



UCVM SURE Research Day 2022

August 25th, 2022

8:25 am - 4:00 pm

Health Sciences Centre Theatre 3 and HRIC Atrium

PROGRAM and ABSTRACT BOOKLET

Message from Dr. Hermann Schaetzl, Associate Dean, Research



Dear summer students, Dear colleagues,

A warm welcome from the Associate Dean, Research. This probably is the first in-person research meeting for many of us after a long, long time. Doesn't this feel good!

The meeting will showcase the wide range of research activities our 2022 summer students were involved in. Over 50 SURE (Summer Undergraduate Research Experience) students registered this year, including eleven students from our DVM program. A special welcome! The range of research activities showcased here is amazingly broad, and of very high quality. We are proud that 56% of SURE students received UCVM-external

stipend support, worth >\$212,000.

We will have 20 oral presentations and 28 poster presentations today, together with a keynote lecture and prizes for best oral and poster presentations. We would like to emphasize that the organizing committee comprises students, postdoctoral fellows, staff, and faculty members. We are confident that this meeting brings together the UCVM community and shows the high quality of research and training activities our faculty is known for. Most importantly, congratulations to all SURE students for their excellent work. They obtained real-life and real-time insight into the fascinating but complex world of research (where things don't always work as expected), an experience that hopefully stimulated their appetite for more in the future.

I would like to thank my co-organizer Brenda Moore from the UCVM Research Office, the members of the organization committee, doing abstract adjudication, session chairing and judging, for their outstanding help and efforts, and everybody else helping to make this meeting a success.

Sincerely,

Hermann M. Schaetzl

Associate Dean Research

Message from Dr. David Hall, Associate Dean, Emerging Scholars



Dear Students and Colleagues,

Welcome to UCVM SURE Research Day, 2022 from the Office of the Associate Dean, Emerging Scholars. Research is a key priority for the Faculty of Veterinary Medicine, and today you will be sharing your results from some of those research projects as well as learning about the discoveries of your peers and research leaders. For many of you this will have been your first opportunity to work on a research project; we hope it has inspired you to consider how your career might include engagement with research.

Today you have the chance to see how broad is the scope of research in veterinary science, ranging from lab bench investigation of cellular activity, to field based epidemiology, to qualitative analysis of latent variables. Despite the wide range of topics, commonality exists through a sense of curiosity, desire to investigate, and enthusiasm for sharing results.

My compliments and gratitude to all of you for your work this summer and for taking the time to share your results with our UCVM community. Enjoy the day and your burgeoning careers in veterinary science!

Sincerely,

David C. Hall

Associate Dean, Emerging Scholars, UCVM

UCVM Sure Research Day 2022

8:25	Welcome and Introductions – Theatre 3: Hermann Schaetzl/David Hall			
Oral Presentation Session #1 – Theatre 3				
0.20	Chairs: Cameron Knight and Angelica Petersen-Dias			
8:30	Raffay Ilyas (Cheng) Anxiety during postnatal development in Fragile X mouse model			
	Lauren Stoffregen (Whitehead)			
	Investigating the seroprevalence of the causative agents of Equine Protozoal Myeloencephalitis in			
	Alberta			
	Aydin Herik (Cobo)			
	Neonatal calves show a constitutively impaired mucin barrier that leads to severe enterocolitis after			
	Cryptosporidium parvum <i>challenge</i>			
	Karlie Neuman (van der Meer)			
	Quantifying virus neutralizing antibodies of bovine respiratory diseases causing BHV-1 in preconditioned			
	and non-conditioned calves using VNT and ELISA			
	Nicole Wilson (Gilch)			
	Cholesterol overload of neuronal cells interferes with de novo prion infection			
9:30	Mini-Break			
	Oral Presentation Session #2 – Theatre 3			
	Chairs: Maria Arifin and Dexter Merenick			
9:35	Anupam Jay (Barkema)			
	Herd-level prevalence of bovine leukosis in dairy herds of Alberta			
	Ihncheol Jung (Poissant)			
	Repeatability of horse body condition scores from photos			
	Hiruni Ranaweera (Careem)			
	Tropism of Delmarva (DMV/1639) Infectious Bronchitis Virus (IBV) variant for lymphoid organs of			
	chickens			
	Kathryn McLellan (Pearson)			
	Cow- and calf-level factors associated with nursing behaviours in beef cattle during the 24-hours			
	following an assisted calving			
	Alexandra Burk (Rosa & Roy)			
	AMR profiling of bacterial clinical isolates and commensal fecal Escherichia coli in Alberta horses			
10:35	Coffee Break - HRIC Atrium			
	Oral Presentation Session #3 – Theatre 3 Chairs: Tahir Ali and Charlotte Bourbon			
11:00	Anisha Jessel (Pang)			
11.00	Comparing loss of posture and loss of righting reflex as indicators of unconsciousness during exposure to			
	carbon dioxide in laboratory rats			
	Carl Dizon (Cork)			
	Modelling the effects of climate change on the geographic distribution of Ixodes pacificus and Borrelia			
	burgdorferi in the Pacific Northwest			
	Monica Bautista (Chu)			
	Constructing a real-time live cell WNT signalling reporter in pluripotent stem cells			
	Aiden McBean (Betancourt)			
	Association between maternal protective behaviour and reactivity to handling of Angus cows			
	Shahaanaa Mohanraj (Soghigian)			
	Distribution and Ecology of Culex Pipiens in Calgary			

12:00	Lunch and Poster Session - HRIC Atrium		
Oral Presentation Session #4 – Theatre 3			
Chairs: Heather Steele and Ben Caddey			
14:00	Nancy Ngo (Kutz)		
	Evaluating the benefits of Traditional Ecological Knowledge (TEK) collection from locally led short		
	interviews in the Ulukhaktok community to build upon classical Western approaches to Arctic wildlife-		
	assessments		
	Louise Caplan (Hall)		
	Hunters' willingness to change their hunting practices in response to zoonotic disease		
	Akeel Faizal (Niu)		
	In vitro evaluation of lytic bacteriophages against Shiga toxin producing Escherichia coli O157:H7 on		
	human intestinal cells		
	Byron Kruger (Schaetzl)		
	Chronic wasting disease (CWD) vaccine development and production		
	Julian Chua (Wasmuth)		
	Computational analysis to identify mimicry proteins produced by myxozoa in fish hosts		
15:00	Keynote Speaker – Theatre 3		
	Dr. Thilo Pfau		
	Applied equine locomotor biomechanics: Technological solutions aiding lameness detection and injury		
	prevention		
15:45	Prizes and Closing Remarks: David Hall/Hermann Schaetzl		

ABSTRACTS

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A phylogenomic perspective on the loss of blood-feeding in mosquitos

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Keywords: Blood-feeding, Phylogenetics, Genomic Tools

Perhaps the most important behavior exhibited by mosquitoes is blood-feeding. However, more than 150 species in three genera are unable to blood-fed. Due to the lack of genetic data available for most mosquitoes, whether this represents a single event in the evolutionary history of mosquitoes, or multiple independent events, is unknown. Advances in sequencing methods have enabled extensive sampling of non-model mosquito species, but the best way to analyze such data has been untested. To simultaneously address the times blood-feeding has been lost, and the best methods with which to analyze orthologous sequence data for non-model organisms, we evaluated two bioinformatic pipelines and sequence data from 59 mosquito species. We hypothesize that the loss of blood-feeding was a single evolutionary event within the common ancestor of the three genera that do not blood-feed, Malaya, Topomyia, and Toxorvnchites. To test this hypothesis, we generated alignments of orthologous sequences from two bioinformatic pipelines, Orthograph and HybPiper. From resulting sequence alignments, we will generate phylogenetic trees to evaluate how many times blood-feeding has been lost in mosquitoes. We compared these pipelines in the number of orthologous sequences each recovered, as well as support values from phylogenetic analyses of sequence alignments. We anticipate that the pipeline utilizing HybPiper will result in more sequence data recovered, and phylogenies with greater bootstrap support values, due to HybPiper retrieving orthologous intronic sequence data. Using the outputs of the pipelines, we will discuss support for a single loss of blood-feeding in mosquitoes, which gave rise to non-biting lineages.

Association between maternal protective behaviour and reactivity to handling of Angus cows

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Keywords: cattle temperament, maternal defense, beef cattle

Cattle reactivity is a behavioural reaction in response to handling, while maternal protectiveness demonstrates care and/or attentiveness towards their calf. Reactive cattle are more difficult to handle and represent risk of injury/death. The behavior of a newly calved cow is also considered one of the greatest threats to the security of producers. Our aim was to evaluate the association between reactivity and maternal protective behaviors of Angus cows. 121 cows were assessed at W.A. Ranches, Alberta, Canada, for their reactivity (flight speed-FS and composite reactivity score-REA) during routine handling in the corral, as well as maternal protective traits (maternal protectiveness-MPS, displacement-DIS, agitation-AGI, and attention-ATT) while stockpersons interacted with their calf within 12h after parturition. The associations among variables were analyzed using factor analysis, with varimax rotation, vielding two factors (PC1 and PC2) with eigenvalues exceeding 1.0. PC1, explaining 33.5% of the total variance, presented higher positive loadings for MPS (0.80), DIS (0.64), AGI (0.78) and ATT (0.58), indicating that those traits are positively related (the more protective the cow, more attentive and engaged). PC2, explaining 28.7% of variance, presented higher positive loadings for REA (0.86) and FS (0.87), illustrating a positive relationship between these indicators. Our results demonstrated that Angus cows' maternal protective behaviours are independent of their reactivity during routine handling. These results contradict our hypothesis that cows who are more defensive of their offspring tend to be more reactive. Instead, expressions of these two behaviors seems to be independent and may result from other characteristics of the cow.

