CUMMINGS SCHOOL
OF VETERINARY MEDICINE AT TUFTS UNIVERSITY

SUMMER RESEARCH
TRAINING PROGRAM AWARDS
2019

ABSTRACT BOOK

2019 Summer Research Proposals

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Student Name	Mentor Name	Year	Project Title	Funding Source
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Mastrostefano, Ava	Dr. Marieke Rosenbaum and Dr. Jonathan Runstadler	V'22	Use of Third Generation Sequencing to Determine Viral Diversity in Trafficked New World Monkeys Confiscated from the Peruvian Wildlife Trade	NIH
Naimark, Alena	Dr. Gregory Wolfus and Dr. Katherine Holmes	V'22	Analysis of Vector-borne Disease Prevalence in Canines in Haryana India	NIH
Nangle, Carly	Dr. Emily McCobb and Seana Dowling-Guyer	V'22	Where Are They Now? An Exploratory Study of Transported Dogs in their Adoptive Homes	Maddies Fund
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Wlodarczyk, Sara	Dr. Christopher Schonhoff	V'21	Sex Differences in Oxycodone-Mediated Alterations in Rat Dendritic Morphology and Axonal Growth in a Primary Neuronal Culture System	Private Funding

Student Name: Riley Aronson V'21

Mentor: Dr. Felicia Nutter

Funding Source: NIH

Title: Use of Computed Tomography (CT) to Determine the Sensitivity to Clinical Signs as a Diagnostic Tool for Respiratory Disease in Rehabilitant Bornean Orangutans (*Pongo Pygmaeus*)

Summary: The aim of this study is to determine the sensitivity of clinical signs in the diagnosis of chronic respiratory disease (CRDO) in the Bornean orangutan (Pongo pygmaus). This will be achieved by comparing observed clinical signs to the definitive results of computed tomography (CT) imaging in a population of clinically affected animals at the Bornean Orangutan Survival Foundation's rehabilitation center in Samboja Lestari, East Kalimantan, Indonesia (BOSF-SL). This center houses 27 animals affected by CRDO, and it has access to CT imaging, making it the ideal location for conducting this important work.

This study will be conducted within broader, comprehensive work in CRDO clinical management that is currently ongoing at Samboja Lestari. By August 2019, this work will have generated a dataset of 30-40 CT scans from the orangutans at the BOSF-SL center. Thorough review of medical records and keeper observations of clinical signs for each individual will be compared to the results of the scans. These comparisons will then be analyzed to determine the accuracy and sensitivity of clinical signs as criteria for presumptive diagnosis of CRDO. It is hypothesized that clinical signs will have high sensitivity for some aspects of CRDO, such as sinusitis, but low sensitivity for other aspects, such as mastoiditis and early bronchiectasis.

 The results of this study are eagerly awaited by veterinarians in all captive orangutan settings. Since most Indonesian zoos and all but one rescue center lack access to CT imaging, it is crucial to establish guidelines for CRDO assessment with low-cost, readily available tools.

There are several logical next steps to this preliminary work. There is a need to establish a severity scale or a symptom-staging rubric, which could in turn be used as a diagnostic tool and prognostic indicator to guide treatment. This is particularly important for maximizing welfare for orangutans in rescue centers in their range country. Additionally, this work will be used to create an imaging atlas of orangutan respiratory pathology, which will serve as a clinical guide to veterinarians around the world and provide an avenue to further research.

By optimizing healthcare and thus enhancing the rehabilitation process, this project can have a direct impact on the conservation of orangutans in their host countries. The rescue centers' limited resources can be used for maximum benefit, and can improve the chance for release to the wild for animals affected by CRDO.

Student Name: Sidney Beecy V'22

Mentor: Seana Dowling-Guyer and Dr. Emily McCobb

Funding Source: Pet Smart Charities

Title: The Effect of Music on Short-Term Stress in Dogs

Summary: Exposing dogs to different types of auditory stimuli such as music can impact a dog's stress level and behavior. Results from studies suggest that kenneled dogs (such as those in shelters) might benefit from calming auditory stimuli, such as classical music (Bowman et al., 2015; Kogan et al., 2012; Wells et al., 2002). The idea has also gained traction outside of academia -- similar to music marketed for humans, retailers now sell music tracks that are promoted to relax dogs experiencing stress (iCalmPet, 2019).

This study will examine the efficacy of music psychoacoustically designed to reduce stress in pet dogs, to explore potential beneficial applications to reduce stress during short-term stressors (such as veterinary visits) and as a preliminary investigation into the potential differences between human and canine perception of music. This will be accomplished by exposing pet dogs to one of three auditory stimuli – psychoacoustically designed dog music, human relaxation music, or no music -- during a short-term separation from the owner. During the exposure, behavior and heart rate variability will be recorded.

Unlike previous studies that exposed shelter dogs potentially experiencing more chronic stress in a kennel environment to auditory stimuli, this study will investigate the utility of music in reducing short-term stress in pet dogs. By comparing the three conditions, it will be possible to determine if psychoacoustically designed dog music truly does reduce anxiety in pet dogs and, if it does so, to what degree compared to other auditory stimuli or no auditory stimuli.

Student Name: Sarah Brattain   V'21

Mentor: Dr. Stanley G. Fenwick and Dr. Felicia Nutter

Funding Source: NIH

Title: Knowledge, Attitudes, and Practices (KAP) Surveys on Antibiotic Use and Antibiotic Resistance among Poultry Producers in Thái Nguyên, Vietnam

Summary: Specific Aims: The purpose of this study is to investigate the knowledge, attitudes and practices on antimicrobial use (AMU) and antimicrobial resistance (AMR) among poultry farmers in Thái Nguyên, Vietnam. Specifically, I aim to 1) provide baseline information on how antibiotics are used on poultry farms and 2) to identify what factors influence farmers' decisions to use antibiotics in their chickens. My long-term goal is to enhance community awareness of AMR and to improve AMU surveillance. The findings from this study will be used to inform policy and to advance the goals of the Vietnam 'National Action Plan on Antimicrobial Resistance'. These objectives will be achieved through the following specific aims:

Specific Aim 1: **Describe antibiotic use on poultry farms in Thái Nguyên, Vietnam.** This specific aim will be achieved through knowledge, attitudes, and practice (KAP) surveys with poultry producers. *Based on previous studies that reveal a high prevalence of AMU misuse and AMR in Vietnamese poultry farms, I hypothesize that antibiotics will be heavily overused in the poultry farms in* Thái Nguyên.

