Acoustoelastography and Shear Wave Imaging: Novel Methods of Quantitative Evaluation of Tendon Strength by Mapping Tendon Elasticity via Ultrasound

Principal Investigator
Adam Biedrzycki, Department of Surgical Sciences

Abstract
Currently, monitoring protocols after tendon injury are based on ultrasonography evaluations of tendon. Unfortunately, these do not provide the clinician with any idea of how well the tendon is healing, it just tells them how well any gaps are filling in. Currently in human medicine, the technology exists to evaluate how hard or soft tissue is based on ultrasound. We aim to evaluate two of these techniques, termed shear wave imaging and acoustoelastography, to determine if we can accurately predict the ultimate tensile strength and stiffness of tendon. Currently, the only way to determine these values is to remove the tendon from the animal and test it in a machine. If successful, these techniques would be highly beneficial in both human and animal patients, where rehabilitation can be matched to how quickly or slowly a tendon heals, minimizing the risk of reinjury, in addition to reducing the number of animals needed for tendon research.

Development of Bone Deformity Profiles Using Computed Tomography and 3D Models for Limb Alignment Correction

Principal Investigator
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Abstract
Orthopaedic growth deformities are common debilitating conditions that affect young growing dogs. These can result in considerable pain, lameness and result in loss of the affected limb if untreated in severe cases. Treatment typically involves a surgical corrective osteotomy of the deformed bones to realign the limb and improve function.
This presents a unique challenge to the orthopedic surgeon, as accurate planning involves advanced CT imaging and measurements of the correction in 3-planes. There are no robust methods of quantifying limb deformity in the forelimb of the dog, which represents the most common limb malformation. This study seeks to develop a novel method of anatomic characterization to quantify limb deformity in dogs. We will use CT imaging, 3D image analysis software, and 3D bone models to accomplish this aim. This project has the potential to greatly improve surgical outcomes in limb deformity correction in small animals.

In-vivo Evaluation of Acoustoelastography (AEG) in the Normal and Abnormal Equine Suspensory Ligament.

Principal Investigator
Sabrina Brounts, Department of Surgical Sciences

Abstract
Damage to the suspensory ligament (SL) is the second most common injury to tendons/ligaments in athletic horses. This injury results in a large degree of morbidity due to a lengthy rehabilitation process and high risk of re-injury. Recovery requires a balance between rest and controlled exercise to maximize function of the healing ligament. Serial ultrasound examinations are performed to monitor for re-injury; however, it is difficult to determine the level of exercise damaged tissue can sustain from anatomic changes alone. Biomechanical understanding of normal and damaged tissue is therefore necessary.

Translation to clinical application requires a noninvasive method to examine mechanical changes. Acoustoelastography (AEG), a novel ultrasound-based method, relates changes in reflected ultrasound waves in stretching tendons to their biomechanical properties. This is similar to changes in pitch of a guitar string when it is tightened. Ultrasound echo intensity has been related to tendon stress and strain, and AEG has accurately modeled strain and stiffness within tendons in vitro and therefore has potential to noninvasively analyze mechanical compromise in injured tissue. Previous experiments have demonstrated that stiffness and strain could be measured reliably in normal equine superficial digital flexor tendons; however this method has not been applied to deeper structures such as the SL.

The goals for this project are to measure stiffness in normal equine SL and measure changes in stiffness in SL with desmopathy using ultrasound-based AEG methods. This study will further the use of AEG for non-invasive evaluation of tendon/ligament function that could vastly improve the ability to detect, monitor, and treat injuries.
The Effects of Acupuncture on Clinical Outcome Measures in a Randomized Controlled Study Using Client-owned Dogs with Hip Osteoarthritis

Principal Investigator
Lindsey Culp-Snyder, Department of Surgical Sciences

Abstract
Osteoarthritis (OA) is a common progressive orthopedic disease of dogs causing intense, often debilitating chronic pain, and leads to damage of the affected joints. Acupuncture (AP) has been used for the treatment of acute and chronic pain in humans for thousands of years. We will evaluate client owned dogs with established OA to determine if AP and electroacupuncture (EA) therapy will improve the clinical outcome measures in dogs with hip osteoarthritis. Through acupuncture’s analgesic effects, we anticipate that the AP/EA treatment group will have better outcome measures than the control group. For dogs with OA that have negative side-effects associated with analgesic drugs, AP may provide a significant improvement to their quality of life. Additionally, if we prove AP to be beneficial in OA, we will provide an adjunct to medical treatment of OA, potentially benefitting a large population of dogs.

