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Health Sciences
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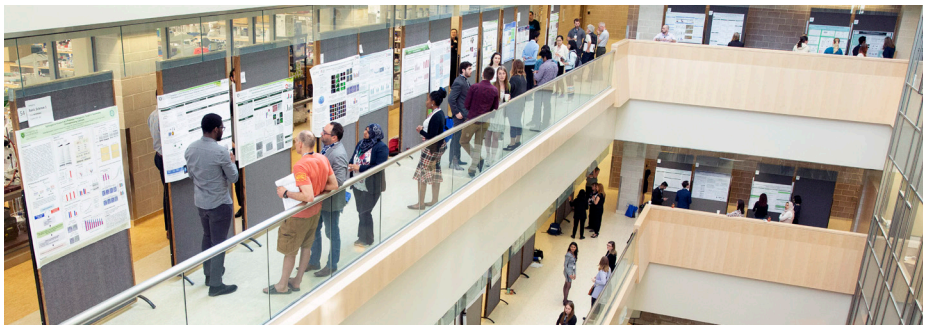
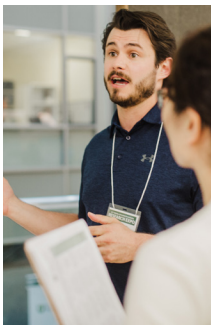
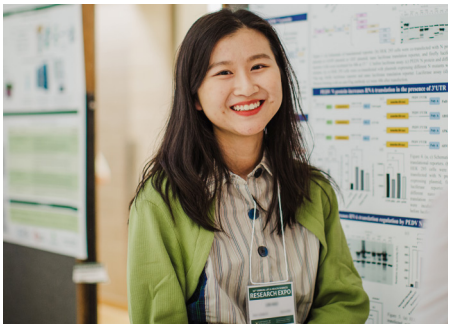
THE 2024 LIFE & HEALTH SCIENCES

RESEARCH EXPO





ACKNOWLEDGING EXEMPLARY RESEARCH AND LEARNING AT THE UNIVERSITY OF SASKATCHEWAN

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Research competition abstracts

Agenda

09:30 – 11:30 a.m.	**STUDENT REGISTRATION*** D-Wing, outside Room 1B21, Health Sciences Building ***Registration and poster setup begin 30 minutes prior to each presentation session.***
10:00 – 11:30 a.m.	**MORNING POSTER SESSION #1** D-Wing Atrium, 2nd floor, Health Sciences Building • Undergraduate Research 1 • Basic Science 1 • Translational, Clinical or Applied Science 2 • Social & Population Health 1
11:00 a.m. – 12:30 p.m.	**AFTERNOON POSTER SESSION #1** D-Wing Atrium, 3rd floor, Health Sciences Building • Undergraduate 2 • Basic Science 2 • Basic Science 4 • Translational, Clinical, or Applied Science 3
12:00 – 1:30 p.m.	**AFTERNOON POSTER SESSION #2** D-Wing Atrium, 2nd floor, Health Sciences Building • Undergraduate Research 3 • Basic Science 3 • Translational, Clinical, or Applied Science 1 • Social & Population Health 2
1:30 – 2:30 p.m.	**NETWORKING BREAK (COFFEE & SNACKS PROVIDED)** B-Wing Foyer (HLTH GB01), Health Sciences Building
2:30 – 3:45 p.m.	**AWARDS CEREMONY AND SPECIAL PRESENTATIONS** HLTH GB03, Health Sciences Building

* Locations listed on the agenda can be found on this map on the USask Health Sciences expo website.

Undergraduate Research 1

1. INVESTIGATING AVASIMIBE AS A NOVEL THERAPEUTIC FOR TARGETING HEPATIC CHOLESTEROL CRYSTALLIZATION

Presenter:	**Maria Zafar**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Collaborators:	May Akl
Supervisor:	Scott Widenmaier, College of Arts and Science

Background:

Dysregulated cholesterol homeostasis is increasingly implicated in steatotic liver disease. Specifically, metabolic- dysfunction associated steatohepatitis (MASH), an increasingly prevalent liver pathology, is linked to excess free (non-esterified) liver cholesterol, which leads to the formation of hepatic cholesterol crystals that are indicated to distinguish MASH from benign steatosis. Intracellular cholesterol crystals are suspected to drive MASH pathogenesis by triggering inflammation however the mechanisms underlying hepatic cholesterol crystal formation remain unclear. Cholesterol esters localize at lipid droplets and Sterol O-acyltransferase (SOAT1) is required for esterification to concentrate cholesterol at the lipid droplet; this may underpin cholesterol crystal formation. Hence, we hypothesized that inhibiting SOAT1 activity, using Avasimibe, will prevent hepatic cholesterol crystallization by inhibiting cholesterol esterification.

Methods:

Here, we employed a previously established accelerated murine model of MASH to investigate the effect of SOAT1-inhibiting Avasimibe on the in vivo development of cholesterol crystals and MASH phenotype. Enzymatic cholesterol assays and polarized light microscopy were used to evaluate cholesterol metabolism and its impact on crystal formation.

Results:

Avasimibe successfully inhibited SOAT1 activity in mice. Decreased cholesterol esterification was found in Avasimibe-treated livers alongside blocked hepatic cholesterol crystallization. Tissue scoring revealed slight improvement in liver phenotype upon drug treatment however this was not statistically significant.

Conclusion:

Avasimibe is an effective SOAT1 inhibitor in vivo. Reduced esterification protects against cholesterol crystal formation. SOAT1 inhibition may therefore serve as a novel target for hepatic cholesterol crystallization observed in MASH.

2. EFFECT OF FILTER STERILIZATION ON THE PROTEOMIC PROFILE OF SALIVA

Presenter:	**Feras Balbous**
USask Affiliation:	**Undergraduate student**
College:	**College of Dentistry**
Collaborators:	Walter Siquiera, College of Dentistry
Supervisor:	Lina Marin, College of Dentistry

Background:
Contamination of sterile human saliva with internal microorganisms poses a significant challenge for in-vitro studies. Filtration is a standard method of removing microorganisms from saliva. Studies have compared saliva filtration with other sterilization techniques, but the impact of different filtration systems on saliva's protein composition is unexplored. Objective: This study assessed the effect of six common filtration systems on the proteomics profile of saliva.

Methods:
Human saliva was collected from five healthy individuals. Filters made from Polyethersulfone (PES), Cellulose Nitrate (CN), and Surfactant-free Cellulose Acetate (SFCA), with pore sizes of 0.2µm and 0.45µm, were evaluated. Filtrate saliva was incubated on Brain Heart Infusion agar to check for sterility. Bicinchoninic acid (BCA) assay and tandem mass spectrometry were used to evaluate any changes to the protein composition of saliva. The experiment was repeated six times (n=6). Kruskal-Wallis and subsequent Dunn's testing were used to assess the results of the BCA assay (α=1%).

Results:
A total of 125 proteins were identified, with six proteins exclusive to Whole Saliva Supernatant (WSS) and only one protein exclusive to filtered saliva (SFCA 0.45µm). A significant decrease in protein concentration was observed ($p<.01$) in both CN filters while also affecting the proteomic profile of human saliva. SFCA and PES filters had little to no effect on protein concentration and composition. All filters, regardless of pore size, achieved effective sterilization.

Conclusion:
This research highlights the role of filtration systems in shaping saliva's protein composition, emphasizing the need for careful selection of the appropriate filtration system. Future research should assess the impact of filtration on the acquired pellicle and focus on comprehensive standardization of saliva collection and analysis methodologies to advance the development of national/international saliva banks for diagnostic application.

3. THE EFFECT OF A1 KNOCKOUT ON SYNAPTIC TARGETS IN A NEURONAL MODEL OF MULTIPLE SCLEROSIS

Presenter:	**Kaitland Fior**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Collaborators:	Hannah Salapa, College of Medicine
Supervisor:	Michael Levin, College of Medicine

Background:

Multiple sclerosis (MS) is a disease of the brain and spinal cord that results in progressive and extensive neuronal loss, or neurodegeneration. Many neurodegenerative diseases, including MS, report synaptic pathology, however the processes underlying synaptic alterations in these diseases are poorly understood. Dysfunction of the RNA binding protein heterogeneous nuclear ribonucleoprotein A1 (A1) is a defining characteristic of MS and its models. Previous RNA sequencing performed by our lab indicates A1 plays an extensive role in RNA metabolism in neurons and is involved in regulating numerous synapse-related RNA targets. We therefore hypothesize that A1 dysfunction will disrupt synaptic processes due to changes in RNA abundance and splicing of synaptic RNA targets, which may contribute to neurodegeneration in MS.

Methods:

Neuro-2a cells, a neuronal cell line, were transfected with a plasmid containing Cas9 and single guide RNA targeting A1 (sgA1) to knockout A1. 96-hours post-transfection, protein and RNA were collected to (i) confirm A1 knockout via western blotting and (ii) examine changes in abundance and splicing of synaptic targets through qPCR and amplicon PCR, respectively.

Results:

Neuro-2a cells that were transfected with the plasmid to knockout A1 showed a greater than 95% decrease in A1 protein expression compared to control cells (****$p<0.001$, unpaired t-test). RT-qPCR results demonstrated that nine synapse-related RNA targets were significantly downregulated, Eno2, Sema6a, Syt5, Atp1a3, Dtnb, Gabbr1, Ppfia3, Aplp1, Dlg4 and two RNA targets were significantly upregulated, Syt1 and Arpp21 (*$p<0.05$ for all) in cells with A1 knockout compared to controls. In addition, three RNA targets were alternatively spliced following A1 knockout, Klhl2, Abat and Stmn4 (*$p<0.05$ for all).

Conclusion:

These results suggest A1 dysfunction impairs processing of numerous synaptic RNA targets through changes in RNA expression and splicing, which may contribute to synaptic pathology reported in MS.

4. METHOD DEVELOPMENT AND VALIDATION OF REVERSE-PHASE HPLC ASSAY FOR QUANTIFICATION OF MYCOPHENOLIC ACID AND PLASMA EXTRACTION PROCEDURES

Presenter:	**Adrienne Nagy**
USask Affiliation:	**Undergraduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Ellen Wasan, College of Pharmacy and Nutrition
	Deborah Michel, College of Pharmacy and Nutrition
Supervisor:	Ellen Wasan, College of Pharmacy and Nutrition

Background:
We have developed modified release formulations of the immunosuppressant drug mycophenolate mofetil (MMF) to simplify dosing regimens for enhancing treatment adherence in organ transplant patients. In preparation for preclinical pharmacokinetic studies, the objective of this project was to develop a validated reverse-phase high-pressure liquid chromatography (HPLC) assay for the quantification of mycophenolic acid (MPA) extracted from rat plasma. Mycophenolic acid is the active metabolite of mycophenolate mofetil.

Methods:
Initial assay conditions were based on a reverse-phase HPLC assay developed for quantification of parent compound mycophenolate mofetil (MMF) in mobile phase. Hypotheses based on the chemical differences between MPA and MMF were formulated to guide method development: decreasing mobile phase pH will improve analyte peak uniformity, gradient flow conditions will improve column retention and timely elution, and acetonitrile as organic mobile phase will improve elution strength.

Results:
An optimized extraction procedure was developed from rat plasma using Captiva Agilent EMR-Lipid 96-Well Plates, with >90% extraction recovery accounting for matrix effect. The validated HPLC assay for quantification of MPA extracted from rat plasma employed a gradient flow mobile phase comprised of TFA 0.03% in water, and acetonitrile on a Phenomenex Luna 5 um C8 100 Å 30 x 2mm column at 37◦C. Median retention time was 2.57 +/- 0.01 min with <1% difference inter and intra-day, and the linear range was1-45 µg/mL with a detection limit of 0.4 µg/mL, and linearity (r2) >0.99. This high sensitivity encompasses the expected range of plasma concentrations anticipated for the planned pharmacokinetic analysis. Inter-day accuracy varied from 92-118% for the lowest limit of quantitation (LLOQ), and 85-114% for quality controls; inter-day precision was <15%.

Conclusion:
This validated HPLC assay will be used to analyze serum concentrations of mycophenolic acid (MPA) in upcoming preclinical pharmacokinetic study providing accurate and sensitive pharmacokinetic data to support future clinical studies of sustained release formulations of MMF.

5. PHARMACOLOGICAL ASSESSMENT OF SYNTHETIC CANNABINOID LIGANDS OF TYPE 1 CANNABINOID RECEPTOR (CB1R) FOR THERAPEUTIC POTENTIAL AS PAIN RELIEVERS

Presenter:	**Andy Kim**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Collaborators:	Alayna Jones, College of Arts and Science
Supervisor:	Robert Laprairie, College of Pharmacy and Nutrition

Background:
Chronic pain is one of the most prevalent complaints amongst patients presenting as an immense burden to the health care system and the economy. The management of pain requires the administration of analgesics, medications to relieve said pain. There is growing evidence that the endogenous cannabinoid system, specifically the type 1 cannabinoid receptor (CB1R), is a target for pain relief. Further evidence supports positive allosteric modulation of the CB1R to have substantial therapeutic potential for pain relief.

Methods:
Therefore, this study aimed to characterize 9 synthetic agonist-positive allosteric modulators (PAMs) of the CB1R both in vitro and in vivo. The inhibition of cAMP accumulation, a critical secondary messenger of synaptic transmission and consequently integration of noxious stimuli, by these compounds were evaluated in vitro assessing agonism and allosteric modulation of the receptor in the presence of an orthosteric CB1R agonist Δ9-tetrahydrocannabinol (THC). The top 2 most potent PAMs were then selected to be evaluated in vivo characterizing 4 phenotypic effects described as the cannabinoid-induced tetrad. These effects are catalepsy (an impaired ability to initiate movement), hypothermia (a decrease in body temperature), analgesia (decrease in pain sensitivity), and hypo-locomotion (reduced movement).

Results:
We observed that all compounds displayed agonism at the CB1R in vitro but GAT 2510 and GAT 2513 were the most potent PAMs. In vivo testing of these compounds are on-going and awaiting results.

Conclusion:
The findings of this study provide valuable information about the untapped potential of CB1R as a therapeutic target for analgesic development. With each result, we are one step closer to fully understanding the complexity of the CB1R and potentially identifying novel pain-relieving drugs.

6. SYSTEMIC GENOME CORRELATION LOSS AS A CENTRAL CHARACTERISTIC OF SPACEFLIGHT

Presenter:	**Anurag Sakharkar**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Collaborators:	Erique Lukong, College of Medicine
	Lauren Sanders
	Sylvain Costes
Supervisor:	Changiz Taghibiglou, College of Medicine
Co-supervisor(s):	Jian Yang

Background:

Spaceflight biosciences and medicine have been a crucial part of space exploration since its inception. The significant effects of spaceflight on the human body were recognized very early on, and efforts were made to ensure astronauts remained healthy during their missions. Human spaceflight is associated with numerous physiological and psychological changes, including bone loss, muscle atrophy, and cognitive decline, which can compromise the health and safety of astronauts during long-duration missions. Recent research has integrated multi-omic analysis methods in individual datasets from space omics databases, with great results. The most notable successes in space health include the development of the space suit, which allowed astronauts to perform extravehicular activities, and the implementation of exercise programs to combat spaceflight-induced muscle and bone loss. However, the global effects of spaceflight on the human genome remain a relatively unexplored area of research. Long-term health monitoring after return from space missions is currently done through the Lifetime Surveillance of Astronaut Health (LSAH) program, and although specific pathways, such as mitochondrial dysfunction and inflammatory stress, have been identified as characteristic of spaceflight, very few genome-wide responses to spaceflight have been investigated so far. Concurrently, it is known that the vast physiological symptoms associated with long-term spaceflight have genomic roots. Identifying these underlying mechanisms and spaceflight biosignatures is critical to developing effective preventive and therapeutic interventions for astronaut health in long-term spaceflight.

Methods:

This study incorporates a multi-omic, integrative analysis pipeline for astronaut health. Firstly, principal component analysis allowed us to verify batch correction methods and ensure fair distribution across our normalized data. Differential expression analysis enabled us to identify genes that were differentially expressed in astronauts compared to control preflight datasets, while genome correlation analysis allowed us to identify co-expressed genes that were potentially involved in similar pathways. Network analysis identified groups of genes that were functionally related, while functional analysis allowed us to identify the biological functions associated with these genes. Interconnectivity analysis allowed us to identify genes that were important in connecting different functional pathways.

Results:

We characterize a massive loss of gene-pair correlation across the entire genome in this study. This stark breakdown in genome correlation is an important step towards identifying gene biomarkers based on gene interaction, rather than individual gene expression levels. This loss of gene pair correlation indicates these astronauts' bodies are losing regulation, working against themselves, wasting energy and possibly even activating conflicting pathways simultaneously. The Gene Ontology

results from our pathway analysis also showcase that most of the heavily affected pathways are large-scale systems in the cell, like gene expression, the cell cycle, apoptosis, and the citric acid cycle. Not only are these processes known to be essential in general cell function and regulation, but the results from this analysis also highlight the large, multi-subsystemic impact of spaceflight on astronauts' bodies, and inform the need for multi-omics analyses of space biology data and the identification of spaceflight biosignature patterns.

Conclusion:
This study uses a comprehensive, integrative multi-omics analysis pipeline to identify gene-pair expression correlation loss as a central biosignature of spaceflight-induced health issues and symptoms in astronauts. This characteristic of spaceflight is proposed to serve as a key cause and regulator of pathological symptoms while in spaceflight. Therefore, it could be used for diagnosis, monitoring, and treatment of astronaut health while in space. Identifying gene correlation loss as a potential biomarker and treatment opportunity highlights the importance of further research and the impact of these findings on space exploration and human health. Future studies with more space datasets, such as samples from the ISS, are proposed to serve as optimization and validation platforms for this biosignature characteristic.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

7. ANATOMICAL PATTERNS IN PARTIAL DISTAL BICEP TENDON TEARS

Presenter:	**Mason Beaulieu**
USask Affiliation:	**Undergraduate student**
College:	**College of Medicine**
Supervisor:	Laura Sims, College of Medicine
Co-supervisor(s):	Haron Obaid

Background:
The distal tendon insertion is at risk of acute injury during an eccentric contraction with the forearm in a supinated position. This can result in a partial or complete tear of the distal biceps tendon. Atraumatic tears have also been described and are thought to result from repetitive use and attritional tearing. The mechanism by which partial tearing occurs as well as the tear pattern is poorly understood.

Methods:
MRI studies of distal biceps tendons will be reviewed, and those with a partial tear of the distal biceps tendon will be included. These images will be re-evaluated to map tear patterns of the distal biceps from the radial tuberosity as well as identify anatomical features associated with partial tears. Data collected from the images include the size of the partial tear; involvement of the short head of distal biceps, involvement of the long head of distal biceps, presence of a confluent vs two discrete distal biceps insertions, radial tuberosity length, radial tuberosity thickness, radioulnar space, radial tuberosity-ulnar space, and presence of enthesophytes.

Results:
Twenty-nine MRI studies from 2013-2023 were included in the final analysis. Five (17.2%), 14 (48.3%), and 10 (34.5%) images were reported as low, middle, and high grade tears respectively. Sixteen (55.2%) had isolated short head tears, 4 (13.8%) had isolated long head tears, and 9 (31%) had components of both. The most common type of tear was a partial tear of the short head with sparing of the long head in the setting of trauma 10 (34.5%). Enthesophytes were observed in 9 (31%) of reports. No significant differences were noted in tuberosity thickness, radioulnar space, nor radial tuberosity ulnar space between traumatic and atraumatic mechanisms.

Conclusion:
Distinct partial distal bicep tear patterns exist. Further research with standardized imaging protocols and larger sample sizes is needed to determine whether anatomical risk factors are correlated with tear patterns.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

Undergraduate Research 2

8. EXAMINING THE JOINT EFFECTS OF MALNUTRITION AND EARLY-LIFE STRESS ON THE BEHAVIORAL AND PHYSICAL DEVELOPMENT OF WISTAR RATS

Presenter:	**Khoi Tran**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Collaborators:	Afzal Javed
Supervisor:	Wendie Marks, College of Medicine

Background:
Malnutrition and stress in early life can cause significant and long-lasting neurodevelopmental and physical issues, but it is unclear whether these variables work together in an additive or synergistic manner.

Methods:
We subjected Wistar rat offspring (N=24 litters) to 4 conditions: (1) control conditions, (2) protein-deficient diet, (3) maternal separation stress, and (4) simultaneously under protein-restricted diet and maternal separation stress. Developmental milestones of rat pups (i.e., body weight, eye opening, ear opening, and fur development) and tests (i.e., righting reflex, negative geotaxis, grip strength) were used to assess physical development. Behavioral development was measured through rat pups' ultrasonic vocalizations (USVs) and their behavior in the open field test.

Results:
Mixed factorial ANOVAs revealed that protein-deficient pups were underweight compared to control pups. Maternally separated pups had significantly delayed ear opening, performed worse on measures of grip strength, and emitted significantly fewer USVs than control pups. Protein restriction and maternal separation stress moderated each other's effects on achievement of eye opening. The majority of evidence did not suggest an additive or synergistic effect of the simultaneous exposure to a protein-deficient diet and maternal separation stress on other variables.

Conclusion:
Overall, these findings underscore the importance of both diet and stress in ensuring healthy physical, behavioral, and social development in rat offspring.

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

9. RETROSPECTIVE ANALYSIS OF BIOPSIES IN A COLLEGE AND SYSTEMATIC REVIEW

Presenter:	**Luke Wandzura**
USask Affiliation:	**Undergraduate student**
College:	**College of Dentistry**
Collaborators:	Felipe Sperandio, College of Dentistry Melanie Hamilton
Supervisor:	Felipe Sperandio, College of Dentistry

Background:
the many different clinical presentations of oral lesions can make diagnosis challenging. Knowing what oral lesions are most frequent in the dental clinic setting could be of great value to both students and educators, as it will ensure that the curriculum correlates to the cases that will be encountered in practice. This study aimed to identify the most common types of oral lesions to facilitate better dental education in the field of oral pathology.

Methods:
a retrospective review of the University of Saskatchewan dental clinic electronic medical records from 2013 – 2023 was performed. All relevant oral pathologies were identified and analyzed in terms of clinical diagnosis and histopathological analysis when available. The oral lesions identified were classified based on Neville's textbook on Oral and Maxillofacial pathology. A systematic review of the most common diagnostic categories and specific oral lesions was completed to provide context and comparison for our retrospective analysis.

Results:
the retrospective chart review revealed 524 patients who presented with oral pathologies, with 107 undergoing biopsy and histopathological analysis. Many clinically identified lesions were not assigned to a specific diagnostic category at time of assessment. Developmental defects and physical and chemical injuries were the most common diagnostic categories. Coated tongue and leukoplakia were the most common clinical diagnoses, and epithelial atypia was the most common histopathological diagnosis. Across the literature, malignant tumors and squamous cell carcinoma were the most prevalent category/diagnosis, respectively.

Conclusion:
The study identified areas in dental education that can be focused on and improved. More consistent diagnostic groupings are needed in the field of oral pathology to better facilitate comparison and use of the research, as well as to guide pre-clinical and clinical teaching. Future studies should examine ways to improve dental students' performance when diagnosing suspected lesions in the clinical setting.

10. A COMPARISON OF GENERIC AND CONDITION SPECIFIC PATIENT REPORTED OUTCOME MEASURES (PROMS) IN PLASTIC SURGERY RESEARCH

Presenter:	**Janan Ashique**
USask Affiliation:	**Undergraduate student**
College:	**College of Medicine**
Supervisor:	Achilles Thoma, College of Medicine

Background:

Patient Reported Outcome Measures (PROMs) assess patients' health outcomes from their perspectives. Generic PROMs tend to reflect a patient's perception of overall health-related quality of life, while condition-specific PROMs consider a single health-related area or intervention. An essential psychometric property of outcome scales is responsiveness to change. External responsiveness reflects the relationship between the changes in a measure and corresponding changes in an external reference measure. Internal responsiveness is the ability of an outcome measure to detect clinically meaningful change after an intervention. Many fields have assessed the internal responsiveness between generic and condition-specific PROMs, but this evaluation has not yet been undertaken in plastic surgery. This review assesses whether generic PROMs provide as much clinically meaningful information following a surgical intervention as condition-specific PROMs. Systematic Review; Level of Evidence: 2

Methods:

This systematic review was performed in accordance with the principles of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement. An electronic search was conducted of both MEDLINE and EMBASE. Studies included used both generic and condition specific PROMs, were randomized controlled trials (RCTs) evaluating a plastic surgery intervention, and assessed an adult population. Mean pre- and post-intervention scores for each critical outcome measure and treatment group were recorded. From this data, calculated the Cohen's d effect size (ES), and a variation known as the Standardized Response Mean (SRM). The magnitude of both the ES and SRM was determined, as defined by Cohen's categories: small <0.5; medium 0.5-0.8; large >0.8. The magnitude of the effect size for each PROM was then compared between generic and condition specific PROMS.

Results:

In total, 7 studies met all criteria for inclusion in the systematic review; 3 of these presented their data as median response values for each PROM. Authors did not respond to requests for additional data to assess mean values, and were excluded from the ES analysis. We calculated ES and SRM for 4 studies and the data from generic PROMs revealed that 70% of the effect sizes were small, 20% were medium and 10% represented a large effect size. Of the condition-specific PROM data, 50% of the ES calculated were small, 0% were medium and 50% were large. According to Cohen, "a medium ES represents an effect likely to be visible to the naked eye of a careful observer". The descriptive data collected revealed that the studies we included predominantly came from the domain of breast surgery and breast cancer research. There were no studies assessing external responsiveness as part of their analysis.

Conclusion:

Condition-specific PROMS capture clinically important changes more profoundly than generic PROMs. If we consider a medium ES to represent a minimally clinically important difference of that PROM, then based on our data it is reasonable to suggest that condition-specific PROMs do provide more information than generic PROMs. Our limited data set, however, makes statistical conclusions difficult to draw. Expanding this systematic review to include cohort studies as well as RCTs is an important next step in further elucidating the relevance of PROMs in plastic surgery research.

11. EXPLORING STERILIZER PERFORMANCE THROUGH EXTERNAL BIOLOGICAL INDICATOR TESTING: A RETROSPECTIVE STUDY

Presenter:	**Bahar Vatanparast**
USask Affiliation:	**Undergraduate student**
College:	**College of Dentistry**
Collaborators:	Juan Buitrago, College of Dentistry
	Michelle Siqueira, College of Dentistry
Supervisor:	Michelle Siqueira, College of Dentistry

Background:
Quality assurance policies mitigate the risk of nosocomial infections from dental office instrument sterilization by assessing sterilizer performance through biological indicator (BIs) testing. This study aimed to evaluate the prevalence of failed sterilization cycles and their causes of failure for a period of eight years through database analysis of a quality assurance laboratory in the province of Saskatchewan, Canada.

Methods:
A database of BIs (198,771) performed by an independent quality assurance laboratory from 2015-2022 was analyzed. Dental offices (n=362) inserted Sporview® Biological Sterility Indicators strips in full sterilizer loads and mailed the processed BI tests to an external quality assurance laboratory for analysis. Samples were assessed for changes in color and turbidity. Failure rates and causes of failure were analyzed with IBM SPSS 28.0.

Results:
The overall failure rate throughout the study was 0.2%, and it decreased gradually from 0.51% (2015) to 0.15% (2022). On average, retests were conducted within 2 days of failure notification. The preferred method of processing was steam sterilization (98%), which had a steadily increasing utilization over time and displays a statistically lower failure rate (0.2%) as opposed to dry heat (1.3%) and chemical vapour (1.4%) sterilizers. Most BI failures were attributable to human error (91.8%), and mechanical malfunctions were significantly more likely to occur with dry heat or chemical vapour sterilizers ($p < .001$).

Conclusion:
This study significantly contributes to the understanding of dental sterilizer performance in Saskatchewan. The low and decreasing sterilizer failure rates over the study period indicate safe dental office procedures and reduced potential for disease transmissions. The study highlights the effectiveness of steam sterilizers with remarkably low failure rates, while human error remains the primary cause of failures. Further research should focus on identifying factors leading to human error and interventions to minimize sterilization failures in dental settings.

12. CHRONIC ADENOSINE A1 RECEPTOR STIMULATION INDUCES NEURODEGENERATION AND BEHAVIORAL DEFICITS

Presenter: **Mahboubeh Pordeli**
USask Affiliation: **Undergraduate student**
College: **College of Arts and Science**
Supervisor: Francisco Cayabyab, College of Medicine

Background:
Adenosine is a purine nucleoside that plays a role in a wide variety of physiological functions, including the regulation of sleep, synaptic plasticity and neuronal excitability, and cardiovascular function. Adenosine is known to bind to four G-protein coupled adenosine receptors (ARs), such as the inhibitory Gi-coupled A1R and A3R that inhibit cAMP production and the facilitatory Gs-coupled A2AR and A2BR that promote cAMP elevation. In the brain, it is widely believed that the extracellular level of adenosine controls neuronal excitability, with tonic A1R stimulation mainly responsible for optimal levels of synaptic transmission. Dysregulation of extracellular adenosine has been implicated in several neurological disorders, including epilepsy, stroke, and neurodegenerative diseases like Parkinson's disease. Therefore, a greater understanding of the regulation of adenosine levels and adenosine actions in the brain should lead to a greater insight into the molecular mechanisms that underlie neuronal damage in these brain diseases. The major transport of adenosine across cell membranes of neurons and glial cells in the brain is mediated by the equilibrative nucleoside transporters (ENTs, encoded by SLC29 gene), of which ENT1 and ENT2 are the most predominant while ENT3 and ENT4 are expressed at lower levels and have intracellular localization. ENTs allow the bidirectional flow of adenosine depending on adenosine's concentration gradient. ENT1 can be phosphorylated at serine 254 (p-ENT1) by the serine/threonine protein kinase casein kinase 2 (CK2), resulting in increased adenosine transport function and ENT1 surface expression. Interestingly, the CK2 levels are downregulated in neurodegenerative diseases and in animal stroke models, where extracellular levels of adenosine are expected to be increased up to 100-fold.