Specific Aim 2: **Evaluate farmers' knowledge of antimicrobial resistance (AMR), understanding of antibiotics, and awareness of safe AMU practices**. This specific aim will be achieved through knowledge, attitudes, and practice surveys (KAP) with poultry producers. *I hypothesize that inappropriate AMU in small-scale poultry production will be positively correlated with poor understanding of AMR among producers.*

Experimental Design/ Methods: Knowledge, attitudes, and practice (KAP) surveys will be administered to 85 poultry farmers in the Thành phố district of Thái Nguyên, Vietnam. Survey participants will be recruited using a random number generator to reduce sample bias. Surveys will include informed consent and local translators from Hanoi University of Public Health. Surveys will be written in English, translated into Vietnamese, and then translated back into English. KAP surveys will be analyzed in StataIC 15.1 using descriptive statistics. Chi-Squared (or Fisher's Exact) will be used to assess crude relationships between dichotomous KAP results and inappropriate AMU (yes/no), which is defined as farms that report giving antibiotics to their flocks prophylactically, more than 2 times a week, and/or without a prescription. Sufficient AMR knowledge (yes/no) will be quantified as a score based on a serious of questions about the implications of AMR to human and animal health. Respondents who are able to answer correctly 75% of the total questions will be defined as having sufficient AMR knowledge. To determine the association between AMU and AMR knowledge, a t-test or non-parametric equivalent will be used to compare AMR knowledge scores between farms that do and do not inappropriately use antibiotics.

Significance: Vietnam has some of the highest antimicrobial resistance levels in the world.[1] The prevalence of multi-drug resistant infections in human hospitals and livestock facilities is steadily rising.[1] Antibiotics are heavily abused in Vietnamese livestock production, especially on small-scale poultry farms.[2] These practices pose serious food safety concerns and increase the risk that AMR bacteria will reach people through the food chain. The Vietnam 'National Action Plan to Combat Antimicrobial Resistance' is an international partnership between the Vietnamese government, WHO, FAO, OIE, and CDC to address these concerns and improve AMR surveillance in Vietnam. Detecting inappropriate antibiotic use and enhancing awareness of AMR among stakeholders is an essential component of the National Action Plan. This study therefore aims to provide baseline information on AMU practices and knowledge of AMR among poultry producers in Thái Nguyên, Vietnam. This information will be used to create advocacy campaigns and risk-based interventions.

Student Name: Kelly Chambers V'21

Mentor: Dr. Annie Wayne and Dr. Claire Fellman

Funding Source: NIH

Title: Antimicrobial Prescription Patterns in Inpatient Dogs at a Veterinary T Tertiary Care Facility

Summary: As antimicrobial resistance continues to threaten the effective use of current antimicrobial drugs, antimicrobial stewardship and surveillance systems have become vital to preserving current antimicrobial therapies. While national and international efforts to counter antimicrobial resistance have flourished in areas of food animal medicine and human medicine, much less progress has been made in companion animal medicine.1 Given that the spread of resistant infections between companion animals and humans has been established, veterinary prescribing practices should be investigated and antimicrobial stewardship efforts in companion animal medicine further developed.

The human medical field has developed standardized methods for measuring antimicrobial use through a national system that provides each hospital with a Standardized Antimicrobial Administration Ratio (SAAR) based on its reported and predicted antimicrobial use. Currently, n similar metric exists in veterinary medicine. The long-term goal of this line of research is to develop a veterinary equivalent of the SAAR system that would allow veterinary hospitals to compare their antimicrobial use to national benchmarks. This study will act as a building block by executing the first steps necessary toward creating a veterinary SAAR system while standing independently as a useful baseline report of antimicrobial prescribing practices at a major veterinary hospital. The short-term objectives of this study are to (1) retrospectively quantify and describe antimicrobial use in dogs at the Foster Hospital for Small Animals, and (2) evaluate patient demographics and risk factors that may contribute to antimicrobial use. We hypothesize that certain risk factors are associated with receiving antimicrobials in inpatient dogs. Additionally, we hypothesize that over 60% of inpatient dogs receive at least one antimicrobial during their hospitalization.

We will conduct a retrospective analysis of antimicrobial prescriptions for inpatient dogs over a twelve-month period by extracting data from the hospital's electronic medical record system. Antimicrobial prescription rates will be calculated using prescription days per patient day and will be categorized based on antimicrobial class. Patient-level data collected will include demographic factors such as age, breed, and sex as well as treatments and procedures received by the patient. The data will be analyzed with statistical software to identify risk factors that correlate with particular antimicrobial prescribing patterns. The findings of the study will be submitted for publication to a peer-reviewed veterinary journal.

Student Name: Asher Feldman V'21

Mentor: Dr. W. Michael Karlin

Funding Source: Boehringer Ingelheim

Title: Development and Validation of a Novel Model to Simulate the Cranial Cruciate Deficient Stifle.

Summary: The most common cause of hindlimb lameness in dogs is cranial cruciate ligament (CrCL) deficiency, which is diagnosed primarily by the cranial drawer test (CDT) — assessing cranial tibial translation relative to the femur. Accurate CrCL deficiency diagnosis is crucial to inform treatment decisions, including early surgical intervention. Veterinary students have limited opportunities for CDT practice, due to the low availability of CrCL-deficient patients and the potentially painful nature of the test. Given canine CrCL deficiency prevalence and the necessity for early and accurate diagnosis, improving the competence and confidence of veterinary students in performing the CDT could improve the welfare of CrCL-deficient dogs. We will address this need by developing realistic models of canine CrCL deficiency.

The specific aims of this study will be: first, to develop and validate two novel, high-fidelity orthopedic models of the canine hindlimb that realistically simulate a cranial cruciate intact and deficient stifle, via the presence or absence of cranial drawer motion; second, to determine if these developed models improve veterinary student confidence and competence in performing the CDT and diagnosing CrCL deficient stifles.

The models will consist of 3D-printed models, from CT data, of the bones of the canine hindlimb, surrounded by silicone to emulate soft tissue and skin. The skin will be formed by a 3D-printed negative mold of the skin surface. An interlocking mechanism will be created to attach the tibia and the femur at the level of the stifle joint, while allowing at least 30° of joint flexion and, in the CrCL-deficient model, 6-8mm cranial tibial translation. The accuracy and realism of the models will be validated by surgeons. We will assess model educational utility by having veterinary students manipulate and diagnose the models, and fill out questionnaires pre and post-evaluation on confidence and competence with the models and CDT.

Upon completion of this study, the developed models will potentially act as new educational tools for veterinary students, optimally increasing confidence and competence in CDT performance and diagnosis of CrCL rupture.

Student Name: Sarah Flick V'21

Mentor: Dr. Fair Vassoler

Funding Source: NIH

Title: Measurement of Differential Expression of miRNAs in Rat Model Following Self-Administration of Oxycodone to Determine mRNA Targets Involved in the Development of Addiction

Summary: The misuse and abuse of prescription and illegal opioid drugs in the United States has been on the rise for decades - mainly due to an increase in prescribed opioids for analgesia purposes. Each day over 130 people in the United States die due to opioid overdose, which has major social and economic impacts.1 The Centers for Disease Control and Prevention (CDC) estimates that in the United States $78.5 billion per year is spent on managing the opioid crisis, some of which goes toward healthcare and criminal justice costs.2 The major driver of the so-called "opioid epidemic" is the highly addictive nature of opioids. Opioid addiction is characterized by a compulsive need to take the drug and frequent relapses after periods of attempted abstinence from drug consumption.