In vitro evaluation of lytic bacteriophages against Shiga toxin producing *Escherichia coli* O157:H7 on human intestinal cells

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Keywords: Bacteriophage, Shiga toxin producing Escherichia coli, human intestinal epithelial cell

Shiga toxin (Stx)-producing Escherichia coli (STEC), particularly the serotype O157:H7, is a significant public health concern worldwide, causing life-threating conditions such as haemorrhagic colitis and hemolytic uremic syndrome (HUS). Unlike other foodborne bacterial infection, antibiotics are not recommended, due to its potential acceleration of Stx release resulting HUS. Given that a few bacteria can cause severe human illness and there is no preferred direct treatment currently available, it is urgent to develop novel STEC control measures. Use of bacteriophages against STEC is one of the options gaining increasing attention. The objective of the proposed work was to compare effectiveness of two lytic phages (AKFV33, AHP24-big plaque and a cocktail of Wv7, AHP24-big plaque and AHP24-small plaque) in preventing STEC attachment onto human intestinal epithelial cells, which is an initial step for STEC invasion. Intestinal epithelial cells were cultured until confluency and the phage treatment was done. After incubation of treated and control cells (1 hour), STEC inoculation was done along with uninoculated controls, the cells were incubated (1 hour) and the adhered and unattached STEC on intestinal cells were enumerated. We observed about 3 log reductions in STEC attachment when the intestinal cells were treated with AKFV33 and AHP24-big plague phages and the cocktail of phages 1 hour before bacterial inoculation. The number of unattached STEC on intestinal cells were also reduced by 3 logs when these phage treatments were done. The results of the proposed work will contribute to the knowledge of the rapeutic outcome of phages against STECO157: H7.

AMR profiling of bacterial clinical isolates and commensal fecal *Escherichia coli* in Alberta horses

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Keywords: Antimicrobial resistance; Horse; Escherichia coli

Antimicrobial resistance (AMR) is a significant wicked problem and current One Health issue. Escherichia coli (E. coli) is commonly used for AMR surveillance because it is a reservoir for resistant genes and contributes to the spread of AMR via fecal shedding. A significant knowledge gap exists regarding bacterial AMR in Alberta horses. This study seeks to address that gap by investigating fecal *E. coli* AMR profiles of various Alberta horse populations, including feral and domestic horses. We hypothesized that the prevalence of AMR in fecal E. coli would correlate with exposure to AMDs with very low prevalence in feral horses (FER) and untreated domestic horses (DOM), and higher prevalence in previously treated (TX) and hospitalized horses (HOSP). Single fresh fecal samples were collected from FER, DOM, and TX. Serial fresh fecal samples were collected from HOSP. E. coli culture and susceptibility testing was performed using the NARMS panel (CMV5AGNF), with human clinical breakpoints used for all AMDs. Reduced susceptibility to chloramphenicol was present in 15, 15, and 8% of FER, DOM, and TX E.coli isolates, respectively. Twenty-seven percent of TX E.coli isolates were resistant to sulfonamides (trimethoprim-sulfamethoxazole, sulfisoxazole), compared to 0% of FER and DOM (p<0.0001; Fisher's exact test). Serial samples from 2 of 3 HOSP revealed development of multi-drug resistance during hospitalisation. These results suggest environmental transmission of chloramphenicol resistance, association between prior treatment and sulfonamide resistance, and high risk of development of multi-AMR in HOSP. This study provides more information on AMR prevalence and transmission in Alberta horses.

Veterinarians' perspectives on implementation of parasite control strategies on cow-calf operations in Alberta

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Keywords: gastrointestinal nematodes, ectoparasites, beef calves

In the beef industry in Alberta, there is a paucity of information available regarding the perceived effectiveness of current pharmaceutical interventions and other practices used for parasite control in pre-weaned calves and their dams. The objective was to describe veterinarian's perspectives of current parasite control strategies and the motivators and barriers that drive the recommendations they make to their clients. A list of 31 veterinarians collaborating with the Canadian Cow-Calf Surveillance network was obtained. Interview invitations were sent by email to all potential participants, which was followed by phone call(s). Interviews were recorded with an audio recorder (Sony IC), then transcribed using OtterAI, and imported into NVivo software where content themes could be extracted. From 4 interviews conducted thus far, the following preliminary themes have been identified as common factors that affect recommended parasite control protocols: cattle handling times, cost, facilities or infrastructure, and a lack of treatment options despite acknowledged drug resistance to macrocyclic lactones. External parasites appear to drive conversations about change in parasite control strategies, corresponding to a perceived increase in the prevalence of itchy cows in the winter despite treatment in the fall. Contrarily, internal parasites are given little consideration, because these are stated to be of low concern for beef producers in Alberta. These findings can help to better direct future research, extension efforts, and policy making for stakeholders in the beef industry by supporting discussions about effective parasite control strategies for beef cattle in Alberta.

Optimisation of electrotransformation efficiency of *E. coli* EDL933 in preparation for transposon-directed insertion site sequencing (TraDIS)

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Keywords: phage, STEC, biocontrols

Shiga toxin-producing Escherichia coli (STEC) serotype O157:H7 is one of the most prevalent pathogens in the food supply chain and has been responsible for numerous food-borne illness outbreaks in North America. Our lab is investigating non-antibiotic therapy in response to an increase in the number of antibiotic-resistant STEC O157:H7 strains. As such, we have isolated a number of STEC bacteriophages including Tequintavirus AKFV33 which can productively infect strains from multiple STEC serogroups, including O157:H7 strain EDL933. We are seeking to better understand which EDL933 genes interact with phage during the infectious cycle. In order to do so, our group will perform a genome-wide phenotypic fitness assay using Transposon-Directed Insertion Site Sequencing (TraDIS) on a transposon mutant library of strain EDL933 treated with AKFV33. To achieve this goal, we will construct a mutant library to assay a sufficient number of genes by Tn5 transposon directed mutation. To develop a protocol that will yield a high number of transformant colonies, we concentrated and electrotransformed EDL933 cultures with pUC19 control plasmid at 1.8, 2.0, and 2.5 kV. 2.5kV replicates yielded the highest transformation efficiency, plating on average 279 colonies on a plate containing a 10^-3 dilution of transformant culture. We believe these experiments will assist us in employing TraDIS to uncover EDL933 genes that could play a role in host-phage interactions, allowing us to better understand how phages can be employed as a biocontrol against STEC in the food supply chain.

Comparing loss of posture and loss of righting reflex as indicators of unconsciousness during exposure to carbon dioxide in laboratory rats

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Keywords: carbon dioxide, loss of posture, loss of righting reflex

The most common euthanasia method for laboratory rodents in North America is overdose with carbon dioxide gas (CO2). As CO2 concentrations increase, unconsciousness occurs, followed by death. Humans report pain at CO2 concentrations between 32.5-55%. It is unknown if exposure is painful to rodents. A first step in exploring the likelihood of pain is to identify when unconsciousness occurs. Definitions of unconsciousness are highly variable in rodent literature. Two common behavioral assays are loss of posture (LOP, an animal is fully recumbent) and loss of righting reflex (LORR, an animal can be placed on its back without correcting its position). The aim of this study was to compare the concentrations at which LOP and LORR occur during CO2 exposure. Adult, Sprague-Dawley rats (n = 28, both sexes), were exposed to CO2 in a motor-driven rotating cylinder (3 rpm). Treatment order was randomized. LOP and LORR were identified using standard definitions (from review of the literature). CO2 concentrations were measured with a calibrated gas analyzer and concentrations recorded, below, at, and above LOP and LORR. Collected data were analysed with probit regression. Escape/aversion behaviours were documented. The EC95 (effective concentration for 95% of the population) for LOP was 23.1% (95%CI 22.4 to 24.3%), a value significantly lower than LORR EC95: 34.3% (95%CI 33.5 to 35.5%). The behavioral outcome (LOP or LORR) used to identify loss of consciousness is important when attempting to identify the likelihood of pain during exposure to CO2. This has important implications for laboratory rodent welfare.

Herd-level prevalence of bovine leukosis in dairy herds of Alberta

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Keywords: Epidemiology, Bovine Leukemia Virus, Dairy Cattle

Background: Bovine leukosis, caused by the bovine leukemia virus (BLV), is an infectious disease of dairy cattle historically widespread among Alberta dairy herds. Although very few cattle develop the clinical disease, cows infected with BLV still experience several adverse effects on their immune function, reproduction, longevity, and milk production, leading to economic consequences for dairy producers and consumers. It is also a growing public health concern as recent evidence affirms a zoonotic potential for BLV. The Cattle Health Surveillance System (CHeSS) project is aiming to comprehensively control major infectious diseases of cattle including leukosis in Alberta. Up-to-date prevalence of diseases is important to devise appropriate control programs. This study aimed to estimate the herd level prevalence of leukosis in Alberta.

Methods: Bulk tank milk samples were collected twice (December 2021 and April 2022) on all Alberta dairy farms (n=493) and stored at -20 °C. Samples were tested for antibodies against BLV using indirect ELISA (Bovicheck BLV). ELISA results were dichotomized (positive or negative) based on manufacturers' cut-off values. Herd-level prevalence was calculated as proportion of positive samples of total tested samples.

Results: Herd-level prevalence of BLV was estimated at 89.5% (95% CI = 86.4 - 91.9%) and 88.7% (95% CI = 85.6 - 91.2%) in December 2021 and April 2022 timepoint collections, respectively.

Conclusions: These results provide up to date information of the disease frequency that will set the basis for further investigation of within-herd prevalence of BLV and help in devising appropriate disease control strategies in Alberta to control bovine leukosis.

Neonatal calves show a constitutively impaired mucin barrier that leads to severe enterocolitis after *Cryptosporidium parvum* challenge

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Keywords: Cryptosporidium parvum, Enterocolitis, Mucosal Immunity

Innate immunity in neonates is underdeveloped and tolerant compared to adults. Indeed, neonatal calves are more susceptible than older calves to infectious diseases, but the mechanism for neonatal calves' susceptibility to aggravated diarrhea is unclear. We hypothesize that the immaturity of gut defences in neonatal calves makes them particularly susceptible to Cryptosporidium parvum, the main cause of diarrhea in cattle, thereby causing worsening of enterocolitis. We explored the mucosal inflammatory responses and mucin barrier in 2-4-day old Holstein calves experimentally challenged with C. parvum and fed commercial colostrum. The ileum and colon of infected calves showed extensive epithelial erosion, increased leukocyte infiltration, and greater hemorrhaging in the lamina propria. Shot-gun proteomic analysis showed two enriched inflammatory pathways in infected calves: necroptosis and neutrophil extracellular trap formation (NETs). The goblet cells in the colons of infected calves contained less N-acetyl-D-glucosamine and sialic acid glycoproteins, and an increased but aberrant accumulation of mucin was present in the lumen. These results indicated that exaggerated neutrophilic inflammation predisposes neonates exposed to C. parvum to epithelial erosion along the gut and a depleted colonic mucosal barrier. Remarkably, even naïve calves displayed incomplete colonic mucosal barriers, and goblet cells were largely restricted to the crypt base. Thus, neonatal calves are highly susceptible to C. parvum enterocolitis at this early life stage, and this vulnerability is likely attributed to yet underdeveloped intestinal mucosal defenses.