Methods:
we used Male Sprague-Dawley rats (30-35 days old) received i.p. injections of the following treatments: Control (DMSO + Saline), CPA (an agonist for A1R), CPA + DPCPX (an antagonist for A1R), and CPA + Istradefylline (A2AR antagonists). All drugs were given daily (3 mg/kg body weight) for 5 consecutive days. Then on day 6 and 7, to assess the effect of chronic stimulation of A1R, injected animals were subsequently tested for cognitive dysfunction (Y-maze test), anxiety (open- field test), and motor function (rotarod test). On day 8, the rats were anesthetized, perfused with 4% paraformaldehyde, and their brains were processed for immunohistochemistry. Analyses were done using the Zeiss software (Carl Zeiss Group Canada) and Fiji (Public Domain). Statistical analyses were done using GraphPad Prism 8 software (San Diego, CA).

Results:
The results showed that chronic A1R stimulation by CPA (A1R agonist) leads to desensitization of A1R and so reduced A1R expression level, potentially contributing to neurotoxicity, alongside decreased CK2 levels that caused to decrease the phosphorylation level of ENT1 (P-ENT1). however, the expression of ENT1 increased after chronic A1R stimulation. Interestingly, this increased ENT1 expression, possibly due to elevated A2AR activity, suggests a mechanism for neurotoxicity via reduced adenosine levels. In contrast, co-administration of the A1R antagonist (DPCPX) prevented CPA-induced effect and so attenuated of ENT1 level compared to the CPA treatment. The results of

behavioral tests highlighted the chronic stimulation of A1R by CPA was accompanied by behavioral abnormalities (cognitive deficits, increased anxiety, increase, decreased motor function), which was attenuated by co-treatments with DPCPX and Istradefylline.

Conclusion:

In conclusion, prolonged A1R stimulation led to reduced A1R levels in the substantia nigra and hippocampus, potentially promoting neurotoxicity. The observed downregulation of CK2 likely contributed to decreased P-ENT1 levels, while the elevation of ENT1 expression, possibly influenced by increased A2AR activity, may exacerbate neurotoxicity through reduced extracellular adenosine. Additionally, behavioral studies elucidated the relative contributions of A1R and A2ARs to cognitive, motor, and mood dysregulation following chronic A1R stimulation.

13. SUBARACHNOID BLOCK: REASONS FOR INSUFFICIENCY

Presenter: **Roya Emadi**
USask Affiliation: **Undergraduate student**
College: **College of Medicine**
Supervisor: Peter Hedlin, College of Medicine

Background:
A subarachnoid block (SAB) using bupivacaine is the most common anesthetic choice for elective cesarean sections due to its reliability, rapid onset, and good postoperative pain control. Current literature suggests SAB failure rates of 0.5-6.4% in cesarean sections. Failed SABs are associated with poor sensory blockade and can result in a conversion to general anesthesia, which can have detrimental effects to the parturient and the neonate. SAB failure can be caused by provider factors (injection technique and dosing of bupivacaine), patient factors (physiological resistance to bupivacaine) and product factors (use of chemically altered bupivacaine).

Methods:
Following local Research Ethics Board approval, a prospective observational study was conducted over 5 years. Consenting eligible participants included patients undergoing elective cesarean sections (with spinal blocks) at JPCH. Patient demographics and procedural data was collected. Buccal swabs, CSF samples, and leftover bupivacaine vials were collected for each participant.

Results:
A total of 5 failed SABs were captured among 205 enrolled patients. Analysis of the collected CSF, buccal swabs, and bupivacaine samples has not yet been conducted.

Conclusion:
Effective spinal analgesia leads to better clinical outcomes and maternal-neonatal bonding. We can better avoid future failed SABs by identifying the most prevalent mechanism for failed SABs.

14. ADVERSE EFFECTS COINCIDING WITH AMANTADINE USE IN PEDIATRIC PATIENTS FOR PSYCHIATRIC INDICATIONS: A RETROSPECTIVE CASE SERIES AND LITERATURE REVIEW

Presenter:	**Breanna Morrison**
USask Affiliation:	**Undergraduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Laura Almas, College of Pharmacy and Nutrition Ashley Clarke Patrick Parkinson
Supervisor:	Katelyn Halpape, College of Pharmacy and Nutrition

Background:
Current pharmacotherapy is unable to control behavioural disorders such as attention-deficit/ hyperactive disorder (ADHD), autism spectrum disorder (ASD), obsessive-compulsive disorder (OCD), and oppositional defiant disorder (ODD) in pediatric patients. Despite limited available evidence, amantadine is used off-label as an adjunctive therapy for pediatric psychiatric conditions. Objective: To identify the number of case pediatric patients who have experienced amantadine adverse effects when it is used for psychiatric indications in Saskatoon, Saskatchewan over the past 15 years.

Methods:
Cases were identified through inpatient pharmacy software and then screened based on the inclusion criteria for patients 17 years or younger with amantadine orders between January 2007 and December 2022 who experienced adverse effects unrelated to another cause. One case was identified through routine pharmacist patient care activities.

Results:
One hundred fifty-seven case charts were initially identified. Twenty patient cases met the study criteria and were included in analysis. The case patients experienced a range of physical and psychiatric adverse effects that coincide with known amantadine adverse effects. The most frequently identified adverse effect was aggression which was observed in 16 (80%) patients. The most severe adverse effect was cardiac conduction abnormalities seen in a patient who overdosed on amantadine and methylphenidate XR.

Conclusion:
This study highlights potential safety concerns regarding amantadine use in pediatric patients. Limitations stem from the retrospective nature such as lack of documentation, confounding factors, and missing information. Additional studies should be performed to draw conclusions regarding amantadine use, dosing, and safety in this patient population.

Undergraduate Research 3

15. 'IT CHANGED ALL OF US': THE EFFECTS OF COVID-19 ON THE ENGAGEMENT OF ALLIED HEALTH WORKERS AND VOLUNTEERS IN LONG-TERM CARE

Presenter:	**Abang Omot**
USask Affiliation:	**Undergraduate student**
College:	**College of Arts and Science**
Supervisor:	Roslyn Compton, College of Nursing
Co-supervisor(s):	Abigail Wickson-Griffiths

Background:

The COVID-19 pandemic significantly disrupted the roles and engagement of allied health professionals and volunteers in long-term care homes, posing challenges to the quality of life for residents, family caregivers, and staff. While the effects of the COVID-19 pandemic on healthcare professionals in long-term care are well-documented in media and literature, there remains a gap in understanding how the COVID-19 pandemic affected the engagement of allied health professionals - workers outside of nursing staff, who also served as frontline workers through their roles in physical therapy, recreational therapy, and spiritual care. Additionally, volunteers assist staff in long-term care with various day-to-day tasks, fill in the gaps of understaffing in long-term care, and socialize with residents. This honours project aims to give a voice to allied health professionals and volunteers in long-term care and to examine the implications of COVID-19 on their engagement in long-term care.

Methods:

Through open-ended interviews, six participants who work with long-term care homes in Saskatchewan were asked how their work, roles, and responsibilities within long-term care were affected by COVID-19. Braun and Clarke's reflexive thematic analysis was utilized to analyze the interviews and how the changes affected the participants.

Results:

Results through thematic analysis revealed three main themes: Top-down decisions: Ever changing rules; Disassembled community: What we had was taken away; and It changed all of us: COVID-19 and mental health.

Conclusion:

The results of this study shed light on the often-overlooked contributions of allied health professionals and volunteers in long-term care. They demonstrate that these individuals play a crucial role in enhancing the quality of life in long-term care homes beyond just caring for residents. Understanding the impacts on their engagement in long-term care provides a broader, more comprehensive picture of the functioning of long-term care toward residents' well-being.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

16. BLOOD LEAD LEVELS AND HAND SURFACE LEAD IN PRIVATE PRACTICE VETERINARY WORKERS

Presenter:	**Yara Al Horoub**
USask Affiliation:	**Undergraduate student**
College:	**College of Medicine**
Collaborators:	Gloria Hooshmand, Western College of Veterinary Medicine Hoda Poorbagher, Western College of Veterinary Medicine
Supervisor:	Niels Koehncke, College of Medicine
Co-supervisor(s):	Monique Mayer

Background:
Veterinary workers regularly utilize lead shielding (gloves, aprons, etc.) when restraining animals during radiographic studies, potentially resulting in lead contamination of hands and their work environment. This potential for oral-dermal exposure can lead to adverse health effects. This study aims to compare lead blood concentrations in veterinary workers using lead shielding with control population. Measure hand surface lead before and after use of shielding, and to quantify surface lead on reception and radiology keyboards.

Methods:
Workers from all veterinary clinics in Saskatoon were invited to complete a survey assessing the frequency of radiology shielding use, with the goal of identifying 50 participants. Workers that have used lead shielding within the last 3 months were place in the exposed group, and workers with no history of lead shielding use in the last 6 months were placed in the control group. Exposed workers with the highest rate of lead shielding were selected for blood lead and hand surface analysis using inductively coupled plasma mass spectrometry analysis. Potential lead transfer was assessed by sampling clinic keyboards at reception and in the radiology area.

Results:
There were 115 workers at 18 clinics that completed the preliminary survey, 31 workers met the control criteria of no history of lead shielding use in the last 6 months, and 19 workers were included in the exposed group. Data collection is still ongoing, preliminary data show significantly higher surface lead on hands post-shielding (mean difference 2.4µg, 95% CI 0.27-4.4µg, $p = 0.03$) compared to pre-shielding.

Conclusion:
Early data analysis demonstrates increased hand surface lead after handling radiology shielding. Blood and hand surface wipe lead measurements will be completed in summer 2024.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

17. RESOURCES TO SUPPORT END-OF-LIFE NUTRITION CARE IN LONG TERM CARE: A SCOPING REVIEW

Presenter:	**Nadia Anvari**
USask Affiliation:	**Undergraduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Heather Alford, College of Pharmacy and Nutrition Erin Yakiwchuk, College of Pharmacy and Nutrition Abigail Wickson-Griffiths, College of Nursing
Supervisor:	Allison Cammer, College of Pharmacy and Nutrition
Co-supervisor(s):	Paulette Hunter

Background:
End-of-life (EOL) can be a challenging time where nutrition and hydration practices for long-term care residents change, shifting the focus of care to emphasize quality of life. Factors that influence decision making for nutrition at EOL include cultural viewpoints, and the beliefs and practices of health care providers. Resources to support decision making are helpful to inform care options. However, little is known about the use of resources to support EOL nutrition care decision-making.

Methods:
A scoping review was conducted to explore what is reported in the literature about tangible resources to support decision-making about nutrition and hydration at EOL in long-term care. Following Arksey and O'Malley's 2005 framework with recommended adaptations by Levac et al. (2010), four databases (MEDLINE, CINAHL, Web of science, Embase) were searched for research published from 2003 to 2023. Articles included peer-reviewed human studies published in English that reported research examining resources used to support decision making about EOL nutrition care in long-term care homes.

Results:
After de-duplication, 1038 abstracts were screened for inclusion, 274 full-text articles were examined for inclusion and appraised for quality, resulting in 15 papers for review. Findings indicated that use of resources resulted in higher reported frequency of conversations about EOL nutrition, increased participant knowledge on options for care, and less decisional conflict for family members. Five themes emerged from thematic analysis of the included articles: conversations about care, evidence-based decision-making, a need for multidisciplinary perspectives, honouring residents' goals of care, and cultural considerations for adapting resources.

Conclusion:
Resources can facilitate the process of choosing EOL nutrition care by providing a foundation of evidence-based knowledge, initiating conversations, and helping to guide the provision of care to meet residents' goals and wishes.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

18. NUTRITION TO PREVENT AND MANAGE DENTAL CARIES: UNDERSTANDING THE PERCEIVED KNOWLEDGE, ATTITUDES, AND PRACTICES OF DIETITIANS IN CANADA.

Presenter:	**Amanda Goncalves Troyack Vanzan**
USask Affiliation:	**Undergraduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Jessica Lieffers, College of Pharmacy and Nutrition Petros Papagerakis Roy Dobson, College of Pharmacy and Nutrition
Supervisor:	Jessica Lieffers, College of Pharmacy and Nutrition

Background:
Dental caries is the most prevalent chronic disease worldwide, and it has a two-way relationship with nutrition. Eating behaviours can influence the risk of developing dental caries and dental caries can impact eating practices and nutritional status. Dietitians and oral health professionals (OHP) have an important role in optimizing nutrition to prevent and manage dental caries. This study aimed at describing and characterizing the knowledge, attitudes, and practices of dietitians regarding nutrition to prevent and manage dental caries in Canada.

Methods:
An interdisciplinary advisory committee guided the study, and a 68-question survey was developed using a multi-step process to address the study objectives. Dietitians from across Canada were recruited to complete the online survey, available in English and French, from June-August 2021. Quantitative and qualitative data were analyzed using SPSSv28 and content analysis, respectively.

Results:
In total, n=235 responses were analyzed. Most respondents (n=198; 84.2%) agreed or strongly agreed that there is a strong relationship between nutrition and dental caries. Barriers about nutrition and dental caries were reported to be experienced by n=128 (58.7%) respondents, and n=167 (85.5%) respondents reported to have never received any training about nutrition and dental caries. Collaboration practices between dietitians and OHP was reported to be conducted in a limited extend, with n=25 (11.5%) respondents reporting to have been consulted or have received a patient/client referral from another health professional regarding nutrition and dental caries.

Conclusion:
The results of this study showed that dietitian respondents demonstrated substantial enthusiasm about the importance of nutrition and dental caries; however, practices concerning the topic are limited and professionals experience barriers working in this area. Furthermore, the results show the need for increased educational programs for dietitians about topics concerning nutrition and dental caries, to ultimately help to decrease the burden of dental caries in Canada and beyond.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

19. PRIMARY CARE, SECOND LANGUAGE PODCAST

Presenter:	**Kevin Entwistle**
USask Affiliation:	**Undergraduate student**
College:	**College of Medicine**
Collaborators:	Retaj Ramadan, College of Medicine
Supervisor:	Helen Chang, College of Medicine

Background:
"Primary Care, Second Language" is a Saskatchewan pilot project, providing healthcare professionals with a podcast resource for improving the care of marginalized patient populations. A significant portion of the Saskatchewan population speaks a language other than English. Lack of familiarity with medical terminology and phrases often presents a barrier to effective communication in healthcare settings, i.e. interactions between healthcare providers and newcomers to Canada. Healthcare professionals (HCPs) who know even a few words or phrases in their patient's language can help bridge this gap in communication, and reaffirm a dedication to creating a safe, welcoming, and inclusive space for patients.

Methods:
We interviewed community members who speak a language other than English and produced several short, educational, and interactive podcasts covering terms and phrases that may be used during a number of common medical encounters.

Results:
The "Primary Care, Second Language" podcast is currently 12 5-10 minute episodes, accessible on multiple audio streaming services, with medical terminology in 8 different languages.

Conclusion:
Our project seeks to address key social determinants of health, including education, literacy, and equitable access to health services by creating a free, interactive and accessible resource for both patients and healthcare providers to bridge gaps in communication. Drawing on the expertise of our own community members in developing this project, we hope that our resource continues to encourage, affirm, and celebrate the lingual diversity within our province.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

20. TAKE-HOME NALOXONE USE AND ACCESS IN OLDER ADULTS LIVING WITH PAIN: A SCOPING REVIEW

Presenter:	**Ryan Chan**
USask Affiliation:	**Undergraduate student**
College:	**College of Pharmacy and Nutrition**
Supervisor:	Katelyn Halpape, College of Pharmacy and Nutrition
Co-supervisor(s):	Erin Yakiwchuk

Background:
Opioids are a common treatment for older adults living with pain. With high rates of polypharmacy and chronic comorbidities, older adults are at risk of experiencing opioid overdose. Take-home naloxone (THN) has evidence to support reduction in opioid-related harms. It is unknown what THN initiatives are available for older adults, especially those living with chronic pain. This study aimed to summarize the literature regarding THN focused on older adults using opioids for pain, including facilitators and barriers to THN access, knowledge gaps, and pharmacist-led initiatives.

Methods:
A scoping review guided by Arksey and O'Malley's framework and PRISMA-ScR guidelines was performed. Search methods involved searching six bibliographic databases, reference harvesting, and citation tracking. Study eligibility was determined by pre-set criteria including age, with inter-researcher consensus for discrepancies. Data were extracted and categorized through thematic analysis.

Results:
Four studies were identified. The mean patient age in the studies was 60.1 to 80.3 years old and primarily female and Caucasian individuals were included. Four studies detailed THN programs in primary care settings with older adults taking opioids for pain management. Two studies highlighted patient-specific risk factors for opioid overdose, including concomitant use of benzodiazepines and/or gabapentinoids, mean morphine milligram equivalents per day greater than or equal to 50, and previous opioid overdoses. Educational programs increased patient interest in THN.

Conclusion:
There is limited literature published about THN for older adults living with pain and no literature on pharmacist-led initiatives in this area. Future research on THN provision in older adults, including pharmacist-led initiatives, could help to optimize care for older adults living with pain.

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

21. EPIDEMIOLOGY OF UPPER LIMB SKIN AND SOFT TISSUE INFECTIONS REQUIRING SURGICAL INTERVENTION IN SASKATOON, CANADA: A RETROSPECTIVE CHART REVIEW.

Presenter:	**Retaj Ramadan**
USask Affiliation:	**Undergraduate student**
College:	**College of Medicine**
Collaborators:	Emma Yanko, College of Medicine
	Rachel Miller, College of Medicine
Supervisor:	Chris Thomson, College of Medicine
Co-supervisor(s):	Gary Groot

Background:
Skin and soft tissue infections (SSTIs) are a leading cause of hospital admission and engagement with the health care system amongst people who inject drugs (PWID). The current study aims to describe the epidemiology of SSTIs requiring surgical intervention in Saskatoon, Canada.

Methods:
This retrospective chart review assessed patients with a primary diagnosis of upper limb SSTIs requiring surgical intervention at St. Paul's Hospital and Royal University Hospital (Saskatoon, Canada) between January 1 and December 31, 2020.

Results:
38 eligible patients with a median age of 34 years and M:F of 21:17 were identified. 31 (81.6%) smoked cigarettes and 19 (50.0%) used intravenous drugs. A majority of SSTIs were unilateral infections involving the hand 22 (57.9%) or upper arm 11 (28.9%). Ten (26.3%) patients had a prior SSTI requiring surgical management. Necrotizing fasciitis was diagnosed in 7 (18.4%) patients, two of which, required amputation of the affected hand or arm. The median length of hospital stay was 6 days (IQR: 4 – 14.5). Ten patients left the hospital against medical advice, before completion of treatment; of these patients, 8 (80.0%) were PWID.

Conclusion:
Harm reduction strategies may help address the rising incidence and recurrence of SSTIs in the injection drug use population. Involvement of addiction services and social work during hospital admission may reduce the rate of patient-directed discharge, facilitating the completion of treatment. Furthermore, increased access to needle exchange programs in the community may reduce the number of SSTIs caused by contaminated injection equipment in the PWID population.

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

22. EXAMINING THE SOCIAL COMPARISON/ACTIVITY ADEQUACY MINDSET RELATIONSHIP: AN EXPERIMENTAL MESSAGING STUDY

Presenter:	**Brittany Krammer**
USask Affiliation:	**Undergraduate student**
College:	**College of Kinesiology**
Collaborators:	Jantz Sawatsky, College of Kinesiology
	Kevin S. Spink, College of Kinesiology
Supervisor:	Kevin S. Spink, College of Kinesiology

Background:
Mindsets have entered the research domain of physical activity (PA) and health in the form of perceived activity adequacy mindset (PAadeq) (Zahrt & Crum, 2020). One factor strongly associated with PAadeq is PA social comparison (PA SC, Gitzel et al., 2023). This prompts the question of whether one can change PAadeq by altering people's perceptions of how they compare with others with respect to PA by lowering the messaging anchor (Tversky and Kahneman,1974). The purpose was to examine whether PA SC would serve as a mediator between messages with different PA anchors and students' levels of PAadeq mindset.

Methods:
University students (N= 114) completed an online survey that assessed demographics, PA SC (Gitzel et al., 2023), PAadeq mindset (Zahrt & Crum, 2020), and MVPA (Fowles et al., 2017). Participants were then randomly assigned to receive one of two PA messages anchored differently (a little versus a lot of PA required for health). Participants then completed PA SC and PAadeq again.

Results:
Both PA SC and PAadeq were found to be reliable measures (cronbach alphas > .90. Results revealed a positive relationship between the anchored messages and PA SC comparison ($p = .004$), controlling for initial PA social comparison. Those receiving the 'little' PA message reported higher PA SC. Further, those reporting the higher PA SC reported higher PAadeq mindset ($p = .02$) when controlling for the anchored PA messages, PAadeq pre message, PA SC pre message, and MVPA.

Conclusion:
This study provides preliminary experimental evidence to suggest that PA SC is a possible mediator of the PA anchored messaging and PAadeq mindset. If these results can be replicated, they may have implications for future PA messaging (i.e., anchoring PA messages around doing 'little' behaviours for health to change mindsets).

23. INCREASING ON-CAMPUS ACTIVITY: CHANGING THE MESSAGE

Presenter:	**Matthew Jarotski**
USask Affiliation:	**Undergraduate student**
College:	**College of Kinesiology**
Collaborators:	Matthew Jarotski, College of Kinesiology
	Kate Korchinski, College of Kinesiology
	Kevin S. Spink, College of Kinesiology
Supervisor:	Kevin S. Spink, College of Kinesiology

Background:
According to current guidelines (150 min weekly MVPA), university students are typically not active enough for health benefits (Scarapicchia et al., 2015). Of interest, WHO (2020) now recommends PA messaging be changed to highlight the health benefits that result from doing any activity, regardless of intensity or duration (i.e., everything counts). This prompts the question of whether PA messaging highlighting an easier target would make the perception of being active for health as more attainable, and thus increase PA levels. The study purpose was to examine whether an 'everything counts' PA message would increase light PA and break up sedentary behaviours for students while on campus.

Methods:
University student participants (N=42) completed two online surveys one week apart. Survey one assessed demographics, current on-campus PA and sedentary behaviours and current MVPA levels (Fowles et al., 2017). Participants were then randomly assigned to receive either an "everything counts" message or control message. Survey two was a reassessment of on-campus PA and sedentary behaviours one week later.

Results:
MANCOVA results examining the effect of messaging on on-campus PA while controlling for pre-message on-campus PA and MVPA revealed a significant condition main effect ($p = .032$). Post-hoc tests revealed only one of the two on-campus PA behaviours (i.e., deliberately taking a longer route) was significant ($p = .008$). Those who received the 'everything counts' message reported taking longer routes to on-campus destinations more often than those receiving the control message. No significant condition main effect emerged for sedentary behaviours ($p = .16$).

Conclusion:
This study provides initial experimental evidence that on-campus PA can be increased using an 'everything counts' message. If replicated, changing to this messaging could possibly increase on-campus PA in the university population.

Basic Science 1

24. ENHANCING NEUROTHERAPEUTIC DELIVERY: SYNERGIZING NEURONAL REPROGRAMMING AND GEMINI LIPID-BASED LIPID NANOPARTICLES FOR TARGETED ASTROCYTE THERAPY

Presenter: **Jonathan Rekve**
USask Affiliation: **Graduate student**
College: **College of Pharmacy and Nutrition**
Supervisor: Ildiko Badea, College of Pharmacy and Nutrition

Background:

Neuronal reprogramming and lipid nanoparticles (LNPs) present novel approaches to treating incurable neurological diseases and effectively delivering gene therapies, respectively. Reprogramming astrocytes into induced neurons (iNeurons) offers a potential to replenish lost neuronal populations and counteract neurodegeneration. Similarly, utilizing gemini lipids in LNPs has shown significant promise in enhancing the delivery of nucleic acid payloads to specific cell types, such as astrocytes, which play a crucial role in brain health and disease pathology.

Methods:

Our investigation synergizes direct neuronal reprogramming with cutting-edge LNP formulations, emphasizing gemini lipids and 1,2-dioleoyl-sn-glycero-3-phosphoethanolamine (DOPE) for specific astrocyte targeting. A cornerstone of our analysis was the application of Small Angle X-ray Scattering (SAXS), providing essential insights into the LNPs' nanostructural organization and lipid molecular arrangement, crucial for enhancing cellular targeting and nucleic acid liberation.

Results:

SAXS investigations revealed that the inclusion of DOPE in our gemini lipid-based LNPs leads to a hexagonal phase formation, integral to achieving an 8-fold increase in nucleic acid delivery efficiency, while maintaining minimal toxicity. This hexagonal configuration signifies a more orderly and stable structure, optimizing the encapsulation, safeguarding, and subsequent release of payloads such as nucleic acids into targeted cells.

Conclusion:

The hexagonal phase structure within DOPE-incorporated LNPs has shown to be pivotal to their enhanced delivery capabilities. These structural insights are crucial for tailoring nanoparticle design for improved interactions with astrocytes, thus elevating delivery success rates. Our research advocates for a hybrid approach, melding cellular reprogramming with nanotechnological delivery enhancements, to forge new pathways in treating neurodegenerative ailments and brain injuries. The ongoing refinement of these techniques stands to introduce novel therapeutic modalities in neurology.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

25. ADAPTING INTERDISCIPLINARY TECHNIQUES TO OPTIMIZE THE PDX MODEL OF BREAST AND OVARIAN CANCERS

Presenter:	**Breanne Bevelander**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Nima Daneshvar Baghbadorani, College of Medicine
	Mira Bisso
	Rown Greene
Supervisor:	Dean Chamberlain, College of Medicine

Background:
Breast and ovarian cancers are some of the most common and deadly cancers for females. With poor prognoses largely attributed to chemoresistance and late stage-of-detection. By modeling patients individually, we can tailor treatments and improve patient outcomes. The gold standard model for cancer research is patient derived xenografts (PDXs), meaning when patient tumours are grown in mice. Modular tissue engineering uses small tissue building blocks to synthesize tissue constructs optimized to meet the goals of the research; these tissue constructs are termed “microtissues”. We are adopting these techniques, along with techniques from cryobiology to improve the utility of microtissues as an in vitro and in vivo model for cancer research.

Methods:
At their most basic form, microtissues are a collagen matrix containing a homogenous distribution of cells. Prior to using patient cells, we are optimizing, characterizing, and complexifying our microtissues using cell-lines. For each cell-line explored, we look to estimate growth rate, do various histological assays throughout their development, and do comparative drug-response assays between 2D cells and our 3D microtissues. To better mimic the complexities of the tumour microenvironment in vivo, we have initiated complexifying our microtissues by adding other cell-types. To improve the utility of our microtissues, we are working to find methods to cryopreserve them. While this optimization continues in vitro, we are commencing animal trials to determine if our basic microtissue model performs better than current xenograft models.

Results:
We have made significant progress in optimizing the synthesis of our microtissues in various cell-lines along with characterizing their growth and development. We have also successfully cryopreserved our microtissues.

Conclusion:
Based on this model’s success in the field of tissue engineering and our preliminary data, our model has promise as a new and more customizable tool for cancer research. As our model progresses to include patient cells, we will have built a model from the ground up that has potential as a precision-oncology tool.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

26. MOLECULAR DIAGNOSTIC METHODS FOR BRUCELLA ABORTUS DETECTION IN WOOD BISON

Presenter:	**Kira Mudrey**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Muhammad Anzar, Western College of Veterinary Medicine
Supervisor:	Tim Dumonceaux, Western College of Veterinary Medicine

Background:
Wood bison, a species at risk in Canada, are threatened by a high prevalence of cattle diseases, such as bovine brucellosis. Brucellosis is a zoonotic bacterial infection caused by Brucella abortus that can lead to infertility, abortions, and arthritis. To support wood bison conservation efforts, B. abortus infection must be controlled and prevented from spreading to healthy herds. However, current diagnostic methods are time-consuming, require specially trained personnel and specialized equipment, or lack specificity. Newer molecular diagnostic methods present a promising alternative to traditional brucellosis diagnostics.

Methods:
We designed portable quantitative polymerase chain reaction (qPCR) and loop-mediated isothermal amplification (LAMP) assays targeting the cpn60 gene and compared their performance characteristics with previously published LAMP and recombinase polymerase amplification (RPA) assays targeting the bcsp31 gene of Brucella abortus. 95% limits of detection (LODs) and analytical specificity were assessed using purified genomic DNA. Diagnostic sensitivity and specificity were estimated by comparing the results of molecular assays to gold standard culture for 45 ram semen samples from a herd infected with Brucella ovis.

Results:
All four assays can be performed with limited equipment, showed good analytical specificity, and did not cross-react with non-Brucella species. qPCR showed the highest analytical sensitivity (21.1 copies), followed by cpn60 LAMP (50.4 copies), RPA (168.6 copies), and bcsp31 LAMP (321.8 copies). The diagnostic sensitivity of all molecular tests was greater than 90%, and diagnostic specificity was greater than 80% compared to gold-standard culture results. When converted to a lateral flow detection format, RPA maintained similar sensitivity and specificity.

