2016 ISU CVM SSRP Mentor Abstract #1

Project Title:
Efficacy and Safety of Topical Nitric Oxide Donors in Lowering Intraocular Pressure in Canine Eyes

Principle Investigators:
Rachel Allbaugh, DVM, MS, DACVO (VCS)
Travis Strong, DVM, MS (VCS)

Abstract:
PURPOSE AND SIGNIFICANCE: The intent of the proposed research is to evaluate the efficacy and safety of topical nitric oxide donors at lowering the intraocular pressure in normal canine eyes. Growing evidence from various animal and human-based studies indicate a multitude of roles for nitric oxide in regulating the intraocular milieu. Of particular note, nitric oxide appears to be critical in the normal physiologic regulation of aqueous humor outflow. Treatment with topical nitric oxide donors in various laboratory animals has been shown to relax trabecular meshwork cells and decrease intraocular pressure. To evaluate the potential for topical nitric oxide donors in common veterinary patients, the ISU Ophthalmology Service is currently studying the expression of enzymes involved in the nitric oxide pathway in normal canine eyes. In addition, we aspire to evaluate the ocular effects of topical nitric oxide donors in healthy canine subjects. Recent preliminary studies conducted by the ophthalmology service with student-owned dogs have demonstrated a predictable decrement in intraocular pressure following treatment with a topical formulation of nitroglycerin. Additionally, no signs of ophthalmic or systemic complications have been observed. Given these promising early results, we strive to more formally evaluate the efficacy and safety of topical nitric oxide donors in healthy canine eyes. MATERIALS AND METHODS: Staff/student-owned dogs will be evaluated in this study. Dogs will be divided into four groups: a control group receiving vehicle and three treatment groups receiving nitroglycerin, hydralazine, or sodium nitroprusside. Prior to treatment, complete eye examinations will be performed and cardiovascular parameters will be assessed. Only dogs with normal eye exams and healthy cardiovascular parameters will be employed in this study. EXPECTED OUTCOME: Canine eyes treated with nitric oxide-donating medications will have demonstrably lower intraocular pressures compared to the eyes of control dogs. No significant ophthalmic or cardiovascular side effects are expected.
Project Title:
Clinical and pathological characterization of ophthalmic disease in a canine model of mucopolysaccharidosis type I.

Principle Investigator(s):
Gil Ben-Shlomo (VCS)
Jodi Smith (VPTH)

Collaborating Investigator:
N. Matthew Ellinwood (AnSci)

Abstract:

The mucopolysaccharidoses (MPS) are a group of lysosomal storage diseases, of which there are many subtypes including MPS I. Mucopolysaccharidosis type I (also Hurler, Scheie, or Hurler-Scheie syndromes) is a rare and potentially fatal pediatric disease caused by a defect in the enzyme α-L-iduronidase. Enzyme deficiency results in glycosaminoglycan (GAG) accumulation in various cell types throughout the body, including ocular tissues. Currently available treatments for MPS I are suboptimal and do not address ophthalmic disease. Affected individuals suffer primarily from corneal disease as a result of GAG accumulation leading to corneal opacity, but other less commonly observed ophthalmic changes include visual impairment, optic disc abnormalities, retinopathy, and ocular hypertension. This research project will focus on the characterization of clinical and pathological changes in the eyes of a canine model of MPS I. This data will serve as the basis for future studies investigating the efficacy of a stem cell-based gene therapy approach using mesenchymal stem cells expressing the missing lysosomal enzyme α-L-iduronidase. The student participating in this project will gain experience in clinical ophthalmology, ocular ultrasound, electroretinography, and ocular histopathology.
Project Title:
The validation of percutaneous testing in horses and comparison of applicator devices.