Chronic wasting disease (CWD) vaccine development and production

<u>Byron Kruger</u>, Kevin Low, Hanaa Ahmed Hassan, Dalia Abdelaziz & Hermann M. Schatzl

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Keywords: CWD, Prion, Vaccine

As chronic wasting disease (CWD) continues to ravish the deer populations in North America with no signs of slowing, the need for a vaccine continues to grow. Our previous studies have demonstrated the efficacy of a prion vaccine, which uses specific recombinant prion proteins (PrP) to overcome self-tolerance and to produce self-antibodies that bind to PrP and inhibit the propagation of prions. This study aims to further test the efficacy of such a vaccine in novel mouse models of CWD infection. Sera was obtained from newly developed knock-in mice that express a cervid PrP immunized with recombinant PrP immunogens and CpG oligonucleotide (CpG) as adjuvant. Using end-point dilution ELISA, the antibody titers against PrP were measured and compared to determine which protein candidate produce the highest antibody titer. While all tested immunogens broke PrP tolerance, a mouse-PrP based immunogen produced the highest antibody titers. Epitope mapping was then done to determine the specific epitopes to which the antibodies react. This experiment was conducted using linear peptide epitopes, which limits it to in vitro analyses as reactivity to discontinuous and conformational epitopes were not tested. Nevertheless, this experiment remains an important metric of reactivity. In parallel, we are optimizing an oral nanovaccine that is based on co-encapsulation of recombinant PrP immunogen and adjuvant into biodegradable polylactide-coglycolide (PLGA) nanospheres. This vaccine will be tested in rodent and cervid models, including reindeer. Our vaccination strategy provides promising prospects to prevent the rising cases of chronic wasting disease.

Modelling the effects of climate change on the geographic distribution of *lxodes pacificus* and *Borrelia burgdorferi* in the Pacific Northwest

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Keywords: Ixodes pacificus, Lyme disease, habitat suitability modelling

Lyme disease is the most common vector-borne disease in the United States and Canada. The primary vector for the causative agent of Lyme disease, Borrelia burgdorferi, in the Pacific Northwest is the western black-legged tick, Ixodes pacificus. Using tick presence data from British Columbia and Washington State, we developed distribution maps for I. pacificus. Using B. burgdorferi data, we developed risk maps for Lyme disease, following a literature review on the ecology and epidemiology of Lyme disease in western North America. Species distribution models were created for I. pacificus using multiple modelling methods, including the Maximum Entropy (MaxEnt) algorithm. The models were used to predict current and future geographic distributions of I. pacificus. Future predictions were made for two Representative Concentration Pathways (RCP) emission scales (4.5 and 8.5). Current suitable habitats for I. pacificus are located along the coasts and inland valleys of British Columbia and the Puget Lowlands of Washington state. For future projections, we expect the total area of suitable habitat for I. pacificus to increase along the northern portions of the study area and contract along western Washington, We anticipate that Lyme disease risk will be situated within areas of very high suitable habitat for I. pacificus, but the risk likely remains low given low infection rates among I. pacificus ticks. The creation of up-to-date predictive maps for I. pacificus and B. burgdorferi can aid Lyme disease surveillance efforts and contribute to current literature on the ecology and epidemiology of Lyme disease in western North America.

Characterization of the putative endosomal pore-forming protein APOL7C in classical dendritic cells

Cassandra M. Wood^{1,2} & Johnathan Canton^{2,3}

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Keywords: dendritic cell; endosomal escape; cross-presentation

Dendritic Cells (DCs), a type of immune cell, sample their surroundings by ingesting extracellular material. Typically, the ingested material, consisting largely of macromolecules, is retained in membrane-bound organelles called phagosomes. Phagosomes deliver their macromolecular cargo to lysosomes for digestion into smaller molecules. In some exceptional instances, however, the internalized material remains undigested and is released directly from the phagosome into the cytosol and processed for inflammatory signaling and antigen presentation. Despite the biological significance of macromolecule escape from the phagosome, the mechanism is largely unknown. We identify a novel endosomal protein uniquely expressed in DCs, apolipoprotein L family member 7C (APOL7C). Although its exact function is unclear, evidence suggests recruitment to phagocytic organelles and implicates APOL7C in macromolecule escape. In this study APOL7C is characterized using immunofluorescent assays (IFA) and confocal microscopy. Preliminary results show that APOL7C can be found on the phagosome and colocalizes with LAMP-1, a late endosome marker. NADPH oxidase inhibition (with DPI treatment) results in decreased APOL7C recruitment ($p \le 0.01$), suggesting that APOL7C requires cytosolic acidification for its recruitment. This is consistent with other proteins in the human APOL family. Furthermore, a pH clamp IFA points to an optimal pH of 6.5 ($p \le 1$ 0.05). Together, these findings suggest that at least two independent factors promote recruitment: pH and a specific protein or lipid. Further work will investigate (1) cytosolic pH by ratiometric fluorescence microscopy using the probe SNARF-1, and (2) determine which regions of APOL7C are required for its recruitment using truncated proteins.

UCVM Sure Research Day 2022

Development of infection model for bovine digital dermatitis

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Keywords: Digital Dermatitis, Bovine Foot Health, Treponema

Bovine digital dermatitis (DD) is an infectious polymicrobial disease causing painful ulcerative lesions on the feet of cattle. Experimental DD models in ruminants have been developed to understand the disease but are limited in their reproducibility. Therefore, the development of a more reproducible experimental model is crucial. Our objectives were to compare the use of peeling agents (urea and salicylic acid) with physical abrasion (tungsten disk) used in previous studies to disrupt the skin of calves and induce lesions using macerated DD lesions from naturally infected dairy cattle. To assess our objectives, four feet of 8 Holstein bull calves were enrolled in the study for a total of 32 feet. Front feet were subject to physical abrasions and rear feet to peeling agents before inoculation with sterile broth (mock-inoculum) and macerated DD lesions (inoculum). Plastic patches were glued using superglue and used as a reservoir for peeling agents, mock-inoculum, and inoculum, and all feet were wrapped to maintain a moist and anaerobic environment. After 14 days post-inoculation, no DD lesions were observed due to inadequate adherence of plastic patches to the calves' feet and dry skin. Thus, disruption of the skin barrier was not enough to induce DD lesions; maintaining a wet environment is also critical for lesion development. Further investigations currently being considered for future trials include modifying and troubleshooting plastic patches for better adherence by reducing their size and modifying their position and orientation, as well as applying a layer of silicone over the patch to minimize leakages.

Developing and testing a new bioinformatic pipeline for the analysis of nanopore nemabiome metabarcoding data from parasitic nematode communities

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Keywords: metabarcoding, nanopore, nematodes

Short-read Illumina sequencing targeting the highly variable ITS-2 region of the rDNA cistron is a well-established method in the Gilleard lab for the relative quantitation of nematode species in fecal samples from domestic ruminants. However, this current method is not suitable for rapid and flexible diagnostic use and has some limitation for distinguishing closely related species. Consequently, we are transitioning to nanopore sequencing, targeting the ITS-1/5.8S/ITS-2 region of the rDNA cistron.

We developed a bioinformatic pipeline analyzing nanopore amplicon sequencing data as the Illumina sequencing Dada2 pipeline is not suitable for this purpose. We tested different programs for each step, such as primer removal and filtering, etc., of the data analysis present in the DADA2 pipeline. To validate our pipeline, we generated nanopore sequencing datasets targeting the ITS-1/5.8S/ITS-2 region using parasite populations harvested from UK and Western Canadian sheep flocks, with pre-existing short-read ITS-2 rDNA nemabiome metabarcoding data to compare and validate species assignments and relative quantitation.

For each step of the pipeline, we compared different program determining the most suitable. From this comparison we decided to use Cutadapt for primer removal, Filtlong for read quality filtering, and Minimap2 for species assignment and quantitation. The pipeline produces an excel file indicating the number of reads assigned to each species reference sequences in our ITS-1/5.8S/ITS-2 database as well as the percentage species composition these reads represents.

This analysis pipeline will further support the development of a more rapid and flexible routine diagnosis metabarcoding tool using nanopore long-read sequencing technology.

Tropism of Delmarva (DMV/1639) Infectious Bronchitis Virus (IBV) variant for lymphoid organs of chickens

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Keywords: Infectious bronchitis virus (IBV), lymphoid tissue, chicken

Recently, the infectious bronchitis virus (IBV) Delmarva (DMV)/1639 strain has increased in prevalence across Eastern Canada. Emergence of new variant IBV strains led to outbreaks of IBV infection in vaccinated flocks. A better understanding of host-virus interactions is a prerequisite for development of sustainable control measures. We hypothesized that IBV DMV/1639 replicates within the lymphoid organs of chickens in addition to respiratory, renal, and reproductive tract tissues. The objective was to confirm if IBV targets lymphoid organs leading to pathological changes in these tissues. In this in-vivo study, twenty-one 1-week-old chickens were infected with the IBV DMV/1639 isolate via oculonasal route maintaining 18 uninfected controls. Seven birds from the infected and six birds from the uninfected control groups were euthanized at 3-, 7- and 10-days post-infection to collect swab samples, blood, and tissues samples from the primary and secondary lymphoid organs including the bursa of Fabricius, thymus, cecal tonsils, and spleen. Severe clinical signs were noted in the infected birds. The IBV genome load was guantifiable in all infected swab and tissue samples at all three time points. Infected birds showed low serum antibody response throughout the entire experiment. Results from immunohistochemistry revealed IBV antigens in all infected tissues. IBV-induced histopathological lesions leading to depletion of immune cells were observed in the infected tissues. Comprehensively, our findings indicate IBV DMV/1639 replication in the primary and secondary lymphoid organs and leads to histopathological lesions which may potentially affect the function of these immune organs, leading to poor immune response.