Conclusion:
Our data indicate that any of the four methods would be appropriate for field diagnosis of Brucella abortus. However, the RPA method provides the fastest and most field-friendly diagnostic option, and therefore, we will evaluate this method further using clinical samples from wood bison herds infected with B. abortus.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

27. LONG-TERM IN-VITRO CHARACTERIZATION OF 3D BIOPRINTED ALGINATE/GELATIN HYDROGEL PATCHES

Presenter:	**Farinaz Ketabat**
USask Affiliation:	**Graduate student**
College:	**College of Engineering**
Collaborators:	Ildiko Badea, College of Pharmacy and Nutrition Reza Gharraei, College of Engineering Alex Guinle
Supervisor:	Xiongbiao (Daniel) Chen, College of Engineering
Co-supervisor(s):	Michael Kelly

Background:

The emergence of three-dimensional (3D) bioprinting technology has revolutionized tissue engineering, particularly for cardiac tissue engineering, enabling the development of more complex and vascularized structures. However, for a successful 3D bioprinting, the printing conditions should be adjusted based on the rheology of cell-laden bioink to avoid disturbing the cells during or after the printing which can cause cell death. In this study, rheological analysis and cell viability assays were employed to optimize the bioprinting process for cell-laden constructs mimicking the heart myofibrils' orientation. Subsequently, the physical, morphological, and cellular viability aspects of 3D bioprinted constructs were comprehensively assessed, contrasting them with cell-free scaffolds.

Methods:

Bioinks containing alginate and gelatin, with or without human umbilical vein endothelial cells (HUVECs) were prepared and their rheological behavior was analyzed. An adopted bioprinting pressure was determined using computational fluid dynamic (CFD) and cell viability assays. Cell-free and cell-laden constructs were 3D (bio)printed in an angular pattern mimicking heart myofibrils' orientation. The cell viability of cell-laden constructs as well as the physical and morphological properties of both cell-free and cell-laden constructs were assessed over a 21-day period.

Results:

Under optimal printing conditions, both constructs exhibited viscoelastic behavior, and cell-laden constructs demonstrated elasticity at lower frequencies. Moreover, cell-laden constructs displayed a lower elastic modulus than cell-free counterparts over 21 days. They also exhibited lower swelling and higher remaining mass percentage. Following bioprinting, the cells demonstrated robust viability, which remained consistently high throughout the initial week and persisted for up to 21 days, with minimal presence of dead cells despite a reduction in the total cell count.

Conclusion:

The 3D printing parameters were successfully adjusted for bioprinting HUVECs and resulted in cell-laden constructs with high stability for up to 21 days. Our findings indicate that the developed 3D bioprinted constructs are appropriate candidates for vascularized cardiac tissue engineering.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

28. THE EPHB4 RECEPTOR TYROSINE KINASE REGULATES CANINE AND HUMAN OSTEOSARCOMA INVASIVENESS AND TUMOR INITIATION

Presenter:	**Jessica Sharpe**
USask Affiliation:	**Graduate student**
College:	**College of Western College of Veterinary Medicine**
Collaborators:	Evelyn Harris, Western College of Veterinary Medicine Tim Strozen, Western College of Veterinary Medicine Behzad Toosi, Western College of Veterinary Medicine
Supervisor:	Behzad Toosi, Western College of Veterinary Medicine

Background:

Osteosarcoma is an aggressive bone cancer in both canines and humans with a high metastatic rate and a poor prognosis. Advances in treatment options are limited, highlighting the need for more effective therapeutic approaches. Recent evidence suggests that the EphB4 receptor regulates the invasion and metastasis of various human cancers. However, the role of EphB4 in human and canine osteosarcoma has been poorly evaluated. Due to the physiological and cellular similarities between canine and human osteosarcoma, we investigate the role of EphB4 in promoting osteosarcoma using a comparative approach.

Methods:

EphB4 expression was evaluated by western blotting in multiple canine and human osteosarcoma cell lines and compared to normal osteoblasts. EphB4 expression was silenced using specific shRNAs and stable cell lines were established. We investigated the effects of EphB4 on canine and human osteosarcoma cell migration and invasion using transwell assays and on propagation of tumor-initiating cells (TICs) using the tumorsphere formation assay. Human osteosarcoma tumorsphere cells were stained with Ki-67 proliferation marker and evaluated by flow cytometry. Tumor initiation was tested using a xenograft model of human osteosarcoma.

Results:

We observed upregulated EphB4 expression in canine and human osteosarcoma cells compared to osteoblasts. Both canine and human EphB4-silenced osteosarcoma cells exhibited reduced migration and invasion. Interestingly, EphB4 knockdown enhanced the expansion of tumorspheres in both canine and human osteosarcoma and increased tumor initiation in mice.

Conclusion:

Our comparative study highlights the pivotal role of EphB4 in both canine and human osteosarcoma. We observed EphB4 promoting osteosarcoma cell migration and invasion, and regulating TIC propagation and tumor initiation in vivo. These findings suggest that EphB4 holds promise as a target for the development of new therapeutic approaches based on its function. Moreover, our research underscores the value of comparative oncology in advancing our understanding of osteosarcoma biology.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

29. A PROTOCOL FOR 3D VISUALIZATION OF MOUSE AND HOMININ MOLARS AND CRYPTS

Presenter:	**Amalya Babayan**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Angeline Leece
Supervisor:	Julia Boughner, College of Medicine

Background:
Lab mouse dental development is an established model of primate tooth development and evolution. Teeth tell a lot about animal's diet and growth. Mammalian molars and bony tooth crypts develop simultaneously via shared developmental processes. Thus, crypts are promising for estimating molar development stage, especially if molars are lost from a fossilized jawbone. Computed tomography (CT) is a noninvasive, high-resolution tool to view developing teeth in 3D. Our two aims were to create protocols to visualize, segment, and study the 3D structures of developing molars and crypts in: 1) mouse micro-CT scans, 2) fossil hominin CT scans.

Methods:
We studied synchrotron radiation micro-CT scans (Canadian Light Source) of postnatal mouse heads (n=4) and CT scans of subadult Paranthropus robustus jaw fragments (n=4). Using Amira 3D visualization software, we segmented molars and crypts in the mouse and P. robustus scans. Amira enables some automation: we used the "blowout" tool to select radio translucent crypt spaces and the "magic wand" tool to select radiodense crowns. Manual selection tools were used when automatic selection lacked accuracy.

Results:
Semi-automatic selection worked well in mouse scans as the densities of mineralized versus soft tissues differed drastically. For P. robustus scans, bone fragmentation, the presence of sediment, and reduced radiodensity due to fossilization challenged Amira's automated selection tools. Instead, manual selection, combined with automated tools, were applied successfully to fossil samples.

Conclusion:
In sum, this CT volume segmentation protocol was successfully optimized for mouse molars and crypts and applied effectively to fossil samples, with some caveats. Next steps include investigating whether molar crypt shape is a reliable proxy for molar development stage. Future work will test the extent to which the processes of modern human molar formation occurred in our fossil relations, shedding more light on the evolution of human maturation and growth.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

30. INVESTIGATING THE ROLE OF INHIBITORY FACTOR-1 IN OZONE-INDUCED LUNG INFLAMMATION.

Presenter: **Mohammad Umar**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Gurpreet Aulakh, Western College of Veterinary Medicine

Background:
Ground-level ozone, one of the toxic environmental pollutants, is an intensely irritating gas, produced above the earth's surface through chemical reactions between pollutants emitted from electric sparks, vehicles, industries, or fuels. It is recognized to have a substantial impact on human health like asthma, ischemia, and stroke, leading to early death. Exposure to ozone injures the epithelial and endothelial barrier in the lungs leading to airway hyperactivity (AHR) or obstruction. The resulting inflammation leads to the accumulation of the neutrophils in lungs. ATP synthase complex V (ATPS-V) is an ATP synthesizing/hydrolyzing mitochondrial motor protein that senses protons. However, during inflammation, ATPS-V can translocate to the plasma membrane and reverse its functions to maintain cell pH at the expense of ATP hydrolysis. The inhibitory factor-1 (IF1) protein binds to the catalytic domain of ATPS-V and inhibits ATP hydrolysis to maintain cell viability during inflammation. The goal of our research is to understand the role of IF1 in neutrophil activation and migration in ozone-induced lung inflammation using an IF1 mimetic, BTB06584, and neutrophil-specific IF-1 knockout (nIF1KO) mice.

Methods:
DARTS, Western Blot, ELISA and Genotyping.

Results:
Our results indicate that BTB06584 interacts with the ATPS-V β subunit and protects it from proteolytic degradation. We now propose that BTB06584 augments IF1 activity by protecting the ATPS-V β subunit from degradation and thus inhibits ATP hydrolysis leading to the accumulation of extracellular ATP (eATP) during inflammation. We have probed the IF1 floxed mice for the presence of IF1 alleles in the floxed mice which will be used to develop nIF1 KO mice using Myeloid Related Protein (MRP8) Cre-IF1Lox technology to understand the role of IF1 in neutrophil activation and migration.

Conclusion:
We conclude that IF1 may play a role in activation and migration of neutrophils, alveolar permeability, and alveolar contractile dynamics in ozone induced lung inflammation.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

31. EARLY INNATE IMMUNE RESPONSE AND EVOLUTION OF A SARS-COV-2 FURIN CLEAVAGE SITE INACTIVE VARIANT IN BAT CELLS

Presenter:	**Kaushal Baid**
USask Affiliation:	**Postdoctoral fellow**
College:	**Vaccine and Infectious Disease Organization**
Collaborators:	Heather Wilson, Western College of Veterinary Medicine
	Qiang Liu, Western College of Veterinary Medicine
Supervisor:	Arinjay Banerjee, Western College of Veterinary Medicine

Background:

SARS-CoV-2 has caused the largest coronavirus pandemic to date and is believed to have evolved in insectivorous bats, likely from the genus Rhinolophus. Although SARS-CoV-2 has not been isolated from bats directly, closely related viruses from the Sarbecovirus lineage circulate widely in different bat species across South-East Asia. Little is known, however, about the evolution of these viruses in its reservoir species and the ability of bats to tolerate viral infections that cause severe disease in spillover mammalian species, such as humans.

Methods:

We investigated SARS-CoV-2-host interactions using human lung and Eptesicus fuscus-derived cells, an insectivorous bat species widely distributed throughout the Americas. Using transcriptomic and proteomic assays, we elucidated global host responses in human and E. fuscus cells infected with SARS-CoV-2. Furthermore, through computational modelling and docking studies as well as biochemical assays, we characterized a novel R685P mutation within the furin cleavage site (FCS) of the SARS-CoV-2 spike protein that was naturally selected for in infected E. fuscus cells.

Results:

Our transcriptomic and proteomic data demonstrate that bat cells mount a more robust and early antiviral response but elicit a dampened pro-inflammatory response upon SARS-CoV-2 infection relative to human cells. Bat cells also drove selection for a SARS-CoV-2 variant with a R685P mutation within the FCS of the spike protein. Data from our computational modelling, docking, and mechanistic molecular studies indicate that the R685P mutation in SARS-CoV-2 spike abolished furin cleavage activity.

Conclusion:

Taken together, our data demonstrate that insectivorous bat cells have evolved a differential antiviral immune response to SARS-CoV-2, likely to mitigate immunopathology that is observed in human patients with severe COVID-19. In addition, our study sheds further light on the evolution of sarbecoviruses in its reservoir host and extends molecular evidence to data from field studies where SARS-CoV-2-related viruses discovered in wild-caught bats lack an intact FCS.

32. CHRONIC DIETARY ARSENIC EXPOSURE DISRUPTS DOPAMINERGIC SIGNALING PATHWAYS AND IMPAIRS COGNITIVE PERFORMANCE IN ADULT ZEBRAFISH (DANIO RERIO)

Presenter:	**Mahesh Rachamalla**
USask Affiliation:	**Graduate student**
College:	**College of Arts and Science**
Supervisor:	Som Niyogi, College of Arts and Science

Background:
The present study was designed to investigate the neurobehavioral effects of chronic exposure to environmentally relevant concentrations of dietary arsenic (As) in zebrafish. Adult zebrafish were exposed to 3 different concentrations of dietary arsenic (30, 60, or 100 As µg/g dry weight; as arsenic) in addition to control for 60 days.

Methods:
The cognitive performance of fish was then examined using a latent learning paradigm in a complex maze, which exhibited a dose-dependent effect of As on the cognitive performance. As-treated fish demonstrated significant impairment of all of the learning parameters tested in the present study when compared to control fish. These behavioral effects were associated with a dose-dependent increase in As accumulation in zebrafish brain. Since oxidative stress is a key driver of neurotoxicity, the antioxidative balance in the brain of experimental fish was assessed. A significant decrease in thiol redox and a significant increase in lipid peroxidation were recorded in As-treated fish relative to control, along with the altered expression of enzymatic antioxidant genes. Dopaminergic neurotransmission in the brain regulates important fish behaviors, including learning, memory, and reward-motivated behaviors.

Results:
In the present study, a significant increase in dopamine level and an altered expression of several dopaminergic genes were also observed in the brain of As-treated fish relative to the control fish.

Conclusion:
Overall, it appears that As causes cognitive impairment in zebrafish, likely by inducing oxidative stress and disrupting dopaminergic signalling in the brain.

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

33. INVESTIGATING THE ROLE OF OXIDIZED PHOSPHATIDYLCHOLINE IN GREY MATTER NEURODEGENERATION IN THE CONTEXT OF MULTIPLE SCLEROSIS

Presenter:	**Ruoqi Yu**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Supervisor:	Jeff Dong, College of Medicine

Background:

Multiple Sclerosis (MS) is a chronic neurodegenerative and neuroinflammatory disease affecting the central nervous system (CNS). In Canada, there are 260 cases of MS per 100,000 population and Saskatchewan has amongst the highest incidence of MS. One pathological hallmark of MS is the presence of lesions characterized by inflammation, demyelination, and neurodegeneration. Although MS is formerly regarded as a white matter (WM) disease, grey matter (GM) atrophy occurs early in the acute phase of MS, and the number and volume of GM lesions expand over time. Examination of lesion pathology with post-mortem tissues revealed a destructive role of microglia and macrophages, exacerbating inflammation and oxidative injury. Indeed, oxidative damage such as oxidized phosphatidylcholine (OxPC) derived from normal phosphatidylcholine are found in both WM and GM lesions. Previous studies discovered a neurotoxic nature of OxPC in vitro. In addition, OxPC in vivo injection in the mouse spinal cord WM induces formation of focal lesions. Within these lesions, demyelination and neurodegeneration are identified, and microglia are recruited to lesion sites where they play protective role against OxPC-mediated damage. However, as OxPC are also discovered in both active and inactive GM lesions, it is important to investigate the effect of OxPC in GM. Based on previous WM studies, we hypothesize that OxPC introduction into the spinal cord GM will induce endogenous lipid oxidation, inflammation, and neuron death, leading to acute and chronic neurodegeneration.

Methods:

OxPC are injected into the mouse spinal cord GM with a stereotaxic machine. Acute lesions, or lesions formed at an early timepoint, were acquired from spinal cord tissues seven days post-surgery. Cellular and molecular features of OxPC-induced lesions are characterized by immunofluorescence staining with different antibodies and markers. For example, neurons are marked by NeuN, and microglia and macrophages are marked by Iba1. Neuronal density changes and microglia recruitment are studied by measuring NeuN and Iba1 fluorescent signals.

Results:

Compared to the contralateral side of OxPC injection, the ipsilateral side contained less NeuN density and increased Iba1 fluorescent signals.

Conclusion:

So far, our results suggest that OxPC injection causes NeuN loss and IBA1 infiltration in the GM. The next steps will be further characterization of acute GM lesions, such as with astrocytes, neuronal degeneration, and pro-inflammatory cytokines, as well as investigation of chronic GM lesions.

Basic Science 2

34. TESTING HERPES SIMPLEX VIRUS 1 (HSV1) TRANSCRIPTION FACTOR BINDING TO CHROMATIN

Presenter: **Laura Kellerer**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Kristen Conn, Western College of Veterinary Medicine

Background:
HSV1 gene expression is temporally regulated during lytic infection. Immediate early (IE) genes are expressed first, then early (E), and finally late (L) genes. The IE protein ICP4 is the only essential HSV1 transcription regulator, therefore ICP4 is required for productive infection and progeny virion production. Although critical for infection, we still don't fully understand how ICP4 functions. ICP4 DNA sequence-specific promoter binding, as for IE genes, represses transcription, whereas DNA sequence-independent promoter binding, as for E and L genes, activates transcription. In addition to transcription regulation, ICP4 promotes chromatin exchange of histones, which destabilizes chromatin. The repeating unit of chromatin is the nucleosome, which is 147 base pairs of DNA wound around a histone octamer composed of two histone H2A-H2B dimers and a histone H3-H4 tetramer. The destabilization by ICP4 is consistent with the highly unstably lytic chromatin assembled with transcription-competent HSV1 genomes. If and how ICP4 chromatin destabilization relates to its regulation of transcription remains unknown. As a first step we will evaluate whether ICP4 binds to chromatin.

Methods:
Chromatin electromobility shift assays will be used to test ICP4-Chromatin binding in vitro. Representative HSV1 IE, E, or L promoters will be assembled into chromatin fragments with purified recombinant histone octamers to test ICP4-DNA sequence-specific or non-sequence-specific chromatin binding. ICP4 may directly bind to histone octamers. Thus, we will also test ICP4 binding to chromatin fragments assembled with a DNA sequence that contains no promoter elements.

Results:
So far, all core histones -except H2A- have been cloned into bacterial vectors for recombinant expression and verified. Conditions for bacterial expression of two histones are established.

Conclusion:
Understanding ICP4 HSV1 Chromatin interactions is critical to understand how HSV1 establishes productive infection to produce progeny virions. Knowledge of such mechanisms will highlight critical factors that may be targeted therapeutically to treat or prevent herpesvirus-associated diseases.

35. EFFECT OF PROPOLIS AGAINST EUROPEAN FOULBROOD DISEASE

Presenter:	**Thanuri Edirithilake**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Sarah Wood, Western College of Veterinary Medicine

Background:
Propolis is composed of plant resins collected by bees which they use to seal the cracks and holes of bee hives. Propolis is suggested to have medicinal properties, including antimicrobial and anti-inflammatory effects. A recent in vitro study demonstrated that propolis collected in Ohio inhibits Melissococcus plutonius, the causative bacterium of European foulbrood (EFB) disease of honey bee larvae.

Methods:
we studied the effect of propolis on EFB in larvae and in culture media. Newly hatched larvae were infected with M. plutonius and reared in plastic wells coated with 1.1 mg/mL or 4.6 mg/mL propolis recovered from either North Dakota, Brazil, or Louisiana.

Results:
We found that propolis treatment did not prevent clinical signs of EFB in honey bee larvae; however, treatment with 1.1 mg/mL Louisiana propolis significantly prolonged larval survival from EFB by 36% relative to the infected group. The minimum propolis concentration that inhibits M. plutonius in culture media for Louisiana propolis was four-fold lower than the effective dose on larvae.

Conclusion:
These results suggest that propolis is not an effective therapy for EFB but may lessen clinical severity.

36. UNRAVELING THE REGULATORY NETWORK OF A CRUCIAL PROTEIN: THE DRIVING FORCE BEHIND LYME BORRELIOSIS

Presenter: **Supriya Ramesh**
USask Affiliation: **Graduate student**
College: **College of Medicine**
Supervisor: Jenny Wachter, College of Medicine

Background:
Lyme disease is a bacterial infection caused by the spirochetal bacterium Borrelia burgdorferi, which is transmitted to vertebrates, including humans, through tick bites. B. burgdorferi exists in an enzootic cycle consisting of a hard tick vector and a susceptible vertebrate host. Lyme disease is the most prevalent vector-borne disease in the Northern hemisphere, with over 400,000 new cases per year in the US alone. Transition between an Ixodes tick vector and a competent vertebrate host requires tight regulation of gene expression and protein profiles in response to the environmental cues encountered in these disparate environments. This is achieved by a regulatory cascade that induces an alternate sigma factor, RpoS, which is a prerequisite for host infection. Previous experiments have demonstrated that the gene, bbd18, encoded on linear plasmid (lp)17 encodes a negative regulator of rpoS/RpoS and directly controls the ability of B. burgdorferi to infect mammalian hosts and survive the mouse-tick infectious cycle. Since RpoS is the critical component for B. burgdorferi's vertebrate adapted response, this proposal seeks to further define the BBD18 regulon and identify critical factors for spirochete viability throughout the enzootic cycle that can serve as future prophylactic targets.

Methods:
Preliminary data strongly suggests that BBD18 is a DNA or RNA binding protein and directly effects the expression of 15 genes. We would like to confirm the ability of BBD18 to bind nucleic acids and will start by performing electromobility shift assays using DNA/RNA probes of the 15 genes found to be differentially regulated in the absence of BBD18. To determine additional targets, substrates, and binding partners of BBD18 we plan to perform co-immunoprecipitation experiments followed by sequencing and mass spectrometry.

Results:
The bbd18 gene was amplified from B. burgdorferi strain B31 genomic DNA and cloned into an expression vector. Work is currently ongoing for expression, isolation, and purification of rbbd18.

Conclusion:
As this plasmid-encoded protein is essential for spirochete survival and infectivity, we seek to further define the regulatory network utilized by BBD18 to control the critical component of the spirochete's adaptive response to vertebrate infection, RpoS.

37. HISTATIN 3 ENHANCES THE PHOTODYNAMIC THERAPY EFFECT ON CANDIDA ALBICANS AND STREPTOCOCCUS MUTANS DUAL-BIOFILM

Presenter:	**Luana Dias**
USask Affiliation:	**Graduate student**
College:	**College of Dentistry**
Collaborators:	Lina Marin, College of Dentistry
	Ana Claudia Pavarina
Supervisor:	Walter L. Siqueira, College of Dentistry
Co-supervisor(s):	Ana Claudia Pavarina

Background:
Candida albicans is the main pathogen responsible for the development of denture stomatitis; however, bacteria such as Streptococcus mutans are also found in biofilms formed on acrylic dentures. Using antimicrobial peptides (AMPs) in saliva in conjunction with Photodynamic Therapy (aPDT) has shown promise in combatting C. albicans and S. mutans single-species biofilms.

Methods:
This study assessed the antimicrobial effect of salivary peptide Histatin 3 (Hist 3) combined with Photodynamic Therapy on Candida albicans and Streptococcus mutans dual-species biofilm formed under acrylic resin. C. albicans (polyene resistant-ATCC 200955) and S. mutans (UA159) were previously standardized (37°C/16h/5%CO2). Aliquots of each microorganism with acrylic resin specimens were submitted to acquired pellicle formation (2h/37◦C/60 rpm/min) with Hist 3 (512µM) followed by biofilm maturation (48h). Then, eight experimental groups were assessed, including: aPDT: application of photoditazine (PDZ) photosensitizer of 200 mg/L) for 20 min (pre-incubation time), followed by LED light irradiation (660nm; 50 J/cm2); aPDT + Hist 3: specimens previously treated by Hist 3 were submitted to aPDT LED light: irradiation by LED light; LED light + Hist 3: specimens previously treated with Hist 3 were irradiated by LED light; PDZ: application of 200 mg/L of PDZ photosensitizer; PDZ + Hist 3: specimens previously treated by Hist 3 were submitted to PDZ; Hist 3: peptide treatment only; Control: no treatment. Total biofilm biomass was quantified using crystal violet (200 µL/1%/570 nm) (n=12/group).

Results:
The highest reduction of dual-species biofilm was found in Hist 3 + aPDT group with 67% decrease in biofilm viability, compared to experimental control group ($p>0.05$). The combined treatment was able to reduce 82% and 75% of S. mutans and C. albicans single biofilms, respectively ($p>0.05$).

Conclusion:
Hist 3 was able to potentialize the effect of aPDT on C. albicans and S. mutans dual-species biofilms formed under acrylic resin.

38. IMPACT OF INTEGRIN ALPHA 11 SUBUNIT DEFICIENCY ON CHONDROGENIC DIFFERENTIATION OF MESENCHYMAL PROGENITOR CELLS: INSIGHTS FROM MOUSE EMBRYONIC FIBROBLAST MODELS

Presenter:	**Blessing Ekwueme**
USask Affiliation:	**Graduate student**
College:	**College of Engineering**
Supervisor:	Andrew Leask, College of Dentistry

Background:

Integrins, crucial cell adhesion receptors, regulate various cellular functions by binding to extracellular matrix (ECM) components. Among these, Integrin Alpha 11 (ITGA11) plays a pivotal role in cell functionality and pathologic conditions, including fibrosis, cancer metastasis, and pluripotency regulation. Previous studies have demonstrated its involvement in pluripotency in osteogenic differentiation, where it interacts with osteogenic growth factors like Osteolectin, essential for bone development and maintenance. However, the role of integrin alpha deficiency in ECM genes regulation and chondrogenic differentiation is yet unknown. This research explores the effect of Integrin Alpha 11 subunit deficiency on ECM gene regulation and its role in chondrogenic differentiation within mesenchymal progenitor cells. Utilizing mouse embryonic fibroblast models and micro mass cultures, it is aimed to elucidate the role of ITGA11 deletion on chondrogenesis and its regulation to ECM genes.

Methods:

To test my hypothesis the first process was to verify the deficiency of ITGα11 protein in ITGA11-KO MEFs and to achieve this a western blot was done. Bulk RNA-sequencing (unbiased approach) was done to identify changes in gene expressions in ITGA11-deficient MEFs. Results: RNA-sequencing identified 244 genes (p-value < 0.05, fold change <1.7) that were downregulated, including those involved in extracellular matrix, cell differentiation, developmental protein, and transcriptional activity. Real-time quantitative polymerase chain reaction (RT-qPCR) was conducted to validate the RNA-sequencing results for specific genes. Results: Deficiency of ITGA11 gene in MEFs causes a downregulation of ECM cartilage and pluripotency markers. The second step was conducting an invitro study using micro mass culture with 1x105cells (mouse embryonic fibroblast, MEFs) in 10ulmedia plated in 24well plate. Incubation of these cells was done for 2hrs after 500ul of differentiation medium (100ng/ml of Bone morphogenic Protein (BMP2) + 15% FBS in DMEM) was added in each well. The Differentiation media was changed after every 2 to 3 days and the cells were cultured for 14dyas. To quantify gene expressions RNA was extracted from both groups and Real-time quantitative polymerase chain reaction (RT-qPCR) was conducted to validate the RNA-sequencing results for specific genes. Alcian Blue Staining on Micro mass cell culture was used to identify proteoglycan formed because of Cartilage formation. After 14days, the plates were washed with PBS, fixed with formalin, and stained with 500ul of Alcian Blue stain (0.5% in 3% acetic acid). All plates were wrapped with Safran paper and kept overnight at room temperature. Then Destaining was done using 70% Ethanol and imaging done. To measure the absorbance, samples were then incubated with 2ml of 6M guanidine HCL for 8hours at RT.

Results:

RNA-sequencing identified 244 genes (p-value < 0.05, fold change <1.7) that were down regulated in the absence of ITGA11, including those involved in HIF-1α pathway, extracellular matrix, cell division, and cellular response to hypoxia. RT-PCR analysis confirmed the downregulation of several genes involved in the extracellular matrix, such as COL2A1, DCN,TNC, ACAN and SOX9, SOX2 genes involved in pluripotency. RT-PCR analysis of Micro mass cultures showed the downregulation of chondrogenesis markers COL2A1, DCN, ACAN and pluripotency markers SOX2, SOX9 due to the deficiency of ITGA11 in MEFs.

Conclusion:

Our findings reveal that deletion of ITGA11 in mouse embryonic fibroblasts resulted in decreased induction of ECM component genes and cartilage gene markers involved in chondrocytes differentiation.

39. REPRODUCIBLE SYNTHESIS OF PLGA NANOPARTICLES VIA SOLVENT EVAPORATION: A METHODOLOGICAL BLUEPRINT FOR PRECISION PARTICLE ENGINEERING

Presenter:	**Arash Amanlou**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Supervisor:	Azita Haddadi, College of Pharmacy and Nutrition

Background:
The purpose of this study was to develop a reproducible method for the preparation of poly(lactic-co-glycolic acid) using the solvent evaporation technique, aiming for a low polydispersity index (PDI), tunable particle size, and zeta potential. This study strived to provide a clear and comprehensive methodology that can be readily adopted by other researchers and displayed some of the often neglected details that can lead to a significant impact.

Methods:
Variables that were investigated in this study were polyvinyl alcohol (Two molecular weights and hydrolysis rates, varying concentrations, two solutions/buffers, concentration validation, preparation method), needle (different gauges and lengths), Injection rate (different injection rates), centrifugation (differential centrifugation, two-step centrifugation at different speeds and duration), wash cycles, sonication cycles (ice bath, generated energy, sample rotation), magnetic stirring bar (different lengths), dynamic light scattering (considerations).

Results:
This study explored the impact of variable adjustments on the physicochemical properties of PLGA nanoparticles. The reproducibility of findings was demonstrated across multiple experiments and replicates. Facile manipulation of variables yielded tunable particle size and zeta potential while achieving a more uniform nanoparticle distribution (low Polydispersity Index). These findings highlight the method's effectiveness and potential for precise nanoparticle synthesis.

Conclusion:
A clear methodology and material section in a PLGA nanoparticle synthesis study will enhance the clarity and comprehensiveness and it will facilitate understanding and implementation of the method by other researchers. The reproducibility of the study is evidenced by obtaining consistent results during the replicates and multiple experiments. Tunable particle size and a low polydispersity index are achieved by understanding the solvent evaporation method and emulsion preparation science.