Principal Investigator:
Darren Berger (VCS)

Collaborating Investigator:
David Wong (VCS)

Abstract:
Equine allergic diseases are common and present with a wide array of manifestations such as: recurrent airway obstruction, urticarial reactions, and excessive pruritus. Currently, identification of potential offending environmental substances in the horse is performed via serologic or intradermal allergy testing (IDT). Percutaneous testing (PT) is the standard technique used in human medicine for confirmation of allergic reactions and is performed using a special applicator, which contains a concentrated amount of allergen to scratch/prick the surface of the skin. PT offers several potential advantages including: time to perform the test, patient comfort, safety, and the use of glycerinated extracts, which provides greater extract stability and decreased costs. In addition to these advantages, percutaneous testing in people has been demonstrated to correlate better with clinical allergy than other testing modalities. The purpose of this study is to demonstrate which commercially available PT applicator is most appropriate for use in the horse and to compare the response of ten clinically healthy horses to PT and IDT performed with control solutions to determine whether results are interpretable and thus whether PT warrants further investigation as a diagnostic test in horses.
Project Title:

Principle Investigator:
Stephanie S. Caston (VCS)

Collaborating Investigator:
Jennifer A. Schleining (VDPAM)

Abstract:

Navicular disease is a common cause of lameness in horses and is a term used to describe pain originating from the distal sesamoid bone (navicular bone) or associated soft tissue structures that make up the ‘navicular apparatus’. The incidence of lameness caused by pain originating from these structures has been theorized to be higher in the Quarter Horse breed. However, a heritable cause has not been identified or investigated in Quarter Horses to our knowledge. Identification of possible genes associated with navicular disease have been described in the Hanoverian warmblood horse. Our study will investigate the incidence of navicular disease in horses presented to Iowa State University Veterinary Medical Center over a 10 year period and assess a possible heritable cause. Search terms including “navicular disease”, “navicular syndrome”, “heel pain” and “caudal heel pain” will be used to select electronic medical records. Inclusion criteria for diagnosis of navicular disease will include: lameness localized to the heel region and radiographic or MRI findings consistent with navicular disease. Electronic medical records will also be evaluated for all horses presented for lameness in the same period to evaluate incidence of navicular disease among our hospital caseload of lameness cases. Signalment, treatments performed, and number of visits will be recorded in a spreadsheet for analysis. In addition, cases with a diagnosis of navicular disease that are listed as Quarter Horses will be separately evaluated and incidence of the condition in these horses will be compared to other breeds. Follow up with owners will be pursued to record outcome and registered name of Quarter Horses (thereby allowing analysis of pedigree). Heritability analysis will be performed using Bayesian analysis. Analysis of variance methods will be used to compare other variables.
Project Title: 
Comparison of post-mortem *Mycoplasma hyopneumoniae* diagnostic tools

Principle Investigator: 
Rachel J. Derscheid (VDPAM)

Abstract:

*Mycoplasma hyopneumoniae* is the etiologic agent of enzootic pneumonia in swine. This disease is a persistent problem in grow-finish swine operations around the world. One of the issues with prevention and treatment is the difficulty in diagnosing the disease within a herd. Diagnostic modalities currently utilized at Iowa State University Veterinary Diagnostic Laboratory (ISU VDL) on post-mortem samples from swine suspected of having *M. hyopneumoniae* are immunohistochemistry (IHC), polymerase chain reaction (PCR), culture, and histopathology. Understanding the correlation between these tests would improve diagnostic outcomes for producer, practitioner, and diagnostician, as well as form the framework for evaluation of additional post-mortem diagnostic assays such as fluorescent in situ hybridization (FISH) as well as ante-mortem diagnostic testing (e.g., serum, nasal swab, or oral fluid PCR, culture, or ELISA). Using stored samples and results from a combination of experimentally inoculated swine as well as ISU VDL cases, IHC, PCR, culture, and histopathology will be evaluated in context to each other.
Project Title:
The comparative medical genetics of an untreatable fatal human neurodegenerative disease of childhood: Mucopolysaccharidosis type III in murine models for developing therapy and understanding pathogenesis.