Repeatability of horse body condition scores from photos

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Keywords: feral horses, body condition, photos

Assessing the health of wildlife is challenging when capture is not feasible. This includes cases where animals are monitored using camera-traps or when capture is logistically challenging or prohibited. One metric of health which is routinely measured in animals is body condition - an index of surplus energy or fat reserves. While estimates of body condition often involve handling, in some species such as horses, condition can also be estimated from photos. However, very little attention has been paid to potential biases and limitations arising when using such a non-invasive technique. In this study, I tested for sources of bias and intra- and inter-observer repeatability for body condition scores obtained from photos in feral horses. Six trained individuals scored the condition of the ribs, spine, and hips of 200 adult Sable Island horses on two separate occasions using the online Zooniverse platform. Horses, which were evenly split between males and females and dark and light-coloured individuals, were each assessed using 4 different photos taken from lateral, lateral-anterior, lateral-posterior, and posterior view angles. Mixed linear models were used to understand the effects of observer, body part, sex, coat colour, and view angle on condition scores and their repeatability. All variables considered had an influence on body condition scores and their repeatability, with particularly important differences observed among body parts and observers. In general, scores obtained from ribs were much more repeatable than those from other body parts. These results indicate that obtaining informative body condition scores from photos in horses is possible, but that biases and repeatability are likely to vary substantially among studies.

Structural characterization of bovine mastitis-derived *Streptococcus* spp. biofilms

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Keywords: Streptococcus, biofilm, CLSM

Bovine mastitis is a bacterial infection of the mammary glands and is the disease responsible for the majority of antimicrobial usage among dairy farmers. Streptococcus spp. is among the most frequently isolated genera of bacteria from cases of clinical and subclinical mastitis. Bacterial biofilms produced by mastitis pathogens have demonstrated a higher tolerance to environmental stressors, such as antimicrobials, when compared to their planktonic counterparts. Currently, knowledge is limited on Streptococcus spp. produced biofilms in terms of biovolume, structural features, and antimicrobial resistance. This study aims to use Confocal Scanning Laser Microscopy (CLSM) to validate our previous biofilm growth assay and characterize the biofilm structures of different Streptococcus species. The isolates will be directly grown on microscope slides with 8-well chambers at 37°C for 60 hours. They will be subsequently rinsed with PBS twice and stained with a LIVE/DEAD viability mixture of SYTO-9 (viable cells) and propidium iodide (non-viable cells). Data collected through CLSM will use ZEN 2.3 SP1 and ImageJ Fiji to produce information on morphology, biofilm thickness, and spatial heterogeneity. We hypothesize that Streptococcus uberis will grow the strongest biofilms because this species is well-known to cause persistent mastitis. Confocal images are currently being processed and will later be analyzed in combination with biofilm-associated genes and clinical information. This will provide valuable insight into the mechanisms associated with persistent mastitis and its enhanced resistance to antimicrobial treatment. Further understanding of these mechanisms will contribute to more opportunities for novel treatment development.

Computational analysis to identify mimicry proteins produced by myxozoa in fish hosts

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Keywords: myxozoa, mimicry, computational

Mimicry of host proteins is a mechanism evolved in certain parasites to promote infection and transmission by evading the host's immune system. Myxozoa are a class of parasites from the cnidarian phylum that primarily infect and kill fish hosts. This parasite is prevalent in Alberta waters and is a major issue in the destruction of the ecosystem, fish populations, recreational fishing, and fish consumption. Despite its large negative environmental impacts, the molecular mechanisms involved in myxozoa evasion of the fish immune system are unknown. This research aims to fill in this gap of knowledge to potentially aid in the development of strategies to mitigate the damage caused by this parasite. We hypothesize that like some parasites, myxozoa utilizes molecular mimicry during infection by producing proteins resembling host immune proteins.

We used a computational approach to identify myxozoa mimicry proteins by comparing protein databases of model organisms to fish host proteomes and myxozoa proteomes. Examination of these comparisons lead to the identification of matching proteins in the fish host and myxozoa exclusive from the protein databases of the model organisms. Most of the putative parasitic mimicry proteins were found to contain amino acids sequences of host immune proteins. In addition, we used sequencing and decontamination software to remove contaminating host sequences and assemble new myxozoa genomes to be later used in the computational analysis. Future directions aim to compare parasite mimicry proteins of myxozoa to other parasites to help understand mimicry functionality and identify novel drug targets to remove parasites.

Combining traditional knowledge and scientific research to promote the health of caribou herds and local communities

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Keywords: Barren ground caribou; Health assessment; Traditional Knowledge

The objective of this project was to bridge traditional and scientific knowledge with Ekwò Nàxoèdee K'è (ENK), an Indigenous-led caribou monitoring program. We created a noninvasive health assessment field guide that directs observations to identify abnormalities and will improve understanding of barren-ground caribou health. The guide is split into four broad categories where abnormalities are commonly seen: head, hair and coat, body condition and behaviour, and the lower limbs. Specifically, this guide highlights how to assess lameness and determine the body condition of caribou. Observers in the field then record abnormalities by briefly describing and marking the location of concern on a caribou outline in field booklets. Finally, we tested the guide to the ENK training workshop in June using photos and videos of healthy and unhealthy caribou. Through this, we received valuable feedback and direct insight of Traditional Knowledge. ENK is in the midst of the monitoring season, so results on the efficacy of the guide will not be available for some time. However, based on the interactions with the organization, I hypothesize that the guide will result in more focused observations that will help provide a more well-rounded understanding of the barren-ground caribou health. Through the inclusion of Traditional Knowledge in wildlife health research, a more holistic perspective can develop to guide recommendations to management to advance conservation efforts of Arctic animals.

Quantifying virus neutralizing antibodies of bovine respiratory diseases causing BHV-1 in preconditioned and non-conditioned calves using VNT and ELISA

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Keywords: Neutralizing antibodies, Preconditioning, BHV-1

Bovine Respiratory Disease (BRD) is the leading cause of morbidity and mortality in feedlot cattle during the first 40 days on feed (DOF). Bovine herpesvirus-1 (BHV-1) is commonly associated with BRD. Preconditioning of calves was utilized to mitigate stress and subsequently reduce rates of BRD. Three groups were incorporated into the study: Preconditioned (PC), Auction derived (AD), and Ranch-Sourced (RS) calves. AD calves were obtained from auction while RS and PC calves were sourced from the same ranch. PC calves underwent a preconditioning protocol before arriving on the feedlot, including vaccinations and weaning, RS and AD calves did not. Serum was collected from all groups at various points throughout the study and used in viral neutralization assays as well as enzyme-linked immunosorbent assays (ELISA) to guantify the presence of neutralizing and total antibodies to BHV-1. Neutralizing antibody titers (nAbt) were calculated using the Spearman-Karber method and were compared to ELISA OD values. A positive correlation between nAbt and OD values was predicted. Spearman's rank correlation was computed to assess the relationship between the variables and showed there was a strong positive correlation between the two, p(114) = .75, p=3.2x10-22. Mean PC nAbt at 40 DOF (225.5 +/- 36.1, n=10) was significantly higher than AD 40 DOF (46.1 +/- 22.4, p<0.01, n=9). Mean RS nAbt at 40 DOF (115.5 +/-74.6, n=10) was not significantly different from PC nor AD (p>0.05). Preconditioning calves holds promise as a method of inducing higher neutralizing antibodies against BHV-1 early during the feedlot phase.

Characterizing biofilm formation of bovine mastitis Streptococcus isolates

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Keywords: Biofilm, mastitis, Streptococcus

Streptococcus spp. are a group of bacterial pathogens that form biofilms in the mammary glands of cattle. Bacterial colonization of these tissues results in bovine mastitis, a disease which reduces both the quality and quantity of milk production in Canadian dairy herds.

The mechanism by which *Streptococcus* establishes biofilms in the bovine mammary gland is poorly understood. Consequently, we investigated 244 *Streptococcus* isolates from the *Mastitis Pathogen Culture Collection* to determine their abilities to form biofilms.

The extent of biofilm formation was determined using a standard microtiter plate protocol: *Streptococcus* isolates were cultured for 24 hours at 37°C in Brain Heart Infusion broth and 0.25% glucose. Cultures were diluted ($OD_{600} = 0.1$), pipetted into 96-well polystyrene microtiter plates, and incubated for 48 hours at 37°C. Microtiter plates were rinsed with phosphate buffered saline (PBS) to remove planktonic cells. Wells were treated with crystal violet, rinsed with PBS, de-stained with 100% ethanol, and the extent of biofilm formation was determined spectrophotometrically (A_{560}). The biofilm forming capability of isolates was defined as strong when $A_{560} \ge 0.550$, moderate when $A_{560} = 0.200 - 0.549$, or weak when $A_{560} = 0.080 - 0.199$.

Of the isolates capable of forming a biofilm on polystyrene, *Streptococcus uberis* appears to form the most extensive biofilms while *Streptococcus dysgalactiae* appears to form the least extensive biofilms. Additional work will be required to determine whether the ability of *Streptococcus* isolates to form biofilms on polystyrene is a good indicator of virulence in the bovine mammary gland.

Cow- and calf-level factors associated with nursing behaviours in beef cattle during the 24-hours following an assisted calving

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Keywords: cow-calf, dystocia, maternal-neonatal bonding

Nursing behaviour is essential following parturition to ensure transfer of passive immunity and development of a strong cow-calf bond. One factor that could modify nursing behaviour is dystocia (i.e., a difficult or prolonged calving that may require assistance); however, little work has documented this effect. The objective was to identify cow- and calf-level factors associated with nursing behaviours in beef cattle for 24hr following an assisted calving. Cows and calves that required an assisted calving were enrolled and video recorded for 24hr to score maternal behaviours. A preliminary analysis (n = 15 pairs) was performed where cow- and calf-level predictors of nursing behaviour (i.e., calf sex, cow parity, calving difficulty, calf presentation, time of calving, measures of calf vigour, and meconium staining) were offered to multivariable general linear models. On average, the first nursing bout occurred at 11.4 + 8.97hr. Calves spent a total of 43.4 + 25.4min nursing and had 24.9 + 16.3 total nursing bouts. Calves nursed earlier if they were born to a multiparous dam compared to a primiparous dam (4.67 + 1.88hr vs. 15.3 + 1.81hr; P = 0.002). Calves with meconium staining nursed earlier (5.21 + 2.17hr vs. 14.8 + 1.69hr; P = 0.008) and had more total nursing bouts (35.3 + 6.10 bouts vs. 17.6 + 4.68 bouts; P = 0.05) than calves with no meconium staining. Results from this study will clarify nursing behaviours after an assisted calving and identify factors for informing management decisions that support cow and calf health and welfare.