While it is accepted that addiction is a complex disease of the brain and body characterized by remodeling of neuronal synapses in the brain due to prolonged drug intake, it is unknown exactly how this brain remodeling takes place. There is emerging evidence that microRNAs (miRNAs) play a role in this process by regulating neuroplasticity associated with addiction behavior and controlling the motivational properties of opioids in reward centers of the brain.4 These miRNAs function to silence messenger RNA (mRNA) via post-transcriptional modification leading to subsequent decrease of gene expression. Learning more about the function of miRNAs in opioid drug addiction could be helpful to future efforts related to opioid addiction treatment.

The specific aim of this study is to measure miRNAs in the brains of rats following self-administration of oxycodone, as well as mRNA targets via PCR. Rats are the model of choice for this study because findings in rodent models can often be translated to humans.5 Jugular catheters will be surgically implanted into 16 rats. Half of them will be able to self-administer oxycodone, a semi-synthetic opioid. The other half will have yoked saline administered via their catheter. Upon euthanasia of the rats, specific brain regions will be dissected and total RNA from the nucleus accumbens (a region known to be involved in reward and addiction)3 will be extracted and subsequently analyzed via qPCR for specific miRNA and mRNA targets. The hypothesis of this study is that the differential expression of specific miRNAs following oxycodone self-administration will lead to the maintained dysregulation of mRNA targets.

Student Name: Gabrielle Fontes V'21

Mentor: Dr. Robert McCarthy

Funding Source: Boehringer Ingelheim

Title: The Effect of Gender on Trauma Survival in Dogs

Summary: *Study Objectives:* To determine if sexual dimorphism effects trauma survival in dogs.

Hypothesis: Intact females will display a survival advantage compared to non-hormonal females, intact males and non-hormonal males. Intact males will display a survival disadvantage compared to intact females, nonhormonal females, and non-hormonal males. *Experimental*

Design and Significance of Proposed Research: Several human studies document a survival advantage in hormonally intact women after trauma, believed to be related to high levels of estradiol associated with estrus. Similarly, hormonally intact men have been reported to have a survival disadvantage linked to high levels of testosterone suppressing cell-mediated immunity.

Other than laboratory rodents, there have been no studies in animals investigating this phenomenon. The aim of this study is to determine if sexual dimorphism in trauma survival exists between four hormonally different groups of dogs (intact male, non-hormonal male, intact female, non-hormonal female) using the VetCOT Trauma database (2013-2017). Results could support future prospective studies employing hormone-based therapy to improve trauma survival in animals.

Student Name: Sarah Gillis-Smith V'21

Mentor: Dr. Megan Mueller

Funding Source: NIH

Title: Perceptions of Support for Pregnancy and Parenting Among Veterinarians Having Children at Different Stages of Their Career

Summary: Significance: In recent decades there has been a marked trend toward having children at an older age, especially among professional groups with extended post-graduate training such as physicians. While this allows parents to establish themselves in their career and achieve greater financial stability to support their children, delaying childbearing increases the risk of pregnancy complications, genetic abnormalities, and having to resort to assisted reproductive technology to conceive. Therefore, it is important from a public health perspective to encourage policies that allow working parents who would like to have children at a younger age to do so. In veterinary medicine, there is data suggesting that veterinarians delay having children, but the extent of this trend has not been described. There is also a lack of description of typical policies surrounding pregnancy and parenting outside of veterinary medical colleges.

Specific Aims: The goals of the study are to quantify when veterinarians have children in relation to career stage, determine the level of support available to them at work when they have children, and summarize the perceived challenges or benefits related to having children at each career stage.

Methods: A quantitative, cross-sectional study of veterinarians in the United States will be conducted using Qualtrics to administer an anonymous, on line survey. IRB approval will be obtained before initiation. The criteria for participation will include anyone with a DVM or VMD degree. Data collected will include demographics, family planning status, timing and number of children, employer size, knowledge of policies related to childbearing, perceptions of support, challenges and benefits of having children at particular career stages, and reasons for nonparents not having children. Data will be analyzed using descriptive and inferential statistics in STATA.

Student Name: Elliot Grossman V'21

Mentor: Dr. Felicia Nutter

Funding Source: Private Funding

Title: Koala Care: Analysis of Causes and Types of Urogenital Tract Disease Due to Chlamydia

Summary: Specific aims: *Specific aim 1*: To extract, organize, and interpret data on how rehabilitation of koalas with *Chlamydia*-induced urogenital disease is conducted at the Australia Zoo Wildlife Hospital.
Specific aim 2: To determine the rate of successful rehabilitation of koalas with Chlamydial urogenital disease.

- *Hypothesis*: The rate of successful rehabilitation will be over 50%.

Specific aim 3: To determine if any signalment, life history, physical examination, diagnostic test, treatment, or other variables during rehabilitation can be used to predict rehabilitation success for koalas.

- *Hypothesis*: There will be data correlates to rehabilitation success.

Background:

Koalas are one of Australia's iconic species, and are currently listed as vulnerable by the IUCN Red List.[12] There are an estimated 300,000 wild koalas remaining, facing threats ranging from climate change and human development activities, to invasive species and infectious disease. Regarding disease, infections by *Chlamydia* are most prevalent and damaging to koala populations. Chlamydial infections can lead to blindness and infertility. Large numbers of koalas with Chlamydial infections are admitted to wildlife rehabilitation hospitals on the east coast of Australia. It is important for veterinarians to determine the effectiveness of treatments and the prognosis for recovery to best allocate resources and time,. This project will help elucidate this important information by retrospectively assessing data from the past 5 years of koala patients with Chlamydial urogenital disease that presented at the Australia Zoo Wildlife Hospital.

Methods:

I will review the past 5 years of koala admissions to the Australia Zoo Wildlife Hospital for cases of *Chlamydia* and urogenital disease. I will then extract, organize, and analyze available data on signalment, date of admission, weight, physical exam findings, clinical signs, lab work and diagnostics, treatment administered, length of hospitalization, and disposition. I will use descriptive statistics and general linear regression models to determine if there are risk factors or protection factors associated with success or failure of the clinical course.

Significance:

This work is important because it will allow a better understanding of the factors associated with clinical success or failure which in turn can be used to pursue a more appropriate course of action in koala rehabilitation. For example, it may decrease wasteful or ineffective diagnostics or treatments and it may improve the welfare of the patients/lead to more positive rehabilitation results.

Student Name: Tatyana Kalani V'21

Mentor: Dr. Marieke Rosenbaum and Dr. Adam South

Funding Source: NIH

Title: Soil, Children, and Wildlife: The Cycle of Lead in the Urban Environment

Summary: Environmental lead contamination in urban areas such Greater Boston is known to impact human, animal, and ecosystem health - being a health risk at even small concentrations. Any organism can acquire lead toxicosis from soil, water, or air sources. Currently, lead research on human health, animal health, and the environment is often conducted in separation. This has led to large bodies of research particularly related to impacts of lead on children, with a lack of data incorporating both non-human animal and environmental components. While lead-based paint is the main focus of exposure for children, there is evidence that lead contamination in soil can also be a source of exposure. In addition, wildlife is likely to be in contact with soil more than humans. Past research conducted by both my mentor and I have shown that animals are living in and frequenting areas of higher lead exposure risk, and that some animals are exhibiting subclinical, elevated blood lead levels when in contact with soil.