Principle Investigator:
N. Matthew Ellinwood (AnSci)

Abstract:
The mucopolysaccharidoses (MPSs) are a group of lysosomal storage disorders resulting in an accumulation of glycosaminoglycans (GAGs). This storage leads to organ dysfunction resulting in both somatic and CNS signs. Our lab primarily works on using animal models (canine and murine) to study different MPS disorders with a focus on MPS III (Sanfilippo syndrome) which is a fatal and untreatable neurodegenerative disease of childhood. We work on characterizing and investigating animal models for MPSs and also developing therapeutic techniques. Our murine colony consists of MPS I, IIIA, IIIB and IIID lines as well as GalNAcT line which lacks complicated gangliosides, a type of lipid critical for normal brain function. Currently we are in the process of generating a double knockout (DKO) mouse model of MPS IIID and GalNAcT to help us unravel new findings on the relationship of white matter pathology in the MPS III disorders. The summer scholar will work on the breeding strategies, colony management, genotyping and behavioral studies of the DKO line. One will also get an experience in biochemical analysis; enzyme assays, GAG assays, HPTLC; as well as in histology and pathology; sectioning and staining the tissues. Hence, one will be able to learn general lab techniques from extracting DNA, PCR, gel electrophoresis, assays, sectioning and staining, perfusions and cell culture. Furthermore, our lab also works with canine models of MPSs, this will give an opportunity to the student to get familiar with research in large animal models including colony management and necropsies. This is an ideal setting in which to both experience varied research techniques and be involved with comparative biomedical medicine research that targets both mechanisms of disease as well as therapy development.
Project Title:
Comparative pharmacokinetics, milk and tissue disposition of Ceftiofur crystalline free acid and its impact on antimicrobial resistance selection in healthy and diseased dairy cows

Principle Investigator:
Patrick Gorden (VDPAM)

Collaborating Investigator:
Johann “Hans” Coetzee (VDPAM)

Abstract:

Mastitis is one of the most common diseases of dairy cattle. Many cases are from coliform bacteria and have a severe presentation. Erskine et al. (JDS, 2002) reported a reduction in the proportion of cows that were culled as a result of severe coliform mastitis when treated with systemic ceftiofur as compared to cows that were not treated.

Ceftiofur, a third generation cephalosporin, is widely used in the dairy industry. Ceftiofur dosage regimens and withdrawal times are established after pharmacokinetic and residue depletion studies conducted in healthy animals. However, veterinarians treat diseased cattle that exhibit significant physiological health derangements compared to healthy animals. In previous work, our group has demonstrated that these derangements significantly alter drug pharmacokinetics of ceftiofur hydrochloride (Gorden, et al., JDS, http://dx.doi.org/10.3168/jds.2015-10239).

In FY14, ceftiofur was cited as the cause of violative drug residues in meat in 44% of the total cull dairy cows with violations, making it the most commonly found violative residue in this production class. The hypotheses for this study are that: 1) ceftiofur will have a higher volume of distribution in cows affected with severe clinical mastitis resulting in altered pharmacokinetics and tissue deposition of ceftiofur crystalline free acid compared to healthy cows necessitating variance in dose regimen and withdrawal periods; and 2) antimicrobial selection pressure applied by ceftiofur crystalline free acid is the same in healthy and diseased cattle and will return to pre-treatment levels following treatment.

Cows with naturally occurring, severe mastitis will be treated with on-label doses of ceftiofur crystalline free acid. At the same time, control cows will be enrolled to provide comparisons in pharmacokinetics and antimicrobial resistance selection. This will provide knowledge of the pharmacokinetics, residue depletion, and antimicrobial resistance of ceftiofur in diseased dairy cattle that is critical for veterinarians to remain in compliance with legislation.

Summer Scholar student responsibilities for the project

The Summer Scholar student for this trial will be responsible, along with other study personnel, for animal enrollment, on-farm animal sample collection, sample analysis, data collection and some general cow care.
Project Title: Identification of modalities to monitor bull mounting behavior on beef cows.