The Effect of Regional Perfusion of the Distal Limb with Amikacin Sulfate on Bioburden in Contaminated Wounds in Horses, and a Descriptive Study of the Varying Effects of Two Common Bacterial Isolates from Chronic Wounds on the Gross and Histologic Features of Granulation Tissue in Horses.

Principal Investigator
Samantha Morello, Department of Surgical Sciences

Abstract
Healing of distal limb wounds in horses can present a serious challenge to veterinarians and owners, especially in the face of infection. Bacterial infections in the distal limb do not always respond well to systemic antibiotic administration, yet topical therapies are often ineffective at penetrating biofilms and deeper tissues of the wound. Regional limb perfusion (RLP) has been used extensively in equine practice to deliver high doses of antimicrobials to bones, joints, and other soft tissues in the distal limb. This study aims to determine the direct, quantitative effects of RLP on bacterial numbers within wound beds. We will simultaneously evaluate the histologic features of wounds chronically infected with the two most common bacterial isolates, Staph aureus and Pseudomonas, in order to try and understand how tissue responds differently to various bacteria, in order to design better treatment strategies.
Genotypic Dissection of Acquired Laryngeal Paralysis in the Dog

Principal Investigator
Peter Muir, Department of Surgical Sciences

Abstract
Opening and closing of the laryngeal cartilages in the throat is an essential part of normal breathing. Acquired laryngeal paralysis (ALP) is an important cause of respiratory distress in dogs, in which the laryngeal muscles become paralyzed. Affected dogs suffer breathing difficulties and many die from airway obstruction. The cause of this condition is not understood. ALP develops by 11 years of age as part of a generalized loss of peripheral motor nerve function (polyneuropathy). ALP is common in the Labrador retriever. The marked breed incidence suggests a genetic basis for the disease. In this project, we will compare the genome of ALP-affected and unaffected Labradors using new genotyping technology. To identify the causative gene mutation, the genomic regions that show association with ALP will be sequenced. A genetic test for ALP can easily be developed from this research that will be broadly applicable to all affected breeds.

Amphibian Analgesia: Evaluation of Safety and Efficacy of Oral Tramadol in White’s Tree Frogs (Litoria Caerulea)

Principal Investigator
Kurt Sladky, Department of Surgical Sciences

Abstract
The goal of this study is to determine the safety and efficacy of tramadol in amphibians using White’s tree frogs (Litoria caerulea) as models. Amphibians are kept as companion animals, displayed in zoos and aquaria, and used extensively in research, but our knowledge of amphibian analgesia is inadequate. Previously published studies were primarily focused on nociceptive pathways, cloning opioid receptors, or developing a non-mammalian, analgesic model. However, few studies have determined effective analgesic dosages in clinical amphibian medicine. Tramadol has not been systematically evaluated in amphibian species, but is a good candidate drug as it is non-controlled, commonly used in most veterinary clinical facilities, and is effective in aquatic reptiles. Tramadol provides analgesia by activating mu opioid receptors and inhibiting serotonin and norepinephrine reuptake. Mu-opioid receptors are present in several frog species. In addition, the oral dosing of tramadol would provide an easy and less stressful delivery alternative to injections.
**Evaluating the Ability of a Small Molecule, Serine-threonine Kinase Inhibitor, to Inhibit Canine Malignant Melanoma Tumor-initiating Cells**

*Principal Investigator*
Timothy Stein, Department of Medical Sciences

*Abstract*
Canine malignant melanoma is a highly aggressive malignancy associated with poor overall survival due to local disease recurrence, highly metastatic nature, and poor response to conventional anti-cancer therapies. Disease recurrence, metastasis, and chemoresistance may be due in part to the presence of a subpopulation of tumor-initiating cells. Recently, tumor-initiating cells have been identified in canine malignant melanoma. The identification of compounds capable of targeting tumor-initiating cells may improve the treatment and subsequent outcomes for dogs with malignant melanoma. We have preliminary evidence suggesting the small molecule serine-threonine kinase inhibitor, 6-bromoindirubin-3'-oxime (BIO), is capable of inhibiting tumor-initiating cells in osteosarcoma. Additionally, our prior work with BIO has demonstrated it has activity against canine oral melanoma cell lines. This project will determine the impact of BIO treatment on canine malignant melanoma tumor-initiating cells. It is possible therapies capable of inhibiting tumor-initiating cells will improve patient outcomes relative to currently available treatments.

