of their primary motor neurons. I expect that embryonic exposure to ACPA for 24 hours will alter primary motor neuron morphology at 5 dpf. I predict that the number and the length of neuronal branches in exposed zebrafish will be increased compared to the control groups. Our research will help shed light on the intricacies of animal physiology and development. It may also be applicable to research on cannabinoid-based treatments for movement disorders because the ECS is the target of cannabinoid therapeutics.

Elucidating the Influence of Cannabinoids on the Acoustic Escape Response in Zebrafish

Hope Giardina, Undergraduate Student [1], Michelle Makila, Declan Ali
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

The use of cannabis for recreational and medicinal purposes is prevalent in today's society, and its use is only increasing. However, we lack a complete understanding of how cannabis acts in the body, and of the endogenous cannabinoid system that cannabinoids act on. What we do know is that cannabinoids can influence locomotive processes and can influence receptors which typically transmit sensory information, such as the transient receptor potential vanilloid 1 (TRPV1) channel. Prior studies have shown that the escape response to sound in zebrafish is affected by brief (30-minute) treatment with the cannabinoids tetrahydrocannabinol (THC) and cannabidiol (CBD). The current study serves to elucidate which receptors could be mediating the change in escape response behaviour following cannabinoid exposure. I hypothesize that TRPV1 channels are involved in CBD and THC disruptive effects on C-start responses to A/V stimuli due to their potential to be acted upon by cannabinoids and their presence in sensory systems, including the systems that sense acoustic stimuli. To test our hypothesis, we will treat 5 days post fertilization zebrafish with the TRPV1 antagonist A-784168 and then use a co-application of 1:2 THC: CBD before testing the acoustic evoked escape response. This research aims to provide a greater understanding of cannabis and its action within developing organisms in order to fully understand the potential and risk of using cannabis as a therapeutic.

Differential Mechanisms of Toxicities of Hypochlorous Acid and Chloramine-T in Oncorhynchus mykiss

Derek Lum, Undergraduate Student [1], Victoria Goncalves, Edyta Jasinska, Arian Farajizadeh, Greg G. Goss
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Accidental environmental releases of chlorines and chloramines into aquatic environments are highly toxic to aquatic life. To date, there are no investigations comparing the relative mechanisms of toxicity of hypochlorous acid (HA) to chloramine-T (CT). This study examined the differences between HA and CT toxicity in Oncorhynchus mykiss by exposing fish to either control, 0.5 or 1.0 mg/L HA or CT, followed by measurement of loss of equilibrium (LOE), methemoglobin formation and

clearance rate, hypoxia-induced LOE, oxidative stress in gill and liver tissues, and histology of gill tissues. To measure time to LOE, O. mykiss were continuously exposed to either 0.5 or 1.0 mg/L HA or CT until LOE occurred. LOE occurred fastest when fish were exposed to 1.0 mg/L HA (0.38 +/- 0.03 h) followed by 1.0 mg/L CT (0.68 +/- 0.07 h), HA 0.5 mg/L (0.87 +/- 0.14 h) and CT 0.5 mg/L (1.38 +/- 0.07 h). To examine post-exposure effects, fish were exposed to either HA or CT for 15 or 30 min, returned to freshwater, and exposed to graded hypoxia, with PO2 at LOE recorded. O. mykiss were more sensitive to HA (at 1.0 mg/L and 0.5 mg/L) than by CT at 1.0 mg/L and 0.5 mg/L. However, metHb formation in red blood cells occurred at a faster rate following to CT exposure at 0.5 mg/L compared to HA at 0.5 mg/L, with both treatments showing similar clearance rates, taking almost 24 h to return metHb to control levels. Both exposures showed significant and sustained increases in oxidative stress in gill tissues as reported by the TBARS assay while liver tissues did not show any significant changes in TBARS.

