Project Title: **Use of a gnotobiotic mouse community to understand host/microbiome interactions**

Principle Investigator(s): **Gregory Phillips, Ph.D.**

Collaborating Investigator(s): **Michael J. Wannemuehler, Ph.D.**

Abstract: (300 words or less): **Recent experimental evidence reveals that the bacterial communities (microbiota) that comprise the mammalian gastrointestinal (GI) tract can have a profound influence on the health of the host. Diseases ranging from colorectal cancer to inflammatory bowel diseases (IBD) have been linked to an abnormal microbiota (dysbiosis) in humans and animal models. Despite the importance of bacteria to the wellbeing of humans and animals alike, how the microbiota influences health and disease is the subject of current research efforts. To better understand how specific bacteria interact with the host, we are using a unique gnotobiotic mouse community, the Altered Schaedler Flora (ASF), which is comprised of animals colonized with only 8 known bacterial species. Despite the low complexity of the microbiota, ASF mouse exhibit normal immune system development and growth. Use of this resource includes monitoring the changes in relative number, spatial distribution and gene expression in response to alterations in diet and following infection with bacterial pathogens. Independent student projects include, but are not limited to, use of quantitative PCR to measure changes in the abundance of individual ASF community members in response to infection with bacterial pathogens, as well as identifying genetic changes in the ASF that occur in response to perturbations to the GI tract. The ASF model also offers the potential to study new results that indicate that the composition of the GI microbiota may actually influence animal behavior. The overall impact of these studies will lead to a better understanding of how the GI microbiota influences human and animal health and disease.**
Methicillin resistant *Staphylococcus* species (MRS) from companion animals.

Funded by USDA NIFA: funding is available to support a scholar’s stipend for the entire 13-week program.

We have not discussed this project with any first- or second-year veterinary students

Principal Investigator: Orhan Sahin, VDPAM

Co-PIs: Qijing Zhang, Adam Krull, Eric Burrough, Paul Plummer, and Grant Dewell

Methicillin-resistant *Staphylococcus* spp. (MRS) (primarily methicillin-resistant *Staphylococcus pseudintermedius* [MRSP] and to a lesser extent methicillin-resistant *Staphylococcus aureus* [MRSA] and coagulase negative *Staphylococcus* spp. [MRCNS]) are among the leading causes of both community and hospital-acquired skin, ear and wound infections in dogs and cats with zoonotic potential. Resistance to methicillin in Staphylococci is typically mediated by *mecA* gene, carried on the staphylococcal chromosomal cassette mec (SCCmec), which encodes PBP2a that has a low affinity for majority of other beta-lactam antibiotics. Similar to the worldwide trend of increasing methicillin resistance in Staphylococci (up to 50% in some regions), we have also seen a substantial increase in methicillin resistance in *Staphylococcus* spp. isolated from diagnostic submissions of dogs and cats at the Veterinary Diagnostic Laboratory of Iowa State University (ISU VDL). Considering the fact that MRS are also frequently resistant to other important veterinary antibiotics such as aminoglycosides, fluoroquinolones, lincosamides, macrolides, tetracyclines, chloramphenicol and trimethoprim-sulfamethoxazole, there is only a limited option to treat such serious infections caused by these pathogens. The goal of this study is to determine prevalence of methicillin resistance in *Staphylococcus* spp. isolated from the clinical specimens of dogs, cats and other companion animals submitted to ISU VDL. The mechanism of methicillin resistance will also be investigated using phenotypic and genetic tests. Furthermore, the genetic relationship of MRS isolates will be determined. Antimicrobial susceptibility testing (AST), latex agglutination assay, PCR and molecular typing methods will be employed to ascertain these attributes. The findings of this study will generate significant knowledge on the prevalence, mechanism and genetic diversity of MRS from pets with severe infections and thus should be valuable for therapeutic and epidemiological purposes.
Project Title: **Experimental strategies for brain rescue and repair**

Principle Investigator(s): Don Sakaguchi  
(http://dssakagu.public.iastate.edu/Sakaguchi/)

Collaborating Investigator(s):

Abstract: This highly interdisciplinary project seeks to develop approaches to facilitate rescue and repair of the damaged and diseased nervous system. As a potential treatment strategy for neurodegenerative diseases and injury, stem cells and progenitor cells have been proposed as unique sources of transplantable cells for a therapeutic approach. A goal of this strategy is for the transplanted cells to provide neuroprotection and/or to replace degenerating cells in the diseased or damaged nervous system. We are implementing these procedures using rodent models of neurodegenerative conditions associated with ocular diseases, severe nerve damage and Parkinson’s disease. Our working hypothesis is that the transplanted cells can provide neuroprotection or replace lost cells and restore function in the diseased/injured nervous system. The intern will have the opportunity to be part of an active research team. Intern will have the opportunity to observe and learn the following techniques: general laboratory procedures, data collection and analysis, mammalian cell culture, cellular reprogramming strategies (genetic and small-molecule approaches), immunocytochemical procedures, fluorescence microscopy, image analysis, and tissue engineering strategies.  
Questions about this research opportunity may be directed to Don Sakaguchi (email: dssakagu@iastate.edu).
Project Title: 16S based metagenomic analysis of the etiology behind turkey cellulitis