40. HOST RESTRICTION FACTORS ZAP AND APOBEC3G WORK TOGETHER TO RESTRICT HIV-1 REPLICATION

Presenter:	**Shreoshri Bhattacharjee**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Amit Gaba, College of Medicine
Supervisor:	Linda Chelico, College of Medicine

Background:

The human APOBEC3 family of cytidine deaminases constitute a cellular intrinsic defense mechanism that is effective against a range of viruses and retro-elements. While these enzymes are well established as antivirals, the possibility of decoding their protein-protein interaction network using affinity purification and mass spectrometry (AP-MS) will help in recognising their range of activity and influence on various molecular/ immune pathways. One such association of APOBEC3G (A3G) with another antiviral factor, ZAP, zinc-finger antiviral protein (also called ZCCHV) has been elucidated. ZAP is a Pattern Recognition Receptor that binds single-stranded RNA (ssRNA) and inhibits viral replication through multiple mechanisms, including targeting RNA for degradation and inhibiting its translation. It does so by binding to CpG dinucleotides present mostly in non-self RNA. The human genome has less CpG dinucleotides than would be expected and accounts for the ability of ZAP to differentiate self and non-self RNA. Both, A3G and ZAP are known to interact with HIV RNA. However, ZAP action against HIV is in the virus producing cell and A3G action against HIV is in the newly infected cell.

Methods:

To determine the protein interaction network for the human APOBEC3 enzymes which uncovered a diverse number of protein-protein and protein-RNA mediated interactions, differentiated by conducting the AP-MS in the presence and absence of RNAseA. Surprisingly, only APOBEC3G (A3G) has been found to interact with another antiviral factor ZAP, zinc-finger antiviral protein (also called ZCCHV). To determine the restriction capacity of these host proteins alone and in combination with each other, we have used cell based transfection system to express A3G and ZAP alone and with each other in HEK 293T Zap knock out cells in presence of HIV-1 transmitted founder virus 77 (CH077). For a better understanding, if the amount of viable virus produced is co relative to infectivity levels of CH077 when A3G and ZAP are together we did real time qPCR assay which lead us to the check the mode of action of restriction by each of these host proteins. The experimental outline and results are in process and will be discussed.

Results:

In this study we have observed a sharp decrease of HIV infectivity in presence of A3G and ZAP together. The lowered rate of infection result is co-relative to lesser production of functional virions in presence of these two antiviral factors. Along with an effect on the viral fitness, does this protein-protein interaction of A3G and ZAP have an affect on each other's mutagenic RNA editing or viral translatory blockage activities are in the process to be determined and discussed.

Conclusion:

Antiviral host factors Zinc Finger Antiviral Protein and APOBEC3G are able to reduce HIV-1 infection.

41. IMBALANCE OF RNA BINDING PROTEINS DRIVES NEURODEGENERATION

Presenter:	**Miranda Messmer**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Hannah Salapa
	Bogdan Popescu, College of Medicine
	Michael Levin, College of Medicine
Supervisor:	Michael Levin, College of Medicine
Co-supervisor(s):	Bogdan Popescu

Background:

Irreversible disability in neurodegenerative disease is caused by neuron cell death and axon damage. RNA binding proteins (RBP) like heterogeneous nuclear ribonucleoprotein A1 (A1) are required for neuron function and survival. A1 dysfunction is characterized by mislocalization from its homeostatic nuclear location to the cytoplasm. Mislocalization results in loss of A1 function in the nucleus and gain of toxicity in the cytoplasm. Both processes cause neurodegeneration, but the cellular mechanisms remain unknown. Expression of NeuN, a neuron specific RBP vital for neuronal structure and function, is reduced with neuronal injury. Thus, we hypothesize that A1 dysfunction impairs NeuN expression and contributes to neurodegeneration.

Methods:

Loss of A1 function was modelled in vitro by CRISPR/Cas9 mediated A1 knock-out in neuronal-like cells. NeuN expression was quantified by PCR and western blot. Gain of toxic A1 function was modelled in vivo by overexpressing A1 in hippocampal neurons by intracranial injection of viral vectors containing A1 RNA. A1 binding to NeuN RNA was evaluated by cross-linking and immunoprecipitation sequencing (CLIP-seq). A1 mislocalization, neurodegeneration, and NeuN expression in situ were analyzed by immunostaining.

Results:

In vitro loss of A1 function led to alternative splicing ($p=0.001$) and reduced NeuN protein ($p=0.017$) but not reduced RNA. CLIP-seq validated A1 binding to NeuN mRNA in vivo. With A1 overexpression, NeuN staining was lost in hippocampal neurons up to 3 weeks post-transduction. In human brain, A1 mislocalization colocalized with axonal injury and correlated to loss of neuronal NeuN staining ($p=0.003$) and reduced neuronal density ($p=0.024$).

Conclusion:

A1 dysfunction post-transcriptionally downregulates NeuN and can chronically reduce NeuN immunoreactivity in hippocampal neurons. Thus, A1 may be required for maintenance of NeuN expression and preservation of neuronal structure and function with injury. This study identifies a novel function of A1 in regulating neuronal health and an underlying neurodegenerative mechanism of A1 dysfunction.

42. IDENTIFICATION AND SEPARATION OF A MAJOR INTERFERENCE IN THE LC-MS/MS QUANTITATIVE ANALYSIS OF PSILOCIN IN MOUSE PLASMA

Presenter:	**Amir Khajavinia**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Deborah Michel, College of Pharmacy and Nutrition
	Randy Purves, College of Pharmacy and Nutrition
	Robert Laprairie, College of Pharmacy and Nutrition
Supervisor:	Anas El-Aneed, College of Pharmacy and Nutrition

Background:

Psilocin is the active metabolite of psilocybin, a psychedelic substance found in some hallucinogenic "magic mushrooms". Psilocin is under study for the treatment of various mental health disorders, such as depression, anxiety, and obsessive-compulsive disorder. The ongoing research on the uses of psilocin requires the evaluation of its pharmacokinetic properties. Therefore, simple, accurate and sensitive analytical methods are needed to quantify psilocin in biological matrices. A rapid analytical technique for the analysis of psilocin was tested, namely flow-injection (FIA)-MS. The analysis showed a major interference in mouse plasma that was not, to the best of our knowledge, reported before. The aim is to identify and separate the interference using analytical mass spectrometry.

Methods:

A quadrupole-linear ion trap (6500 QTRAP®) instrument equipped with an electrospray ionization (ESI) source was utilized in the positive ion mode, using multiple reaction monitoring (MRM). Several chromatographic conditions and column chemistries reported in the literature, including C-18, Phenyl, and Phenyl-hexyl were applied and failed to separate the interference. A high-resolution mass spectrometer, namely Quadrupole-Orbitrap MS (Q Exactive®) was used to obtain the exact mass and MS/MS spectrum of the interfering compound.

Results:

Exact mass measurement and MS/MS analysis showed the structure of the interfering compound as Tryptophan. Therefore, chromatographic columns with hydrophilic interaction chemistries were tested due to the hydrophilic nature of psilocin and the interference. Reversed phased columns like C-18, phenyl, or phenyl-hexyl were not able to retain and separate the compounds using isocratic or gradient mobile phase conditions. Among the six monitored MRM transitions, tryptophan shared five transitions with psilocin; m/z 205.1 → m/z 58.1 was psilocin's only unique transition. However, in order to have an accurate and selective method, at least two transitions are required since ion ratio is a key criterion that is used to ensure peak purity in animal samples. The two compounds were separated capitalizing on the pH of the mobile phase and the chemistry of the stationary phase.

Conclusion:

A major interference in the LC-MS/MS quantification of psilocin in mouse plasma was identified and separated for the first time.

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43. PROFILING CANCER STEM CELLS USING MODULAR TISSUE ENGINEERING DESIGN

Presenter:	**Nima Daneshvar Baghbadorani**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Mira Bosso, College of Medicine
	Rowen Greene, College of Medicine
Supervisor:	Dean Chambeerlain, College of Medicine

Background:
The tumor microenvironment (TME) and its features, such as hypoxia, extracellular matrix, immune cell surveillance are more effective drivers of cancer stem cells (CSCs) than intrinsic oncogenic mutations. However, one of the major challenges for characterization of CSC populations and their properties is development of in vitro models capable of maintaining these external drivers. Modular tissue engineering is one of the methods that allows us to develop cancer microtissues composed of cancer cells embedded in collagen hydrogel as the ECM, which can be enriched with various types of stromal cells to better resemble additional features of a functional TME. We hypothesize to observe expansion of CSC populations in our 3D model compared to 2D-cultured cancer cells.

Methods:
The preliminary analyses were based on the triple negative breast cancer cell line HCC1806 cultured in microtissues. This included assessment of morphology by H&E, proliferation by immunohistochemical staining of Ki-67, viability by live/dead staining, and CellTiter-Glo® 3D assays, and hypoxia by hypoxiTRAK staining. Additionally, the CSC markers were analysed by flow cytometry.

Results:
Our preliminary data showed that microtissue-cultured HCC1806 cells remain viable and proliferative with minimal cell death. Moreover, the establishment of a hypoxic core has also been shown to start developing by day 10. The potency of both paclitaxel and doxorubicin was reduced in killing cells grown in microtissues, while paclitaxel efficacy was also notably diminished. The flow cytometric analysis of CSC populations showed upregulation of CSC markers in microtissues-cultured HCC1806 cells compared to 2D-cultured ones.

Conclusion:
Our preliminary results proved the capacity of this model to maintain a proper environment for growth and proliferation of cancer cells, and are also in accordance with our hypothesis regarding expansion of CSC population, showing the potential of this model to mimic the drivers of CSCs present in an actual TME.

44. MYELOID NEOPLASMS ASSOCIATED DDX41 HELICASE IS ESSENTIAL FOR P-BODY FORMATION

Presenter:	**Lacey Winstone**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Ananna Bhadra Arna, College of Medicine
	Ravi Shankar Singh, College of Medicine
	Shizhuo Yang, College of Medicine
Supervisor:	Yuliang Wu, College of Medicine

Background:

Myeloid neoplasms (MNs) are clonal disorders that arise through the acquisition of genetic mutations leading to excessive proliferation and defective differentiation. The most frequent mutations are SF3B1, TET2, ASXL1, SRSF2, DNMT3A, RUNX1, U2AF1, ZRSR2, STAG2, TP53, EZH2, CBL, JAK2, BCOR, IDH2, NRAS, and NF1 genes. Mutations in DDX41, both germline and somatic, have been recently associated with MNs, particularly myelodysplastic syndrome (MDS) and acute myeloid leukemia (AML). MDS is categorized by unsuccessful hematopoiesis and peripheral blood cytopenia, whereas the spread of immature myeloid cells categorizes AML in the bone marrow, and one third of MDS patients will progress over their lifetime to AML. DDX41 is a DEAD-box helicase, which contains the conserved DEAD (Asp-Glu-Ala-Asp) sequence. Helicases are ubiquitous enzymes that transduce the chemical energy generated from nucleoside triphosphate hydrolysis to the separation and displacement of oligonucleotide strands. They are involved in virtually all aspects of nucleic acid metabolism, including replication, repair, recombination, transcription, chromosome segregation, and telomere maintenance. Despite the increasing number of DDX41 clinical mutations, how DDX41 dysfunction results in MDS/AML is still not clear. Processing bodies (P-bodies) are cytoplasmic ribonucleoprotein granules comprised primarily of decapped mRNAs in complex with various proteins associated with translation repression and 5′ to 3′ mRNA decay. P-bodies exist in normal conditions but increase in size and frequency when cells undergo stress. Many proteins, including DCP1A (decapping mRNA complex), EDC4 (decapping complex component), LSM14A (mRNA decay factor), and 4E-T (translational repression factor), are vital for P-bodies formation. P-bodies follow a dynamic of assembly and disassembly according to their local cellular condition, and their dysregulation has been associated with various cancers and diseases.

Methods:

Cell culture and treatment. Using the CRISPR technology, HeLa DDX41 knockout (KO) cell line has been established. DDX41 KO and wildtype (WT) cells were grown in DMEM (10% FBS and penicillin/streptomycin). Cells were treated with sodium arsenite (0.5 mM) or glucose-free DMEM for 1h and collected at various time points (0-8h). Western blot analysis. Proteins were separated on 10% SDS-polyacrylamide gel and transferred onto a PVDF membrane. The membrane was incubated with 5% BSA for 1h at room temperature, then a primary antibody (1:500) at 4°C overnight. The following day, the membrane was incubated with a secondary antibody (1:3000) for 1h, and washed, treated with Clarity Western ECL Substrate and visualized with ChemiDoc Imaging System (Bio-Rad). Immunofluorescence. Cells were grown on slides, fixed with 100% methanol, and blocked with 3% BSA. Cells were incubated with a primary antibody (1:1000) overnight at 4°C, washed with PBS, and incubated with a fluorescent secondary antibody (1:1000). Cells were washed and mounted using Prolong Diamond antifade containing DAPI (Invitrogen) and cured at room temperature in the dark for 24h. Immunofluorescence was conducted on a Zeiss laser scan microscope with a 63×oil objective.

Images were captured and analyzed using Zeiss. GFP-tagged-DCP1A plasmid was transfected to HeLa cells using Lipofectamine 3000 (Invitrogen). Cells were collected 24h post-transfection and treated as described above and observed directly under an Zeiss microscope. Quantitative real-time PCR. RNA was extracted using TRIzol and reverse transcribed using SuperScript II Reverse Transcriptase (Invitrogen). mRNA expression was measure by qRT-PCR using Luna universal qRT-PCR master mix (NEB) on a StepOnePlus Real-Time PCR system (Applied Biosystem). GAPDH was used as an internal reference. Relative quantification was calculated using $2-\Delta\Delta Ct$.

Results:

Using P-body markers EDC4 and DCP1A, we observed P-bodies formed within the cells, peaked at 2h, and disassembled after $NaAsO_2$ or glucose starvation treatment. Compared to the WT, we found that the KO cells had fewer P-body foci. Our results indicate that DDX41 is required for P-body formation under stress conditions. To determine if DDX41 is localized in the P-bodies, we co-stained cells with P-body marker EDC4 and DDX41 and found that DDX41 was predominantly localized in the nucleus, whereas the P-bodies are in the cytoplasm. Hence, DDX41 does not directly interact with the P-bodies. To understand the mechanisms how DDX41 regulates P-body formation, we conducted Western blot analysis to investigate the expression of the P-body markers EDC4, 4E-T, LSM14A, and DCP1A. Our blots revealed that DDX41 KO samples had reduced expression of EDC4 and 4E-T compared to the WT. However, DDX41 did not influence LSM14A and DCP1A. Next, we transfected the WT and DDX41 KO cells with an GFP tagged-DCP1A plasmid and observed the P-bodies followed the same dynamic as the endogenous P-bodies. However, we found that the foci frequency was comparable between the KO and the WT. Therefore, DDX41 may have a role in splicing as endogenous P-body proteins are affected but not exogenous proteins. To determine DDX41's potential role in the splicing of the P-body genes, we performed qRT-PCR. Our preliminary results showed that the mRNA levels of EDC4, 4E-T, and DCP1A genes were lower in the KO cells compared to the WT cells, indicating DDX41 has a potential role in the splicing for these genes. We will investigate the alternative variants of these genes to verify this result. Lastly, we are conducting similar Western blot and immunofluorescence experiments on WT and Ddx41 conditional KO mouse cells and DDX41-mutated MDS/AML patient cells.

Conclusion:

We have found that DDX41 is required for P-body formation under various stress conditions; however, DDX41 is not localized in P-bodies. DDX41 deficiency causes reduced levels of some P-body essential proteins, but not all. DDX41 also shows potential for requiring splicing of the P-body genes since it does not affect exogenous P-body formation. These results will be verified in mouse primary cells and patient cells. Our results will shed light on new molecular pathogenesis and potential targets for diagnostics and treatments for DDX41-mutated MDS/AML.

45. THE GENETIC SYMPHONY ORCHESTRATING TOOTH DEVELOPMENT

Presenter: **Nooshin Shekari**
USask Affiliation: **Graduate student**
College: **College of Medicine**
Supervisor: Julia Boughner, College of Medicine

Background:
Understanding how jaws and teeth develop independently in vertebrates is important to understand the proper growth of animal faces. The p63 gene regulatory network (GRN), is essential for tooth development in vertebrates including rodents and fishes. This project focuses on zebrafish, which have pharyngeal teeth but naturally lack oral teeth, and medaka, which have both pharyngeal and oral teeth. We hypothesis the p63 GRN is active similarly in the pharyngeal tissues of both fish species and active only in the oral tissues of medaka. This comparison between teleost fishes studies the p63 GRN's role on dentitions located in two different types of jawbone.

Methods:
Wildtype medaka (stage 40) and zebrafish (4.5 days post-fertilization) were dissected to collect oral and pharyngeal prominence from each fish (n=3 replicates per species). RNA was extracted, sequenced, and aligned to the reference genome. Differential gene expression analysis identified genes with significant differences between oral and pharyngeal tissues, and potentially between fish species.

Results:
RNA-sequencing of oral and pharyngeal prominences in zebrafish and medaka revealed differential expression of p63 network genes (Cbln1, Cldn23, Pltp, Krt15). These genes were more highly expressed in the toothless zebrafish oral prominence compared to the toothed pharyngeal arch prominences. However, in toothless oral tissue of zebrafish compared to pharyngeal primordia the data showed higher expression of genes including dkk1a/b which inhibit tooth development.

Conclusion:
The observed p63 GRN expression pattern was similar in both fishes, suggesting evolutionary conservation of GRN function across teleost fishes. Several p63 network genes were expressed at higher levels in the toothless oral prominences compared to the toothed pharyngeal arch prominences. Some of these genes mirrored the expression levels in mice and some not, this can be due to different development stages and can be further examined in the future.

46. DEGRADATION OF HISTATIN 5 BY PORPHYROMONAS GINGIVALIS AS A NOVEL BIOMARKER OF PERIODONTAL DISEASE

Presenter:	**Andrea Escalante**
USask Affiliation:	**Graduate student**
College:	**College of Dentistry**
Collaborators:	Lina Marin, College of Dentistry
	Walter Siqueira, College of Dentistry
Supervisor:	Walter Siqueira, College of Dentistry
Co-supervisor(s):	Lina Marin

Background:

Periodontal disease, a silent threat worldwide, compromises oral health, leading to tooth mobility and diminished quality of life. Dysbiosis, disrupting the host-microorganism balance, fosters the proliferation of specific oral microbiota in defined environments, culminating in periodontitis. Porphyromonas gingivalis is considered the key etiological agent in the pathogenesis and progression of the inflammatory events that occur during periodontal disease development. Histatins, salivary proteins susceptible to degradation by oral proteolytic enzymes, undergo varied rates and modes of degradation in healthy and periodontitis-afflicted individuals, indicating that proteases produced by periodontopathogenic microorganisms may play a crucial role in this degradation process. This study aimed to determine if the key periodontopathogen, Porphyromonas gingivalis, degrades histatin 5, to determine its degradation rate and mode, identify the possible enzymes that cause the degradation and compare to other oral microorganisms such as Fusobacterium nucleatum, Streptococcus mutans and Aggregatibacter actinomycetemcomitans.

Methods:

To evaluate the degradation rate and mode of histatin 5, 100 µL of each bacterial inoculum with varying concentrations (108, 107, 106) were mixed individually with synthetic histatin 5 (0.5 µg/µL) for 0, 2, and 6 hours. The negative control was prepared by reconstituting synthetic histatin 5 in 100 µL of PBS, to a final concentration of 0.5µg/µL. Each suspension having P. gingivalis and F. nucleatum was incubated in anaerobic conditions at 37°C, while the bacterial suspension having S. mutans and A. actinomycetemcomitans was incubated in a water bath at 37°C. At each time point, aliquot samples were boiled for 5-min to stop any proteolytic activity and then processed for cationic polyacrylamide gel electrophoresis (PAGE). The percentage of histatin 5 degradation was determined by comparing the resulting band densities with a reference control having 0.5µg/µL of histatin 5 in PBS. The experiment was done in triplicate. The supernatant of the samples was taken and desalted before Liquid Chromatography–Electrospray Ionization–Tandem Mass Spectrometry (LC-ESI-MS/MS) analysis to identify the enzymes that are naturally produced by bacteria. The acquired MS/MS spectra were queried against the bacteria database (Swiss Prot and TrEMBL) using the SEQUEST algorithm in Proteome Discoverer 2.4 software, filtering by natural peptides.

Results:

The results showed that histatin 5 was degraded only by P. gingivalis at all time points evaluated and exhibited faster degradation with increased bacterial concentration. On the contrary, F. nucleatum, S. mutans and A. actinomycetemcomitans did not degrade histatin throughout the experiment. The supernatant analysis revealed 61 proteins, with gingipains, P. gingivalis' main virulence factors, being particularly relevant. Gingipains are the main virulence factor of P. gingivalis; they exhibit proteolytic

activity and are directly related to periodontal tissue destruction. Some gingipains were found at all time points.

Conclusion:
Altogether, these findings demonstrate that P. gingivalis plays a key role in the degradation of histatin 5. In higher concentrations, the enzymes released by P. gingivalis can degrade histatin 5 more quickly and produce unique fragments. The degradation of histatin 5 by P. gingivalis may represent a novel, innovative and promising biomarker of periodontal disease and could change how we diagnose and track periodontal health.

47. CHARACTERIZING SUBSTRATE BINDING PROTEINS OF A PUTATIVE MALTOSE UPTAKE SYSTEM IN GARDNERELLA SWIDSINSKII

Presenter:	**Agnes Truc Nguyen**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Janet E Hill, Western College of Veterinary Medicine

Background:
The MusEFGKI system is known to be involved in the uptake of glycogen breakdown products such as maltose and malto-oligosaccharides by bacteria, and MusEFGKI operons have been detected in Gardnerella species from the human vaginal microbiome. This transport system includes substrate binding proteins (SBPs) that recognize and bind to specific ligands, facilitating the uptake process. Unlike other Gardnerella species that have only one MusE SBP, G. swidsinskii encodes two MusE SBPs. We hypothesize that these MusE SBPs differ in ligand-binding properties providing G. swidsinskii with competitive advantages in accessing glycogen breakdown products by broadening the range of ligands. This study aims to biochemically characterize specificity and affinity of the two G. swidsinskii MusE SBPs for maltose and malto-oligosaccharides.

Methods:
Phylogenetic analysis of representative MusE SBPs in all Gardnerella species was performed. Plasmids containing codon-optimized sequences for G. swidsinskii SBPs of interest (annotated as MusE 1345 and MusE 1346) fused with a 6-Histidine tag and/or a GST tag to increase protein expression and ease of purification were constructed. E. coli was transformed with plasmids and induced for protein expression.

Results:
The two G. swidsinskii SBPs share only 62.2% protein sequence identity. While MusE 1346 clustered with SBPs from other Gardnerella species in the protein sequence phylogeny, MusE 1345 formed a separate clade. Production of both SBPs in E. coli was successful. Despite initial difficulties in expressing and acquiring soluble proteins, both SBPs are now under the process of purification.

Conclusion:
Two phylogenetically distinct MusE SBPs in G. swidsinskii were successfully expressed and will be purified for isothermal titration calorimetry experiments to measure specificity and affinity for maltose and malto-oligosaccharides.

48. INVESTIGATING THE CYTOTOXIC POTENTIAL OF PORCINE GAMMA-DELTA T CELLS

Presenter: **Leonie Bettin**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Volker Gerdts, Western College of Veterinary Medicine

Background:
Pigs could be an excellent biomedical model for human diseases due to their similarity in size, anatomy and physiology. However, the porcine immune system is surprisingly understudied and, therefore, difficult to compare to humans. One peculiarity of the pig is its high number of gamma-delta (γδ) T cells in circulation. Approximately 50% of circulating T cells are γδ T cells in young pigs, which is 6-12 times more than in humans. Despite this abundance, the functions of porcine γδ T cells are mostly unidentified. In humans, activated γδ T cells exhibit broad innate cytotoxic activity against stressed, infected and cancerous cells through death receptor-dependent and perforin/granzyme-dependent pathways. However, so far, it is unknown, if porcine γδ T cells can perform cytolytic functions.

Methods:
In this study, we performed a detailed phenotypic characterization of porcine γδ T cells by flow cytometry followed by a series of in vitro co-culture assays to investigate their potential to kill target cells.

Results:
Our results show that only CD2+ γδ T cells express cytotoxic markers (CD16, NKp46, perforin) with higher perforin and NKp46 expression in γδ T cells isolated from lung and nasal mucosa. To further analyze γδ T cell's cytolytic potential, cytotoxicity assays were performed using purified γδ T cells as effector cells and virus-inoculated or mock-inoculated primary alveolar macrophages as target cells. We find that γδ T cells exhibit cytotoxic responses but do not show selectivity for virus-infected cells. Nevertheless, the cytotoxic responses by γδ T cells are cell-cell contact dependent and seem to involve perforin production.

Conclusion:
Overall, this data shows that porcine γδ T cells (CD2+) express cytotoxic markers and can kill other cells, potentially involving the perforin/granzyme-dependent pathway. This may indicate an important role of porcine γδ T cells in stress surveillance, which is a known function of human γδ T cells.

49. EXAMINING THE EFFECTS OF HNRNP A1 KNOCKOUT IN MOUSE CORTICAL NEURONS IN VIVO

Presenter: **Fariba Karami**
USask Affiliation: **Graduate student**
College: **College of Medicine**
Supervisor: Michael Levin, College of Medicine

Background:
Incomplete understanding exists regarding the role of dysfunctional RNA binding proteins (RBPs) like heterogeneous nuclear ribonucleoprotein A1 (A1) in the pathogenesis of neurodegenerative diseases. While A1 is known to play a role in various RNA metabolic processes in neurons, such as transcription, splicing, mRNA transport, and translation, the precise mechanisms linking its dysfunction to neurodegeneration are not fully elucidated. In vitro studies suggest that loss of A1 induces transcriptional changes leading to neurodegeneration, however, studying the effects of A1 knockout in vivo are hindered by its embryonic lethality. To address this gap, we developed a new mouse model allowing for inducible, controlled A1 knockout in cortical neurons.

Methods:
For this, we generated a mouse with Cre-inducible Cas9 expression under the control of the neuron-specific Thy1 promoter. Following tamoxifen injections to induce Cas9 expression, mice are intracranially injected with an adeno-associated virus (AAV) containing single guide RNA targeting A1 (AAV-sgRNA(A1)) to knockout A1 in Cas9 expressing neurons.

Results:
First, we confirmed successful Cas9 expression in Thy1+ neurons following tamoxifen injections. Here, we found Cas9 expression throughout the brain, including in cortical layer V neurons. Subsequently, we validated the efficacy of our sgRNA targeting A1 in vitro, which demonstrated significant knockout efficiency ($p<0.05$). Finally, intracranial injections of AAV-sgRNA(A1) into these mice resulted in A1 knockout in cortical neurons in vivo, thus validating our novel animal model.

Conclusion:
Overall, our novel mouse model provides a platform to investigate the effects of A1 loss in neurons. By elucidating the mechanistic links between A1 dysfunction and neurodegeneration in vivo, we aim to enhance understanding of the pathogenesis of neurodegenerative diseases.

50. DIFFERENT EFFICIENCIES OF APOBEC3 DEGRADATION BY HIV-1 AND HIV-2 VIFS

Presenter: **Maria Yousefi**
USask Affiliation: **Graduate student**
College: **College of Medicine**
Supervisor: Linda Chelico, College of Medicine

Background:
APOBEC3 (A3) enzymes play a crucial role in the intrinsic antiviral response against retroviruses, such as HIV. If A3 enzymes avoid Vif, the HIV Vif protein that induces their degradation through the proteasome, the A3 enzymes can encapsidate into newly forming HIV virions and upon infection deaminate Cytidine (C) to Uracil (U) on the (-) DNA when it is single-stranded during reverse transcription. This causes G to A mutations in the (+) DNA. Since A3-induced mutations are stochastic and determined by the ability to resist Vif, high levels of mutations inactivate the proviral DNA, but lower levels of mutation may cause viral evolution. HIV has two types: HIV-1 and HIV-2. A3 enzymes contain seven members, four of which can restrict HIV-1 (A3D, A3F, A3G, and A3H). HIV-1, the causative agent of most infections worldwide, is more virulent and infective than HIV-2. The Vifs from HIV-1 and HIV-2 both induce degradation of A3 enzymes but have only 30% amino acid similarity and HIV-2 Vif induces degradation of A3 enzymes more efficiently than HIV-1 Vif. However, when A3 enzymes are expressed in combination, they can interact with each other and resist HIV-1 Vif-mediated degradation. It is not known if A3-A3 interactions also enable resistance to HIV-2 Vif.

Methods:
Cell culture techniques and molecular biological techniques, such as DNA extraction, PCR, quantitative PCR DNA cloning and sequencing are considered basic laboratory techniques in my research area. My research is focused on HIV. Research on this virus should be conducted using biosafety level 2/3 containment (Biosafety level 2 plus). Biosafety level 2 plus requires different safety measurements to work safely with life-threatening viruses. Computational analysis plays a significant role in interpreting our data and making it understandable. Some techniques like DNA sequencing require further simplification to draw more meaningful conclusions.

Results:
We found that A3-A3 interactions do not enable high resistance to HIV-2 Vif in comparison to HIV-1 Vif.