Investigating Abnormal Morphology and the Role of Movement in Rescuing Development of the CHRNA1 Zebrafish Mutant

Anna Marks, Undergraduate Student [1], Gabriel Dillenburg De Oliveira, Zach Hall, PhD
[1] Department of Physiology, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Zebrafish (Danio rerio) are a common model organism used for genetic and developmental research for several reasons. One difficulty encountered when performing experiments on this species is that after 3 days post-fertilization (dpf), zebrafish start swimming. In recent years, a mutation in the chrna1 gene has been proposed as a convenient method to immobilize zebrafish as this mutation paralyzes the animal. Any limitations of the chrna1 mutant have yet to be reported in the literature, even though models are born without the ability to move which has a high likelihood of negatively affecting development. Chrna1 null mutants will be generated in zebrafish with Cas9-CRISPR mutagenesis. All fish will be repeatedly imaged on days 2, 4, and 6 post-fertilization. With these images, eight morphometric measurements will be taken of the body and eyes. Analysis will also be performed on data collected from polarized light microscopy to assess muscular integrity in both controls and mutants. During the second round of data collection, fish will be placed on a rotator protocol from 3-6 dpf. This will aim to simulate movement during the period in which zebrafish naturally start to move and allows us to study the effects of reintroducing movement. During our first round of experiments, chrna1 mutants exhibited significantly different morphometric measurements from controls in all but body length at 6 dpf. Experimentation will continue to assess differences in muscular integrity and response to the rotator protocol. This project will primarily focus on assessing the suitability of the chrna1 zebrafish mutant as a model for future research. In addition to characterizing defects observed in mutants, we will also be able to compare measurements obtained from chrna1 mutants before and after introducing movement, which allows for further investigation into how movement and movement-dependent sensory input are vital for the normal development of vertebrates.

The Role of Soluble Epoxide Hydrolase Genetic Deletion on Cellular Senescence Following Myocardial Infarction

Andy Byeongin Lee, Undergraduate Student [1], Liye Fang, Ala Yousef, John Seubert
[1] Department of Physiology, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Aging is a risk factor for cardiovascular disease. As individuals age, some vulnerable cells will undergo senescence, defined as the irreversible arrest of the cell cycle. The development of cellular senescence can be induced by accumulated cellular stress with aging, such as oxidative damage and mitochondrial dysfunction. In the heart, senescent cardiomyocytes are characterized with mitochondrial dysfunction and exhibit senescence associated secretory phenotype (SASP). Recently, it has been demonstrated that myocardial infarction following ischemia can lead to cellular senescence in the heart, thus contributing to worsened cardiac function. N-3 and N-6 polyunsaturated fatty acids are metabolized by CYP epoxygenases into bioactive lipid mediators, epoxylipids, which provide a protective response toward cardiac injury. These epoxylipids are readily metabolized by soluble epoxide hydrolase (sEH) into less bioactive molecules. Previous evidence demonstrates the genetic deletion of sEH protects hearts in aged mice from ischemic injury. However, our understanding of the protective mechanisms remains limited. We propose the deletion of sEH will attenuate cellular senescence and limit cellular stress following ischemic injury. In this study, we used wildtype and sEH null male and female mice aged 15 to 21 months old. The mice underwent permanent ligation of the left anterior descending artery to induce ischemic injury, and were sacrificed 7 days later to collect tissues. Real-time qPCR was used to assess gene expression in RNA isolated from hearts. Results indicate that senescence markers, p21 and p16, are attenuated with sEH genetic deletion in aged male but not female mice following injury. Furthermore, sEH genetic deletion reduced the characteristic SASP phenotype found in cardiac senescent

cells, with lower expression of MCP-1 and IL-6. This study indicates an adverse role of senescence in ischemic injury which is attenuated by the genetic deletion of sEH.