The purpose of this project is to use a One Health approach to understand the relationships between soil, child, pet, and wildlife lead levels. More specifically, we aim to determine if child blood lead levels, which are routinely collected by all pediatricians and reported to the state of Massachusetts for children under 3, can be used to predict environmental lead contamination and lead exposure to animals in urban areas. We propose a study utilizing the prevalence of childhood lead exposures in Boston to identify eighteen census tracts with high or low prevalence. Within each census tract, soil samples will be collected and analyzed by x-ray fluorescence for lead. Additionally, traps will be placed to trap and collect blood from house sparrows at the site of soil collection. Statistical analyses will be conducted to determine if blood lead levels in children are predictive of lead in soil, and lead levels in urban wildlife.

Student Name: Kelley Kilpatrick V'21

Mentor: Dr. Elizabeth Rozanski

Funding Source: Boehringer Ingelheim

Title: Effectiveness of the a-1 Antagonist, Prazosin, in the Prevention of Recurring Urethral Obstructions in Male Cats

Summary: *Study Objectives:* To investigate whether the use of the alpha-1 blocker,

prazosin, in post-obstructive treatment for cats presenting with a urethral obstruction decreases the rate of a recurrent urethral obstruction before discharge and at 14 days after initial unblocking. Prazosin is used in an effort to relax the urethral smooth muscle, and limit urethrospasm, which may contribute to re-obstruction of the urinary system.

Hypothesis: Cats treated with prazosin as a post-obstructive treatment after initial unblocking when presented to an emergency room **will not** have a difference in the rate of recurrence of urethral obstruction during hospitalization as well as within 14 days after initial unblocking.

Experimental Design and Significance of Proposed Research: Urinary obstruction (UO) is a common clinical problem in cats; while initial therapy is straightforward, including sedation, urinary catheterization, and supportive care, somewhere between 15-40% of cats re-obstruct after the initial urinary catheter is removed. Many clinicians advocate the addition of prazosin, as a mechanism to reduce urethrospasm, which is considered to lead to re-obstruction. Prazosin may contribute to hypotension, adds cost to care, and requires medicating cats which may be challenging. Limited peer-reviewed information exists concerning the utility of prazosin. According to Reineke *et al*, in a prospective study, it was found that prazosin compared to placebo did not show a significant difference in the rate of a recurring UO within 24 hours and within 30 days. However, the study was underpowered and it was determined in *post-hoc* analysis, that about 1900 cats would be required rather than 47 cats that were enrolled. Retrospectively, Hetrick and Davidow (2013), found that prazosin had a significant benefit in the reduction of the rate of recurrence of cats that present with UO, although a control group was lacking. In clinical practice, including at Tufts University, >75 % of cats with UO are currently administered prazosin, but **without** any evidence of efficacy. This study aims to provide an evidence based recommendation on the utility of prazosin in UO.

Student Name: Hayley Klosowski V'22

Mentor: Dr. Elizabeth Byrnes

Funding Source: NIH

Title: RXFP3 Expression in Post-Stroke Rats

Summary: Specific Aims: To test the hypothesis that ischemic injury will result in the down-regulation of RXFP3 receptors in the pre-frontal cortex. This study will utilize both male and female middle-aged rats, examining receptor expression and hypothalamic pituitary adrenal (HPA) activity 1 or 10 weeks post ischemic injury.

Significance: Stroke is the third leading cause of death world-wide and has significant effects on its surviving population, which is estimated to reach 70 million people by the year 2030. Post-stroke depression affects one third of stroke survivors, making it the most common psychiatric disorder facing this population [1]. PSD is characterized by all-encompassing sadness, apathy,

and changes in eating or sleeping patterns following a stroke [3]. Relaxin-3 (RLN3) is a neuropeptide that is part of a broad arousal system in the brain and plays a role in modulating higher brain functions such as cognition and behavior [4]. Studies suggest the RLN3 system has roles in regulating depressive-like behaviors and motivated responding. As such, further research is needed to investigate the RLN3 system and its potential role in PSD.

Method: We will be examining the effects ischemic injury has on the expression of the RLN3 receptor, relaxin family peptide receptor 3 (RXFP3) and HPA activity in middle-aged, male and female rat subjects. To induce an ischemic stroke, rats will be infused with endothelin-1 and control subjects will be infused with artificial cerebral spinal fluid. RXFP3 expression in the prefrontal cortex will be measured by the number of RXFP3 positive cells using immunohistochemistry (IHC) with double-labeling of anti-RXFP3 (Sigma) and anti-HuC/HuD (neuron-specific antibody). To determine effects on the HPA axis, plasma corticosterone and adrenocorticotropic hormone (ACTH) will be measured using enzyme linked immunoassays (ELISAs).

Student Name: Kendall Leet-Otley V'22

Mentor: Dr. Annie Wayne and Dr. Claire Fellman

Funding Source: NIH

Title: Evaluation of Antimicrobial Prescribing Patterns in Cats in a Veterinary Tertiary Care Facility

Summary: *Study Objectives:* The objective of this study is to evaluate the prevalence and patterns of antimicrobial prescribing in a small animal hospital by quantifying the current rates of antimicrobial use in inpatient cats and documenting factors associated with the receipt of antimicrobials. This study is an important addition to the veterinary literature on its own but is also the first essential step of a larger project which aims to develop methods for tracking antimicrobial use, creating a platform analogous to human medical reporting systems. This research will contribute to the ultimate goal of creating evidence-based protocols, inciting stewardship interventions, and optimizing antimicrobial prescribing practices for cats.

Hypothesis: We hypothesize that this retrospective review will allow us to identify important demographics and risk factors that are significantly associated with antimicrobial prescribing in cats. Furthermore, we hypothesize that greater than 50% of all inpatient cats will have been prescribed at least one antimicrobial during their hospital stay.

Experimental Design: This study will be a retrospective analysis of cat inpatient records at Tufts Foster Hospital for Small Animals for a period of twelve consecutive months. Using descriptive statistics, we will calculate the percent of inpatient cats prescribed antimicrobials as well as recording antimicrobial class, duration, and the prescription of multiple agents simultaneously. Demographic descriptors, medical indications, and risk factors associated with receipt of antimicrobials will also be analyzed. This data will be quantified by prescription days per patient day and statistically analyzed using Microsoft Office Excel and SAS. The student researcher will validate the data through the hospital medical record, StringSoft. We, the

student researcher and mentors, will create and submit a scholarly manuscript of our findings to a peer-reviewed veterinary medical journal.