Conclusion:
These data suggest that HIV-2 Vif is likely to incur less proviral DNA mutations than HIV-1 Vif. The implications of these differences on viral evolution shed some light on Virus-host interaction.

51. ELECTRON PARAMAGNETIC RESONANCE SPECTROSCOPY TO DETECT REACTIVE OXYGEN SPECIES IN ZEBRAFISH

Presenter:	**Mitra Sabetghadam**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Eli Wiens
	Ramaswami Sammynaiken
	Sébastien Gauvrit
	Michelle M.Collins, College of Medicine
Supervisor:	Michelle M. Collins, College of Medicine

Background:
Electron paramagnetic resonance (EPR) is an excellent choice for the detection of reactive oxygen species (ROS) in vivo. Biologically relevant ROS are extremely short-lived and cannot be detected directly, emphasizing the need for an appropriate compound to generate stable adducts that can be measured by EPR. Spin trapping using 5,5-dimethyl-1-pyrroline N-oxide (DMPO) is often used for ROS detection within biological tissues. However, there are numerous drawbacks associated with this method. The cell-permeable cyclic hydroxylamine probe 1-hydroxy-3-methoxycarbonyl-2,2,5,5-tetramethyl pyrrolidine (CMH), offers a more advantageous means for quantitatively assessing ROS.

Methods:
Here we present a protocol for utilizing EPR using a CMH spin probe to detect ROS, specifically O2·– in zebrafish intact heart tissue and larvae.

Results:
This study focuses on the zebrafish heart and larvae, providing a method to assess in vivo oxidative stress through the application of the EPR spin probe.

Conclusion:
Together, our data shows that EPR using the CMH probe is a reliable method to detect ROS in zebrafish pathologies linked to oxidative stress, such as cardiovascular diseases.

52. GENETIC ANALYSIS OF THE PHENOTYPIC PLASTICITY OF MYCOBACTERIUM TUBERCULOSIS

Presenter: **Nandini Nandini**
USask Affiliation: **Graduate student**
College: **School of Public Health**
Supervisor: Neeraj Dhar, College of Medicine

Background:
Tuberculosis, caused by Mycobacterium tuberculosis (Mtb), is one of the leading causes of mortality responsible for 1.6 million deaths every year. Mtb has evolved mechanisms to persist in the host and survive against immune and antibiotic stress. Also, Mtb exists in a latent form in 25% of the world's population. This latently infected population serves as a reservoir potentially leading to reactivation and spread of the disease and hampers our efforts towards eradication. Understanding the mechanisms of persistence, latency and reactivation can help us develop novel strategies to target this pathogen. Towards this I propose two broad approaches: 1) We hypothesize that fluctuations in specific pathways in Mtb mediate the latency-reactivation, cell-fate decisions. For example, in case of HIV, fluctuations in Tat protein were found to be strongly correlated with latency. Therefore, in an unbiased approach to identify pathways that are phenotypically noisy and driving heterogeneity in Mtb, I propose to construct a genomic scale promoter library. Phenotypic noise in the generated strains will be quantified using flow cytometry. 2) In bacteria, two component systems (TCS), composed of a sensor kinase and a response regulator, mediates cellular responses to external stimuli. Mtb genome encodes for 14 sensor kinases and 16 response regulators. In a targeted approach to identify if some of these TCS play a role in latency and/or reactivation, I propose to study the transcriptional dynamics of these TCS in vitro at the single cell level and in ex vivo models of latent TB.

Methods:
Approach 1: 1. Isolation and digestion of Mtb genomic DNA to generate promoter library fragments. 2. Construction of promoter library plasmid 3. Cloning of promoter library fragments and electroporation of library into Mtb 4. Screening of promoter library grown under axenic conditions using flow cytometry 5. Evaluate phenotypic noise upon exposure to immune-related or antibiotic stress. Approach 2: 1. Construct transcriptional fusion strains for each of these TCSs. 2. Study gene expression dynamics of each of these TCSs in vitro at the single cell level using time-lapse imaging. 3. Capture the response kinetics of these TCSs in response to antibiotic and host environment associated stresses. 4. Construct promoter mutants or deletion strains to validate the in vitro observations.

Results:
Till now I have successfully cloned the Promoter Library in E.coli. and now I'm cloning it in Mtb and simultaneously optimizing the flow cytometry conditions for my library screening. Early indications from the optimization are promising, fluorescence signals are detectable.

Conclusion:
My optimization data suggests successful expression of the reporter proteins driven by the various promoter fragments within the library.

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53. EVOLUTION OF INTERFERON BETA SPECIFICITY IN HUMANS AND BATS MAY BE RESPONSIBLE FOR DIFFERENCES IN VIRAL PATHOGENESIS.

Presenter: **Rita Maria Quintela Tizon**

USask Affiliation: **Graduate student**

College: **Western College of Veterinary Medicine**

Supervisor: Arinjay Banerjee, Western College of Veterinary Medicine

Background:

Bats are being increasingly recognized as evolutionary hosts of multiple emerging zoonotic viruses, including betacoronaviruses (βCoVs) such as SARS-CoV-2, and MERS-CoV that can cause severe disease and death in humans and livestock. Conversely, bats infected with these viruses do not show clinical signs of disease, making them a fascinating model to study the evolution of virus-host interactions. The innate immune response is the first line of defense against viral infections. Type I interferons (IFNs) are among the first cytokines to be released when a viral infection is detected by cells. IFNs bind to the interferon-α/β receptor (IFNAR1/2) in host cells and stimulate the production of interferon stimulated genes (ISGs) that possess several antiviral properties. Type I IFNs are critical in the early stages of viral infection. Therefore, some βCoVs have evolved mechanisms that impair the activity of type I IFNs in humans, making us more vulnerable to infection. However, little is known about how type I IFN signals in bat cells and whether bats have evolved more efficient processes to better tolerate viral infections compared to humans.

Methods:

We have identified that wildtype (WT) bat IFNβ do not protect human cells and vice versa, suggesting species-specific mechanisms. To explore this specificity further, we performed a positive selection analysis on the IFNβ sequence of different mammals, including humans and several species of bats. Two amino acids were found to be positively selected for in the bat IFNβ sequence. Based on these two sites, we have developed human and bat (Eptesicus fuscus, Pteropus alecto) mutant recombinant functional IFNβ by changing the amino acid in the bat sequence for the orthologous amino acid in the human sequence, and vice versa. I have developed functional recombinant human and bat IFNβ through stable expression in Drosophila S2 cells. To test the antiviral efficacy of the mutated IFNβ compared to its wildtype counterpart, I have performed infection studies on IFN-treated cells with a prototypic IFN-sensitive RNA virus, vesicular stomatitis virus (VSV).

Results:

Preliminary VSV infection studies show that in both human (A549) and Eptesicus fuscus bat (Efk3B) cells treated with their species matched IFNβ, mutated IFNβ induces a weaker antiviral protection compared to WT IFNβ. Furthermore, no protection is observed when human cells are treated with mutant bat IFNβ containing human amino acid residues, and vice versa. This result mirrors the lack of cross-species protection also observed in infected cells treated with WT IFNβ.

Conclusion:

The differential antiviral protection observed in my antiviral assays suggests that our identified amino acid residues are key determinants of IFNβ-mediated protection. My study will demonstrate why mammalian species have evolved such tightly regulated type I IFN-mediated antiviral processes and how we may harness these adaptations to develop new antiviral therapies.

54. TOO HOT TO HANDLE: DEVELOPMENT AND VALIDATION OF A 4°C RAPID EQUILIBRIUM DIALYSIS (RED) ASSAY TO ASSESS PLASMA PROTEIN BINDING BY HEAT SENSITIVE COMPOUNDS IN EARLY DRUG DEVELOPMENT

Presenter:	**Alexandra Cullen**
USask Affiliation:	**Graduate student**
College:	**College of Arts & Science**
Collaborators:	Brian Sterenberg
Supervisor:	Jane Alcorn, College of Pharmacy and Nutrition

Background:

In drug development, lead compounds are selected for optimization if they exhibit favourable pharmacology under physiological conditions. Phosphopyricin (PPR) is a novel compound with high antibacterial activity but poor thermal stability. To assess its fraction unbound in plasma (fu), a 4°C rapid equilibrium dialysis (RED) assay was developed.

Methods:

RED plates were loaded with 350 µL buffer and 100 µL spiked plasma. Propranolol (PL) and timolol (TL) served as highly and poorly bound controls, respectively; plates were incubated 4 hours at 37°C and 24 hours at 4°C. PPR plates were incubated 24 hours at 4°C only. Analyte concentrations in buffer and plasma were determined by LC-MS/MS for fu calculation.

Results:

Experimental fu for PL and TL were consistent across incubation conditions and agreed with literature: at 37°C, PL fu = 0.21 and TL fu = 0.70 and at 4°C, PL fu = 0.25 and TL fu = 0.76. For PPR, fu ≈ 0.0 due to buffer sample PPR concentrations falling below the method limit of quantitation (4 ng/mL). This is likely an underestimate due to PPR precipitating in buffer.

Conclusion:

The 4°C RED method represents a quick, easy approach to assessing fu of heat sensitive compounds in early drug development. This can facilitate the characterization and further optimization of thermally unstable compounds that otherwise display favourable pharmacological activity. The system can also be further validated with other drug species.

55. FROM LARVA TO ADULT: IN VITRO REARING PROTOCOL FOR HONEY BEE (APIS MELLIFERA) DRONES

Presenter:	**Marina Carla Bezerra da Silva**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Sarah Wood, Western College of Veterinary Medicine
Co-supervisor(s):	Elemir Simko

Background:
Recently, there has been a dramatic increase in understanding of the optimal conditions to rear honeybees in vitro. Considering that honeybee reproductive castes are also exposed to pesticides, it is crucial to have an in vitro rearing methodology for toxicological risk assessment. In the past decades, studies have attempted and failed to rear honeybee drones in vitro. Accordingly, the objective of this study was to optimize a protocol to rear honeybee drones in vitro from larva to adult stage.

Methods:
Using an established honeybee worker in vitro protocol, we modified the grafting day, diet volume, pupation orientation, and absorbent tissue. Further, developmental characteristics such as wing abnormalities, body weight, testes weight, and abdominal area were measured and compared with age-match drones from the field colonies. We grafted larvae five days after egg laying and fed them the worker diet described in the literature increasing the daily volume. In addition, the pupal orientation was changed (horizontal or vertical).

Results:
We found that honeybee drone pupa reared horizontally with WypAll® absorbent tissue had a mean survival of 74%± 3.47 (SEM) until adulthood. The horizontal pupation with Kimwipe® absorbent tissue had a significant survival increase of 90.9% relative to vertically with Kimwipe®. Pupation horizontally with WypAll® had a significant 23% increase in survival relative to horizontally with Kimwipe®. All newly emerged in vitro drones from vertical pupation had wing abnormalities. The in vitro honeybee drone's total body weights, and adult testes weight were lower than those of the in vivo. In comparison to the in vivo drones, the in vitro adult drone had a smaller abdominal area.

Conclusion:
We successfully developed a honeybee drone in vitro rearing protocol with a mean survival of 74%. This developed protocol has the potential for future toxicological risk assessment of xenobiotics on immature drones.

56. OH-SYNAP! THE ROLE OF SYNAPTOTAGMIN 11 IN OSMOREGULATION.

Presenter:	**Kirk Haan**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Thomas Fisher, College of Medicine
Supervisor:	Thomas Fisher, College of Medicine

Background:

Humans and other mammals carefully regulate the osmolality of their body fluids, which is a measurement of the concentration of salts in those fluids. Large deviations from normal osmolality can lead to deleterious consequences. Regulation of body osmolality occurs through conscious (e.g., behavioural thirst and salt appetite) and unconscious mechanisms. Elderly people and the chronically ill are especially prone to deviations in osmolality due to polypharmacy, reduced mobility, and reduced thirst. With an aging population, this issue is precipitating a larger burden on healthcare systems and intensifying the need for the development of better therapeutics and treatment strategies to ameliorate this issue. Specialized neurons in the hypothalamus called MNCs release a hormone called vasopressin/antidiuretic hormone (VP/ADH) in response to increases in osmolality (e.g., from drug interactions or insufficient water intake), which causes renal water reabsorption and buffers further increases in osmolality. MNCs undergo marked structural and functional changes in response to increases in osmolality, among which is a substantial increase in cell size, which is unique to this cell type. Though hypertrophy appears to be necessary for osmoregulation, the mechanisms surrounding MNC hypertrophy remain understudied. We previously found that a mechanosensitive ion channel (dN-TRPV1) translocates to the plasma membrane during sustained increases in osmolality (e.g., longer than one hour), and that this process was dependent on phospholipase C (PLC) activation, Ca2+ influx through L-type Ca2+ channels, and SNARE-mediated exocytosis. We sought to further investigate the mechanisms surrounding MNC channel translocation and mechanisms of osmoregulation, and we now show that a specific isoform of part of the SNARE complex, called synaptotagmin 11 (Syt11) is critical both for channel translocation and osmotically evoked hypertrophy.

Methods:

We utilized stereotaxic viral vector injections paired with live-cell immunocytochemistry techniques to visualize isolated supraoptic neurons using advanced confocal microscopy.

Results:

We found that reduced expression of Syt11 significantly reduces translocation of ion channels as well as osmotically evoked hypertrophy.

Conclusion:

Syt11 plays a significant role in osmoregulation in isolated cells. We now plan to expand our experiments to include other ion channels, as well as to determine the identity of the vesicles that Syt11 and various ion channels are contained in.

57. OPTIMIZING THE CONDITIONS TO IDENTIFY THE DDX41 INTERACTOME BY BIOID

Presenter:	**Ananaya Charaya**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Supervisor:	Yuliang Wu, College of Medicine
Co-supervisor(s):	Erique Lukong

Background:

DDX41 (DEAD-box 41 protein) belongs to the helicase superfamily 2 of RNA helicases. It is involved in crucial processes, including genome stability, mRNA splicing, ribosome biogenesis, translation, cell differentiation, and innate immunity. Mutations in DDX41 cause myeloid malignancies, especially myelodysplastic syndrome and acute myeloid leukemia (MDS/AML). Previous studies in Wu lab have shown that basal protein expression of DDX41 experience a dynamic change when the cells are exposed to distinct stress conditions, including oxidative stress (sodium arsenite), microbial infection (HSV-1), and radiation exposure (infrared radiations). This dynamic expression is accompanied by the formation of P-bodies (marker EDC4) and a shift in the sub-cellular localization of DDX41 from the nucleus to the cytoplasm on being treated with exogenous dsDNA (poly dA:dT). However, the molecular mechanisms regulating DDX41's expression and sub-cellular localization are unknown. Previous studies have reported DDX41 to be found in the spliceosomal complex, nevertheless its interactome has not been reported yet. We, therefore, hypothesize that DDX41-interacting proteins may regulate the dynamic expression and cellular localization of DDX41.

Methods:

To identify the interacting proteins of DDX41, we have utilized proximity-dependent biotin identification (BioID). For BioID, we constructed DDX41-BioID fusion vectors tagged with FLAG at N-terminus and the TurboID ligase at the C-terminus.

Results:

We have observed successful overexpression and sub-cellular localization of control and DDX41-containing BioID fusion proteins. A successful observation of efficient biotinylation activity of TurboID ligase as well as the presence of potential interacting proteins of DDX41 has been made. We have also been successful in purification of BioID fusion protein along with its interactome by immunoprecipitation.

Conclusion:

Furthermore, the mass spectrometric analysis of purified BioID proteins will facilitate in identification of the interactome proteins of DDX41 and the validation of the top DDX41 interacting proteins will lead to a better understanding of the regulatory pathways and potential druggable targets for the treatment of DDX41-related blood cancer.

58. EVOLUTION AND ROLE OF BETACORONAVIRUS TRANSCRIPTION REGULATORY SEQUENCE (TRS) IN DISEASE PATHOGENESIS

Presenter:	**Arkadeb Bhuinya**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Arinjay Banerjee, Western College of Veterinary Medicine

Background:
Variation in case fatality rates in highly pathogenic Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2, 0.4%-15.4%) and Middle East respiratory syndrome coronavirus (MERS-CoV, ~36%) infection suggest potential differences in disease pathogenesis. However, molecular determinants for differential pathogenesis of these two Betacoronaviruses remain unknown. Transcription regulatory sequence (TRS) is a short motif present in coronavirus genomes that regulate viral transcription. Although SARS-CoV-2 and MERS-CoV proteins contribute directly to disease pathogenesis by inhibiting cellular antiviral response, little is known about role of these TRSs on viral protein expression.

Methods:
To study coronaviral TRS, I have developed a Betacoronavirus non-infectious replicon reporter system with wildtype and mutated TRS. Quantification of the reporter nano-luciferase (nluc) expression accounts for role of TRS in viral gene transcription. A plasmid-based reverse genetics system of SARS-CoV-2 will be used to rescue weakened replicating viruses with attenuated TRS followed by characterization in mammalian cell lines and the Syrian hamster animal model of COVID-19.

Results:
Deleting the viral TRS in the replicon reduced nLuc expression, which confirms the TRS' role in viral gene regulation.

Conclusion:
Indeed, identifying characteristic properties of CoV TRS will enable us to predict low and highly pathogenic CoVs that currently circulate in wild reservoir species.

59. COMPARISON OF BIOLOGICAL EFFECTS PRODUCED BY RADIOACTIVELY LABELED ANTIBODIES IN HUMAN CANCER CELLS AND FUNGAL CELLS

Presenter:	**Jonathan Bonet Ramirez**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Kevin Allen, College of Pharmacy and Nutrition
	Mackenzie Malo, College of Pharmacy and Nutrition
	Connor Frank, College of Pharmacy and Nutrition
Supervisor:	Ekaterina Dadachova, College of Pharmacy and Nutrition

Background:

Cancer patients are immunocompromised, therefore they can get infections from an opportunistic microorganism such as cryptococcus neoformans. Leukemia is one of the most common types of cancer. AML is the most lethal leukemia subtype. It is characterized by an increased rate of immature myeloid cells in the marrow. Cryptococcus neoformans is an encapsulated pathogenic yeast characterized for being very opportunistic, and for causing meningitis, it is responsible for 90 percent of deaths from this disease among immunocompromised.

Methods:

A yeast called Cryptococcus neoformans (a mutant version) was used, and treated with a radiolabeled antibody called 400-2 antibody against 1, 3-β-glucose. An acute myeloid leukemia cell line, called OCI-AML3, treated with radiolabeled antibodies called HuM-195 antibody against CD33 was employed, radioisotopes with an EDTA for Lutenium-177 and DTPA for Actinium-225 were utilized as a nonspecific group, and radiolabeled antibodies mentioned above with different doses of Lutenium-177 and Actinium-225 served as the specific group, and untreated cells as control. The cells were incubated with the radioactive material for 24 hours, then the subsequent assays were performed; clonogenic survival assay, γH2A/X immunofluorescent staining assay, internalization assay, and micronuclei assay.

Results:

Based on the results obtained from the treatment with 177-Lutetium either with the human cancer cells or the yeast, demonstrate that the radiolabeled antibodies are better for killing the cells. Comparatively, the cells treated with the radiolabeled antibodies showed more foci (double-strand breaks) when compared to the other groups. The antibody Hum-195 was internalized with time. While 400-2 did not internalize. Moreover, the treatment with 177-lutetium was efficient in producing micronuclei.

Conclusion:

This project is still in progress, consequently, more data will be collected. 222-Actinium needs to be tested using the same assays as described before, so far the result indicates that radiolabeled antibodies are good for treating both diseases using the same radiation dose.

60. DISCOVERING AND CHARACTERIZING BAT RESTRICTION FACTORS

Presenter:	**Sauhard Shrivastava**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Linda Chelico, College of Medicine
Supervisor:	Arinjay Banerjee, Western College of Veterinary Medicine

Background:

Bats, as natural reservoirs of numerous viruses, including retroviruses, exhibit remarkable resilience to the negative effects of viral infections. Investigating the mechanisms underlying bats' tolerance to retroviruses holds promise for uncovering novel insights with potential implications for human disease management.

Methods:

We identified TRIM5α-like genes within E. fuscus and P. alecto bats, characterizing their expression and function in bats. This involved comparative analysis with human TRIM5α assessing their expression patterns in response to stimuli, examining gene regulation and subcellular localization within bat cells, and establishing stable cell lines expressing bat TRIM5α-like genes.

Results:

Our findings demonstrate that bat TRIM5α-like genes exhibit inducibility by Interferon and Poly I:C, akin to human TRIM5α. Notably, E. fuscus TRIM5α induction surpasses human TRIM5α by approximately 100-fold in response to Poly I:C stimulation, mirroring RNA virus infection scenarios. Subcellular localization studies reveal cytoplasmic localization of bat TRIM5α-like proteins.

Conclusion:

Our study elucidates the role of bat TRIM5α-like proteins as Interferon Stimulated Genes (ISGs) within bat cells, akin to their human counterpart. The observed similarities prompt further investigation into the comparative restriction activities of bat TRIM5α-like proteins against different retroviruses relative to human TRIM5α.

61. CANONICAL AND NON-CANONICAL FEATURES OF THE BAT INTERFERON RESPONSE

Presenter:	**Victoria Gonzalez**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Linfa Wang
	Darryl Falzarano, Western College of Veterinary Medicine
	Andrew Doxey
Supervisor:	Arinjay Banerjee, Western College of Veterinary Medicine

Background:
Bats are reservoirs of emerging viruses that cause severe and often fatal disease in humans. Remarkably, infected bats do not demonstrate clinical signs of disease. In mammals, type I IFNs, like IFNβ, are induced upon viral infection to protect infected and neighboring cells through the induction of interferon stimulated genes (ISGs).

Methods:
To elucidate the bat IFN response, recombinant IFNβ from Pteropus alecto, Eptesicus fucus, and humans was generated. P. alecto (PaKiT03), E. fuscus (Efk3B), and human (A549, RPTEC) cells were then treated with their species-matched IFNβ for various lengths to assess protection against vesicular stomatitis virus (VSV) and Middle East respiratory syndrome coronavirus (MERS-CoV). The IFNβ signaling pathway was also profiled by RNA-sequencing.

Results:
Treatment of bat and human cells with IFNβ led to complete protection against MERS-CoV in bat cells, while a reduction in viral load was observed in human cells. The transcriptional response to IFNβ was further profiled using bulk RNA-sequencing. Both bat species upregulated a canonical set of ISGs, with transcription factors STAT1/STAT2 differentially expressed in bats. Immunoblotting and confocal microscopy demonstrated similar phosphorylation and translocation patterns for STAT1/STAT2 in P. alecto and human cells, while basal levels of nuclear STAT1/STAT2 are higher in unstimulated E. fuscus cells. Knocking down STAT1/STAT2 led to loss of antiviral protection in P. alecto cells, while STAT2 alone was able to protect E. fuscus cells against VSV. Furthermore, MERS-CoV infection inhibited STAT1/STAT2 expression and phosphorylation in human cells while minimal effect was observed in bat cells.

Conclusion:
We observed a potent and divergent IFNβ response in bats which rapidly protected against VSV and MERS-CoV infection. We are currently evaluating the differential activity of non-canonical ISGs against a range of viral isolates to gain further insights into how these may have evolved in bats to further control viral pathogenesis.

62. DEVELOPING COMBINATION THERAPIES FOR TELOMERASE-OVEREXPRESSING CLEAR CELL CARCINOMA OF THE OVARIES

Presenter:	**Vincent Maranda**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Frederick Vizeacoumar, College of Medicine
	Yue Zhang, College of Medicine
Supervisor:	Franco Vizeacoumar, College of Medicine
Co-supervisor(s):	Andrew Freywald

Background:
Ovarian clear cell carcinoma (OCCC) is the second most common type of ovarian cancer affecting 10-13% of women with ovarian tumors. OCCC is also the predominant histologic type of gynecological malignancy that harbors activating mutations within the telomerase reverse transcriptase (hTERT) promoter, causing its increased expression. The telomerase (hTERT) gene is hyperactive in many cancers and has been identified as a potential target for treatment. Although this led to multiple telomerase-targeting approaches, disappointingly, none have been successful in clinics. To circumvent this concern, our team has applied a genetic approach called synthetic dosage lethality (SDL), to exploit hTERT overexpression to identify potential targets to treat cancer. SDL is a genetic concept, where a normally non-lethal gene inactivation kills cells only in the context of overexpression of another gene like hTERT.

Methods:
Our laboratories have used lentiviral-based, pooled CRISPR/Cas9 and pooled shRNA-screening platforms to systematically query the genome and recently identified several SDL partners of hTERT. These potential partners have been validated using a novel CRISPR-based strategy in an in vivo pooled screen.

Results:
In collaboration with the AtomWise company, we have developed novel inhibitors for a target identified by these screens. We have also performed a large-scale screen of FDA-approved small molecule inhibitors with the goal of repurposing these drugs to target hTERT-overexpressing cancers. Subsequently, we plan to apply these therapies in combination to amplify the efficiency of treatment against cancers.

Conclusion:
The results will identify new targets exploiting hTERT overexpression and provide preclinical evidence to support the development of novel OCCC therapies.

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63. RECLAIMING WELLNESS: INCORPORATING MÉTIS PERSPECTIVES INTO SEXUAL HEALTH EDUCATION FOR YOUTH

Presenter:	**Katie Tolley**
USask Affiliation:	**Graduate student**
College:	**School of Public Health**
Supervisor:	Amanda Froehlich-Chow, School of Public Health

Background:

The enduring impact of colonization includes the pervasive influence of heteropatriarchy and sexism rooted in Christian principles, leading to the enforcement of colonial gender norms, marginalization of non-conforming individuals, and intergenerational consequences that affect the sexual health and well-being of Indigenous youth today (Wesley, 2015). Currently, Indigenous youth experience a disproportionate representation in HIV and other sexually transmitted and blood-borne infections. My research, nested within the ATOHTITUM project, investigates the direct and indirect impacts of colonization on the gender identity and sexuality of Métis youth in Saskatchewan, and how it influences their decisions surrounding sexual health. It aims to identify gaps in knowledge, attitude, and practice of culturally appropriate sexual health among Métis youth. Additionally, it seeks to explore how Métis youth access sexual health services in conventional healthcare settings and identify barriers that may adversely impact their experience. Wesley, D. L. (2015, April). Reimagining two-spirit community: Critically centering narratives of urban two-spirit youth. http://www.proquest.com/docview/1826318201?pq-origsite=primo

Methods:

The research method employed in this study is the conversational method. This approach prioritizes the relationship between researchers and participants, emphasizes orality as a means of knowledge transmission, and adheres to the specific tribal epistemology and protocols of the Indigenous communities involved (Kovach, 2010). Kovach, M. (2010). Indigenous Methodologies: Characteristics, Conversations, and Contexts, Second Edition. University of Toronto Press.

Results:

n/a

Conclusion:

Missing from the middle years and high school curricula are opportunities for students to better understand diverse First Nation and Métis perspectives on sexuality, sexual identity, and sexual health. As such, the findings of this research will help to inform the development of an arts-, land-, and culture-based wellness program for Métis youth that compliments existing public-school curriculum.

Self-declaration of research alignment with additional themes (optional):

Indigenous Health Research

64. GOLD NANOPARTICLE AND CATIONIC POLYMER SAFETY FOR RADIOFREQUENCY ABLATION APPLICATIONS

Presenter:	**Michelle Vargas Fernández**
USask Affiliation:	**Graduate student**
College:	**College of Engineering**
Collaborators:	Wenjun Zhang, College of Engineering Michael Moser, College of Medicine Ildiko Badea, College of Pharmacy and Nutrition
Supervisor:	Ildiko Badea, College of Pharmacy and Nutrition
Co-supervisor(s):	Wenjun Zhang

Background:

In 2022, the Canadian Cancer Society projected that 3,500 Canadians would receive a liver cancer diagnosis. Radiofrequency ablation (RFA) offers a minimally invasive treatment option for advanced stages of hepatocellular carcinoma (HCC). While demonstrating efficacy in creating large ablation areas (>5cm), achieving uniform cell death necessitates a multidisciplinary approach. This study seeks to enhance current techniques using 50 nm gold nanoparticles (AuNPs) functionalized with two cationic polymers: Poly-L-Lysine and the Gemini Surfactant (16-3-16).

Methods:

The cytotoxic effects of AuNPS+Poly-L-Lysine and AuNPS+PBS in HepG2 cells were evaluated using the ATP (CellTiter-Glo™ 2.0 Assay kit (Promega) and MTT assays. 10-day spheroids (1x105 cells/spheroid) were treated with AuNPS+Poly-L-Lysine, and AuNPS+Gemini surfactant (16-3-16). 24 hours post-treatment, cell viability was assessed through a Live/Dead assay kit and imaged with a confocal microscope Nikon ECLIPSE Ti2.

Results:

MTT did not provide reliable data due to cell viability values >100%. The ATP assay showed that cell viability for both treatments was always higher than 60%. 0.500 µM is a concentration that exhibited a safe use of AuNPS+Poly-L-Lysine (One-way ANOVA with Sidak's correction for multiple comparisons 0.500 ± 7.47 vs. 0.031± 5.19, p= 0.0014) and for AuNPS+ PBS 3.938E+07 particles/ml (3.938E+07 ±3.14 vs. 2.461E+0±4.68, p=0.0077). Confocal Microscope images did not show adverse effects of AuNPS+Gemini Surfactants in HepG2 spheroids in the proliferative cells and inner cells of the spheroids.

Conclusion:

The MTT reagent possibly interacted with the treatment conditions causing an interference leading to unreliable results. The ATP assay proved to be a reliable assay for the current experimental conditions thus becoming a more adequate option. Poly-L-Lysine+ AuNPS in HepG2 cell viability was found to be higher than 60%, making them a viable option without compromising cell survival.