Principle Investigator: David K Hardin (UNL)

Collaborating Investigator: Richard Randle, Brad White, Robert Larson, and Kaitlynn Abel (UNL)

Abstract:

The predominant factors determining revenue in a commercial cow-calf operation are the number of calves sold, their weight and sale price per pound (Garrick and Golden, 2009). To maximize revenue, high conception rates early in the breeding season are required, (Funston, 2012) along with the ability to pass on genes for superior performance of the offspring. Therefore, high fertility in cows and bulls is essential. It has been known for some time that socio-sexual behavior of bulls in a multi-sire breeding pasture influences the sexual activity and reproduction performance (Blockey, 1979) of the individual bulls. Due to labor intensiveness, very little research has been conducted in this area since the 1980’s. Recent advancements using DNA to determine parentage and electronic sensors to study animal behavior have created a renewed interest in this area. Parentage data recently obtained from US MARC on 33 bulls that were placed with cows in 5 multi-sire breeding pastures, was recently evaluated for individual bull breeding performance. The number of cows per bull was proximately 23:1 with the number of bulls per pasture ranging between 6-7 bulls. The top two bulls in each breeding pasture sired 90 calves (64%); 51 calves (53%); 102 calves (72%); 37 calves (56%), and 82 calves (66%) respectively. In contrast the bottom two bulls in each breeding pasture sired 4 calves (2%); 17 calves (17%); 6 calves (4%); 6 calves (10%) and 8 calves (7%) respectively. The mean number of calves sired per bull varied widely, ranging from 66 calves to 1 calf (unpublished). Studies conducted spring and fall of 2015 have demonstrated the feasibility of using electronic sensors to monitor bull mounting behavior in a controlled (small paddock) environment. The project objective for this summer is to evaluate the system under field conditions using three bulls running with 80 cows. The long term objective is to develop a model that can be used to study the behavioral differences between high performing and low performing bulls leading to the development of selection and/or management strategies that would improve overall bull performance, thus allowing for higher cow to bull ratios and adding superior genetics to the gene pool.
Project Title:
Role of gastrointestinal dysbiosis in feline intestinal neoplasia

Investigators:
Albert Jergens, Kayode Garraway, Chad Johannes, Todd Atherly (VCS)

Abstract:
The mammalian gastrointestinal (GI) tract hosts a complex and structured community of microorganisms colonizing the small and large intestines with over $10^{14}$ bacteria existing in a symbiotic environment. These bacteria are believed to be associated with maintenance of GI mucosal homeostasis and immunologic tolerance. This same microbial community is also involved in the intricacies of progressive inflammation, mediation of oncogenic effects and suppression of tumourneogenesis. Accumulating evidence in human and animal models of intestinal inflammation suggests that imbalances (abnormal and disproportionate bacterial populations) in the composition of the intestinal microbiota contribute to dysregulation of host immune response, chronic or recurrent inflammation and eventual progression to cancer. Therefore it is believed that there is enrichment of select bacterial species associated with deleterious immune modulation leading to tumor development.

The aim of the proposed study is to identify changes in the mucosal microbiota composition and aberrant immunologic mechanisms contributing to development of GI cancers (lymphoma, adenocarcinoma, mast cell) in cats. Identification of the potential negative effects of microbiota dysregulation may provide insights for implementation of early preventative measures of gastrointestinal neoplasia.

The student scholar will perform molecular studies on archived and recently collected endoscopic biopsies of intestines in cats diagnosed with gastrointestinal lymphoma, adenocarcinoma and mast cell cancer. More specifically, the student will perform culture-independent 16S rRNA techniques (fluorescence in situ hybridization [FISH]) on tissues identifying the mucosal microbiota of feline alimentary cancer. The project will allow for direct interaction with veterinary internists, oncologists and house officers relative to the diagnosis and management of gut cancer in clinical patients. Participation in gastrointestinal endoscopic procedures (using endoscopic equipment to observe the gastrointestinal tract in real time on anesthetized cats) at the CVM and collection of fresh gastrointestinal biopsy samples for analysis is anticipated and encouraged.

A. Endoscopic erosions; B. H&E of homogenous lymphocytic infiltrate; C. IHC immunophenotype (CD3+ T-cells are >90% and suggest intestinal lymphoma; D. FISH analysis of GI microbiota.