Significance of Proposed Research: Currently, there is minimal data describing the percentage of veterinary cases that are prescribed antimicrobial agents. To limit the development of antimicrobial resistance and prolong the use of these life-saving drugs, we seek to create a data set documenting the current rates of antimicrobial use in a tertiary care small animal veterinary hospital. The results from this study will provide a benchmark against which other small animal hospitals can evaluate their own antimicrobial usage and contribute to the development of a system parallel to existing human reporting systems, providing a foundation for subsequent antimicrobial research, guidelines, and interventions.

Student Name: Daniela L. Lopez Goicochea V'21

Mentor: Dr. Marieke Rosenbaum and Dr. Megan Mueller

Funding Source: NIH

Title: Understanding Sociocultural Differences in Knowledge, Attitudes and Experiences related to Veterinary Medicine between Latinx and non-Latinx Pre-Health Undergraduate Students

Summary: Although the second largest minority and one of the fastest growing demographics in the US, the Latinx community continues to be highly underrepresented in veterinary medicine even after increased recruiting efforts in this community. Diversifying the veterinary workforce is important for increasing access to care, as students from underrepresented backgrounds are more likely to provide services to underserved communities. Diversity in health care is also important for increasing client satisfaction, improving doctor – client communication, and developing cultural competence in the health-care system to create more inclusive environments. The AVMA estimates that 62.5% of Latinx households have pets therefore there is a need for veterinarians that serve this segment of the population. Additionally, there is an incredible need for bilingual veterinarians to increase effective communication between doctors and clients, as it is estimated that 13 million pets are owned by limited English proficient Spanish speaking pet owners. Although there are known factors and barriers that keep underrepresented students from accessing higher education, there are not many studies that explore the reasons why students who are interested in medicine and who have the resources to access a medical education chose not to enter the veterinary profession. Through a mixed-methods approach, this hypothesis-generating study attempts to understand the sociocultural differences in knowledge, attitudes and experiences related to veterinary medicine between Latinx and non-Latinx pre-health undergraduate students. An online Qualtrics survey adapted from a Midwestern University study about students' perceptions of the veterinary profession will be distributed to pre-health students in undergraduate universities that have or are associated with an accredited college of Veterinary Medicine in the US. The survey will be modified to include demographics and to consider other factors such as past experiences with family pets and other animals, importance of role models in the field, past interactions with veterinarians, perceived economic status as a veterinarian,

perceived feasibility of pursuing the career, cultural and parental perceptions of the profession, and perception of the impact that veterinary medicine has in their communities. Additionally, in-depth interviews will be conducted with pre-health students that self-identified as Latinx in the online survey to better understand barriers to considering a career in veterinary medicine as well as cultural perceptions of the profession and its impact. Identifying the factors that are keeping Latinx students from pursuing a career as a veterinarian could assist colleges of Veterinary Medicine in designing more effective, targeted recruiting and community programs to help develop the diverse workforce that our diverse population needs.

Student Name: Ananya Mahalingam-Dhingra V'22

Mentor: Dr. Melissa Mazan

Funding Source: USDA

Title: Examining the Effect of Nebulized Lidocaine on Clinical Signs, Pulmonary Function, and Lung Inflammation in Equine Asthma

Summary: Equine asthma is one of the most prevalent diseases in stabled horses of all ages and environments and is a leading cause of poor performance. The disease is characterized by airway inflammation and hyperreactivity, with clinical signs such as chronic cough and poor performance. Despite the pervasiveness, an economical and effective treatment for this disease is lacking. The current pharmaceutical treatments for equine asthma include corticosteroids and beta-2 receptor agonist drugs, both of which have negative side effects. Moreover, corticosteroids delivered via a pressurized metered dose inhaler (pMDI) are often too expensive for most equine owners to afford. Therefore, there is an urgent need to find methods of treatment for horses with equine asthma that are not only safe and readily available, but cost-effective as well.

Equine asthma is characterized by pro-allergic Th-2 and Th-1 immune mediated responses as well as increases in levels of inflammatory cytokines (Il-4, Il-17, IFN-gamma) and decreases in anti-inflammatory cytokines (IL-10). This project looks to use nebulized lidocaine as a treatment for these symptoms. Lidocaine is an amine-derivative that exerts its anesthetic by blocking fast voltage-gated sodium channels and can also aid in reversing the immune mediated Th-2 and Th-1 responses, thus normalizing levels of inflammatory and anti-inflammatory cytokines. Nebulized lidocaine has been used in human and feline models and resulted in ameliorating chronic cough and improving lung function, respectively. We therefore hypothesize that nebulized lidocaine treatment will result in improved clinical signs and cough, decreased bronchoalveolar inflammation (BAL), and decreased airway reactivity in equine asthma. More specifically, treatment with nebulized lidocaine will increase levels of the anti-inflammatory cytokine, Il-10, and decreased levels of the Th-2 cytokine, IL-4, the Th-1 cytokine, IFN-gamma, and the neutrophil-activating cytokine, IL-17 in bronchoalveolar lavage fluid (BALF).

The study will be performed by identifying and subsequently recruiting horses for the study that have veterinarian-identified inflammatory airway disease (IAD). We will perform a physical

examination, clinical and cough scoring, histamine bronchoprovocation (HBP), and cytological analysis of BALF to further characterize the degree of hyperreactivity and inflammation occurring within the airways of these horses. They will then be treated with a solution of 4% lidocaine (2mg/kg) *via* nebulization twice a day for 2 weeks, and then re-tested on day 14. We will also examine the effect of nebulized lidocaine on the production of inflammatory cytokines using commercial equine multiplex assays. We will compare these data with that of a control group (receiving a placebo) and look for significant differences between the two groups.

The results of this project not only have the potential to play a large role in future treatments for equine asthma, but also could play an important role in establishing the horse as a model for improved treatments for human asthma as well, therefore expanding its potential to multiple species.

Student Name: Ava Mastrostefano V'22

Mentor: Dr. Marieke Rosenbaum and Dr. Jonathan Runstadler

Funding Source: NIH

Title: Use of Third Generation Sequencing to Determine Viral Diversity in Trafficked New World Monkeys Confiscated from the Peruvian Wildlife Trade

Summary: The Amazonian region of Peru is among the most biodiverse regions in the world, home to over 57 taxa of non-human primates, or NHPs. In addition to its high degree of biodiversity, Peru also has the highest rate of wildlife trafficking in the world, with many species of wildlife illegally trafficked and sold as pets, for meat, for religious rituals and for tourist attractions. Every year, thousands of NHPs are confiscated from the Peruvian wildlife trade and are brought to rescue centers. Both in the wildlife trade and at the rescue centers, these NHPs have extensive interaction with humans, other species of NHPs and various other species of trafficked and rescued wildlife. This increased level of interspecies interaction puts confiscated NHPs at a high risk for contracting zoonotic pathogens. Release of NHPs carrying zoonotic disease poses major conservation threats to wild populations. Preliminary data obtained by our research group demonstrates that rescue centers prioritize the health of the NHPs when considering reintroduction to the wild, however these organizations lack information and guidelines for medical screening before release. The goal of this study is to examine viral diversity in semi-captive spider monkeys and howler monkeys. This protocol will employ third generation sequencing and metagenomic analysis using the MinION third generation sequencer. The MinION specifically has been used successfully as a cost-effective way to examine large populations of viruses with high levels of diversity commonly missed by short-read sequence methods. RNA extracted from oral swabs collected from NHPs under sedation will be used to synthesize and amplify cDNA, which will be used to prepare 12 barcoded sequencing libraries. These 12 libraries will be run simultaneously on a single flow cell, resulting in an effective and efficient protocol reducing time, cost and reagent use. The MinIT module will carry out data acquisition and real time basecalling in the field. The MinION data will be analyzed at the

laboratory in Lima to examine strain-level, diversity, and viral-genome alignment. These results will inform guidelines for viral screening of spider monkeys and howler monkeys before reintroduction into the wild.