Characterizing the functions of *Tequintavirus* AKFV33's lateral tail fibers and receptor binding proteins during phage adsorption process to Shiga toxin-producing *Escherichia coli* O157

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Keywords: lytic phage, phage adsorption, functions of AKFV33's binding proteins

Shiga toxin-producing Escherichia coli (STEC) is the cause of many of North America's foodborne infections for humans and feedlot animals. Lytic phages are viruses that can guickly kill specific bacterial pathogens without affecting commensal bacteria. Antimicrobial resistance is a problem: thus, phage therapy is a potential alternative. AKFV33 is a *Tequintavirus* highly lytic and specific to common O157:H7 strains of STEC. The infection process starts with AKFV33's lateral tail fibers (Ltfs) reversibly binding to the O antigen of the host lipopolysaccharide, then followed by the phage's receptor binding protein (RBP) irreversibly binding to the host's outer membrane proteins. The detailed mechanism of how AKFV33 executes phage adsorption has not been extensively studied. For example, does co-recognition between RBP and Ltfs occur? This project aims to verify the functions of AKFV33 Ltfs (Ltfa, LtfB, and orf136) and RBP in phage adsorption. The target genes were overexpressed. Through colony PCR, sequencing, and plasmid extraction, overexpression plasmids were constructed. They were then expressed in *Escherichia coli* BL21 *i*. The target proteins will be purified, and their function will be evaluated by phage adsorption inhibition assay. This study is essential since this interaction can be used to kill STEC in food sources, infected people, and animals. This study can pave the way for future studies on the functions of Ltfs and RBP of other phages.

Investigating the seroprevalence of the causative agents of Equine Protozoal Myeloencephalitis in Alberta

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Keywords: Equine Protozoal Myeloencephalitis, Horse, Neurological

Equine Protozoal Myeloencephalitis (EPM) is caused by Sarcocystis neurona and Neospora hughesi. There are no studies investigating the seroprevalence of these protozoa in Albertan horses. The geographic range of the definitive host for S. neurona excludes Alberta meaning the expected seroprevalence is low. Anecdotal reports of horses showing clinical signs consistent with EPM warrants investigation into its presence in Alberta. The objective was to investigate the seroprevalence of S. neurona and N. hughesi in Albertan horses by testing serum antibody levels. In this cross-sectional study, 61 banked serum samples were tested by indirect fluorescent antibody test (IFAT) to determine their titres for S. neurona and N. hughesi antibodies. Sample size was calculated for an expected prevalence of 2% and later expanded to all samples whose owners indicated the horse had always resided in Alberta. Testing demonstrated that 32.8% and 36.1% of horses had titres indicating exposure to S. neurona and *N. hughesi* respectively. Surface antigen ELISA ruled out cross reactivity to other protozoa found in Alberta. The sensitivity and specificity of the IFAT using serum from horses showing neurological signs is 83.3% and 96.9% respectively. The sensitivity and specificity of the IFAT using serum from apparently healthy horses are undocumented, so the interpretation of titre values is complicated. The results indicate that seroprevalence for the causative agents of EPM is higher than expected. Considering this, Albertan veterinarians should include EPM when developing differential diagnosis lists for horses with neurological signs and test cerebral spinal fluid to serum ratio by IFAT.

Hunters' willingness to change their hunting practices in response to zoonotic disease

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Keywords: Hunting practices, brucellosis, tuberculosis, behaviour change, bison

The North American Wildlife Conservation Model (NAM), which began in the early 20th century, posits that wildlife are held in the public trust and hunting is a positive force for conservation. We investigated potential influences on Alberta hunters' stated willingness to change (WTC) their hunting practices in response to a hypothetical case of a zoonotic disease identified in a wildlife species they regularly hunt. We anticipated that significant predictors would include demographics (e.g., age, education), perception of risk, and knowledge of zoonotic disease. A link to a questionnaire was distributed in a seasonal newsletter update sent to more than 100,000 licensed hunters by Alberta Environment and Parks, exploring Alberta hunters' opinions on potential ways to manage the health of wood bison in Wood Buffalo National Park; 139 useable responses were evaluated for this study. Hunters were asked how close an animal infected with tuberculosis (TB) or brucellosis could be before they would change their hunting practices (e.g., regular hunting area). Risk awareness was calculated as an aggregated score from questions addressing TB and brucellosis impact on health and economic livelihood; knowledge was similarly based on questions evaluating knowledge of TB and brucellosis. Preliminary results of multiple variable linear regression models show significant predictors of WTC include knowledge of zoonotic disease and perception of risk to hunting opportunities and economic livelihood. Our research provides important findings addressing potential policy support that engages hunters in wildlife conservation and their willingness to engage in the particular supportive behaviours.

Using a One Health approach to identify drivers of and barriers to improving antimicrobial stewardship across various professions in Canada

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Keywords: Antimicrobial Resistance, One Health, Qualitative

Antimicrobial resistance (AMR) represents an ongoing global health threat that highlights the need for effective and practical mitigation strategies. As antimicrobial use is a major driver of AMR development, antimicrobial stewardship (AMS) is vital to preserve antimicrobial efficacy. Although Canadian AMS programs exist, AMR prevalence continues to increase. Therefore, we must improve understanding of AMS drivers and barriers with a One Health approach involving stakeholders from human, animal, and environmental sectors. A qualitative and quantitative questionnaire was developed and conducted with various Canadian professionals (n = 81)participating in the 2021 One Health Antimicrobial Stewardship Conference and their networks to understand AMS perceptions, motivators, and barriers and analyzed using template analysis (n = 61 veterinary sector, n = 15 human, n = 2 agricultural, n = 2 human and veterinary, n = 1 undefined). Regarding AMS barriers, common themes were access to knowledge, timely diagnostics, prescribing guidelines, agreement among experts, economic factors, as well as lack of motivation to change, sustained funding and skilled personnel. Regarding AMS responsibility, a common theme was shared responsibility, in addition to antimicrobial prescribers and users, government, industry, professional associations, researchers, diagnosticians, and educators. Some participants placed blame for AMR barriers or lack of progress on others within their profession, other sectors, existing industry structures, and cultural norms. Therefore, personal AMS responsibility should be leveraged to advance initiatives. Without an understanding of cross-cutting and sector-specific barriers to AMS. effective change is unlikely, highlighting the importance of social science in AMR research and mitigation.

Investigating the signaling requirement to maintain pig iPS cells in feeder-free conditions

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Keywords: Induced pluripotent stem cell, self-renewal, media

Introduction: Induced pluripotent stem (iPS) cells can proliferate indefinitely *in vitro* while retaining the ability to differentiate into any cell in the body, creating an essentially unlimited resource for cell-based therapies. Due to the similarity of organ size, physiology, and anatomy, pigs pose a prime model organism for studying the safety and efficacy of cell-based therapies prior to human use. This process has previously used complex culture conditions relying on mouse embryonic fibroblast (MEF) feeders to supplement custom media. Our goal was to simplify culture conditions of feeder-free pig iPS cells to allow for effective maintenance of pluripotent cell self-renewal.

Materials & Methods: We altered the presence of ActivinA, bovine serum albumin (BSA), IWR1, and CHIR99201. To monitor the effects of media alterations, we used qPCR, cell morphology, flow cytometry, differentiation, and immunostaining.

Results: Following the media modifications, we noted that removal of ActivinA resulted in high rates of cell death. BSA was not required for cell survival, although small amounts helped maintain cell morphology. IWR1 was necessary for maintenance of iPS cells but had negative effects when present in combination with CHIR99201 which was not necessary for pluripotency maintenance.

Conclusions: We concluded our work with a simplified media recipe that allowed us to maintain the pig iPS cells in a more chemically defined state without the need for MEF feeder cells.

Endogenous cathelicidin regulates neutrophil viability during homeostasis

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Keywords: cathelicidin, neutrophils, viability

Neutrophils eliminate pathogens and dead cells via inflammatory functions, while their timely clearance via constitutive apoptosis maintains homeostatic balance. Certain lytic modes of neutrophil death (e.g., necrosis) can exacerbate inflammation and tissue damage. How neutrophils regulate their own death to favor clearance actions and control inflammation remains elusive. It has been shown that mice deficient in cathelicidin (cathelicidin knock-out; Camp^{-/-})—an antimicrobial peptide abundantly stored in neutrophil granules have decreased leukocyte infiltration in the colon during inflammation, and that Camp is upregulated during early neutrophil maturation. To determine if endogenous cathelicidin modulates neutrophil life and predisposes death mechanisms, we examined the viability of wildtype (*Camp*^{+/+}) and *Camp*^{-/-} mouse bone marrow (BM)-derived neutrophils, and their genetic profiling in the colon under homeostasis. We determined via RNAseq that the expression of leukocyte survival-related genes was not different between colons of both mice strains. However, Camp ^{/-} BM neutrophils, compared to Camp^{+/+} counterparts, displayed spontaneously decreased viability (8 h, 37°C in serum-supplemented media) as determined by flow cytometry detection of cell membrane structural changes in apoptotic and necrotic neutrophils with annexin V (AV) and propidium iodide (PI) fluorescent dyes. Camp-/-BM neutrophils showed a predisposition for early apoptosis (4 h) and a later tendency for necrosis (8 h). Premature death of Camp-^{/-} BM neutrophils, largely caused by necrosis, was also detected by IncuCyte in live cells stained with AV / PI. Thus, endogenous cathelicidin could regulate neutrophil viability, predisposing their survival during homeostasis by preventing necrotic death.