Self-declaration of research alignment with additional themes (optional):

Indigenous Health Research

65. HOUSING AND MENTAL HEALTH AMONG ON-RESERVE FIRST NATIONS ADULTS IN SASKATCHEWAN: EXPLORING GENDERED EXPOSURES AND VULNERABILITIES

Presenter:	**Prashikchhya Parajuli**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Tina Nelson
	Larry Burgess
Supervisor:	Bonnie Janzen, College of Medicine

Background:
Research has shown associations between housing characteristics and mental health outcomes such as anxiety, depression, loneliness, and psychological distress. Housing also intersects with other social locations such as Indigenous identity and gender to influence mental health. This study explored the relationship between housing characteristics and mental health among First Nations adults living on reserve in Saskatchewan, Canada and whether the nature of those associations differ by gender.

Methods:
The source of data was the First Nations Sleep Health Project (FNSHP), a study conducted in partnership with two First Nations communities in rural Saskatchewan. Two phases of data collection (2018/19 and 2021/22) from the FNSHP were combined for this cross-sectional study, resulting in a sample of 808 adults. Data for this quantitative study were collected by self-report questionnaire on a broad array of health conditions and determinants, including those related to housing and self-rated mental health (SRMH).

Results:
Preliminary results indicate women were more likely than men to report fair/poor SRMH. Regarding housing exposures, men were more likely than women to report exposure to second hand smoke, whereas women were more likely to indicate exposure to household dampness, mold, and noise.

Conclusion:
Although preliminary, results suggest that First Nations women may be more likely than their male counterparts to report fair/poor SRMH and greater exposure to potentially harmful housing characteristics. Multivariable analyses are in progress to confirm these findings and to determine which housing characteristics are related to mental health and whether those relationships differ for women and men.

Self-declaration of research alignment with additional themes (optional):
Indigenous Health Research

66. CONTRIBUTION OF LAND BASED KNOWLEDGE OF CUMBERLAND HOUSE TO ENSURE WATER SECURITY, FOOD SECURITY AND ECOLOGICAL JUSTICE.

Presenter:	**Kaneta Eya Lam Lam**
USask Affiliation:	**Graduate student**
College:	**College of Education**
Supervisor:	Cari Dawn McIlduff, College of Medicine

Background:
Located in the Saskatchewan River Delta, the Cumberland House Village is the oldest community in western Canada. The community was the hub of the fur trade for trappers, making this a prime location for Canadian historic designation. This wetland ecosystem contains diverse and abundant plant, fish, and wildlife species and provides valuable goods and services for Indigenous communities located near it (Abu, 2017).

Methods:
Community-Based Participatory Research (CBPR) is an orientation to research that alters researcher-community relationships. CBPR emphasizes collaboration and engagement with the community throughout the research process, aligning with the goals of understanding Traditional Ecological Knowledge (TEK) and water security in Cumberland House.

Results:
Traditional Land Based Knowledge can bring solutions to the problem

Conclusion:
Reconciliation with science and traditional lad based knowledge is required.

Self-declaration of research alignment with additional themes (optional):
Indigenous Health Research

67. THE ASSOCIATION OF CANADIAN DIETARY PATTERN AND PHYSICAL ACTIVITY WITH BONE PQCT MEASUREMENTS - A PROPENSITY SCORE MATCHING ANALYSIS

Presenter:	**Parisa Jandaghi**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Hassan Vatanparast, College of Pharmacy and Nutrition Punam Pahwa, College of Medicine Philip Chilibeck, College of Kinesiology
Supervisor:	Hassan Vatanparast, College of Pharmacy and Nutrition

Background:

Osteoporosis affects a significant proportion of adults, with approximately 12% in Canada. Several lifestyle factors play an important role in bone health including a healthy diet, regular exercise, avoiding smoke and alcohol, and healthy weight. We aimed to investigate the association of dietary patterns and physical activity with bone parameters among Canadian adults.

Methods:

We used the 2015-2016 Canadian Health Measure Survey (CHMS), a nationally representative cross-sectional survey. The sample represents 26,756,508 Canadians aged 19-79 years. Peripheral Quantitative Computed Tomography (pQCT) bone measurements included cortical cross-sectional area (CSA), cortical bone mineral density (BMD), and stress-strain index (SSI) at the tibia. We used the Propensity Score Matching (PSM) method with frequency weights to determine the association between dietary patterns (extracted from cluster analysis), and physical activity with bone indicators adjusting for potential confounders.

Results:

We classified participants into two age groups: 19 to 49 and 50 to 79 years old, with the proportion of 55.6% and 44.4% respectively, and a roughly equal distribution of males and females. An unhealthy-mixed dietary pattern which comprised 67.1% of the sample was associated with a lower cortical cross-sectional area, cortical bone mineral density, and stress-strain index compared to a healthy-plant-based-high-protein diet, with an average treatment effect (ATE) of 4.71 mm^2, 2.62 mg/cm^3, and 95.81 mm^3 respectively (p-value<0.001). Low physical activity was also associated with lower cortical cross-sectional area and stress-strain index, but higher cortical bone mineral density compared to high physical activity with average treatment effects (ATEs) of 6.38 mm^2, 52.09 mg/cm^3, and 2.98 mm^3 respectively (p-value<0.001).

Conclusion:

Our findings provide significant insights into the impact of healthy-plant-based-high-protein dietary pattern and high physical activity on bone strength among Canadian adults, potentially leading to more effective strategies for osteoporosis prevention and treatment.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

68. ACCEPTABILITY TESTING OF A MOBILE APPLICATION PROTOTYPE FOR TAILORED PATIENT EDUCATION AND SELF-MANAGEMENT ALONG THE TRANSPLANT JOURNEY

Presenter:	**Taylor Raiche**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Allison Cammer, College of Pharmacy and Nutrition Nicola Rosaasen Nathaniel Osgood, College of Arts and Science
Supervisor:	Holly Mansell, College of Pharmacy and Nutrition

Background:

The organ transplant journey is arduous and demands that patients and their care partners adopt multimodal strategies to preserve long-term wellbeing. Tailored, digital education and self-management interventions may help. This study explores the prospective acceptability of the prototyped Health Education and Learning Platform (HELP), a mobile application for transplant education and self-management developed via user-centered design.

Methods:

A cross-sectional electronic survey based on the Theoretical Framework of Acceptability (TFA) was distributed via purposive and snowball sampling to transplant patients and care partners. Participants watched a 7-minute video illustrating the prototype prior to completing the survey. Likert-scale questions garnered prospective acceptability within the seven TFA component constructs of Affective Attitude, Burden, Ethicality, Intervention Coherence, Opportunity Costs, Perceived Effectiveness, and Self-efficacy. Open-ended questions enabled participants to provide qualitative feedback. Data was collected using REDCap and analyzed using descriptive statistics and content analysis to deductively code free-text responses to TFA component constructs.

Results:

One hundred seventy-eight responses were received, of which 169 contained demographic information. All Canadian provinces were represented by at least one participant. Approximately half (51.6%) of participants were transplant patients. The majority were aged 31-50 (62.7%) and had completed at least some level of post-secondary education (76.2%). Overall, more than 70% of respondents agreed or strongly agreed with acceptability items related to each of the TFA constructs. The highest affirmed TFA constructs were Intervention Coherence (83.3%) and Self-efficacy (80.3%). Disparity between patients (83.9%) and care partners (53.1%) was observed within the Affective Attitude construct. Participants expressed overall positive regard for the prototype, including its design and novelty.

Conclusion:

Participants endorsed an understanding of how the HELP app is designed to help, belief it would improve their ability to manage their health, and confidence that they could use it. Confirmation of prospective acceptability suffices to progress the prototype to beta testing.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

69. A STUDY OF HEALTH-RELATED BEHAVIORS OF SENIORS IN RESPONSE TO MINOR HEALTH CONDITIONS

Presenter: **Parinaz Amirimoghadam**
USask Affiliation: **Graduate student**
College: **College of Pharmacy and Nutrition**
Supervisor: Jeff Taylor, College of Pharmacy and Nutrition

Background:
Minor ailments are prevalent across all age groups and health conditions, significantly impacting healthcare expenditures and resource utilization. The elderly population, in particular, experiences a high frequency of minor ailments due to physiological changes, comorbidities, and polypharmacy. Understanding the health behaviors of seniors in managing minor ailments is crucial for optimizing healthcare resources and improving patient outcomes.

Methods:
The objective of this study is to investigate correlations between the nature of symptoms, intra-personal factors, and health behaviors among the elderly. A validated tool, PSM-OTC, will be used to analyze health behaviors through patients' health diaries over a one-month period. This mixed-methods approach will provide insights into decision-making processes, symptom management strategies, and intervention outcomes.

Results:
At present, our study awaits ethics approval, and therefore no results are available. However, we anticipate that our findings will shed light on the complexities of seniors' health behaviors in response to minor ailments.

Conclusion:
Pending ethics approval, this study aims to fill a critical gap in understanding the health behaviors of seniors in response to minor ailments. By investigating correlations between symptom nature, intra-personal factors, and health behaviors, we aim to inform the development of tailored services and resources in community pharmacies for the elderly population. Through optimized resource allocation and enhanced patient experiences, we anticipate contributing to the ongoing improvement of healthcare delivery in community pharmacy settings.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

70. GLOBAL SURVEILLANCE OF ANTIBIOTIC RESISTANT PAENIBACILLUS LARVAE USING COMMERCIALLY AVAILABLE HONEY SAMPLES

Presenter:	**Oleksii Obshta**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Tayab Soomro, Western College of Veterinary Medicine
	Marina C.B. Silva, Western College of Veterinary Medicine
	Thanuri L.K. Edirithilake, Western College of Veterinary Medicine
Supervisor:	Sarah Wood, Western College of Veterinary Medicine

Background:

American foulbrood (AFB) is a fatal infectious disease of honey bee brood caused by the endospore-forming bacterium, Paenibacillus larvae. P. larvae spores are resilient in the environment and thus colonies showing clinical signs of AFB are often destroyed by burning to eradicate the causative agent. To prevent outbreaks of AFB, oxytetracycline metaphylaxis is widely used in North America, resulting in sustained selective pressure for oxytetracycline resistance in P. larvae isolates. In contrast, there is a ban on use of antibiotics in beekeeping in the EU and New Zealand.

Methods:

To investigate the suitability of honey as appropriate sample for surveillance of antibiotic resistant P. larvae and to assess the P. larvae spore contamination, 174 honey samples from 39 countries were purchased online through Amazon. Honey samples were cultured on selective media with and without antibiotics to screen for antimicrobial resistance to the three antimicrobials approved for AFB. Antimicrobial-resistant isolates were further characterized by the broth microdilution technique to determine the Minimum Inhibitory Concentration (MIC) of antibiotic, as well as by PCR to evaluate for the presence of tetracycline resistance genes.

Results:

55% of the 174 tested samples contained P. larvae at spore concentrations ranging from 0.12-9163. 4 samples from the USA (Tennessee, Colorado, South Dakota and Florida) and two from Canada (MB and BC) contained oxytetracycline-resistant P. larvae with MICs ranging from 64-128 µg/mL; tetracycline resistance genes PCR test results are pending.

Conclusion:

Overall, presented results of this pilot study highlight the suitability of honey as a matrix for continuous surveillance and monitoring of antimicrobial resistance in P. larvae and support evidence-based decision making regarding antimicrobial use in commercial beekeeping operations.

Self-declaration of research alignment with additional themes (optional):

Sustainable Progress

71. SIGNALING MOLECULES IMPLICATED IN ORAL MANIFESTATIONS OF SYSTEMIC SCLEROSIS: A SCOPING REVIEW

Presenter: **Asmaa Fadl**
USask Affiliation: **Graduate student**
College: **College of Dentistry**
Supervisor: Andrew Leask, College of Dentistry

Background:
Systemic sclerosis (SSc) is a chronic autoimmune disease characterized by vascular abnormalities, immune dysregulation, and progressive fibrosis. SSc affects the skin and internal organs, causing organ dysfunction or, often, death. Oral manifestations significantly affect 80% of SSc patients. These manifestations, such as restricted mouth opening (microstomia), dry mouth, periodontitis, and gingival inflammation, significantly impair their abilities to eat, swallow, and speak, thus adversely affecting their quality of life. With no disease-modifying treatment available, SSc, especially its oral manifestations, presents a critical unmet medical need. Despite their substantial impact, oral manifestations of SSc are frequently overlooked and poorly understood. Therefore, understanding the molecular mechanisms underlying these manifestations is crucial for advancing clinical management strategies.This scoping review aims to synthesize current literature on the oral complications of SSc and associated signaling molecules, while also identifying research gaps for future exploration of oral biomarkers and molecular targets to enhance diagnostic and treatment strategies, ultimately improving the management of oral symptoms and the quality of life of SSc patients.

Methods:
A scoping review using the PubMed database was conducted for relevant original research articles published between 2000 and 2024 and focused on identifying molecular markers involved in oral manifestations in SSc patients. The search strategy included keywords related to SSc (systemic sclerosis, scleroderma), terms specific to oral complications (oral, orofacial, facial, and mouth) and (symptoms, manifestations, complications), as well as signalling molecules-related terms (cytokine, interlukin, chemokine, signalling molecule). Studies were included if they met these criteria: they involved human participants diagnosed with SSc and exhibited oral manifestations, or they examined sera or various oral samples from SSc patients and focused on biomarkers or signaling molecules related to oral symptoms of SSc. The exclusion criteria included animal studies, case reports, and review articles. Relevant data from eligible research articles was extracted and summarized.

Results:
Eleven studies met our inclusion criteria. These studies demonstrated an association between oral manifestations in SSc, mainly microstomia and periodontitis, and increased levels of some signaling molecules in oral samples or sera from SSc patients. These studies provided insights into the involvement of interleukin-6 (IL-6), IL-1β, and transforming growth factor-β1 (TGF-β1) in stimulation of collagen deposition and myofibroblast differentiation, which promote fibrosis and microstomia associated with SSc. Furthermore, these studies proposed a potential correlation between the elevated levels of IL-6, matrix metalloproteinase-9 (MMP-9), chemokine (C-X-C motif) ligand 4 (CXCL-4), and tumor necrosis factor-α (TNF-α), in oral samples from SSc patients with periodontitis and its underlying mechanisms involving vascular endothelial dysfunction and pathological extracellular matrix remodeling. Interestingly, two studies demonstrated elevated IL-6 levels in the gingival crevicular fluid of SSc patients; however, these levels did not correlate with any clinical parameters of periodontitis. This observation could potentially reflect the early phase of SSc, supported by IL-6's

elevated serum levels in early-stage SSc patients, thus indicating its promise as a diagnostic marker in this particular phase.

Conclusion:

Studies included in this review have identified key signaling molecules like IL-6, IL-1β, MMP9, TNF-α, and CXCL4, indicating their involvement in the inflammation and fibrosis associated with oral symptoms in SSc. Future extensive studies with larger patient cohorts, diverse oral sample sources, targeted molecular pathways, and a broader spectrum of oral manifestations are necessary to fully elucidate the intricate interplay of these molecular pathways in the pathogenesis of SSc. Consequently, these molecules could be proposed as potential therapeutic targets or diagnostic markers, underscoring the importance of translating research findings into clinical practice to enhance the management of SSc-related oral complications.

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72. PHYSICAL THERAPY AND HEALTH PROMOTION IN CHRONIC CONDITIONS: A SCOPING REVIEW

Presenter:	**Rosmary Martinez-Rueda**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Catherine Boden
	Katie Crockett, School of Rehabilitation Science
	Brenna Bath, School of Rehabilitation Science
Supervisor:	Brenna Bath, School of Rehabilitation Science

Background:
Physical therapists (PTs) have considerable potential to contribute to improve health and wellness through health promotion interventions, but little is known about the overall scope of those practices.We conducted a scoping review to identify and describe the health promotion practices conducted by PTs regarding lifestyle risk factors and chronic health conditions.

Methods:
Following the Joanna Briggs Institute's guidelines for scoping reviews, we searched OVID MEDLINE, CINAHL, OVID Embase, Scielo, PEDRO and LILACS for English and Spanish references from January 2002 to January 2023. Title, abstract, and full-text screening for inclusion criteria was completed by two independent researchers with a third reviewer engaged for conflict resolution. Four independent researchers extracted data.

Results:
Of the 9907articles found, 286 studies were included. Most of the studies were conducted in North America (USA=50, Canada=9), and the majority of them were randomized controlled trials (56.3%). The studies mainly focused on physical activity/exercise as an intervention, and balance/fall prevention (24%) as well as on chronic pain management/prevention (20%) as the main conditions addressed.

Conclusion:
Physical activity/exercise is the most common health promotion intervention delivered by physical therapists across a wide range of chronic conditions. There was little evidence of health promotion practices related to nutrition, sleep hygiene, or tobacco cessation. The health promotion interventions included in this study focused on the development of personal skills rather than other health promotion action areas such as promoting healthy policies, supporting healthy environments, strengthening community action or reorient health services.

73. SEE US, UNDERSTAND US. EXPLORING NEWCOMER'S MATERNITY CARE EXPERIENCES IN SASKATOON: A CASE STUDY.

Presenter: **Isabelle Dena**
USask Affiliation: **Graduate student**
College: **College of Medicine**
Supervisor: Nazeem Muhajarine, College of Medicine

Background:
Notwithstanding Canada's universal health care system, disparities exist in access to and in the quality of maternity care experienced by people of immigrant backgrounds, in turn impacting birth outcomes. Mitigating these disparities requires understanding the specific structural and social determinants of newcomer maternity care experience in the current context of Saskatchewan. This study aims to explore newcomer women's maternity care experiences in Saskatoon by examining their access to and the quality of care received.

Methods:
This qualitative case study recruited eight newcomer women, seven maternity care providers, and three health system leaders. Eighteen virtual and telephone semi-structured interviews were conducted in English from May 2021 to March 2023, averaging 20-50 minutes, except for two interviews via Arabic interpretation. All participants provided verbal consent. Interview data were audio-recorded and transcribed. Thematic analysis followed using NVivo software, starting with organizing and familiarizing with data. Re-reading data multiple times helped to categorize the codes into subthemes and then refining the process to identify main themes. The theoretical framework of intersectionality guided the analysis.

Results:
The analysis generated three main themes: (1) Navigating the intersections of language, power, & culture in newcomer-provider interactions. This theme highlighted barriers and facilitators of communication, autonomy and implicit bias. (2) Influence of cultural and religious beliefs on maternity care. This theme focused on newcomer's health beliefs impacting acceptance of perinatal treatment options. (3) Missed opportunities by providers to uphold respectful maternity care standards. This theme called attention to aspects of disrespectful care, including privacy issues and ignoring newcomers' embodied maternity care experiences.

Conclusion:
Most newcomer women in this study reported a positive experience of maternity care. However, the findings revealed implicit bias among providers, health system constraints related to limited language interpretation services, workforce issues and inadequate culturally appropriate care to promote respectful maternity care practices.

74. SCHOOL FOOD: AN ANALYSIS OF SCHOOL FOOD ENVIRONMENT, FOOD AND NUTRITION-RELATED KNOWLEDGE, ATTITUDES, AND PRACTICES AFTER A TWO-YEAR UNIVERSAL, COMPREHENSIVE ELEMENTARY SCHOOL LUNCH PROGRAM

Presenter:	**Tachlima Chowdhury Sunna**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Supervisor:	Rachel Engler-Stringer, College of Medicine

Background:
Child nutrition influences health, well-being, and learning across the lifespan. The quality of the diets of Canadian children during school hours is poor across the socio-economic spectrum. School lunch programs can help to maintain nutrition during the school years. This study assessed the changes in the school food environment and food and nutrition-related knowledge, attitudes, and practices among elementary school students enrolled in a two-year curriculum-integrated school lunch program compared to students without the intervention.

Methods:
This study is a part of the larger Good Food for Learning project, conducted in two intervention and control Saskatoon Public Schools Division elementary schools. A staff survey was conducted with 94 and 20 participants at baseline and endpoint. The student survey was a self-administered online questionnaire in grades 4-6 at baseline and 6-8 at the endpoint. Student participation in the survey was 185 at baseline and 192 at endpoint.

Results:
Progress in food and nutrition-related initiatives in the school context occurred between baseline to endpoint. At baseline, school gardening, composting, food related activities, healthy food, food-related initiatives with community, food preparation activities, and traditional food were found to be low, while by endpoint there was improvement in almost all areas of the school food system. No significant differences in terms of time (baseline and endpoint) and types (intervention and control) have been found so far regarding students' food and nutrition-related knowledge, attitudes, and practices. The total food insecurity score has decreased for the intervention group, though it has increased for the control group from the baseline to the endpoint.

Conclusion:
The study results will be helpful to understand better how a school food program can impact the overall school food environment and student nutrition. The findings can provide recommendations for the development of a national school food policy and program.

75. STATISTICAL LEARNING TECHNIQUES EMBEDDED FOR IMPROVING PREDICTION AND PARAMETER ESTIMATION IN A LOGISTIC BOX-COX MODEL

Presenter:	**Jing Wang**
USask Affiliation:	**Graduate student**
College:	**School of Public Health**
Collaborators:	Shiyu Xu, College of Arts and Science
	Xing Li, College of Arts and Science
Supervisor:	Li Xing, College of Arts and Science

Background:
Nonlinear relationships are common in statistical analysis in epidemiology, posing challenges in model development. Current nonlinear models, such as logistic regression, Generalized Additive Models, natural splines, and polynomial models, have varying limitations in capturing nonlinear patterns.

Methods:
To address these limitations, we introduce the Logistic Box-Cox (LBC) model, a modified version of logistic regression using Box-Cox transformation to handle nonlinearity. The LBC model estimates parameters based on the original maximum likelihood method. To enhance its performance, we employ ensemble learning methods and multiple starting points in the maximum likelihood estimation process as the enhanced LBC models separately.

Results:
We simulate common exposure-disease relationships in epidemiology and train with current nonlinear models, the original LBC model, and enhanced LBC models. We evaluate the models' performance using deviance. The results show that the deviance difference between the original LBC model and current nonlinear models is small. Comparing the original LBC model with the enhanced LBC models, we find that the ensemble learning LBC model performs the best.

Conclusion:
While there is no significant difference between the LBC model and current nonlinear models, the shape parameter unique to the LBC model provides good interpretability. Additionally, the ensemble learning LBC model can provide more stable and reliable results. The multiple starting points LBC model increases the chances of finding the global optimum of the log-likelihood function.

76. PROTECTING AGAINST ANXIETY: PHYSICAL ACTIVITY VERSUS PERCEPTIONS OF PHYSICAL ACTIVITY

Presenter:	**Bailey Gitzel**
USask Affiliation:	**Graduate student**
College:	**College of Kinesiology**
Collaborators:	Darren Nickel, College of Medicine
Supervisor:	Kevin S. Spink, College of Kinesiology

Background:
Higher levels of physical activity (MVPA) have been shown to be protective against the emergence of anxiety (Schuch et al., 2019). In explaining this protective effect, conversations often defer to dose-response thinking (i.e., more MVPA, less anxiety). However, recent research suggests that PA perceptions might also influence this relationship. In this study, we examined three PA perceptions: 1. Perceptions that the amount of PA one engages in is adequate for health (PA adeq). 2. Perceptions about PA levels compared to similar others (PA SC). 3. Perceptions of sitting behaviour as a threat to health (SITT). The purpose was to examine whether MVPA would be more related to anxiety than PA perceptions.

Methods:
University students (N= 656) completed an online questionnaire assessing MVPA (Fowles et al, 2017), PA adeq (Zahrt & Crum, 2020), PA SC (Gitzel et al., 2023), SITT (developed for this study) and generalized anxiety symptoms (GAD-7, Spitzer et al., 2006).

Results:
The PA perception measures were found to be reliable measures as Cronbach alpha values were all > .90. Results from a regression analysis with all predictors included revealed that MVPA was not significantly related to anxiety ($p = .939$). In terms of the PA perceptions, two were significantly related to anxiety – PA adeq ($p = .01$) and SITT ($p < .001$). Those who perceived their PA as more adequate for their health and perceived their sitting as less of a threat to their health reported less anxiety symptoms. Socially comparing one PA levels to others was not significantly related to anxiety ($p = .961$).

Conclusion:
This study provides preliminary evidence to suggest that one's PA level may be less related to anxiety when an individual's PA perceptions are included. If replicated, these results may provide new PA messaging targets.

77. IMPACT OF HIV DRUG COVERAGE PROGRAM IN SASKATCHEWAN

Presenter: **Ann Babu**
USask Affiliation: **Graduate student**
College: **College of Pharmacy and Nutrition**
Supervisor: David Blackburn, College of Pharmacy and Nutrition

Background:
In 2018, the Ministry of Health in Saskatchewan implemented a universal coverage program that eliminated all costs of antiretroviral medications (ART) for people living with HIV (PLWHIV) who were beneficiaries of the provincial drug plan. The purpose of this study was to measure the program's impact.

Methods:
A retrospective cohort study was conducted on individuals with at least one HIV diagnosis from either the physician fee-for-service database or the hospital discharge database who were provincial drug plan beneficiaries. The prescription drug database was used to capture data on ART claims to measure aggregate ART drug utilization in each four-month interval, starting three years before and three years after the program. Interrupted time series analysis using a linear regression model was used to test the program's impact.

Results:
We identified 4,994 individuals with at least one diagnosis of HIV; of these, 1,828 satisfied all inclusion criteria. Almost two-thirds (57%) were male and 33.5% were aged between 20-39. The vast majority (71.6%) received no ART claims during the study period. Before the policy was implemented, only 1% to 2% of eligible PLWHIV received ART medications during each four-month exposure interval. Following the implementation of the coverage policy, the number receiving ART increased above the projected/pre-policy rate by approximately 10 additional ART users per 1,000 PLWHIV. Thereafter, the rate continued to rise by approximately 3 ART users per 1,000 PLWHIV every four months. This improvement appeared to be restricted to males only (Figure X).

Conclusion:
The vast majority of PLWHIV in Saskatchewan do not receive ART medications every four months. Although the province's coverage policy resulted in a statistically significant improvement in ART utilization, the clinical significance of the effect remains unknown. Moreover, the policy may not have benefitted all equally.

78. INVESTIGATING BELIEFS AND THEIR INFLUENCE ON MEDICATION ADHERENCE IN CHRONIC DISEASE MANAGEMENT

Presenter: **Chidimma Umeaghadi**
USask Affiliation: **Graduate student**
College: **College of Pharmacy and Nutrition**
Supervisor: David Blackburn, College of Pharmacy and Nutrition

Background:
Patient beliefs are one of the most important determinants of future medication persistence. In a previous study, we developed a concise three-item beliefs scale that showed good prediction performance. However, definitive negative beliefs were infrequently expressed by respondents. Individuals were more likely to express uncertainty regarding their new medications. This study's aim was to evaluate the impact of uncertainty on the risk for medication non-persistence.

Methods:
Data for this study was obtained from a questionnaire to Saskatchewan residents who were newly prescribed a chronic medication. Beliefs items comprised three items assessing perceived threat of the condition, drug importance, and potential harms. We described the frequencies of responses categorized as positive, negative, and uncertain. Logistic regression analysis was performed to assess the impact of “uncertainty” on the risk for non-persistence.

Results:
Data were available from 3,029 completed questionnaires. Uncertainty was observed least often for the question asking if their medical condition was a threat to respondent’s health. Definitive negative beliefs (disagree or strongly disagree) was reported by 4.9% (n=148), while 6.7% (n=202) were uncertain. However, when asked about the importance of the medication to their health, 10.9% (n=329) were uncertain compared to 1.8% (n=55) expressing definitive negative beliefs. The highest level of uncertainty was observed for the question asking if their new medication would do more harm than good; over one-quarter selected “not sure” (23.8%, n=720) while 10.2% (n=309) expressed a definitive negative response. Odds ratios for the effect of uncertainty on non-persistence across the three belief questions ranged between 1.9 to 2.6 and were all statically significant.

Conclusion:
Patients frequently exhibit uncertainty regarding medications that have been prescribed to them. Uncertainty is a clear risk factor for medication non-persistence, albeit with a lower effect size compared to definitive negative beliefs.