Student Name: Alena Naimark V'22

Mentor: Dr. Gregory Wolfus and Dr. Katherine Holmes

Funding Source: NIH

Title: Analysis of Vector-borne Disease Prevalence in Canines in Haryana India

Summary: India is home to a wide variety of climates zones that create environments for pathogen-carrying vectors such as mosquitos and ticks to thrive. In this study, data will be collected about the prevalence of six vector-borne pathogens *Dirofilaria immitis, Borrelia burgdorferi, Ehrlichia canis, Ehrlichia ewingii, Anaplasma phagocytophilum,* and *Anaplasma platys* in a population of canines sampled from the Delhi National Capital Region (NCR), India. It is hypothesized that the vector-borne pathogens tested for using the SNAP® 4Dx® Plus Test will be present in the surveyed population of canines.

This project has four aims:
Aim 1: Determine the prevalence of the six vector-borne pathogens screened for in the SNAP® 4Dx® Plus Test in the sample population of canines.
Aim 2: Evaluate relationships between disease occurrence, sex, and BCS.
Aim 3: Provide recommendations for the prevention and treatment of disease identified in the sample population based upon test results.
Aim 4: Provide recommendations for the screening, prevention, and treatment of the evaluated vector-borne pathogens to the general canine population in the Delhi NCR.

The population of canines sampled in this study live at All Creatures Creatures Great and Small (ACGS), an animal sanctuary in Haryana, India, that houses a rescue population of canines from the Delhi NCR. Venipuncture will be performed on each canine (n=400), and the blood samples will be hematologically analyzed using SNAP® 4Dx® Plus Tests. Signifiers including sex, coloring, and body condition score will be recorded. The percentage of dogs that test positive for exposure to each pathogen will be reported. The statistical relationships between the prevalence of the six screened pathogens and sex, as well as each disease prevalence and body condition score (BCS) will be analyzed. A study counterpart will assess hematologic changes in the sample population to further assess evidence of disease.

The goal of this pilot study is to assess and determine the prevalence of the six vector-borne pathogens identified in the SNAP® 4Dx® Plus Test in the sample canine population, surveillance to date of which has been greatly limited. *D. immitis, B. burgdorferi, E. ewingii,* and *A. phagocytophilum* all have zoonotic potential, and thus pose a threat to both canine and

human health. The limited existing data about vector-borne pathogen and disease prevalence in the Delhi NCR suggests that canines in this region are likely to be infected with the vector-borne pathogens being screened for. The results of this study will provide implications for disease screening, treatment, and prevention in the sample population as well as in the general population of canines living in the greater Delhi region.

Student Name: Carly Nangle V'22

Mentor: Dr. Emily McCobb and Seana Dowling-Guyer

Funding Source: Maddie's Fund

Title: Where Are They Now? An Exploratory Study of Transported Dogs in their Adoptive Homes

Summary: Many animal health professionals and animal welfare advocates believe canine transportation is a life-saving procedure that can mitigate dog overpopulation problems that still exist in some parts of the United States. Though approximately 778,000 dogs are transported each year, canine transportation is still a controversial topic among animal welfare advocates ("MSU College," 2017). Shelter dog transport is a topic that has thus far been underexplored despite its increasing prevalence, with only two studies looking at dog transport programs to date (McCobb & Dowling-Guyer, 2017; Simmons & Hoffman, 2016). In order to properly analyze the success of canine transport more information on all aspects of the process is required. The goal of the proposed study is to gather data about the impact of dog transportation on adopted dogs and their owners.

An exploratory study conducted by the Center for Shelter Dogs in 2016 determined that staff members of organizations that did not participate in canine transport programs saw transport in a more negative light and believed that transported dogs had more problems post adoption than local dogs (McCobb & Dowling-Guyer, 2017). Therefore, we are interested in testing the assumption that   transported dogs are less successful than local dogs post-adoption because of behavior, health, or other problems. Both studies conducted on canine transport programs thus far have primarily focused on industry practices. In contrast, this study will look at the outcome of adoption by evaluating the success of transported dogs in their adoptive homes and comparing it to the success of non-transported dogs in their adoptive homes. Adoption outcome will be determined through a survey of 600 dog adopters who have adopted a dog from one of our partners over the last five years. We aim to include 300 adopted local dogs and 300 transported dogs. The survey will determine the dog's current status (still in home, returned, deceased, etc.), medical and behavioral history while in the home, and the owners experience and satisfaction with the dog. While exploratory in nature, we expect to find that the majority of adopted dogs go on to be successful dogs regardless of whether they are local or transported.

Student Name: Nicole Nelson V'21

Mentor Name: Dr. Nicola Hill

Funding Source: USDA

Title: Predicting the Effects of Ocean Warming and Food Stress on an Influenza Reservoir: Will Epidemics in Gulls Change in Frequency, Timing and Amplitude?

Summary: Background/Significance: My project objective is to determine how periods of food stress due to rising sea surface temperatures can impact Glaucous-wing gull (*Larus glaucescens*: GWGU) populations and influenza A virus (IAV) dynamics in Cordova, Alaska. Glaucous-wing gull populations are a natural reservoir for IAV and are therefore a useful model species to understand how environmental changes can impact disease ecology. Strains with low pathogenicity can evolve into highly pathogenic viruses that threaten human and animal health. GWGU are commonly found close to human settlement and respond strongly to changes in food availability caused by commercial fishing, climate change, or pollution. One of these environmental factors, sea surface temperature, has been found to increase cannibalism and decrease reproductive success among GWGU owing to critical food shortages during breeding. However more research is needed to understand how ocean warming will impact the dependency that gulls have shown toward fisheries and whether the effect will be synergistic causing enhanced interaction between gulls and fisheries. If gull populations are more dependent on fishery offal during period of food stress, congregating at the canneries before fall migration, epidemics of influenza are also likely to shift in frequency and intensity. This research project will take advantage of a long-term dataset to address these information gaps by testing whether ocean warming accurately predicts host density and influenza prevalence within and between years.