Addressing Functional Redundancy in a Set of Genes Necessary for Female Meiosis II in C. elegans

Muneeb Rehman, Undergraduate Student [1], Martin Srayko
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Gene duplication is an important mechanism for introducing new material for evolutionary forces to act upon. The initial state of these duplicates is complete functional redundancy, in which one gene can compensate for the loss of the other. However, the genes can diverge from each other and result in novel functionality. The Srayko lab identified a set of highly similar genes called memi-1, memi-2, and memi-3 (meiosis-to-mitosis defective) within Caenorhabditis elegans. The memi gene family is maternally expressed and the MEMI proteins are present during female meiotic cell divisions but are degraded prior to the subsequent first embryonic mitosis. A deletion strain of memi-1 alone has no phenotype, but combinations with memi-2 and memi-3 deletions exhibit embryonic phenotypes, suggesting the family is functionally redundant. For example, loss of memi-1, 2, and 3 function resulted in embryos that skip meiosis II entirely and progress straight into unregulated mitosis. However, a temperature-sensitive, gain-of-function mutant called memi-1(sb41), resulted in normal meiosis II, but failure to transition from meiosis II to the first embryonic mitosis. Thus, it was suggested that memi-1(sb41) disrupts the natural degradation of MEMI-1 proteins during the meiosis-mitosis transition, leading to embryonic lethality. Despite the high sequence similarity between memi genes, it is still not clear whether they are functionally equivalent. For example, some combinations of memi deletions are more lethal than others. Furthermore, it is not clear whether the memi-1(sb41) mutant phenotype is unique to memi-1. To address this, the memi-1(sb41) mutation was recreated in memi-2 and memi-3 through CRISPR/Cas9 methods. We predict that these mutants will be phenotypically similar to memi-1(sb41) due to predicted functional redundancy. The results of this experiment could yield important information on the scope of redundancy within the MEMI family and serve as an illustration of how to study functional redundancy via guided mutagenesis.

Poster Presentations in Immunology and Infection

Investigating the Role of DNA G-Quadruplexes in Anti-tumour Immunity in Colon Cancer

Natalie Lapa, Undergraduate Student [1], Shayla Mosley, Kristi Baker
[1] Department of Cell Biology, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Colorectal cancer (CRC) is the second leading cause of cancer-related deaths in Canada. The two major subtypes of CRC, chromosomally instable (CIN) and microsatellite instable (MSI), are respectively characterized by genomic instability and mismatch repair deficiencies. MSI colorectal cancer is associated with better patient prognosis compared to CIN, which may be due to the greater immunogenicity of MSI tumors. MSI tumors recruit higher numbers of cytotoxic T cells than CIN tumors. Previous research in our lab indicated that this enhanced T cell recruitment depends on cGAS/STING-mediated chemokine release from cancer cells and immune cells within the tumor. The cGAS/STING pathway can be activated by endogenous DNA that has leaked out of the nucleus, termed cytosolic DNA (cyDNA). Preceding experiments in our lab demonstrated that cyDNA isolated from MSI CRC cells is more stimulatory to the cGAS/STING pathway in vitro than cyDNA from CIN CRC cells. Sequencing the cyDNA from both subtypes revealed that MSI cyDNA contains a greater number of putative secondary structures, including G-quadruplexes (G4s). Using immunofluorescence microscopy, we aimed to determine if G-quadruplexes are forming within CRC cells. We also aimed to isolate G4-containing cyDNA from CRC cells via immunoprecipitation. We evaluated the stimulatory capacity of these sequences on DCs using in vitro stimulations and subsequently tracking gene expression via qPCR and Western blotting. Our preliminary results indicate that G4s are detectable in the cytosol of cancer cells. We have optimized the immunoprecipitation of G4s and confirmed the protocol's specificity for these sequences using synthetic G4 oligos. Our initial stimulation of DCs with G4-enriched DNA indicate that these sequences induce higher expression of inflammatory chemokines than non-enriched cyDNA. Our findings point to one explanation for the differing immunogenicities of the CRC subtypes, and could thus inform the design of therapeutics for CIN tumors.