79. EXPLORING STANDARD PRECAUTIONS FOR INFECTION PREVENTION AND CONTROL READINESS IN SIX LOW- AND MIDDLE-INCOME COUNTRIES: INSIGHTS FROM A NATIONAL CROSS-SECTIONAL SURVEY

Presenter:	**Md Abdullah Al Jubayer Biswas**
USask Affiliation:	**Graduate student**
College:	**School of Public Health**
Collaborators:	Scott J. Adams, School of Public Health
	Li Xing, School of Public Health
	Michael Szafron, School of Public Health
Supervisor:	Scott J. Adams, Li Xing , Prosanta Mondal, Michael Szafron

Background:

Despite the significant morbidity and mortality caused by healthcare-associated infections worldwide, especially in low- and middle-income countries (LMICs), there is a lack of understanding of the readiness to apply standard precautions for infection prevention and control (IPC) in healthcare facilities across different LMICs and the associated factors influencing this readiness

Methods:

We analyzed nationally representative publicly-available health system data from the Service Provision Assessment surveys, focusing on six selected LMICs, including Afghanistan, the Democratic Republic of the Congo, Haiti, Nepal, Senegal, and Bangladesh. We selected nine tracer items of standard precautions and recoded them into binary elements using guidelines from the World Health Organization's Service Availability and Readiness Assessment manual. We calculated the total score of these items per service delivery area. Then, we determined an overall percentage score named readiness index across all nine service delivery areas for each healthcare facility. Likewise, we computed the readiness index of all healthcare facilities in a specific country. Through a survey-weighted multivariable generalized estimating equation, we identified the factors associated with the readiness index

Results:

Our analysis of data from 6,159 healthcare facilities across six countries revealed that 47.3% (95% CI: 46.0, 48.6) of the necessary standard precaution amenities were present across all countries, ranging from Bangladesh at 43.9% and Senegal at 57%. Across six countries, readiness varied by service area; the tuberculosis services area was the lowest equipped at 33.1% availability, while the general outpatient services area was the highest equipped at 57.7%. Healthcare facilities in Bangladesh and rural areas had lower readiness index, with mean differences of -2.5 (95% CI: -4.3, -0.7) and -4.0 (95% CI: -5.5, -2.6) compared to Nepal and urban areas

Conclusion:

Our study reveals significant deficiencies in standard precautions within healthcare facilities across six LMICs, notably in rural areas. The findings underscore an urgent need for targeted interventions to improve IPC strategies, particularly in domains like tuberculosis care

80. BIOINFORMATICS-DRIVEN MASS SPECTROMETRY-BASED PROTEOMICS UNVEILS THE PROGRESSION FROM HEALTHY PERIODONTIUM TO GINGIVITIS AND PERIODONTITIS: A META-ANALYSIS

Presenter:	**Paras Ahmad**
USask Affiliation:	**Graduate student**
College:	**College of Dentistry**
Collaborators:	Walter L. Siqueira, College of Dentistry
Supervisor:	Walter L. Siqueira, College of Dentistry

Background:
The current review comprehensively summarizes the existing body of literature concerning clinical mass spectrometry (MS)-based proteomics research focused on periodontal diseases. It extensively delves into the salivary and gingival crevicular fluid (GCF) proteins serving as biomarkers along with their gene ontology (GO) analysis. Moreover, the review explores experimental proteomics design, assessing protein-protein interactions (PPIs).

Methods:
Eligibility criteria involved adult human studies analyzing saliva and/or GCF proteome via MS, with control (healthy) and experimental (gingivitis and/or periodontitis) groups. A literature search was conducted across Scopus, Web of Science, and PubMed. Data extraction was standardized and verified. Differentially expressed proteins were identified and subjected to bioinformatics analysis, followed by meta-analysis for synthesis.

Results:
The review identified significant differences in protein expression between healthy individuals and those with gingivitis and periodontitis. In GCF, 247 proteins were upregulated and 128 downregulated in periodontal diseases. Saliva analysis revealed 79 upregulated and 70 downregulated proteins. There were distinct protein profiles between gingivitis and periodontitis, with 159 and 31 unique upregulated proteins in GCF, respectively. Meta-analyses confirmed significant upregulation of various proteins in periodontitis, including Albumin and Matrix metalloproteinase-9, while Cystatin B and Glutathione S-transferase P were downregulated. α-Amylase and α-1-antitrypsin were upregulated in periodontitis saliva. Hemoglobin subunit delta was upregulated in gingivitis GCF, while β-Defensin-3 was downregulated. PPI analysis revealed complex networks of interactions among differentially expressed proteins. GO and KEGG pathway analyses provided insights into biological processes and pathways associated with periodontal diseases.

Conclusion:
In conclusion, ongoing MS-based proteomics studies emphasize the need for a highly sensitive and specific diagnostic tool for periodontal diseases. Multiplex measurement of multiple biomarkers simultaneously and large study cohorts are crucial for validity. Clinician acceptance of the eventual diagnostic method relies on its ability to provide superior or complementary information to current clinical assessment procedures.

81. MEALTIME EXPERIENCES OF FAMILIES LIVING WITH DEMENTIA IN LONG-TERM CARE

Presenter:	**Heather Alford**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Allison Cammer, College of Pharmacy and Nutrition
	Heather Ward, College of Medicine
	Jason Perepelkin, College of Pharmacy and Nutrition
Supervisor:	Allison Cammer, College of Pharmacy and Nutrition

Background:
Relationship-centred mealtime interventions improve staff-resident relationships and reduce the risk of malnutrition for residents living in long-term care settings. Less is known about the nature, goals, or outcomes of family-resident relationships at mealtimes, particularly for residents living with dementia. This constructivist grounded theory study aimed to explore what is needed to support meaningful family relationships and engagement at mealtimes with residents living with dementia in long-term care homes.

Methods:
Individual interviews were conducted with 13 family caregivers of people living with dementia in long-term care homes in Saskatchewan. Interviews were recorded, transcribed verbatim, and analyzed using constant-comparison method.

Results:
Three themes characterized family member participants' experiences of mealtimes: 1) Changing Ways of Connecting, 2) Nutrition as a Reflection of Dementia Progression, and 3) Adjusting Expectations.

Conclusion:
Mealtimes are important opportunities for supporting family-resident connection and can facilitate 'acceptance' of the illness trajectory in dementia among caregivers. By conceptualizing their mealtime involvement, this study advances understanding of the essentiality of family caregivers for enhancing relationship-centred practices in long-term care settings. By explicating the structure, function, and experience of family involvement in nutritional care, this study addresses the current gap in understanding of how families living with dementia build and preserve relationships through nutritional care and dining.

Translational, Clinical, or Applied Science 1

82. EVALUATING THE EFFECT OF AGE AND SEX ON UPPER LIMBS KINEMATICS AMONG SASKATCHEWN FARMERS DURING FARM TASKS

Presenter:	**Opeyemi Akinluyi**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Angelica Lang, College of Medicine
	Dennise Balough, College of Medicine
Supervisor:	Angelica Lang, College of Medicine

Background:

Musculoskeletal disorders (MSDs) are one of the leading causes of disabilities and work-related injuries among farmers [1]. Farmers are exposed to numerous risk factors and experience MSDs due to the nature of the job. The consequences of MSDs can be clinically and economically significant among farmers and farm workers.[2]. However, there is a need to better define the demands of high-risk farm tasks to understand how specific tasks may contribute to upper limb MSDs. In Saskatchewan, 85.6% of farmers reported having musculoskeletal pain in at least one body part over the past year [3]. A recent study of rural farmers reported that the upper extremities are the most affected body parts (75%) for farmers with musculoskeletal complaints [4]. While there is a strong association between arm elevation posture and MSDs, no studies have measured farmers' full shoulder kinematics during work-related farm activities to understand other aspects of upper limb postural exposures. The objective of this study is to investigate humeral and scapular kinematics among male and female Saskatchewan farmers during previously identified high-risk farm tasks. Specifically, this study will examine if sex and age affect upper limb kinematics during farm tasks to assess how they may influence postural risk factors for injury.

Methods:

Four farmers (all male, age 18 years and above, free from upper limbs disorders) from rural Saskatchewan farms have participated. The recruitment goal is 20 farmers (10 males, 10 females), and data collection will resume in the spring. Participants' demographics were recorded, and each farmer completed the Quick Disability of the Arm, Hands, and Shoulder (QuickDASH) questionnaire to assess upper limb abilities. Participants were measured as they performed four typical, high-risk farm tasks on their respective farms: climbing the seed drill, operating power tools overhead (i.e. a drill), shoveling gravel, and yard maintenance (i.e. pruning trees or bushes). Movements of the bilateral humeri, scapulae, and torso were tracked with XSens Awinda IMUs (XSens Technologies, Enschede, The Netherlands) during these tasks. Scapular and humeral angles were extracted and calculated using a custom Matlab script following ISB standards [5]. The data from all four participants was averaged for each task and the descriptive statistics were calculated for each task, including the mean, maximum, and standard deviation of the dependent variables, i.e., the scapular and humeral angles in each task.

Results:
Preliminary data from the four completed participants suggests that all participants have similar maximal humeral elevation when performing climb, drill, pruning, and shoveling tasks (80 – 100°) (Figure 1). These values indicate the humeral elevation demands of all these tasks are high. As the participants lift their arms to perform a task, there is an increase in scapular upward rotation, as expected, but scapular upward rotation was higher when climbing and shoveling than in drilling and pruning tasks, despite similar, or even lower (shovel), arm elevation levels in all tasks. The higher relative upward rotation indicates the farmers may be adopting a more "shrugged" shoulder posture in these tasks, possibly through increased upper trapezius activation, which could contribute to neck and shoulder fatigue and injuries.

Conclusion:
The results from the study provide new knowledge about the scapular and the humeral movements during farm tasks. All measured tasks (climbing the seeder, pruning, drilling, and shoveling) pose a high demand for humeral elevation and scapular positioning may contribute to shoulder injury risk. High humeral elevation and atypical scapula upward rotation can predispose farmers to injuries.

Self-declaration of research alignment with additional themes (optional):
Indigenous Health Research

83. REGULATION OF INTESTINAL MUCOSAL IMMUNITY TO JOHNE'S DISEASE IN CATTLE THROUGH PARENTERAL AND ENTERIC VACCINATION

Presenter:	**Itzel Aguilar**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Amy H. Lee, Jeroen De Buck
Supervisor:	Antonio Facciuolo, Western College of Veterinary Medicine

Background:

First described by Johne and Frothingham in bovine cattle, the causative agent of Johne's disease (JD), Mycobacterium avium ssp. paratuberculosis (MAP) has prevailed in every major dairy and beef cattle producing country. Its economic impact is mainly related to decreased milk production and reduced slaughter value. Young calves are most susceptible to infection and infected cattle can remain asymptomatic for years while sporadically shedding MAP bacteria in feces. Current diagnostic tests fail to reliably detect many infected animals. In the absence of effective vaccines, hygiene and management strategies are the best tool currently available to control JD. Despite this, the estimated prevalence in Canada is 5% in beef herds and 70% in dairy herds. MAP transmission is primarily through the fecal-oral route. Once ingested, MAP is taken up by M cells and subepithelial macrophages associated with Peyer's patches (PP). Ruminants uniquely possess two functionally and morphologically distinct types of PP: discrete PP (DPP) in the jejunum and continuous PP (CPP) in the ileum. Our lab previously found marked differences in MAP persistence and mucosal immune responses associated with parenteral vaccination between DPP and CPP. However, it remains to be determined which vaccine delivery routes are effective in priming protective and durable immune responses at these specific sites.

Methods:

We will use a commercial JD vaccine and an experimental oral vaccine together with our unique surgical model to target MAP infection to each distinct PP in young calves to analyze enteric mucosal immunity following parenteral and oral vaccination. We will also characterize phenotypic and functional changes in mucosal leukocytes from each PP using flow cytometric and gene expression analyses.

Results:

Our results will expand our knowledge immunogenicity, effectiveness, and limitations of different vaccine delivery routes in stimulating intestinal mucosal immunity within the most susceptible population to this infection.

Conclusion:

These data will determine MAP protein candidates to determine actual vaccine candidates and delivery routes against MAP infection in young cattle.

Self-declaration of research alignment with additional themes (optional):

Interdisciplinary/Interprofessional Collaboration

84. SITE-SPECIFIC DIFFERENCES IN BONE AREA, DENSITY, AND MICRO-ARCHITECTURE BETWEEN MONOZYGOTIC TWINS DISCORDANT FOR TYPE 1 DIABETES: A CASE-CONTROL STUDY

Presenter:	**Ali Rezaei**
USask Affiliation:	**Graduate student**
College:	**College of Kinesiology**
Collaborators:	Munier Nour
	James Johnston
	Saija Kontulainen
Supervisor:	Saija Kontulainen, College of Kinesiology

Background:
Type 1 Diabetes (T1D) is known to have detrimental effects on bone, however, confounding factors such as genetics, environmental exposures and lifestyle differences influence bone development. Twin studies offer the ability to control for genetic influences on bone development, however, are challenging in T1D due to high concordance of developing T1D in monozygotic twins.

Methods:
We report a case-control study on a pair of monozygotic twins, obtaining data through High-Resolution peripheral Quantitative Computed Tomography. We calculated Z-scores for each bone outcome using reference data and described differences between the twin with T1D and control twin (without T1D).

Results:
The twins, both male, were 12.1 years old. The twin with T1D had lower radius total area than the control, as well as relative to the mean of the reference population (Z-score = -1.77 for T1D and -0.28 for TD, respectively), while tibia total area was comparable between the twins (Z-score = -1.67 for T1D and -1.31 for TD, respectively). Although the twin with T1D had a higher tibia trabecular number compared to his sibling (Z-score = -0.58 for T1D and -1.77 for TD, respectively), his tibia trabecular thickness was markedly lower (Z-score = -0.85 and Z-score = 0.17). Additionally, we observed relatively lower tibia cortical porosity (Z-score = 0.87 for T1D and 2.35 for TD, respectively), along with higher radius cortical bone mineral density in the twin with T1D (Z-score = 4.57 for T1D and 3.01 for TD, respectively).

Conclusion:
The findings suggested site-specific effects of T1D on bone area, density, and micro-architecture, characterized by lower total area and higher total and cortical mineral density in the radius, as well as higher trabecular number and lower trabecular thickness and cortical porosity in the tibia.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

85. INDUCTION OF IMMUNE RESPONSE AGAINST NECROTIC ENTERITIS BY A SINGLE INTRAPULMONARY LIVE CLOSTRIDIUM PERFRINGENS VACCINE AT HATCH: UNCOVERING THE PRINCIPLES OF GUT-LUNG AXIS

Presenter:	**Hemlata Gautam**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Susantha Gomis, Western College of Veterinary Medicine

Background:
Clostridium perfringens (CP) in poultry is an emerging threat to animals and public health. Restriction on the use of antibiotics of human importance in Canadian poultry industry led to a considerable increase in the infections of necrotic enteritis (NE) specially in the chickens raised without antibiotics (RWA). Moreover, CP residing in the gut of broiler chickens is capable of causing significant illnesses in human via consumption of contaminated poultry meat. Developing an effective vaccination strategy against NE, preferably delivering a single dose of vaccine at hatch to protect broiler chickens against NE without a booster vaccine is an enormous challenge.

Methods:
Therefore, the objective of this study was to induce mucosal immunity in the intestines against NE by intrapulmonary (IPL) delivery of a live CP vaccine at hatch using the gut-lung axis (GLA) concept in vaccine delivery following in ovo delivery of cytosine-phosphorothioate-guanine oligodeoxynucleotides (CpG-ODN) to induce immune cell maturation in the lungs. Live CP vaccine was delivered by the IPL route following in-ovo delivery of CpG-ODN or saline. Groups of broiler chickens (n=35) were vaccinated as:1) no vaccine, no CP challenge; 2) CP challenge only; 3) in-ovo CpG-ODN +live CP IPL; 4) in-ovo saline + live CP IPL. Additional studies were conducted to measure serum IgY, mucosal IgA and histopathology of lungs following vaccination.

Results:
Delivery of live CP vaccine by the IPL route with or without in ovo delivery of CpG-ODN provided a significant protection against NE ($p<0.0001$). Systemic IgY and mucosal IgA against CP were corelated with protection against NE. There was no inflammation or any necrosis in the pulmonary parenchyma following live CP delivery by the IPL route. A significant influx of ($p<0.001$) of CD8 + T cells and macrophages were noted in the lungs two days following live CP delivery by the IPL route.

Conclusion:
In conclusion, delivery of a single dose of live CP at hatch by the IPL without a booster dose protected broiler chickens against NE. This study demonstrated utility of gut-lung-axis (GLA) concept in vaccine delivery in broiler chickens.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

86. PROTECTION OF BROILER CHICKENS FROM LETHAL BACTERIAL INFECTIONS BY TRAINED IMMUNITY INITIATED BY CPG-ODN

Presenter:	**Iresha Subhasinghe**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Susantha Gomis, Western College of Veterinary Medicine

Background:
Bacterial diseases like Escherichia coli and Clostridium perfringens have severely impacted the poultry industry leading to significant economic losses. Similarly, these bacterial infections are among the major causes of human foodborne illness. Short term immunoprotection of oligodeoxynucleotides containing cytosine phosphodiester guanine (CpG-ODNs) using a single administration has been proven in broiler chickens. Immune cells undergo metabolic changes while attaining memory characteristics to augment protection. We hypothesize that multiple CpG-ODN administrations can protect broiler chickens during the production cycle beyond the first week of life by inducing innate immune memory.

Methods:
To investigate this possibility, neonatal broiler chickens were injected with CpG-ODN multiple administrations, then mitochondrial oxidative phosphorylation (OXPHOS) and cellular glycolysis in peripheral blood mononuclear cells (PBMC) were measured using Seahorse XFp. In another experiment, birds were injected with two administrations of CpG-ODN at days 1 and 4 of age and challenged with lethal doses of E. coli at day 27 of age. CpG-ODN was administered by in ovo route, then chicks were vaccinated by pulmonary route (IPL) on the day of hatch, followed by booster at day 9 of age against necrotic enteritis (NE) and cellular metabolic pathways were investigated.

Results:
We observed that immune cells switched to mitochondrial OXPHOS for energy demand from cellular glycolysis which indicated a changing metabolic phenotype during attaining memory characteristics (21 days period). Remarkably there was a significant protection of broiler chickens ($p < 0.05$) against E.coli at day 27 of age following twice CpG-ODN administrations at day 1 and 4. Similarly, chickens were significantly protected ($p < 0.05$) and enhanced mitochondrial OXPHOS when administered CpG-ODN before the IPL vaccine against NE.

Conclusion:
This indicates that immune cells shift metabolism from glycolysis to mitochondrial OXPHOS resulting induction of trained immunity. In conclusion, CpG-ODN has induced trained immunity in broiler chickens, effectively controlling bacterial diseases during the production cycle.

Self-declaration of research alignment with additional themes (optional):
Interdisciplinary/Interprofessional Collaboration

87. ONCOGENIC FUNCTIONS OF THE EPHA2 RECEPTOR IN HUMAN AND CANINE MELANOMA

Presenter:	**Shabnam Abdi**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Arata Matsuyama, Western College of Veterinary Medicine
	Kaylee Jo, Western College of Veterinary Medicine
	Behzad M. Toosi, Western College of Veterinary Medicine
Supervisor:	Behzad Toosi, Western College of Veterinary Medicine

Background:
Melanoma in humans and dogs is a deadly type of skin cancer since it spreads rapidly throughout the body. Despite current treatment options including tumor removal combined with chemotherapy and/or radiotherapy, survival in both species is short, highlighting the need for more effective treatments. The EphA2 receptor, a member of the erythropoietin-producing hepatocellular (Eph) receptor family, is upregulated in various types of human malignancies and has been associated with poor prognosis and tumor aggressiveness. Therefore, this receptor may serve as a target for cancer therapies that inhibit its activity, although its expression status is unknown in neither canine or human melanomas. The biological and clinical similarities between human and dog melanoma make it an ideal model for comparative oncology research. This comparative study examined EphA2 expression in canine and human melanoma cells, and its role in the invasive characteristics of melanoma cells.

Methods:
The expression of the EphA2 receptor in canine and human melanoma cell lines was evaluated by Western blotting. Melanoma cells were transduced with lentiviral particles encoding EphA2-targeting shRNAs for silencing EphA2 expression. Transduction with non-silencing scrambled shRNAs was performed to generate non-silencing controls Silencing was confirmed by Western blotting and immunofluorescence. The effect of EphA2 silencing on melanoma cell survival, invasion, colony formation, and the propagation of melanoma tumorspheres was analyzed.

Results:
Both canine and human melanoma cell lines had high EphA2 expression levels compared to normal melanocyte cells. Moreover, stable silencing of EphA2 significantly and consistently decreased colony formation, invasion and tumorsphere formation in both human and canine melanoma cells. In addition, EphA2 silencing induced increase in apoptosis of canine melanoma cells.

Conclusion:
Our data provide functional evidence that the EphA2 receptor contributes to the malignant biological behavior of melanoma cells in both humans and dogs. This suggests that EphA2 inhibition could potentially aid in suppressing melanoma invasiveness

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

88. THYROID STIMULATING HORMONE IS NEGATIVELY ASSOCIATED WITH CORTICAL BONE VOLUME AND MICRO-ARCHITECTURE IN CHILDREN WITH TYPE 1 DIABETES

Presenter:	**Zahra Ghafouri**
USask Affiliation:	**Graduate student**
College:	**College of Kinesiology**
Collaborators:	Munier Nour, College of Medicine
	Saija Kontulainen, College of Kinesiology
Supervisor:	Saija Kontulainen, College of Kinesiology

Background:
Thyroid hormones are essential for bone growth and development. Childhood hypothyroidism may cause delayed skeletal development, retarded linear growth and impaired bone mineral accrual. Multiple imaging modalities have demonstrated altered bone development in children and youth with type 1 diabetes. It is unknown if variations in thyroid hormone metabolism are associated with bone micro-architectural, density and morphologic differences in children with T1D. Thus, we explored correlations between thyroid hormones and bone outcomes in children with T1D.

Methods:
We obtained measures of thyroid stimulating hormone (TSH; mIU/L) and free thyroxine (fT4; pmol/L) from clinical health records of a total of 54 participants of the Bone Strength Development Study in children with T1D, (N= 22 male, median age = 12.4, SD = 2.2). High-Resolution peripheral Quantitative Computed Tomography (HR-pQCT) scans of the distal radius and tibia were obtained and evaluated using standard evaluation and advanced cortical analysis to quantify bone measures. We tested nonparametric partial correlation between thyroid hormones and bone outcomes of HR-pQCT after accounting for sex using IBM SPSS 27. Significance was set to $p < 0.05$.

Results:
The correlation analysis in children with T1D between TSH and distal radius and tibia cortical total volume, cortical bone volume, cortical area, and apparent cortical thickness was negative $r(49) = -0.3$, $p = 0.03$. There was no significant correlation between fT4 and bone outcomes in either site.

Conclusion:
The negative association between TSH and cortical bone volume, area, and thickness in the sample of children with T1D indicates that alterations in thyroid hormone metabolism may alter cortical bone segmental size outcomes. Further longitudinal research is needed to elucidate the clinical impact, underlying mechanisms and establish causality.

Self-declaration of research alignment with additional themes (optional):
Sustainable Progress

Translational, Clinical, or Applied Science 2

89. INVESTIGATION OF PROTEIN BOUND-METABOLITES OF NITROFURAZONE BY NUCLEOPHILIC SUBSTITUTION

Presenter:	**Sedigheh Barzegar**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Collaborators:	Anas El-Aneed, Bryn O. Shurmer, Randy W. Purves
Supervisor:	Randy W. Purves, College of Pharmacy and Nutrition
Co-supervisor(s):	Anas El-Aneed

Background:
Nitrofurazone, a synthetic antibiotic, has been used in animal feed to treat gastrointestinal and dermatological infections in animals as well as to promote growth. This antibiotic is now banned for use as veterinary drug due to the potential harm it can cause to consumers through its residues in food animal products. Nitrofurazone metabolizes rapidly and the majority of the metabolites covalently bind to cellular proteins, which leads to very low free metabolite levels. Current nitrofurazone residues detection focuses on monitoring the side chain, semicarbazide (SEM), released from the protein-bound metabolite. However, SEM detection, which may also originate from non-nitrofurazone sources, requires time consuming overnight hydrolysis and derivatization steps. Since protein-bound metabolites persist in tissues for long periods, a rapid liberation of specific residues from these protein-bound metabolites would offer a reliable and sensitive approach for monitoring these veterinary drug residues (VDRs) in animal-derived foods.

Methods:
S9 fractions prepared from bovine liver tissues were used for studying the in-vitro metabolism of nitrofurazone (both nitrofurazone and 13C15N3-labeled nitrofurazone were used). Several buffers were investigated for use in protein substitution reactions with different nucleophiles. Ultra performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) was used for detection of the nucleophile-metabolite complexes produced through protein substitution.

Results:
The protein substitution process with the various nucleophiles was monitored, and in some cases, within 15 min the cyano-metabolite (both labeled and non-labeled) conjugated with the nucleophile was identified. The stability of the metabolite-nucleophile complexes is currently under investigation and preliminary results will be presented.

Conclusion:
The release of protein bound metabolites back into solution as a metabolite-nucleophile complex enables a sensitive and selective analysis of trace nitrofurazone residues using UPLC-MS/MS. This efficient and time-saving method offers an improvement over traditional detection methods for trace nitrofurazone residues.

90. UNLOCKING THE PHOSPHOPROTEOME: ADVANCED ENRICHMENT TECHNIQUES FOR COMPREHENSIVE ANALYSIS OF LOW-ABUNDANCE SALIVARY PROTEINS"

Presenter:	**Ahmed Eldib**
USask Affiliation:	**Graduate student**
College:	**College of Dentistry**
Supervisor:	Walter Luiz Siqueira, College of Dentistry

Background:
Phosphoproteins are vital in the oral cavity for enamel protection, remineralization, and microbial defense, thereby playing a significant role in oral health maintenance. However, characterization of phosphoproteins has challenges due to unstable chemistry of phosphor-Ser/Thr residues and phosphate group disassociation during mass spectrometry. Thus, this experiment aims to optimize and explore different techniques for enrichment of single, double, and multiply phosphorylated peptides, in order to unveil the comprehensive phosphoproteome of low-protein samples and enhance our understanding of their critical functions in the oral ecosystem.

Methods:
Our approach incorporated a sequential metal oxide affinity chromatography (SMOAC) method. The initial sample consisted of one hundred micrograms of digested parotid saliva. For the primary phase of phosphopeptide enrichment, we employed titanium dioxide (TiO2) chromatography. The flow-through and wash fractions obtained from the TiO2 stage were then combined and subjected to Fe-NTA chromatography for further enrichment. We then applied the same technique to pooled samples of the acquired enamel pellicle, collected from various individuals, totaling approximately sixty micrograms. Results were compared to control samples that had not undergone enrichment

Results:
The SMOAC approach significantly enriched multiply phosphorylated peptides and notably increased the detection of single and double phosphorylated peptides. This enrichment resulted in identification of a diverse range of phosphoproteins, previously undetected in low-protein oral samples, thereby providing a more comprehensive view of the oral phosphoproteome. Utilizing SMOAC has allowed for a detailed analysis of the phosphoproteome associated with both parotid and AEP samples, highlighting its significant utility in comprehensive phosphoproteomic studies

Conclusion:
The SMOAC method is an effective strategy for the enrichment and detection of phosphoproteins from low protein samples. Funding support from supervisor grants: Canadian Institutes of Health Research #PJT-159760, #MOP-106657 Natural Sciences and Engineering Research Council #RGPIN-2020-06119 Canada Foundation for Innovation #37442.

91. EFFECTS OF INCREMENTAL DOSES OF VAPORIZED OXALIC ACID ON HONEY BEE WORKERS AND QUEENS

Presenter:	**Emilio Enrique Tellarini Prieto**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Marco Pietropaoli
	Ylona Camus
	Marcelo Polizel Camilli, Western College of Veterinary Medicine
Supervisor:	Sarah Wood, Western College of Veterinary Medicine
Co-supervisor(s):	Elemir Simko

Background:

The honey bee ectoparasitic mite, Varroa destructor, is one of the main causes of honey bee colony loss worldwide. Synthetic acaricides are the most commonly used strategy for Varroa control, however, resistance to these acaricides is emerging. Consequently, the use of organic acids for Varroa control is gaining more interest among beekeepers. For example, oxalic acid (OA) is a natural compound that has been shown to be an effective acaricide against Varroa mites, however, the potential toxicity of OA to adult bees and queens is poorly understood. Beekeepers often use higher OA doses than those recommended on the product label (1 g OA) due to concerns about efficacy of the label dose. We hypothesize that high doses of OA may negatively affect bee health and colony productivity.

Methods:

Therefore, the objective of the study was to evaluate the toxicity of incremental doses (0, 5, 10 and 20 g) of vaporized OA on honey bee workers and queens. We focused on investigating short-term effects of OA on adult bee mortality and colony strength, including brood production and population size. Next, we aimed to understand the residual effects of OA applications on both worker bees and queens. Regarding workers, we investigated their ability to rear new queens. As for queens, we evaluated critical aspects such as acceptance, performance, and sperm quality.

Results:

We found that colonies treated with 20 g OA (20 times the label dose) had a statistically significant increase in worker bee mortality, with a non-significant, 25% decrease in brood relative to controls. No significant differences were observed in queen performance nor sperm quality.

Conclusion:

Overall, our results support that higher-than-label doses of OA, up to 10 g, are a safe for honey bee colonies undergoing treatment for varroa mites.

92. COLONIZATION OF PROBIOTICS IN THE INTESTINE OF CHICKEN EMBRYOS FOLLOWING A COARSE SPRAY APPLICATION ON INCUBATING EGGS

Presenter:	**Mihiprabha Rathnayake**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Susantha Gomis, Western College of Veterinary Medicine

Background:
Innovative technologies are priority in the poultry industry to control diseases and to minimize antimicrobial use. This study explores, a novel technique of introducing probiotics to developing embryos as a coarse spray on hatching eggs, to minimize pathogenic bacterial infections of newly hatched chicks. The objective of this study was to investigate the colonization of probiotics in chicken embryos following application as a coarse spray on eggshells.

Methods:
Bacterial broths of Enterococcus faecalis, Bifidobacterium gallinarum, Pediococcus acidilactici and Lactobacillus salivarius were sprayed on embryonating specific pathogen free (SPF) eggs as a coarse spray. Probiotic solutions were maintained at 10OC and applied to the eggshell via spray continuously for 30 sec at 15 and 17 days of incubation (DOE). The negative control group left unsprayed. At 20 DOE, intestines of embryos were cultured on selective media. Matrix assisted laser desorption-time of flight (MALDI-TOF) and whole genome sequencing were used to determine the migration of probiotics through the eggshell into the intestines.