Specific Aims:

1. To determine if the abundance of gulls at the fisheries in Cordova reflects colony collapse triggered by a decline in natural forage fish that GWGU consume during breeding.
2. To analyze the relationship between sea surface temperature (SST) data and gull abundance at the fisheries – a key driver of influenza prevalence within the population.
3. To measure the prevalence of influenza virus in samples collected between May (arrival of GWGU) and September (departure of GWGU) in 2019 using reverse transcriptase polymerase chain reaction (RT-PCR) and ELISA testing

Methods:

Historical gull count and influenza prevalence data from 2011-2018 as well as results from RT-PCR and ELISA testing of samples collected between May and September of 2019 will be used in statistical modeling. In addition, sea surface temperature (SST) data for Cordova, Alaska will be obtained from International Comprehensive Ocean-Atmosphere Data Set release

3.0. Generalized linear modeling will be used to test my hypothesis in order to determine if there are predictors of colony collapse, gull density, and influenza prevalence.

Student Name: Jillian Nolan V'22

Mentor: Dr. Jonathan Runstadler

Funding Source: NIH

Title: Characterization of the interferon-induced transmembrane protein gene family in the gray and harbor seal genomes

Summary: Influenza A virus (IAV) infections in harbor seals (Phoco vitulino)and gray seals (Holichoerus grypus) along the North Atlantic coast have been observed to have disparate outcomes for these populations (1). These species are closely related and have historically shared and continue to share substantial portions of their habitat so this apparent discrepancy in response to IVA infection is unexpected and is likely to have some basis in their respective genetic composition.

Influenza infection and the immune response in a given affected species is highly complex and involves numerous innate and adaptive immune factors (2). Recently, studies in humans and mice have demonstrated the importance of one family of innate immune factors collectively called interferoninduced transmembrane proteins (IFITMs), which restrict the ability of certain viruses to enter cells and replicate (3). Human carriers of specific IF|TM3 alleles have also been shown to be more likely to have a severe outcome in the presence of influenza infection (4). The Runstadler lab has assembled novel genomes for the harbor seal and the grey seal (unpublished data). We propose to identify and annotate the IFITM gene family in these genomes to characterize any significant difference between the seal species. To understand and quantify the diversification and evolution of this gene family, we will conduct cross-species and interspecies phylogenetic analyses, identify the ratio of synonymous versus non-synonymous mutations and compare the amino acid composition and characteristics of each protein in the family. We expect to identify orthologs or genes with key similarities to human IF|TM3 and we will investigate the expression of these genes in harbor and grey seal samples of known influenza status with the expectation that our potential correlate of susceptibility or resistance will be elevated in influenza positive seals.

Student Name: Alene Pohly V'22

Mentor: Dr. Nicholas Robinson

Funding Source: Private Funding

Title: Surveillance of canine vector-borne pathogens from the Delhi National Capital Region, India

Summary: Objective: Aim 1: Determine prevalence of vector-borne pathogens within canines from the Delhi National Capital Region, India.   **Aim 2: P**rovide recommendations in writing for the prevention/treatment of diseases.

Animals: Population of 400 canines living in a sanctuary in Haryana, India, rescued from Delhi National Capital Region (NCR).
Procedures: Analyses of blood will be conducted including packed cell volumes, total solids, and smears to evaluate leukocyte estimate and differential, cell morphology, platelet estimate, and presence of infectious organisms. Information for preventing the contraction of vector-borne diseases will be provided verbally.

Relevance: The prevalence of vector-borne diseases of canines in India is not an area that has been widely surveyed. Some of the pathogens are zoonotic and can be spread to human hosts. Animals infected with vector-borne pathogens that are not treated act as a reservoir for the disease and pose a threat to public health. Since dogs can act as reservoirs, hematological analysis for the prevalence of vector-borne diseases (*D. immitis, B. burgdorferi, E. canis, E. ewingii. A. phagocytophilum, and A. platys)* would provide evidence that vector-borne diseases are prevalent in the Delhi region of India. Currently, there are limited studies regarding these issues and the studies that have been done, focus on only one species of vector-borne disease in various regions. Few, if any, studies look at a variety of species in one region in India. Identification of the prevalence of these diseases is important in being able to advocate for prophylaxis in India, preventing the spread of the diseases among canines. The practice of prophylaxis in canines may also prevent the transmission of the diseases into the human population. Providing information on disease treatment and prevention would aim to prevent the future spread vector-borne diseases.

Student Name: Peri Prendergast V'22

Mentor: Dr. Rachel Gately

Funding Source: NIH

Title: Influence of extender, temperature, and time on post-thaw sperm motility in ram semen *(Ovis aries)*

Summary: The objective of the experiment is to compare total and progressive sperm motility in ram semen before and after cryopreservation when using a soybean-based extender (AndroMedÒ) versus an egg yolk-based extender (TriladylÒ) versus a liposome-based extender (OPTIXcellÔ) in various extension protocols. The importance of this experiment is not only to compare the media in which the semen is diluted but also the technical preparation methods used in common practice. Over the course of a century, numerous investigations have been made into the composition of these extenders, chiefly regarding their base. The appropriate choice of commercial-made extender for ram semen has not yet been determined and is of particular concern due to the current low post-thaw motility of sperm seen in today. Commercially made extenders, like those being tested in this experiment, streamline the dilution process, are readily available, and come from reliable manufacturers ensuring quality control in the composition of the product. Extenders provide a source of energy and protection for the spermatozoa during freezing, allowing for longer storage times, and further manipulation in the protocol will also help improve post-thaw motility. For example, the time the sperm and media spend equilibrating as well as the temperature in which the diluted semen is packaged into the semen straws will also be explored as possible enhancements to the freezing protocol. More importantly, this study aims to identify synergistic effects on post-thaw motility between the extender and variables tested, optimizing protocol and eventually fertility rates in the sheep industry.

In this study, semen will be collected from 5 reproductively sound Dorset rams during the month of June via artificial vagina. Prior to accepting an ejaculate to the study, semen will be evaluated for acceptable volume, sperm concentration, progressive motility, and abnormal sperm count. Accepted ejaculates will be pooled before processing and total motility and progressive motility will be measured with the iSperm Analyzer, an economically feasible analyzer of sperm parameters. Three commercial extenders will be tested, representing soybean-, liposome-, and egg yolk-based extenders. For each extender, temperature at which the samples are loaded into the straws (23°C vs 4°C) and duration of equilibration (2h vs 4h vs 12h) will be manipulated. A total of 18 cryopreservation protocols will be tested.
After thawing of the extended semen, total motility and progressive motility will again be measured with the iSperm Analyzer as well as with CASA equipment, computer-assisted sperm analysis equipment.

The overall goal is to increase post-thaw motility of ram sperm in effort to contribute in the design of a cryopreservation protocol that will eventually increase fertility rates. The data collected from this study will contribute to the improvement of assisted reproductive techniques in sheep.