**Serum Biomarkers of Histopathologic Severity in Canine Liver Disease**

*Principal Investigator*
Lauren Trepanier, Department of Medical Sciences

*Abstract*
Chronic liver diseases are common in dogs, with varying levels of necrosis, inflammation, and fibrosis on liver biopsy. Response to treatment is ideally monitored by serial liver biopsies, which can be invasive and expensive. In humans, several serum biomarkers have been correlated on liver histopathology with the degree of necro-inflammatory activity and fibrosis, which are two key targets of therapeutic intervention. These biomarkers include C-reactive protein (CRP), interleukin 6 (IL-6), CCL2 (chemokine ligand 2), and the ratio of serum AST:ALT (two liver enzymes found on routine biochemical panels). We hypothesize that these same biomarkers will correlate with the severity of histopathologic findings in dogs with liver disease. To address this hypothesis, we will correlate serum CRP, IL-6, CCL2, and the AST:ALT ratio with the degree of necro-inflammatory activity and fibrosis in 100 dogs undergoing liver biopsy by laparoscopy or laparotomy, using the METAVIR system for liver histopathologic scoring. We expect that, as in humans, CRP will correlate with both necro-inflammatory activity and fibrosis on liver biopsy. We anticipate that IL-6 will be significantly higher in the presence of cirrhosis on biopsy even when controlled for the presence of hepatic
encephalopathy. We further hypothesize that increased CCL2 concentrations will be associated with higher necro-inflammatory activity scores on biopsy. Finally, we expect that an AST:ALT ratio > 1.0 will correlate with higher fibrosis scores (F3/F4), and may have high specificity for the presence of cirrhosis. The goal of this study is to provide clinically useful tools for monitoring treatment protocols targeting necrosis, inflammation, and fibrosis in dogs with liver diseases.

The Use of Circulating Biomarkers of Oxidative Stress as an Assessment of the Redox Status of the Liver in Dogs with Naturally Occurring Liver Disease

Principal Investigators
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Sarah Dowling, Department of Medical Sciences

Abstract
Liver disease is a common problem within the dog population. There is increasing evidence that oxidative stress plays an important role in acute and chronic liver disease. The main intracellular antioxidant produced by the liver, glutathione, is decreased in both experimentally induced and naturally occurring liver disease. It is unknown whether circulating glutathione levels correlate with liver tissue concentrations. The medical management of liver disease includes antioxidant supplementation (ie Denamarin®, Denosyl®, vitamin E, etc.), however there is no validated way to measure response to therapy. The objective of this study is to determine whether circulating antioxidant levels can be markers of liver glutathione tissue levels. The results of this study will further our understanding of the oxidative status of dogs with liver disease and help to determine whether blood levels can be used to evaluate the need for, and response to, antioxidant supplementation in dogs with liver disease.

Evaluation of the Effects of Time, Temperature, and Sample Storage on In Vitro Lactate Concentrations

Principal Investigator
Julie Walker, Department of Medical Sciences

Co-Investigator:
Jonathan Bach, Department of Medical Sciences

Abstract
Lactate is produced by the body’s tissues in times of insufficient oxygen delivery. In the clinical setting, lactate is used as a marker of poor tissue perfusion. The measurement of blood lactate levels is used to guide intravenous fluid therapy and serve as an indicator of prognosis in various diseases. Red blood cells may continue to produce lactate after a
blood sample has been collected, which may contribute to falsely elevated results. While short-term room temperature storage of blood samples has been shown to falsely elevate lactate in humans and cats, the extent to which this occurs in dogs is currently unknown. Accurate measurement of lactate is vital when critical decisions are to be made regarding prognosis or therapy. Our study will evaluate various techniques in blood sample handling following collection to better understand the factors that cause artifact during lactate measurement in dogs.