Constructing a real-time live cell WNT signaling reporter in pluripotent stem cells

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Keywords: WNT signalling, pluripotent stem cells

<u>Introduction:</u> The WNT signalling pathway is critical for human development and tissue homeostasis. As such, dysregulation is associated with disorders and cancer, but how this occurs is not fully understood. Thus, this project aims to construct a faithful reporter to elucidate the pattern of WNT signalling in pluripotent stem cells during maintenance and differentiation.

<u>Materials and methods:</u> We subcloned a NanoLuc luciferase (encodes a luminescent protein) under the control of a M50 Super 8X TOPFLash response element (contains 7 TCF/Lef binding sites and a mini-promoter). This construct was integrated into H1 and H9 human embryonic stem cells (hESCs) via a piggyBac transposon system. We tested how this reporter responded to the treatment of CHIR99021 (a WNT signalling pathway activator) in hESCs with various durations and concentrations.

<u>Results:</u> In both H1 and H9 hESCs, the undifferentiated samples (no CHIR99021) released the least luminescence, samples differentiated for 4 hours (3uM of CHIR99021) released intermediate luminescence, and samples differentiated for 24 hours (3uM of CHIR99021) released the highest luminescence. During the dosage test, reporter activation increased as CHIR990021 concentration increased, with peak luminescence at 5uM. However, at concentrations of 10uM and above, cell death occurred, impeding reporter activity.

<u>Conclusion</u>: In this project, the NanoLuc reporter provides high utility, allowing for robust luminescence activation over a ranger of CHIR99021 concentrations and durations. In the future, tests should be done to optimize this reporter. This may include altering the response element by changing the number of binding sites or the size of space sequences.

The effects of chronic PD treatment on a juvenile and adult mouse model with Fragile X Syndrome

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Keywords: Fragile X Syndrome, anxiety-like behaviors, PD drug

Fragile X Syndrome (FXS) is a neurodevelopmental condition associated with intellectual and behavioral disabilities such as hyperactivity and anxiety-related symptoms, and is the leading monogenic cause of autism. It is caused by mutations in the fmr1 gene. The ERK signaling pathway plays a crucial role in brain development; upregulated ERK signaling is observed in FXS patients. A highly selective drug called PD was found to inhibit ERK activation in the brain and reverse impairments in the adult fmr1 knockout (KO) mouse model of FXS with acute treatment. Here, we chronically treated fmr1 KO mice with PD and tested at both adolescent and adult stages to determine the influence of developmental stages on treatment effect. After weaning, mice were given either vehicle (control) or PD in drinking water. The amount of water intake and individual body weights were regularly recorded. The mice then underwent behavioural tests at both postnatal day 40 (P40) and P70. The light/dark test assesses the duality of avoidance of light areas and the innate desire to explore novel areas. Our results showed that there is no significant difference between the treatments at P40 and P70. Further, the results of the open field test, which measures exploratory behavior while also gauging locomotion, shows that there is a significant difference related to sex of the mice at P70. Analyzing the effects of chronic PD treatment at various developmental stages will help to identify whether ERK pathway inhibitors can be used as effective therapeutic approaches for FXS.

Evaluating the benefits of Traditional Ecological Knowledge (TEK) collection from locally led short interviews in the Ulukhaktok community to build upon classical western approaches to Arctic wildlife-assessments

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Keywords: Traditional Ecological Knowledge, wildlife health assessment, Community-based monitoring

Muskoxen and Dolphin Union (DU) caribou are ecologically important and are central to the food sovereignty and culture of the Inuit. The recent population declines in DU caribou and muskoxen on Victoria Island has put emphasis on the importance of population monitoring to inform management decisions. However, conducting research in the remote Arctic is challenging due to the extreme costs of resources, the lack of infrastructures, and logistical challenges. Moreover, locals are seldom given the opportunity for direct involvement in wildlife monitoring. In my summer project, I explored how Traditional Ecological Knowledge (TEK) could complement Western approaches in monitoring muskox and caribou populations. I transcribed and analyzed eleven short harvester interviews conducted in the community of Ulukhaktok in 2020. Through GIS mapping, semiguantitative statistics and gualitative thematic content analysis, I extracted information on the species' seasonal distribution, population trends, population structure, and population health. Additional observations (i.e., predators, individual health) were also beneficial as they offered insight on potential concerns for the communities and the species of interest. Important findings from those interviews include (i) health status of muskoxen subpopulations differed, (ii) DU caribou groups remained on Victoria Island during the winter instead of moving south to the mainland, and (iii) perceived drivers/themes of caribou and muskoxen populations were established such as health, environment, and human pressure. This work highlights how invaluable TEK of the land and of arctic species held in Indigenous communities could enhance classical wildlife monitoring approaches, bring novel perspectives, and improve data collection from isolated regions.

What are our gaps in knowledge regarding Indigenous perspectives on humananimal relationships, western Veterinary Medicine, and traditional Indigenous animal care/medicine and how may these concepts act as barriers for Indigenous students to apply for fields in veterinary medicine?

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Keywords: human-animal relationships, ethnoveterinary, Indigenous Peoples

The ethnic diversity in the Canadian veterinary profession remains limited and this may hinder the profession's ability to serve clients from diverse cultural backgrounds and understand diverse community needs. This scoping review aims to identify and understand gaps in knowledge regarding traditional Indigenous animal care practices, perceptions surrounding Western veterinary medicine, animal health care needs, and barriers faced by Indigenous Peoples regarding veterinary education. This research systematically reviewed peer-reviewed articles and several grey article sources (podcasts, blogs, etc). Specific search strings and data extraction protocols yielded 132 articles and book chapters for initial abstract screening, of which 93 were fully reviewed. 54 final literature pieces with relevant information on the concepts of this project were included. Three grey article sources were also identified and included. Our results indicate that literature on Indigenous human-animal relationships is generally available, but the literature rarely addressed Indigenous perceptions of Western veterinary medicine and traditional ethnoveterinary practices. Furthermore, information on the barriers faced by Indigenous students interested in veterinary medicine was scarce, but similar difficulties were identified in articles focusing on medical admissions. This work will improve our understanding of Indigenous relationships with animals and perceptions of Western animal medicine, which could help create a bridge between Indigenous students and the veterinary profession. Furthermore, improving our understanding of the relationships that various cultures have with animals could help improve the cultural/racial representation of veterinarians in practice and our ability to serve diverse clients and better address animal health needs.

The impact of the experimental route of challenge on the pathogenesis of the Canadian Delmarva (DMV/1639) Infectious Bronchitis Virus (IBV) infection in laying hens

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Keywords: Infectious bronchitis virus (IBV); Delmarva (DMV)/1639 Strain; Oculo-nasal route of infection; Intratracheal route of infection; Chicken

Infectious bronchitis virus (IBV) impacts the poultry industry worldwide. Recently, the Delmarva (DMV)/1639 IBV variant was isolated from Canadian laying hens with egg production problems. Previous studies exploring the pathogenicity of different IBVs have varied in their route of experimental challenge. We hypothesized that the intratracheal route of infection would increase the pathogenicity of the Canadian IBV DMV/1639 in laying chickens. The objective was to investigate the impact of the route of challenge on the pathogenesis of this virus in laying hens. Fifteen specific-pathogen-free layers were divided into three groups: oculo-nasal challenged (ON), combination of oculo-nasal and intratracheal challenged (ON/IT), and mock-infected control. The groups were observed for 12 days post-infection (dpi), at which point they were euthanized. Both IBV-infected groups displayed mild respiratory distress. The egg production of the ON/IT group started to decline at 4 dpi, compared to 9 dpi in the ON group. A significant decrease in mean body weight and oviduct length at 12 dpi was only detected in the ON group when compared to the control group. Necropsy revealed regressed ovaries, ruptured or hemorrhagic ova, and atrophied oviducts in both challenged groups. The IBV genome load in various tissues, histopathological scores, and humoral antibody responses were comparable between challenged groups. Overall, the virus replicated efficiently in the tissues of both infected groups. However, the ON/IT group showed a drop in egg production earlier than the ON group. We believe that these results will enable more informed decisions on the experimental route of IBV challenge.

Antimicrobial activity of bacteriophage-derived endolysins on *Mannheimia haemolytica* strains 587 and 330

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Keywords: Endolysin, HEWL antimicrobial activity, Mannheimia haemolytica

Mannheimia haemolytica (MH) is a major cause for bovine respiratory disease, creating significant economic losses in Canadian beef and dairy industries. The use of bacteriophagederived endolysins to treat gram-positive bacteria has been successful, however the treatment of gram-negative bacteria such as MH is challenging due to its additional outer membrane. We hypothesized that applying bacteriophage-derived endolysins and membrane-destabilizing factors to MH cells will inhibit bacterial growth. Endolysins 3-2 (pBAD-185) and P-2 (pBAD-PCNP-L-185), previously cloned in pBAD plasmid and expressed in *E.coli* BL21, were expressed and confirmed by SDS-PAGE. Antimicrobial assay was performed to test antibacterial activity of purified endolysins against MH strains MH330 and MH587. This was guantified using the formula $\log_{10}(N_0/N_i)$; N₀ represents the number of untreated cells (MH cells diluted with PBS), and N_i represents the number of residual cells after treatment with endolysins and HEPES or EDTA with HEWL as positive control. 3-2 and P-2 were successful in inhibiting the growth of MH587 and MH330 cells when combined with EDTA (membrane-destabilizing agent) showing no growth of MH cells. Both endolysins were less successful when combined with HEPES (buffer used to stabilize pH). 3-2 showed positive log values when treating both MH strains, and P-2 showed positive log values when treating MH330 cells and negative values when treating MH330 cells. The combined use of endolysins and a membrane-destabilizing agent has shown to be capable of inhibiting the growth of MH587 and MH330 cells, whereas the use of endolysins without the agent has shown inconsistent and less successful results.