Student Name: Cameron Pryor V'21

Mentor: Dr. Eric Brum and Dr. Marieke Rosenbaum

Funding Source: NIH

Title: Rabies Detection in Rural Bangladesh

Summary: In 2010, Bangladesh initiated a national campaign to eliminate dog-mediated rabies from the country.   At the time, rabies, which is both 100% fatal and 100% preventable, was estimated to kill 2,100 people annually in Bangladesh. Under the Rabies Elimination Program (REP), National and District Rabies Prevention and Control Centers (NRPCC and DRPCC) were established, mass dog vaccination campaigns conducted, and surveillance, research and public engagement prioritized. As Bangladesh pushes towards its goal of rabies elimination, the importance of accurate surveillance increases. Surveillance provides much needed feedback on the effectiveness of prevention and treatment initiatives, can detect new outbreaks early and be used to inform decisions regarding future efforts and targets for resources. Surveillance presently relies upon reporting through treatment and control centers, but rabies is a disease of disproportionate consequence in low-income and rural areas where treatment is more difficult to access and therefore reporting is incomplete.

Prior to initiation of the REP, studies in Bangladesh established the high impact but low reporting of rabies in rural areas. Through the REP, Bangladesh has made progress towards elimination, but underreporting has not been evaluated and remains a threat to public health. It is unknown how many people continue to be bitten by dogs or die of rabies outside of the NRPCC and DRPCCs, leaving a lack of knowledge concerning the effectiveness of rabies reporting in Bangladesh. Therefore, a cross-sectional study has been designed with the objective of identifying regions of Bangladesh where rabies remains undetected or unreported. Key informants will be interviewed to identify dog-bite cases, and those patients will be invited to participate in a standardized questionnaire that will collect information regarding reporting, treatment, and factors that affected their decisions to seek treatment. Rabies reporting in Bangladesh is dependent upon patients accessing rabies control centers for treatment. Factors that impede treatment and therefore hinder rabies reporting will be identified so that future dog vaccination campaigns and treatment resources can be delivered to areas where the need is greatest. Finally, results of this study will be used to generate recommendations for improving rabies surveillance in Bangladesh.

Student Name: Jonathan Stone V'22

Mentor: Dr. Jonathan Runstadler

Funding Source: NIH

Title: Refining disease diagnosis through microbial profiling of the 2018 New England Unusual Mortality Event

Summary: Emerging infectious diseases, which are attributed to habitat loss and climate change are on the rise in both humans and wildlife. Marine mammal mass mortality events attributable to infectious disease are being increasingly observed and are raising concern for both conservation efforts and public health. Beginning in July 2018, an unusual mortality event (UME) along the northeast coast of the United States killed 1,400 seals. Preliminary screening determined the primary cause was phocine distemper virus (PDV). However, only 47.3% of seals screened were PDV positive and the results indicate a number of additional pathogens may be involved. We hypothesize that secondary infections played a major role in this UME. Specifically, this study aims to i) perform metagenomic analyses to identify potential secondary pathogens and ii) determine if a correlation exists between the presence of predicted pathogens and healthy seals versus seals that died in the UME.

During the initial UME response, samples were collected by Marine Mammals of Maine and sent to the Runstadler lab for testing. Of these samples, four were selected for exploratory sequencing from two harbor and two grey seals. We propose to analyze and compare these datasets using bioinformatic approaches to predict potential cofactors. RNA-seq datasets will be analyzed using multiple metagenomic programs including Kraken, QIIME, and Clark, as recent studies have shown a high degree of variability between bioinformatic algorithms. Primary descriptive statistics, including alpha diversity, will be calculated and qualitatively compared to identify potential pathogens. Any potential pathogens identified will be confirmed using reference-guided assembly.

Then we propose to test for predicted pathogens in samples from healthy seal pups and animals that died in the UME. Genes from predicted pathogens of interest will be selected and specific primer sets will be designed using primer-BLAST. A total of 120 seals will be tested using reverse transcription polymerase chain reaction (RT-PCR) and gel electrophoresis: 30 UME samples positive for PDV, 30 UME samples negative for PDV, and 60 samples from healthy seals. In addition to the UME samples collected by Marine Mammals of Maine, samples were collected from healthy gray seal pups in Cape Cod from 2015 to 2019 and samples from healthy harbor seals were collected from wildlife rehabilitation centers prior to their release. Inferential statistics will be used to determine if the prevalence of the predicted pathogen(s) is significantly higher in UME seals as compared to controls and if the pathogen(s) are involved in significant UME coinfection.

Student Name: Sophia Upton V'22

Mentor: Dr. Stephanie Borns-Weil

Funding Source: Elizabeth A. Lawrence Endowed Fund

Title: Factors That Influence the Comfort Level of ER Clinicians Deciding to Euthanize Aggressive Dogs

Summary: Specific Aims: To determine what factors, such as age, gender, years since veterinary school, and formal behavioral training, affect the comfort level of emergency room clinicians when requested by clients to perform euthanasia on a dog presenting for aggression.

Methods/Experimental Design: An electronic survey will be sent out to veterinary emergency rooms across the country. Multiple choice questions will be used to gather data on personal and educational background, including any behavioral training that the clinician experienced in veterinary school or as continuing education. Data will be analyzed to determine whether correlations exist between their background and their comfort level in responding to provided situational questions.

Significance:
There is currently no data on what factors play a role in clinicians making the decision to euthanize a dog for aggression. Aggression is a public health concern, however, some animals respond well to behavioral modification and do not need to be euthanized. It is important for veterinarians to make the right decision for the dog and the public. As not all veterinary schools in the United States provide formal behavioral training, this study will expose potential deficits in the veterinary school curriculum and any inconsistencies that exist in the decision making of the clinicians.

Student Name:	Sara Wlodarczyk V'21
Mentor:	Dr. Christopher Schonhoff
Funding Source:	Private Funding
Title:	Sex Differences in Oxycodone-Mediated Alterations in Rat Dendritic Morphology and Axonal Growth in a Primary Neuronal Culture System

Summary: Using primary hippocampal neuron cultures from rats, this study will explore the sex specific effects of oxycodone on dendritic arborization and axonal growth. The use of opioids has increased over the past two decades, due to its effectiveness in treating pain. Unfortunately, as a result, the number of children that have been prenatally exposed to opioids has risen as well. There has been a lack of data outlining the effects of oxycodone on dendritic morphology or axonal growth. In addition, there are sex-specific differences in brain structure, neuronal cytoarchitecture and synaptic connectivity, which is why it is important to look at sex specific impacts of opioids. Studying the sex specific effects of oxycodone on neuronal development will illuminate whether or not children are neurologically affected by prenatal opioid exposure, and if one sex is more impacted than the other.

The first experiment will look at the sex differences in dendritic arborization under the effects of oxycodone. Primary hippocampal neurons will be treated with three different concentrations of oxycodone over varying lengths of time. Neurons will be illuminated with fluorescence, and their morphology will be observed under a fluorescent microscope. The second experiment will determine if there are sex differences in axonal lengths under different concentrations of oxycodone. Hippocampal neurons will be exposed to oxycodone for 3 hours and will then be observed for any sex-specific differences in axonal length. Immunostaining the neurons with an antibody for Tau-1 will allow for observation of the neurons, and axonal lengths will be determined from neuronal images. Ultimately, it is suspected that there will be sex-specific

differences of dendritic arborization and axonal length in hippocampal neurons after oxycodone exposure.