Principle Investigator(s): Yuko Sato, Adam Krull

Collaborating Investigator(s): Ganwu Li, Bailey Arruda, Orhan Sahin, Paul Plummer, Jeff Zimmerman, Ju Ji, Nancy Cornick

Abstract: (300 words or less):

Turkey cellulitis (TC) is a condition which causes mortality and plant condemnation in market age turkeys. It is a nationwide health concern and ranks as the third most important issue facing the turkey industry\(^1\). When long-term antimicrobial therapy such as penicillin is initiated to control this mortality, overall costs of treatment and bird loss may be as high as $16,000-$18,000 per affected flock. *Clostridium septicum* is one of the most common pathogens associated with TC (84.1% in WI/MN study\(^2\)), although other bacteria have been isolated. Our goal is to evaluate the etiology of the disease by using 16S based metagenomic community profiling to look at affected vs unaffected birds at commercial turkey sites. The source of infection has been hypothesized to be bacteria contaminating the environment and serving as a source for oral infection\(^3\). In this study, we intend to look at 1) role of “flock-level” and environmental microbiota in its contribution to clinical disease and interactions with oral penicillin, 2) intestinal microbiota comparison between affected and unaffected birds, and 3) environmental microbiota comparison between affected and unaffected barns.

The selected student will be working closely with several commercial turkey producers in Iowa to collect weekly cloacal and fecal samples from 10-15 week old turkeys, as well as litter samples from the same barns. In addition, the student will be monitoring flocks for the development of TC lesions with the help of the flock supervisors, performing post-mortem examination, and tissue sampling from affected turkeys. When the proposed research is completed, we will have a better understanding of the complete etiology of TC, microbiota changes within the gut of affected turkeys to help determine risk factors for disease development, and evaluate the effect that oral penicillin therapy has on changing the gut microbiota and aiding in resolution of disease.


\(^2\)Bedford PD, Wells SJ, Hennessey M, Oliveira S, Costa M, Been C, Porter R. Descriptive Epidemiology of Turkey Cellulitis in Minnesota and Wisconsin Turkey Flocks.

Project Title: **Incidence and characterization of lameness in beef cattle**

Principle Investigator(s): Jennifer Schleining, DVM, MS, DACVS-LA

Collaborating Investigator(s): Amanda Kreuder, DVM, PhD, DACVIM (LAIM)
Joe Smith, DVM, MPS, DACVIM (LAIM), CVMA

Abstract: (300 words or less):

Lameness is one of the leading causes of decreased production and culling in cattle operations. While there is extensive research and literature describing the economic impact, welfare concerns, and characterization of lameness in dairy herds, relatively little information exists on the incidence and characterization of lesions in commercial beef cattle operations. Lameness is one of the leading causes of presentation of beef breeds to the Iowa State University Food Animal and Camelid Hospital. Our heavy caseload thus provides a valuable, yet currently underutilized, resource in regards to identification of the most common causes of lameness in beef cattle. Identification of the most common causes of lameness in beef cattle will then allow for improved treatment and management strategies to be developed specifically for beef herds. The goal of this project is to identify and characterize the causes of lameness in beef cattle through conducting a 15-year retrospective analysis (2001-2016) of the medical records of all lame beef cattle presenting to the Iowa State University Food Animal and Camelid Hospital. This analysis will serve three purposes – 1. Identify and characterize the causes of lameness in beef cattle to contribute to the very small existing body of evidence regarding beef lameness, 2. Provide a complete database of lesions that we can further utilize to determine treatment outcomes for specific disease, and 3. Help drive the development of a contemporary medical record system to better characterize lameness diagnoses and foot lesions in cattle to use in prospective lameness studies.
Project Title: Towards a better understanding of corneal sequestrum and symblepharon in cats: pre-clinical and clinical evaluation.

Principle Investigator: Dr. Lionel Sebbag

Collaborating Investigator: Dr. Rachel Allbaugh

Abstract:

**Corneal sequestrum** is a common disorder in cats and occurs due to chronic irritation from feline herpesvirus-1, non-healing corneal ulceration, or entropion. The condition is characterized by an area of corneal degeneration with a brown-to-black discoloration. Determining the source of the coloration could lead to a method of preventing sequestrum formation after chronic corneal ulceration or other ocular irritation, thus avoiding surgery or lengthy medical management. Unfortunately, the source of coloration remains an enigma to date. In humans, long-term use of topical epinephrine is reported to cause corneal discoloration attributed to oxidation of epinephrine to an adrenochrome pigment. We hypothesize that a similar process is occurring in cats, and the study proposes to collect and analyze tears of normal cats and cats affected with corneal sequestra.