Results:
At 20 DOE colonization of E. faecalis was revealed from 75% of embryos. Whole genome sequencing results revealed 100% nucleotide identity between sprayed and recovered bacteria.

Conclusion:
This study demonstrated probiotic delivery onto eggshells of incubating eggs colonize the intestines of chicken embryos without interfering with hatchability and health, which is a noninvasive industry feasible technique of delivering probiotics to chicken embryos. We hypothesize this technique will colonize probiotics in the intestines to reduce yolk sac infections in neonatal chickens.

93. MICROBUBBLE ASSISTED DRUG DELIVERY FOR GLIOBLASTOMA

Presenter:	**Isabella Zittlau**
USask Affiliation:	**Graduate student**
College:	**College of Pharmacy and Nutrition**
Supervisor:	Ildiko Badea, College of Pharmacy and Nutrition
Co-supervisor(s):	Steven Machtaler

Background:

Glioblastoma is the most common and malignant brain tumour in adults; moreover, the blood-brain barrier (BBB) is a significant obstacle in the effective delivery of anti-cancer therapies. Microbubble (MB) is a colloidal particle that consists of a gas core surrounded by a protective shell composed of biocompatible materials, used as contrast agents and, when combined with low-intensity focused ultrasound, is a promising technique to cross physiological barriers. Gemini lipid is a new class of surfactants, presenting a unique structure and properties compared to conventional surfactants. A versatile non-viral gene delivery system that, if incorporated into MB, could bring advantages to the formulation. Throughout mechanical vibrations, High-intensity focused ultrasound (HIFU) forms an ultrasound wave which propagates across tissues, causing alternating cycles of increased and reduced pressure. MBs loaded with drug or gene therapy vectors, if applied with HIFU, can be used to deliver, and release the transport of substances into tissues of hard access.

Methods:

The formulation of MBs combined with Gemini Lipid has been tested at various charge and molar ratios of the constitutive elements, in addition to component concentration exploration. After the preparation of MB, these will be characterized as size measurement, zeta potential and stability. Also, conjugation and lipid arrangement will be evaluated by small-angle X-ray scattering.

Results:

Until abstract submission, no significant results involving the MBs formulation have been achieved. However, the HIFU technique has been tested previously, and its functionality has been proven.

Conclusion:

This project focuses on synthesizing MBs combined with Gemini Lipids to increase chemotherapeutic cross BBB into glioblastoma, maintaining low toxicity and reducing side effects. Moreover, the ability of MBs to disrupt biological barriers and permeate the BBB, evaluating the delivery efficiency and toxicity in vitro astrocytes, will be tested through a small animal HIFU system.

94. ASSESSMENT OF THE TOTAL METABOLOME AS A METHOD OF NORMALIZATION IN URINE BIOMARKER RESEARCH IN COMPARISON TO CREATININE

Presenter:	**Emma Finch**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Deborah Michel, College of Pharmacy and Nutrition
Supervisor:	Darryl Adamko, College of Medicine
Co-supervisor(s):	Anas El-Aneed

Background:
Respiratory illnesses like COPD and asthma require significant healthcare system resources. Effective diagnosis is a major limitation for the treatment of respiratory illnesses. A transition towards personalized medicine through biomarker research is a promising approach given the current limitations of traditional testing methods. Urine is an easy to collect metabolite rich biofluid for biomarker discovery. Liquid Chromatograph Mass Spectrometry (LC-MS) is a powerful biomarker quantification tool; however, it requires a method of normalization for study subjects' hydration status. The commonly used normalization method, creatinine, lacks the ability to normalize all patients such as those with kidney dysfunction. Total metabolome is a new method of normalization that uses the overall signal of a representative sub-metabolome to normalize urine data.

Methods:
Dansyl chloride was used to derivatize amine and phenol containing metabolites in urine and the unified signal acts as representative sub-metabolome. 24 derivatized metabolite standards were used to create a standard curve to quantify urine samples. Pooled healthy urine was collected and used to determine matrix effects and create quality control samples. The total metabolome of 50 samples will be used to normalize previously determined biomarkers and the data compared with creatinine normalized values.

Results:
Optimization of urine derivatization was carried out on the pooled urine. LC-MS was used to confirm full derivatization of the metabolome in standards and pooled urine prior to validation. A 3-day validation is being performed to determine accuracy and precision.

Conclusion:
Our findings will indicate if the total metabolome is a robust method that can be used to quantify a representative portion of the total metabolome in patient urine samples instead of creatinine. The method may allow for the inclusion of patient samples excluded in past studies when creatinine was used.

95. CHARACTERIZING NUCB1 EXPRESSION IN DROSOPHILA MELANOGASTER GASTROINTESTINAL TRACTS

Presenter: **Yona Al-Tahir**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Adelaine Leung, Western College of Veterinary Medicine

Background:
Nucleobindin-1 (NUCB1) is a protein observed to have anorexigenic (appetite suppressing) effects within a wide variety of animals. Despite its conserved function in regulating energy metabolism across different animal species, the molecular mechanism of how NUCB1 functions remains elusive. Unlike other species, such as mice, whose NUCB1 expression profile has been observed co-expressed with NUCB2, Drosophila melanogaster (fruit flies) are utilized in this study as they only express NUCB1. Accordingly, this research aims to determine how NUCB1 evokes its anorexigenic effect through the manipulation of NUCB1 in Drosophila melanogaster to localize and characterize NUCB1 expression.

Methods:
The expression of a reporter protein is used to observe the expression profile of NUCB1 in the gut. Immunochemistry using primary and secondary antibodies is utilized on dissected gastrointestinal tracts from these flies to generate fluorescence. When imaged under a confocal microscope, NUCB1 expression can be localized.

Results:
Preliminary results successfully show NUCB1 expression across the gastrointestinal tract with higher concentrations in the midgut regions. Experiments are ongoing to complete the aims of this study.

Conclusion:
This research has localized NUCB1 expression within the fruit fly gastrointestinal tract, prompting inferences of its role within the fly. The next stages of this research include utilizing specific Gal4 lines within the Gal4-UAS system to determine which cells within the gut express NUCB1. These experiments will lead to a deeper understanding of the mechanism in which NUCB1 plays its appetite suppressing role and its potential effects on other systems within Drosophila melanogaster.

96. RAPID DETECTION OF POULTRY DISEASES USING METABOLOMICS

Presenter: **Asha Ranaraja**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Susantha Gomis, Western College of Veterinary Medicine

Background:
Metabolic markers are increasingly important in the rapid detection of diseases, risk assessment, and prognosis. Although there is enough documentation on the application of these biomarkers in human healthcare, they are still emerging in veterinary medicine, especially in avian disease diagnosis. The poultry industry demands the need for diagnostic tools that can detect infections early before clinical symptoms appear such as within 1-2 days post-infection, which will help to minimize huge economic losses faced by the broiler chicken industry in Canada due to infectious diseases. It is also essential to address subclinical and concurrent bacterial and viral infections as well. Therefore, this research was conducted to identify metabolic markers for the rapid detection of viral and bacterial infections in broiler chickens.

Methods:
Blood samples were collected from broiler chickens exposed to avian pathogenic Escherichia coli and avian reovirus, as well as samples from unexposed control birds, 24 hours after the challenge. Serum metabolites were analyzed using liquid chromatography-mass spectrometry in conjunction with multiple statistical techniques such as univariate, and multivariate analyses, feature selection, machine learning, and pathway analysis with metaboanalyst 4 software.

Results:
The metabolite profiles of infected and control birds differed significantly. Pathway analysis revealed a substantial downregulation in purine metabolism in infected birds compared to controls.

Conclusion:
Purine metabolism is critical for energy production, DNA synthesis, and signaling. During the early phases of infection, the body undergoes reprogramming, which includes enhanced glycolysis for ATP and biosynthesis. This is necessary for damage repair and defensive responses, as well as epigenetic changes associated with immune cell functions. Therefore, the downregulated of purine derivatives can be a potential biomarker for early identification of E. coli and reovirus infections in broiler chickens. This research marks a significant step forward in applying metabolic markers for the expedited diagnosis of avian diseases in veterinary science.

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97. ENGINEERING NEXT-GENERATION LIPID NANOPARTICLES FOR MESSENGER RNA DELIVERY BY INHALATION

Presenter:	**Naga Suresh Kola**
USask Affiliation:	**Graduate student**
College:	**School of Public Health**
Collaborators:	Adam Leontowich Neeraj Dhar, College of Medicine Yan Zhou, Western College of Veterinary Medicine
Supervisor:	Aneesh Thakur, College of Medicine

Background:

Messenger ribonucleic acid (mRNA) vaccines are heralded as a revolutionary modality in vaccinology, owing to their safety and efficacy. Yet, delivering mRNA for protein synthesis in the cells is challenging due to its vulnerability to degradation. Lipid nanoparticles (LNPs) emerge as a promising delivery system that ensures safe and effective mRNA delivery. Despite advancements in LNP development, effective mucosal delivery of mRNA remains elusive, posing a significant research problem. Developing efficient mucosal vaccine delivery systems could enhance antigen-specific mucosal and systemic immunity, which is crucial for combating intracellular pathogens at their initial entry sites.

Methods:

We designed LNPs by screening constituent lipids via quality-by-design principles and formulated mRNA-LNP complexes by microfluidic mixing. To characterize the nanoparticles, various analytical methods were employed such as photon correlation spectroscopy (PCS) for size and polydispersity, cryo-transmission electron microscopy (Cryo-TEM) for morphology, and fluorescent-based assays for determining mRNA entrapment and ionization potential (pKa) of LNPs. Synchrotron-based small-angle X-ray scattering (SAXS) was used to determine the lipid self-assembly process. To assess the efficacy of LNPs for mRNA transfection, we conducted in vitro studies using different cell lines and in vivo bioluminescent imaging in mice following subcutaneous injection and pulmonary administration by vibrating mesh nebulizer.

Results:

Nebulization significantly influenced LNP's physicochemical characteristics. LNPs displayed distinct in vitro transfection efficiencies, indicating the influence of intrinsic nanoparticle properties on cellular uptake and mRNA release. Furthermore, the ionization potential of LNPs (pKa range of 6 to 7) affected lipid phase transitions. SAXS measurements revealed higher-order structures at pH 5, attributed to lipid ionization behavior. Cryo-TEM analysis depicted spherical oligolamellar structures, consistent with PCS-reported sizes. Luciferase expression assays in mice showed sustained protein expression until 5 days.

Conclusion:

The developed mRNA-LNPs exhibited physicochemical, structural, and functional properties consistent with the literature. However, nebulization significantly influenced the physicochemical properties of mRNA-LNPs.

98. ANTIMICROBIAL RESISTANCE SURVEILLANCE AND INDENTIFICATION OF RESISTANCE GENERS IN E.COLI CAUSING CANINE URINARY TRACT INFECTIONS IN SASKATCHEWN, BETWEEN 2013-2022

Presenter:	**Yaasin Dulymamode**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Collaborators:	Joe Rubin, Western College of Veterinary Medicine
Supervisor:	Joe Rubin, Western College of Veterinary Medicine

Background:

E. coli is the most common cause of urinary infections in dogs. Antimicrobials are the primary treatment for these infections. However, the emergence of antimicrobial resistance globally is resulting in limited therapeutic options available to veterinarians and physicians. Since 2013, we have conducted a longitudinal passive surveillance project targeting E. coli causing UTIs in dogs isolated by a regional diagnostic lab. The objectives of this study were to describe the antimicrobial susceptibility of E. coli isolates from Oct 2018-Oct 2022 (n = 593) and the frequency of epidemiologically important resistance genes.

Methods:

Laboratory records were reviewed to ensure that only one isolate per dog was included. Antimicrobial minimum inhibitory concentrations (MICs) were determined by broth microdilution and agar dilution. Molecular sequencing techniques are used to identify resistance genes and sequence types of resistant isolates.

Results:

Overall, 76.2% of the isolates were pan-susceptible while 4.3% were multidrug-resistant (resistant to 3 or more drug classes). Ampicillin resistance, identified in 14.0% of isolates, was the most common. Resistance to amoxicillin-clavulanic acid, cefazolin, chloramphenicol, nalidixic acid, and tetracycline individually was between 5-10%. Fewer than 5% of isolates were resistant to cefoxitin, ceftriaxone, cefepime, ciprofloxacin, gentamicin, amikacin, tobramycin, or trimethoprim-sulfamethoxazole. Based on the susceptibility profile, isolates were screened for broad-spectrum β-lactamases (ESBL and AmpC enzymes) and plasmid-mediated quinolone resistance determinants (PMQRs) by PCR, amplicons were then sequenced to identify gene alleles. Isolates possessing these genes were strain typed by MLST to identify resistant clones. Broad-spectrum β-lactamases were identified among fewer than 5% of the isolates; the AmpC type enzymes were most common. Three isolates possessed a PMQR determinant. Two isolates possessed qnrS8 and a single isolate possessed aac(6')-Ib-cr. Over the 9-year study period, no significant changes in the frequency of resistance to ampicillin, amoxicillin-clavulanic acid, cefazolin, nalidixic acid, tetracycline, or chloramphenicol were identified. However, significant increases in the MICs of ampicillin, nalidixic acid, and chloramphenicol were found, similar trends were not identified for other drugs.

Conclusion:

This study demonstrates that although E. coli causing canine UTIs remain susceptible in this region, temporal increases in the MICs to some drugs may indicate the emergence of resistance. These results support the use of first-line therapies such as amoxicillin or trimethoprim-sulfamethoxazole recommended by the ISCAID guidelines for the region of Saskatchewan, Canada. Finally, continued surveillance is warranted to identify emerging resistance trends and guide future empiric therapy.

99. DEVELOPMENT OF NEXT-GENERATION RNA VACCINES USING SELF-AMPLIFYING RNA

Presenter:	**Dhruv Patel**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Anil Kumar, College of Medicine
	Ekaterina Dadachova, College of Pharmacy and Nutrition
	Arinjay Banerjee, Western College of Veterinary Medicine
Supervisor:	Aneesh Thakur, College of Medicine

Background:
A major limitation of messenger RNA (mRNA)-based vaccines is the requirement of multiple immunizations to achieve desired protective efficacy and at a higher mRNA dose. Self-amplifying mRNA (saRNA) based vaccines that encode a self-replicating machinery within it could be an attractive approach to tackle these issues. saRNA produces multiple mRNA copies inside the body, generating a similar immune response but using much lower doses than mRNA. Therefore, the project aims to develop a saRNA vaccine that can induce sustained antigen expression and eliminate the requirement for multiple immunizations.

Methods:
Firefly luciferase (Fluc) protein-encoding mRNA and saRNA were selected to evaluate in vivo protein expression. Lipid nanoparticles (LNPs) encapsulating mRNA or saRNA were prepared by the microfluidic mixing technique. The physicochemical characterization of LNPs was performed through dynamic light scattering (DLS) and transmission electron microscopy (TEM). Quantification of RNA was performed with Quant-it™ RiboGreen RNA assay kit. In vivo protein expression was compared between mRNA and saRNA by injecting the RNA-loaded LNPs in BALB/c mice subcutaneously.

Results:
Dynamic light scattering results demonstrated the size of nanoparticles smaller than 150 nm and polydispersity index (PDI) less than 0.15. RNA quantification results showed the RNA entrapment greater than 80% for both mRNA and saRNA. TEM images showed round and spherical nanoparticles. In vivo protein expression in mice showed higher protein expression ($p<0.005$) with saRNA-LNPs at multiple time points as compared to mRNA-LNPs. Whereas luminescence at the site of injection completely disappeared at and after day 7 with mRNA-LNPs, saRNA-LNPs continued to express protein after day 7.

Conclusion:
This study demonstrated a significant difference in in vivo protein expression between mRNA and saRNA-encapsulated LNPs. An extended protein expression can be obtained using saRNA, which can be used to formulate vaccines inducing sustained expression of any antigenic protein such as spike protein in the case of SARS-CoV-2.

100. EXPLORING SEX-BASED IMMUNE RESPONSES IN SWINE AFTER INTRAMUSCULAR VACCINATION

Presenter:	**Kezia Fourie**
USask Affiliation:	**Graduate student**
College:	**Vaccine and Infectious Disease Organization**
Collaborators:	Pooja Choudhary
	Alison Jeffery, Western College of Veterinary Medicine
	Dylan Chand, School of Public Health
Supervisor:	Heather Wilson, Western College of Veterinary Medicine

Background:
In humans, sex can influence the immune response to vaccination. These sex-influenced responses to vaccination have been well explored in humans but less so in swine. Accordingly, many studies use mixed-sex groups to account for possible sex influence. The question remains, however, if sex-based differences are observed in sexually immature swine.

Methods:
Systemic and mucosal antibody-mediated and cell-mediated immune responses after vaccination were evaluated. Vaccines 1 and 2 were formulated with the same adjuvant (A1) but different proteins – F, O, and G or C and M. Vaccines 3 and 4 were formulated with different adjuvants (A2 or A3) but the same proteins – F, G, and Y. For Vaccines 1 and 2, pigs were vaccinated on days 0, and 14 with blood collection on days 0, 14, and 27 post-vaccination. For Vaccines 3 and 4, pigs were vaccinated on days 0, and 21 with blood collection on days 0, 21, and 42 post-vaccination. Samples for mucosal antibody-mediated immunity were collected on day 45 for Vaccines 1 and 2 and on day 42 for Vaccines 3 and 4. Antigen-specific antibody titres and interferon gamma (IFNγ) concentrations were measured.

Results:
Analysis of antibody titers showed no influence of sex on systemic and mucosal antibodies regardless of vaccine formulation or time point. As well, cell-mediated immune responses as quantified through IFNγ was not influenced by sex for any formulation.

Conclusion:
This initial analysis suggests that in sexually immature swine, sex-based differences in response to vaccination are not observed. However, more work is needed to increase the power of the study but to also examine the effect of age on sex-based differences.

101. A ONE-DOSE STREPTOCOCCUS ZOOEPIDEMICUS VACCINE DECREASES SEVERITY OF CLINICAL SIGNS AND LESIONS POST-CHALLENGE

Presenter: **Ana Norte**
USask Affiliation: **Graduate student**
College: **Western College of Veterinary Medicine**
Supervisor: Matheus Costa, Western College of Veterinary Medicine

Background:
Streptococcus equi subsp. zooepidemicus (SeZ) is an emerging pathogen that causes septicemia and high mortality in pigs. A previously developed two-dose live attenuated vaccine prevented 100% of the clinical signs and mortality post-challenge. This study aimed to evaluate clinical signs and mortality following an one-dose regimen.

Methods:
Eight 3-week-old pigs were allocated into two groups: control (sham vaccinated, n=4), or one-dose vaccinated (n=4). One-dose group received 1 mL (106 CFU/mL) orally 21 days pre-challenge. Sham vaccinated group received 1 mL of sterile culture broth, orally. All pigs were challenged orally with 1 mL (106 CFU/mL) of virulent ST-194 SeZ strain. Following challenge, pigs were monitored daily for responsiveness, respiratory effort, and rectal temperature. Animals were euthanized either due to welfare concerns or 7 days post-challenge. Necropsy was performed to assess gross lesions. Differences in clinical scores between groups were assessed by Wilcoxon rank-sum test. Differences in rectal temperature between groups were analyzed by T-test. Risk ratio was assessed by Fisher's exact test.

Results:
Data is shown as mean and standard deviation. The control group had a higher rectal temperature 24 hours post-challenge (hpc) (41.42°C±0.22) than the vaccinated group (39.92°C±0.58), suggesting fever (>40°C) ($t=2.39$, $p=0.053$). The control group was significantly less responsive ($z=2.646$, $p=0.028$) and had increased respiratory effort ($z=2.530$, $p=0.0286$) when compared to the vaccinated group at 24 hpc. Control pigs (4/4) were twice more likely to die following challenge than vaccinated pigs (2/4, $p=0.428$, 0.750 - 5.328 CI). Post-mortem examination revealed a higher frequency of gross lesions in control animals than vaccinated ones: polyserositis (50% vs 0%), tonsilitis (100% vs 25%), gall bladder edema (50% vs 25%), spleen infarction (50% vs 25%), and hemorrhagic submandibular lymph nodes (100% vs 25%).

Conclusion:
The one-dose regimen decreased the severity of clinical signs and lesions and increased survival rates compared to unvaccinated pigs.

102. INFLUENCE OF TIME SINCE PAIN ONSET AND AGE ON SCAPULAR KINEMATICS DURING AN OVERHEAD REACHING TASK

Presenter:	**Lauryn Campbell**
USask Affiliation:	**Graduate student**
College:	**College of Engineering**
Collaborators:	Sophia Abiara, College of Kinesiology
Supervisor:	Angelica Lang, College of Medicine

Background:
Rotator cuff disorders and aging are associated with kinematic alterations in the shoulder, but results assessing both are often inconclusive. Time since pain onset is crucial, yet often overlooked, in understanding biomechanical consequences. Few studies have explored how it affects shoulder motion, potentially contributing to long-term disorders. The objective of this study is to explore the effects of time since pain onset and age on scapular kinematics in individuals with rotator cuff disorders. We hypothesize that biomechanical alterations will present along a continuum.

Methods:
Sixty participants were pre-screened to ensure the presence of chronic shoulder pain. Movement was tracked with motion capture during an overhead reach task. Scapular orientations and humeral axial rotation at 30° increments of humeral elevation were extracted. Robust linear regression models ($p<.05$) examined the relationship between scapular and humeral angles, time since pain onset (reported in years), and age.

Results:
Scapular upward rotation was influenced by the interaction of time since pain onset and age at 60° of humeral elevation during the overhead reach. Younger participants demonstrated substantially decreased upward rotation over time while upward rotation slightly increased over time for the older participants ($p <.001$, $r^2 = .36$). The older population displayed protective movement patterns as time since pain onset increased. While the protective compensation may be beneficial, other harmful movement patterns associated with aging could prevent recovery. Conversely, younger individuals exhibited harmful scapular kinematics with a sharp decline in upward rotation suggesting potential for further tissue damage over time.

Conclusion:
Different alterations in scapular kinematics over time, mediated by age, may shed light on why shoulder pain continues over time, and highlight potential different mechanisms for injury depending on age. Ongoing analyses with are exploring the effects of other factors on kinematic alterations over time in individuals with rotator cuff disorders to better understand these findings.

103. INVESTIGATING TRUNK AND HUMERAL POSTURAL DEMANDS ACROSS A URANIUM MINE SITE

Presenter:	**Denise Balogh**
USask Affiliation:	**Graduate student**
College:	**College of Medicine**
Collaborators:	Angelica Lang, College of Medicine
Supervisor:	Angelica Lang, College of Medicine

Background:

The low back and shoulder are common sites for injury in workers exposed to occupational risk factors, including non-neutral postures, prolonged overhead work, and repetitive humeral and trunk movement [1]. At a mine site operation, many workers, from miners to those in a support role (ex. site-services), may be exposed to these ergonomic risk factors to varying degrees. Various studies have used naturalistic methods, surveys, or observational ergonomic assessment tools to assess the musculoskeletal risk of mine site workers [2,3]. In a cross-sectional study of 459 stone, sand, and gravel mine site workers, 57% of workers experienced musculoskeletal symptoms at the low back, while 38% of workers experienced musculoskeletal symptoms at the shoulder within the past year [2]. There is a clear need for further investigation into the trunk and shoulder postural demands required on a mine site. Inertial measurement units (IMUs) allow for kinematics to be directly measured in-field; consequently, IMUs may allow for a higher resolution definition of postural demands than what is provided by traditional ergonomic assessment methods [4]. However, IMUs have yet to be taken onto a mine site for the purposes of kinematic analyses. The purpose of this study was to utilize IMUs for a more precise and comprehensive description of the humeral and trunk postural demands in occupations necessary for the operation of a mine site.

Methods:

Thirteen workers, including 2 miners, 2 mechanics, 2 housekeepers, 1 baggage handler, 1 electrician, 1 kitchen staff, 1 fire safety technician, 1 plumber, 1 pipefitter, and 1 process operator, stationed at a northern Saskatchewan uranium mine site participated in this study. Three IMUs sampling at 100Hz (Xsens, Awinda, Xsens Technology, NL) were placed on the sternum and bilateral humeri of workers to capture short snapshots of upper body kinematics during 1-4 natural tasks per worker. Due to disturbance by ferromagnetic material in the mine environment, magnetometer data was discarded; joint angles relative to gravity were calculated using the accelerometer data through a custom code (MatLab 2023a, MathWorks, MA). Right humeral flexion (+ values) as well as trunk flexion (+ values) and extension (- values) were descriptively compared between work tasks using the 10th, 50th and 90th percentiles (PTLs). Additionally, the range of movement (90th PTL -10th PTL), the percent time in neutral (0-20^{o}) and extreme (>60^{o}) flexion, as well as the frequency of movement changes per minute (FMPM; expressed as the number of times the humerus or trunk moved ±20^{o} away from the mean flexion angle) for each task were calculated.

Results:

A total of 27 different tasks performed by the various mine site workers were captured, resulting in 9.4min ±7.6min of kinematic data per work task. Humeral flexion across all tasks was an average of 13.0^{o} ± 3.5^{o}, 68.2^{o} ± 19.5^{o}, and 117.6^{o} ± 27.4^{o} for the 10th, 50th and 90th PTLs respectively; the mean range was 104.6^{o} ± 24.5^{o}. The greatest range of humeral elevation was 137.1^{o}, which was performed by a housekeeper during a ‘make bed’ task. The mean % time spent in extreme humeral flexion was 19.2% ± 15.9%. A process operator performing a ‘filter press operation-bins’ task spent the greatest %

time in extreme humeral flexion (54.8% of time). The mean humeral FMPM across all tasks was 51.7 ± 22.7. The greatest FMPM was 72.8 and performed by a plumber during a 'pipe cutting' task, indicating that this task involved highly repetitive humeral movement. Trunk flexion across all tasks was an average of -6.3^{o} ± 15.9^{o}, 36.1^{o} ± 13.9^{o}, and 78.9^{o} ± 20.0^{o} for the 10th, 50th and 90th PTLs respectively, with a mean range of 85.1^{o} ± 23.4^{o}. The mean % time spent in extreme trunk flexion was 8.5% ± 9.7%, while the greatest % time spent in extreme trunk flexion was 38.6% and performed by a plumber during a 'water pump repair' task. Finally, mean trunk FMPM across tasks was 24.3 ± 11.6; the highest FMPM was 29.6, performed by a miner during a 'drilling' task.

Conclusion:

This study was one of the first investigations to use IMUs to quantify postural demands on a mine site. Across all tasks, the highest levels of humeral and trunk flexion range, % time in extreme posture and FMPM were all observed in different occupations, indicating that risk factors for shoulder and trunk injury are spread across an array of jobs at a uranium mine site. References [1] van der Windt et al.,2000. Occup Environ Med. 57(7):433–442 [2] Balogun and Smith, 2020. Int. J. Environ. Res. Public Health, 17(10), 3512 [3] Weston et al., 2016. J Saf Health Environ Res.12(1): 274–283 [4] Schall et al., 2022. J. Occup. Environ. Hygiene. 19(9):501-508 [5] Kazmierczack et al., 2005. App. Ergonomics. 36(3):263-27

104. ACUTE INTERMITTENT HYPOXIA AND NEUROPLASTICITY AFTER SPINAL CORD INJURY

Presenter:	**Nima Khalili Tanha**
USask Affiliation:	**Graduate student**
College:	**Western College of Veterinary Medicine**
Supervisor:	Bhezad Toosi, Western College of Veterinary Medicine
Co-supervisor(s):	Gillian Muir

Background:
Spinal cord injury (SCI) is a debilitating condition leading to significant motor deficits. Despite medical advancements, restoring neural activity post-SCI remains challenging. This study explores the therapeutic potential of acute intermittent hypoxia (AIH) treatment in enhancing neuroplasticity and motor recovery, using a murine model of partial cervical spinal injury.

Methods:
Healthy male C57BL/6 mice underwent spinal hemisection at the 3rd cervical spinal segment or a sham surgery (n=24 per group) followed by one week of recovery. Mice were then treated with daily AIH (10, 5-min episodes of 11% inspired O2 with 5-min intervals of 21% O2) or normoxia (continuous 21% O2) following either a short-term (7 days) or long-term (7 days plus 5 times a week for an additional 3 weeks) protocol (n=6 per group). Motor recovery was assessed using the narrow beam walking test, and the expression of the neuroplasticity marker, BDNF, was analyzed using Western blotting and immunohistochemistry to identify BDNF-expressing cells.

Results:
The spinal hemisection resulted in significant motor deficits, detectable with the narrow beam walking test. Long-term AIH treatment improved motor recovery in mice with spinal injury. Interestingly, enhanced BDNF expression was only detectable after 7 days of AIH treatment, suggesting that changes in plasticity markers precede improvements in motor functions. Additionally, an increase in the number of glial cells and astrocytes at the site of injury was observed, as indicated by immunohistochemistry for Iba-1 and GFAP, markers of these cells, respectively.

Conclusion:
We have successfully developed a mouse model of SCI to study AIH-induced neuroplasticity. Our findings suggest that AIH treatment can enhance motor recovery following SCI, potentially through the upregulation of BDNF. These results provide a promising foundation for further investigation into the therapeutic potential of AIH in SCI recovery.

Special thanks

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Thanks to their commitment to supporting the next generation of health professionals, the expo has been able to continue its mission of bridging disciplinary gaps and enhancing discovery while championing efforts that can improve the health of people across Saskatchewan, Canada, and the world.

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