Determining the Role of the Transcription Factor Helios on T Cell Development and Tolerance

Nikhil Bhatnagar, Undergraduate Student [1], Julia May, Troy Baldwin
[1] Department of Medical Microbiology and Immunology, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Proper T cell development is conditional on T cell (Tc) receptor interactions with self peptide MHC complexes. Recognition of self MHC leads to positive selection, while too strong of an interaction leads to negative selection by clonal deletion or anergy. Both positive and negative selection are characterized by changes in gene expression, such as the induction of various transcription factors; these act to regulate T cell differentiation. Helios is one such transcription factor, being upregulated specifically in single positive Tc that strongly bind to self antigens. This induction suggests it may have a role specific to the process of negative selection. We are interested in the role Helios has in developing and maintaining Tc tolerance. Our method included creating OT-1 > Rip-mOVA Helios wildtype (WT) or knockout (KO) bone marrow chimeras into irradiated mice. The purpose is to study the function of Helios in naive Tc, and whether it has an effect on the development of tolerance. We also adoptively transferred both WT and Helios KO CD8+ Tc into RIP recipients that express high affinity self antigen (OVA) to monitor the effect of Helios on maintaining tolerance of mature Tc. If Helios does contribute to negative selection and maintenance of tolerance, we should see the emergence of diabetes in the recipient mice. At the current time, there has been no change in the blood glucose levels of the recipients, indicating no emergence of diabetes. Our study aims to further elucidate the function of Helios in the process of developing and maintaining Tc tolerance.

The Pathogenicity of a Truncated Influenza A Virus PB1-F2 Protein

Angelina Botros, Undergraduate Student [1], Ximena Fleming-Canepa, Katharine E. Magor
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Polymerase basic 1 frame 2 (PB1-F2) is a non-structural influenza A virus protein that interacts with the mitochondrial antiviral-signaling protein (MAVS) pathway to inhibit interferon production and induce cell death. The PB1-F2 gene transcript in many mammalian influenza A virus strains contains premature stop codons, resulting in a truncated PB1-F2 protein. It is currently unknown if a truncated PB1-F2 protein would properly induce interferon inhibition in the MAVS pathway. In our hypothesis, a truncated PB1-F2 protein would be insufficient to induce an inhibitory interferon response because it lacks the basic C-terminus amphipathic helix, which targets the MAVS protein. To investigate, we used western blot analysis to determine which reading frames of PB1-F2 were being expressed, followed by luciferase assays to measure levels of interferon inhibition. Our preliminary results show that the truncated PB1-F2 protein possesses a degree of interferon inhibitory capability despite the absence of the basic C-terminal amphipathic helix. We expect subsequent results to demonstrate the expression of different reading frames within PB1-F2, which may provide an explanation of this phenomenon. Understanding how PB1-F2 truncation affects influenza A pathogenicity may provide insight into its evolutionary history and an understanding of additional alternate pathways PB1-F2 may take to inhibit interferon production.

Investigating the Functionality of Natural Immune Fighter Cells

Chloe Church, Undergraduate Student [1], Luisito Padua, Shokrollah Elahi
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Innate fighter cells of the immune system are the Natural killer (NK) cells, killing abnormal cells within the body such as virus-infected cells or cancerous cells. These cells release cytotoxic substances, such as perforin and granzymes to target and destroy infected and cancer cells. NK cell function is sophisticated and tightly regulated by its cell surface receptors including VISTA, Gal-3, and PD-1. This project investigates the frequency of NK cells expressing these molecules within the spleen, bone marrow, and lungs of adult and neonatal mice. In particular, each of these cell subsets were subjected to flow cytometry staining which measures the proportion of surface and intracellular proteins associated with NK cell function. To evaluate the functionality of NK cells, different activation markers were used (CD25, KLRG1 and Sca-1). Our findings have provided novel insights into the role of VISTA, Gal-3, and PD-1 in the functionality of NK cells. Investigating the variation in the functionality and phenotype of NK cells between neonatal versus adult mice offers insight into the development and potency of the immune system. As well, divergences found between the immune performance of male and female mice suggests possible reasoning for gender heterogeneity in human therapeutic responses. This novel information can potentially provide a new direction in the development of immune treatments for various ailments.