Support for data management and quality assurance/quality control toward public release of health-related water quality data

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Keywords: Water, quality, data

The management of water data, and its quality assurance and control are necessary components of reporting and surveillance within the public health system. In line with the Government of Alberta's Open Government Policy Initiative, Alberta water quality, in terms of drinking water, beaches, pools and hot pools, are a critical data source that should be publicly accessible and shared across multiple jurisdictions. Open-source water quality data is important for protecting the health of Albertans, shaping public health guidelines and policies, and providing transparency to the public. However, the public distribution of water quality data must overcome obstacles, such as, the concerns of a breach of privacy with the release of personal and well location data for individuals who submit water samples. The goal of this study is to; a) conduct interviews with key stakeholders to capture and characterize their needs for access to data and surveillance tools; and b) survey stakeholders and the public to understand their perceptions around open-source water quality data.

Cholesterol overload of neuronal cells interferes with de novo prion infection

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Keywords: prion, cholesterol, prion infection

Prions are fatal, infectious, and currently incurable pathogens that cause neurodegenerative diseases in humans and animals. They form upon the misfolding of the cellular prion protein PrP^C into its infectious isoform PrP^{Sc} and cause widespread neuronal death upon accumulation in the nervous system. Studies suggest that cholesterol-rich sites in the membrane are favourable regions for PrP^{Sc} conversion, supported by findings showing that reducing membrane cholesterol interferes with prion propagation. Interestingly, prion-infected neuronal cells showcase elevated cholesterol synthesis. This project aims to determine whether elevated cholesterol induced by prion infection acts as a positive feedback mechanism to further aggravate prion propagation. Therefore, Cad5 and N2a mouse neuronal cells were infected with 0.2% prion-infected mouse brain homogenate and exposed to media containing 300 ug/mL of cholesterol either pre, post, or pre and post infection. One group of cells was infected with prions but not exposed to cholesterol (positive control), and another group was neither exposed to prions nor cholesterol (negative control). Cells were then continuously passaged, and PrP^{sc} content indicative of prion propagation was analyzed from several passages using Western Blot. Our preliminary results showcase that Cad5 and N2a cells overloaded with cholesterol after prion infection did not generate PrPSc whereas positive control cells efficiently propagated PrPSc. Cad5 cells exposed to cholesterol before and after infection also contained no PrP^{Sc}. These data suggest that elevated cholesterol synthesis observed in cells upon prion infection may serve as a mechanism to rescue neuronal cells from PrP^{Sc} conversion.

ERK activation during postnatal development in a mouse model of Fragile X Syndrome and wildtype mice

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Keywords: Western blot, PD, FXS

Fragile X Syndrome (FXS) is associated with dysregulated mRNA translation in the brain leading to phenotypes including cognitive impairment, hyperactivity, anxiety, and seizures. FXS is caused by a mutation in the fragile X messenger ribonucleoprotein1 (*fmr1*). Extracellular signal-related kinases (ERK) are overexpressed in FXS, disrupting normal mRNA translation. The result of the kinase signalling cascade causes phosphorylation or activation of ERK itself and downstream effectors such as ribosome protein S6 (pS6). Adjusting the levels of phosphorylated ERK (pERK) could potentially correct the dysregulated mRNA translation and associated phenotypic abnormalities in FXS. The first part of my project explores the efficacy of the PD 0325901 drug, an inhibitor of the ERK pathway, in the cortex of the *fmr1* knockout mouse model of FXS using western blot. The expression levels of ERK, pERK, S6 and pS6 were semi-quantified. The second part of my project was to determine pERK, ERK, S6 and pS6 expression in neonatal, juvenile and adult mice in the cortex, of male and female FXS and wildtype (WT) mice, to determine the relative expression in different stages of development.

Currently, results are being collected for both projects. The first project will determine the efficacy of the PD drug on ERK and S6 activation levels. The second project will help identify the relative activation levels of ERK and S6 in FXS and WT mice in different developmental stages. These results could provide more understanding of future therapeutic targets and the developmental stages of potentially using ERK pathway inhibitors to treat FXS.

A molecular genetic investigation into the origins of anthelmintic drug resistance in *Ancylostoma caninum* in pet dogs across the USA

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Keywords: Ancylostoma caninum, amplicon sequencing, genetic diversity

The canine hookworm, Ancylostoma caninum, is a highly pathogenic and prevalent intestinal nematode of domestic dogs. Our previous work revealed it is resistant to multiple drug classes in kenneled greyhounds across the USA and that the benzimidazole resistance is associated with mutations at codons 134 and 167 of the isotype-1 β-tubulin gene (encoding the drug target). Both these mutations are now widespread in A. caninum, not only from greyhounds, but also from pet dogs in the USA. We have hypothesized that resistance originally emerged due to intense drug selection in kenneled greyhounds and subsequently spread to pet dogs via environmental contamination by retired greyhounds rehomed across the USA. This summer project tests the specific hypothesis that there has been more intense selection at the A. *caninum* isotype-1 β-tubulin locus in greyhounds than in pet dogs. Deep amplicon sequencing data is already available for the isotype-1 β -tubulin locus, and we have undertaken additional deep amplicon sequencing of the cox-1 and nad-1 mitochondrial genes to use as neutral markers to assess the overall genetic diversity of the parasite. Primers were designed to variable regions of the cox-1 and nad-1 genes and PCR amplicons sequenced at depth from 120 and 50 A. caninum samples from pet dogs and greyhounds, respectively. We will present a genetic analysis of the sequence data and test the prediction that the isotype-1 β -tubulin genetic diversity is lower in A. caninum from greyhounds than pet dogs, but that no such difference occurs for the neutral mitochondrial markers.

Testing a live-attenuated vaccine against Johne's disease in dairy cattle

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Keywords: Johne's disease, live-attenuated vaccine, dairy calves

Johne's disease is a chronic enteritis in ruminants caused by *Mycobacterium avium* subsp. paratuberculosis (MAP). Commercial Johne's disease vaccines are ineffective at preventing the spread of MAP, and none are approved in Canada due to cross-reactivity with tuberculosis diagnostics. The ideal vaccine should prevent MAP shedding and disease spread. For this trial, 24 dairy calves were collected from farms with low JD prevalence and randomly assigned into four groups of six calves. One group was an uninfected and unvaccinated control. Two groups were orally inoculated at two weeks of age with 10⁹ CFU of live-attenuated vaccine, with a challenge in one group at five weeks of age with 2x10⁹ CFU of the wild-type MAP strain. The last group was unvaccinated and orally challenged with the wild-type strain at five weeks of age. Fecal and blood samples were collected from groups every two weeks, with tissue sample collection occurring at 18 weeks of age. Blood was used for IGRA assay to evaluate a cellmediated immune response to MAP infection. Fecal samples were cultured, followed by DNA extraction and qPCR to evaluate MAP shedding in feces. Initial qPCR analysis on samples taken before inoculation confirmed that animals were from JD negative farms, while gPCR results on samples taken 12 hours after inoculation in infected groups confirmed passive shedding of MAP in feces. Further analysis should ideally indicate a decreased MAP level in vaccinated calves. Quantifying MAP in feces and tissue samples is a good indication of protection provided by a live-attenuated vaccine.

Highly pathogenic avian influenza surveillance

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Keywords: avian infleunza, outbreak, surveillance

Avian infleunza normally circulates within wild populations, but it tends to only result in mild levels of sickness in wild birds. However, the 2022 HPAI outbreak within North America has not only threatened the health of domestic poultry operations across Alberta but has also resulted in wild bird populations experiencing serious health concerns. Furthermore, mammals, such as skunks and other bird species (eagles, hawks) have been more readily infected in comparison to previous outbreaks. Consequently, a surveillance project was created to gain more information regarding the unique characteristics of this outbreak, such as how HPAI is spreading within farms and the wild bird-domestic bird interface. A literature review was conducted to determine the information that is currently known regarding the epidemiology, transmission, risk factors, symptoms, and existing suggested biosecurity protocols of HPAI. Following the literature review, a surveillance questionnaire was created within Qualtrics and will be sent out to poultry facilities across Alberta to gain a better understanding of the specific components of each operation (e.g., biosecurity protocols, waterfowl contact, demographics, feed, and equipment). Following data collection from the questionnaire, data will be analyzed to determine the similarities and differences between this outbreak and previous outbreaks. The results of this surveillance project will provide data for identifying the unique factors of the 2022 HPAI outbreak which has resulted in differences in wild bird infection level and spread. This will provide more information for poultry operations within Alberta and North America regarding how to prevent and manage future HPAI outbreaks.

Anxiety during postnatal development in Fragile X mouse model

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Keywords: Fragile X Syndrome, Anxiety, Behaviour Test

Fragile X Syndrome (FXS) is a congenital disorder caused by the inactivation of the Fragile X Messenger Ribonucleoprotein 1 (FMR1) gene found on the X chromosome. FXS is associated with autism and pronounced in males compared to females due to the presence of the single X chromosome. Individuals with FXS experience heightened anxiety-related symptoms. However, the relationship between postnatal development and anxiety in individuals with FXS remains poorly understood. To explore the relationship between anxiety and postnatal development, we used the FRM1 knockout (KO) mouse model of FXS (N=25), and the wildtype as a control (N=29), to study how anxiety-like behaviour alters at postnatal day 40 (P40) and day 70 (P70). The experiment was conducted using the Light and Dark (LD) test, followed by the Open Field Test (OFT) and freezing test. The LD test measures anxiety-like behaviour in mice based on their natural preference for dark area, and their tendency to explore a new place. The OFT was designed to assess gross motor activity and anxiety-related behaviour in mice. Finally, the freezing test via fox urine was used to measure innate fear response (freezing) to predator's smell. The results were quantified via two-way ANOVA. Our results showed that KO mice displayed increased anxiety-like behaviours when compared to controls, which were more pronounced at P70. Additionally, we observed that females displayed less anxiety-like behaviours than males did at both developmental stages. Overall, during adulthood, male FXS mice displayed increased anxiety to behaviour test when compared to wildtype and female mice.