Poster Presentations in Health Sciences

An Investigation of Biomarker and Common, Chronic Disease Relationships

Jiaxuan Wang, Undergraduate Student [1], Gane Ka-Shu Wong, David Wishart
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Biomarkers are intrinsic physiological indicators crucial in identifying pathological states within the body. They can take the form of proteins, nucleic acids, lipids, carbohydrates, or small metabolites. Each type serves as a "molecular fingerprint," offering insights into the biochemical landscape and its association with disease processes. These biomarkers are pivotal in predicting or diagnosing diseases, yet their relationship with disease outcome and progression is complex and non-linear. This complexity is compounded by the multifaceted nature of many diseases, variations in scientific understanding, and inconsistent definitions within the literature. This project aims to explore the relationship between the top 10 biomarkers for five common, chronic diseases. A key goal is to synthesize and consolidate a better understanding of both the disease in question and the biomarkers themselves. We intend to categorize these biomarkers according to their diagnostic, prognostic, and predictive roles in clinical applications. Our analytical approach employs the receiver operating characteristic (ROC) curve to assess diagnostic accuracy, odds ratios (OR) for evaluating association strengths, and applying Cox proportional hazards models to determine prognostic values, prioritizing biomarkers based on the credibility of the supporting research. Overall, this research aims to identify critical biomarkers for disease prediction and diagnosis, offering potential targets for drug discovery and therapeutic intervention. By focusing on these significant biomarkers, we aim to facilitate the development of more effective treatments and enhance patient care in managing chronic diseases.

N-Of-1 Short-Term Metabolomic Response to Various Stressors

Kellie Yang, Undergraduate Student [1], Dorsa Rayat, Tammy Zheng, Eponine Oler , Fatma Elessawy, Ashley Zubkowski, David Wishart
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

There is an escalating need for innovative approaches to combat our multifaceted healthcare crisis. This crisis is due to a culmination of inflammatory diets, alcohol and tobacco use, sedentary lifestyles, social and environmental factors, and exacerbated by inaccurate lab results/interpretations. The inadequacies of the current system necessitate a shift towards personalized approaches, since the traditional reliance on population baselines often leads to late diagnoses and symptomatic management. Broadly speaking, this study proposes metabolomics (the study of small molecules) as a powerful tool in establishing precise personal baselines inexpensively with low volumes of biofluids. Doing so could enable us to identify disease onset, progression, and risk factors for an individual on a molecular level. The biggest barrier in using metabolomics to assess our health however, is that many metabolites tend to fluctuate widely and rapidly even within the same individual; thus, this study investigates the short term metabolomic changes in response to various stressors including fasting, sleep deprivation, dietary changes, heat, and exercise stress. This was done by taking samples of my own blood using a self-administered Tasso every 1.5 hours from 9:30am to 7:30pm, one day per stress condition. The samples were then prepared using the TMIC MEGA assay, which detects 645 metabolites of various chemical classes (such as amino acids, biogenic amines, organic acids, lipids, acylcarnitines etc.) using liquid chromatography–mass spectrometry (LC-MS). Early results demonstrate that many metabolites follow a distinct trend, a.) throughout the day and b.) in response to stressors. These data imply that it's possible to explain and correct for intrapersonal variation in metabolite concentrations that may otherwise be too noisy to provide an overall picture of our health.

Development of a Novel Fluorine-18 Labeled Heparanase Targeting Radiotracer for PET of Cancer

Livia Mangin, Undergraduate Student [1], Travis Kronemann, Ralf Schirrmacher
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Heparanase is an enzyme that degrades heparan sulfate chains, leading to disassembly of the extracellular matrix and alterations in cellular integrity. Although heparanase is rarely expressed under normal conditions, it has a near universal overexpression in cancerous tumors. This overexpression of heparanase increases tumor growth and aggressiveness by facilitating angiogenesis, metastasis, and chemoresistance, which all contribute to worsened patient prognosis. Accordingly, much research has been done on synthesizing heparanase inhibitors. A leading candidate among these inhibitors is pixatimod (formerly PG545), a heparan mimic that's currently in stage III of clinical trials for the treatment of various cancers. It is a persulfated tetrasaccharide bearing a steroidal aglycone, with a 12.0 nM IC50, making it suitable for tracer development. Fluorine-18 is an attractive radioisotope for PET as its half-life of 110 min allows for prolonged image acquisition, and its low positron emission energy (250 keV) provides highly resolved images. We aim to introduce a silicone-fluoride acceptor (SiFA) group to pixatimod, allowing for rapid fluorine-18 incorporation via an isotopic exchange reaction. The steroidal aglycone is an optimal site for SiFA introduction as it may potentially improve the functions of the aglycone through added lipophilicity. Once the tracer is obtained, heparanase binding affinity will be assessed on a novel assay which will take place in tandem with fluorine-18 radiolabeling optimizations in preparations for assessments in animal models. If successful, this radiotracer has great potential to help with diagnosis, staging, and monitoring of cancer to improve patient care quality. A number of steps remain in the synthesis of the proposed radiotracer, though several reactions along the route were optimized this summer, working with a more cost-effective model, B-D-maltose monohydrate. Future steps will include continued reaction optimizations, followed by work on the actual tetrasaccharide, radio-labeling, and assessment in animal trials.

Relative Size of Forebrain-Cerebellar Pathways in Corvids

Mansi More, Undergraduate Student [1], Lauren Appel, Cristián Gutiérrez-Ibáñez, Andrew N. Iwaniuk, Douglas R. Wylie
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Parrots and corvids (crows, ravens, rooks, magpies) are two groups of birds that have evolved complex cognitive abilities similar to primates. In birds and mammals, convergent evolution of cognitive abilities is associated with similar changes in the brain, specifically, the increase in whole cortex/telencephalon size. In primates, an increase in the cortex size has been associated with an increase in the size of the cerebellum and concomitant enlargement of areas that connect these two structures, particularly the pontine nuclei. Previous research has shown that in parrots, like in primates, there is an increase in the relative size of structures that connect the forebrain to the cerebellum. Specifically, the medial spiriform nucleus (SpM), a structure not found in mammals, is enlarged in parrots. Interestingly, the pontine nuclei of parrots are not enlarged. Given the similar cognitive abilities and a similar increase of forebrain size between parrots and corvids, in this study we test if structures that connect the forebrain with the cerebellum are enlarged in corvids. Utilizing an existent collection of sectioned and nissl stained samples, coronal sections of the PM, PL, and SpM were photographed and relative size of the structure was measured. The collection consists of 80 specimens, with 4 corvid species and 40 alternative songbird species. Due to the ongoing nature of this project, results and analyses of the 4 corvid specimens will be presented. Preliminary results show no enlargements of the SpM or pontine nuclei in corvids. Completion of this project will provide an enriched understanding of how telencephalic-midbrain-cerebellar pathways of birds have an important role in controlling fine motor skills and complex cognitive functions.

DG9-Pmo Conjugates Boost Cellular Uptake, Nuclear Localization, and Prolong Benefits in a Murine Model of Spinal Muscular Atrophy

Melissa Kohut, Undergraduate Student [1], Umme Sabrina Haque, Toshifumi Yokota
[1] Department of NeuroScience, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

Introduction: Spinal muscular atrophy (SMA) is an autosomal recessive neuromuscular disease caused by mutations in the SMN1 gene, leading to reduced levels of the survival motor neuron (SMN) protein. Splice-switching antisense oligonucleotide (ASO)-based therapies have emerged as a promising strategy for SMA treatment. Although FDA-approved, limitations such as invasive spinal injections, high expenses, and limited multi-organ impact exist. We recently identified a novel class of cell-penetrating peptide named DG9 that significantly improved the systemic effectiveness of ASO-based therapy when conjugated with phosphorodiamidate morpholino oligomers (DG9-PMO), outperforming both FDA-approved ASO called 2'-O-methoxyetyl (MOE) and benchmark peptide R6G in a severe mouse model of SMA. However, the exact