Tick surveillance globally: A scoping review

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Keywords: Ticks, Surveillance, Pathogens

Ticks sit at the interface of human, animal, and environmental health. Furthermore, ticks are a reservoir for bacteria, viruses, and parasites causative agents of diseases such as Lyme disease and Rocky Mountain spotted fever. Climate change is increasing and changing the range of many tick species, allowing ticks to occupy new ecosystems and exposing previously unexposed hosts to the many pathogens that ticks carry. This puts tick surveillance in a pivotal role, as knowing the distribution and range of ticks, and what pathogens they are carrying becomes integral to the prevention of disease. We conducted this scoping review following PRISMA guidelines, using MEDLINE, CAB Abstracts, BIOSIS Previews, and the Web of Science Core Collection databases within the Covidence platform. Two reviewers, blinded to each other, performed titles and abstract screening which eliminated 1868 articles; full text reviews eliminated 400 articles leaving 220 articles for extraction. Only English articles published after 1960 that referenced a surveillance system including ticks, not just tick-borne diseases, conducted for a minimum of two years were included. Data collected includes the following: the place of surveillance, surveillance methodology, study population, the tick species under surveillance, number of ticks collected, and any tick-borne disease studied. This data is being pulled to determine trends in surveillance techniques in different areas. This will allow us to identify the focus of tick-surveillance occurring globally, as well as to identify knowledge gaps for further exploration.

Therapeutic evaluation of cathelicidin derivatives in Citrobacter rodentium colitis

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Keywords: cathelicidin, colitis, inflammation

Small host defence peptides, initially postulated as microbicidal, have shown abilities to modulate inflammatory and immune responses. Innate defence regulator (IDR-) 1018, a synthetic peptide derived from bovine neutrophil bactenecin, increases chemokine synthesis, and polarises cellular differentiation in leukocytes while reducing pro-inflammatory characteristics. IDR-1018 enhances wound healing in mice and protects against Staphylococcus aureus, multidrug resistant Mycobacterium tuberculosis, herpes virus, and inflammatory disorders, including cerebral malaria and neuronal damage in a pre-term birth model. However, whether IDR-1018 suppresses inflammation or increases antibacterial defenses in infectious colitis is unknown. In this study, mice were orally challenged by Citrobacter rodentium—a model for attaching/effacing Escherichia coli—and systemically administered IDR-1018 (intraperitoneal at 1-, 4- and 6-days post challenge). Challenged mice that received IDR-1018 had higher faecal C. rodentium shedding (CFU counts) than mice that did not receive peptide injections. Lipocalin, a neutrophil gelatinase- associated protein, marker of intestinal inflammation, guantified with the use of ELISA, was seen to be increased in feces at 8 days post challenge in all infected mice, but was higher in those treated with IDR-1018. Microscopically, infected mice showed leukocyte infiltration in lamina propria with hemorrhage and epithelial erosion, but those hallmarks were observed to be more prominent in IDR-1018 group. The cathelicidin derivative (IDR-1018) did not mitigate *Citrobacter rodentium* colitis. Other administration routes, more applicable to the type of challenge (e.g., oral) could reveal the immunomodulatory effects observed in other inflammation models.

Identifying potential microRNAs targeting the avian corona virus, infectious bronchitis virus

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Keywords: Infectious Bronchitis Virus (IBV); microRNA; Viral Control

Avian Infectious Bronchitis Virus (IBV) is a highly infectious Gammacoronavirus causing respiratory infection in chickens leading to significant economic losses for the poultry industry. Like other positive-sense single-stranded RNA viruses, IBV is characterized by high mutation and recombination rates, diminishing the effectiveness of vaccines and by extension, the control of IBV infection. One way to develop novel control strategies is to study the role of microRNAs in the context of IBV infection. MicroRNAs are small non-coding RNAs that regulate gene expression of complementary messenger RNAs by base pairing. This results in gene silencing through translational repression or target degradation. In this study, we **aimed** to identify the microRNAs targeting 3 different IBV serotypes (DMV, 4/91 and MASS) endemic in Canada through an *in-silico* analysis. We *hypothesized* that specific microRNAs can modulate viral replication upon IBV infection in chickens. To this end, we retrieved all the Gallus gallus mature microRNA sequences and the IBV viral genomes from miRbase and GenBank, respectively. Using our target prediction tools, mirDB, miRanda, and RNAhybrid, we identified a total of 198 candidate microRNAs targeting DMV, 175 targeting 4/91 and 165 targeting MASS. The number of overlapping microRNAs between the prediction tools was found to be 26 for DMV, 23 for 4/91, and 20 for MASS. We conclude that Gallus gallus microRNAs are predicted to target the IBV genome. Ultimately, we have identified a list of candidate microRNAs which will be used in in vitro and in vivo experiments to test microRNAs functions against IBV infection.

UCVM Sure Research Day 2022

Developing the de novo genome of Aedes triseriatus

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Keywords: Mosquito, Genome, Vector

La Crosse encephalitis is a leading cause of vector-borne pediatric encephalitis in North America and is caused by La Crosse virus (LCV). Aedes triseriatus is the main vector of LCV but its sister species, Ae.hendersoni, is unable to transmit LCV. Previous research has shown that physiological differences in the salivary glands prevent transmission by Ae. hendersoni. Despite knowledge of a genetic basis for this salivary gland barrier, the lack of genomic resources for either species has prohibited further interrogation of the genomic basis of this barrier. To address this lack of genomic resources, we sequenced and assembled the genome of Ae. triseriatus from a single adult female using circular consensus high-fidelity (HiFi) sequence data generated on the PacBio sequel IIe. We sequenced three flow cells, resulting in 35Gb of raw Hifi Data. We compared three different assembly programs, HifiASM, IPA, and NextDenovo, and assessed the performance of each using measures of genome completeness and contiguity. HifiASM performed better than other assemblers we evaluated, resulting in the highest N50 value (7.50Mb) with a genome size of 1.69 GB. Additionally, HifiASM had the highest number of complete BUSCO genes (97.32%), with the lowest number of duplicated BUSCO genes (6.15%). Our assembly of Ae. triseriatus is of comparable quality to existing reference genomes of Aedes mosquitoes, and is the largest mosquito genome sequenced to date, consistent with estimates from flow cytometry. Our results show that HiFi data can yield highly contiguous and accurate genome sequences from single mosquitoes.

Distribution and ecology of *Culex Pipiens* in Calgary

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Keywords: mosquito, invasive species, disease vector

Culex pipiens is a global invasive mosquito and is a vector of many parasites and pathogens, including, West Nile virus and Wuchereria bancrofti. Cx. pipiens has two subspecies found in North America: Cx. pipiens pipiens, which prefers being active above ground, feeding on birds and mammals and Cx. pipiens molestus, which prefers being active below ground, feeding predominantly on mammals. In 2018 Cx. pipiens was detected in Edmonton, Alberta. This coincides with previous literature that utilized climate change as a determining factor to predict the expansion of *pipiens* habitat to encompass majority of southern Alberta. Subspecies and distribution outside Edmonton are unknown. To understand this invasive species in Alberta, we received samples of Cx. pipiens from Edmonton and worked with the City of Calgary to assess if Cx. pipiens is present here. We found this mosquito is widespread throughout Edmonton and used PCR-based methods to assess which subspecies is found in Edmonton. We expect that this population will be Cx. pipiens pipiens. To assess if this mosquito is found in Calgary, we used larval surveys and adult trapping throughout the city of Calgary, Alberta. Larval collections revealed that a minimum of twelve distinct species are found within the city limits of Calgary. We did not detect Cx. pipiens during our sampling. As Cx. pipiens was not found, it indicates that the species is rare or not present at all. The later situation in turn also implies pipiens was introduced to Edmonton via anthropogenic causes rather than climate change.

Locomotion scoring as a diagnostic tool for foot rot and digital dermatitis in feedlot cattle

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Keywords: Lameness, Cattle, Diagnosis

Lameness is a major concern for the feedlot industry from health, welfare, and production perspectives. Lameness is any alteration in normal stance or locomotion caused by pain or mechanical dysfunction. Foot rot (FR) and digital dermatitis (DD) are among the most prevalent and potentially painful infectious foot lesions found in feedlot cattle. Locomotion scoring (LS) can be broken down into gait-related traits including weight bearing, stride length, head bob, and back arch. Studying the differences in LS and related traits between FR and DD affected cattle could inform diagnostic strategies as treatment for both diseases differs. Visual inspection of the lifted foot is the most accurate option for diagnosis and differentiation between FR and DD however, it is laborious and potentially dangerous. In the present study, cattle from four different feedlots suspected of having FR or DD by the crew, were filmed freely walking, before entering the treatment barn. Once restrained, the affected foot was lifted, and the lesions closely assessed. The first twenty animals were enrolled in summer 2022 and videos are being evaluated for LS. LS will be done using a 4-level scoring system where levels are differentiated by the presentation of gait-related traits. The LS and diagnosis will be compared to investigate whether LS or any combination of gait-related traits is associated with foot pathology. It is hypothesized that comparison of LS and more specifically gait traits of cattle diagnosed with DD, FR, unclear, and healthy could function as predictive of the type of foot lesion present.

EEG as a non-invasive biomarker in a mouse model of Fragile X Syndrome

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Keywords: Biomarker, EEG, PD

Fragile X Syndrome (FXS) is the leading cause of autism due to single-gene mutations. The Extracellular Signal-Regulated Kinase (ERK) pathway in the brain plays a key role in autism and FXS development. An inhibitor, PD, targets ERK activation. Previous acute PD treatment trials in Dr. Cheng's lab reversed behavioural deficits and decreased power in the high gamma frequency band of electroencephalogram (EEG) signal in a mouse model of FXS. My project built upon these findings and determined whether EEG is a reliable and non-invasive biomarker for tracking improvements in behaviour after PD treatment in individual animals of the FXS mouse model. The objective was to examine if and what changes in EEG signal correlate behavioural improvements in individual animals after PD versus vehicle control treatment. An Open-Source Standalone Electrophysiology Recording system for Rodents (OSERR) was used. We recorded EEG both when the subject was active in the home cage and undergoing an open field test. The power of EEG signals in different frequency bands was computed, and the activity during the open field test was measured. Results were compared between the conditions when the mouse was treated with PD versus vehicle. The treatment sequence was counterbalanced. We are in the process of completing data collection. The project provides insight into the use of EEG as a reliable and non-invasive biomarker for FXS, which could be further tested on human subjects. It is important to have a trustworthy method to track behavioural improvements in patients without requiring invasive or distressing procedures.

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2022 SURE Research Day Organizing Committee

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Session Chairs:

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Session Judges:

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