mechanisms by which DG9 improves its effectiveness were unclear. Here, we investigated the ability of DG9-PMO to enhance cellular uptake and nuclear localization, and assessed its therapeutic effects in the murine model of SMA across three treatment schedules. Methods: A neuroblastoma cell line was used to examine DG9's ability to increase cell infiltration and distribution to the nucleus with immunocytochemistry and live cell imaging. Severe SMA mice were injected with either the DG9-PMO or MOE once subcutaneously on postnatal day (PD) 0 for group 1, PD 0 and 2 for group 2, or four times on PD 0, 2, 28, and 56 for group 3. Body weights, survival, and functional testing were used to analyze motor improvement. Results: DG9 peptide improved the PMOs' uptake, endosomal escape, and nuclear localization. Group 3 DG9-PMO treatment showed increased survival, body weight, and motor capabilities compared to other groups. The DG9-PMO treatment surpassed MOE in increasing tail length and delaying necrosis onset. Discussion: Overall, these experiments indicate the potential of using DG9-PMO as a therapeutic for SMA without invasive intrathecal injections and the possibility of conjugating DG9 for other therapeutics.

Investigating Sharp-Force Trauma on Cervus canadensis Limbs through Pyro-Forensic Analysis

Tristan Mula, Undergraduate Student [1]
[1] Department of Biological Sciences, Faculty of Science, University of Alberta, Edmonton, Alberta, Canada

This study addresses the challenges associated with identifying patterns of sharp-force trauma in burned skeletal remains. With a rising trend in violent crimes involving sharp objects, the research focuses on the impact of heat-induced alterations on bone structures subjected to mechanical sharp-force trauma. The study used Cervus canadensis (elk) limbs as surrogates for human bones, investigating the kind of trauma produced by three blade types—hatchet, circular saw, and crosscut hand saw. The bones undergo controlled burning; qualitative and quantitative analyses are conducted using the Keyence microscope and naked-eye observations. Findings reveal persistent features in cut marks after burning, offering opportunities for identifying blade types. The Mann-Whitney U-Test and Kurskal-Wallis tests show no significant differences in median reduction of cut length or width based on bone or weapon type. This research contributes to forensic protocols, emphasizing the importance of considering bone and weapon types in interpreting burned skeletal remains and enhancing the credibility in legal proceedings.

Conflicts of Interest

There are no conflicts of interest.

Authors' Contributions

LRM: served as 'Primary Organizer' for the conference, helped draft the conference abstract booklet, reviewed the abstract submissions and ensured that they adhered to correct formatting standards, and gave final approval of the version to be published.
CLL: served as 'Student Presentations' volunteer for the conference, helped draft the conference abstract booklet, reviewed the abstract submissions and ensured that they adhered to correct formatting standards, and gave final approval of the version to be published.
AE: served as 'Student Presentations' volunteer for the conference, helped draft the conference abstract booklet.
PW: served as 'Student Presentations' volunteer for the conference, helped draft the conference abstract booklet.

Acknowledgements

Thank you to the entire R.E. Peter Biology Conference volunteer team including: Sam Tan (Secondary Organizer), Dana Guhle and Laura Manerus (Finance), Paris Brown and Ilia Promenov (Conference Mechanics), Connor Stewart and Jenelle McCuaig (Judges and Prizes), Sofia Panzetta and Calla McKague (Conference Events), Sophia Huang, Brittany Sauter and Sissi Yuli (Keynote Speakers), Caitlin Hall (Social Media), Evan Amyotte (Graphic Design).

Funding

University of Alberta Department of Biological Sciences, University of Alberta Faculty of Science, University of Alberta Faculty of Graduate Studies and Research, University of Alberta Graduate Students' Association, Alberta Professional Biologists Association, Genetics Faculty Library Endowment: Created in 1986 by donations from the Faculty members in the Department of Genetics to support Genetics Research and Education, University of Alberta Campus Sustainability Grant Program, Arbour, Safeway.

Article Information
Managing Editor: Jeremy Y. Ng
Article Dates: Received Feb 29 24; Published Mar 13 24

Citation
Please cite this article as follows:
MacLeod LR, Lopez CL, Etuk A, Wickramasinghe P. Richard E. Peter 15th Annual Biology Conference (2024). URNCST Journal. 2024 Mar 13: 8(3). https://urncst.com/index.php/urncst/article/view/585
DOI Link: https://doi.org/10.26685/urncst.585