**Symblepharon** is another ocular surface disease in cats, especially in kittens affected by feline herpesvirus-1. The virus causes ulcerations of the cornea and conjunctiva, and the ulcerated areas can form permanent adherences to one another. Simply breaking down the adherences can result in transient relief, but recurrence rates are very high. As a collaborative effort between the Primary Care and Ophthalmology departments, we propose to evaluate a novel surgical therapy for symblepharon in cats, involving the use of buccal mucosa and amniotic membrane grafts. The selected student will help with pre-clinical evaluation, anesthesia and post-operative follow up of cases.
Project Title: Blood-derived topical therapy for ocular surface diseases: preclinical and clinical evaluation

Principle Investigator: Dr. Lionel Sebbag

Collaborating Investigator: Dr. Rachel Strauss

Abstract: (300 words or less):

Blood-derived products such as serum and platelet-rich plasma (PRP) have gained popularity as an adjunctive treatment for several ocular surface diseases in humans, including dry eye, neurotrophic ulcers and chemical burns. In addition to lubricating properties, serum and plasma contain many growth factors that support corneal cell homeostasis, growth and migration. For example, nerve growth factor has been shown to improve corneal sensitivity and healing of chronic ulcers, while epidermal growth factor reduces re-epithelialization time. PRP is a novel approach in regenerative medicine and could have significant impact in the field of veterinary ophthalmology. Although PRP is effective, the cost of its preparation using commercially available kits is very high, which could be a barrier to its widespread use in veterinary patients.

The first aim of the study is to develop and optimize a cost-effective method to produce PRP eye drops in dogs. The second aim is to determine optimum storage conditions for blood-derived eye drops by measuring the concentration of several growth factors as a function of storage temperature and storage duration. Last, blood-derived topical therapy will be used in clinical patients presented to ISU Ophthalmology service and the study will describe their outcome.

In sum, the proposed project will expose the selected student to a variety of tasks including blood collection, serum/plasma extraction, growth factor quantification (ELISA), clinical evaluation, therapy and follow-up on patients.
Project Title: Metabolic adaptations of *Staphylococcus aureus* to intermediate antibiotic resistance

Principle Investigator(s): Greg A. Somerville, Ph.D. (UNL)

Abstract:

*Staphylococcus aureus* pose major health risks and cause significant economic hardships to livestock producers, food industries, and human and veterinary medical industries. To combat the health risks and mitigate the economic hardships of bacterial infections, antibiotic usage has increased in humans and animals. A consequence of increased antibiotic usage is increased bacterial antibiotic resistance, which is a major problem for veterinarians and physicians that can lead to treatment failures. In 2015 in response to the growing problem of antibiotic resistance, the FDA revised the 1996 Animal Drug Availability Act to “…include eliminating the feed and water use of medically important antimicrobial drugs for production purposes in food-producing animals and bringing all remaining therapeutic uses under the oversight of licensed veterinarians.”

Understanding how bacteria gain resistance to antibiotics, allows you to converse with clients about the magnitude and the hyperbole of antibiotic resistance. The first step in bacterial resistance is often a process called adaptive resistance, where bacteria adapt their metabolism and physiology to permit growth in the presence of intermediate concentrations of antibiotics. We study these metabolic adaptations and develop ways to reverse these them and re-sensitize bacteria to antibiotics. The goal of this work is to extend the usable life of new antibiotics and to make older antibiotics more efficacious in an environment where veterinarians are increasingly constrained in therapeutic options.
**Project Title:** Enhancing natural immunity through ligation of FcγR1 on equine macrophages with molecularly cloned and expressed equine IgG1

Principal Investigator(s): Brett Sponseller

Collaborating Investigator(s): Doug Jones, Adam Barb

**Abstract:**

While antimicrobial drugs remain the most common approach for treatment of intracellular infections, the increasing emergence of multidrug-resistant-organisms requires the need for alternative methods of bacterial control. One spontaneous naturally occurring bacterial disease model is pneumonia caused by *Rhodococcus equi* evidenced by immunocompromised humans and, especially, foals less than six months of age. *R. equi* is a common and devastating pathogen causing high morbidity and mortality from severe bronchopneumonia in young foals; however, adult horses are not susceptible to this intracellular pathogen. Given the neonates’ age-related susceptibility to disease, the need for alternative antimicrobial therapy is imperative. Evidence demonstrates that plasma products reduce the incidence and severity of pneumonia in foals but the mechanism largely remains unknown. We speculate that soluble immune complexes (sIC), present in plasma products, interact with surface FcγRs on macrophages and may contribute to the success of equine plasma products in prevention of clinical *R. equi* pneumonia. Our studies suggest that non-specific activation of macrophage FcγRs by sICs results in increased reactive intermediates of oxygen and nitrogen. Additionally, our collaborators (DJ lab) have demonstrated a reduction in the in vitro load of *Leishmania amazonensis*, a macrophage-tropic intracellular pathogen, through sIC stimulation. These results support the hypothesis that soluble immune complexes activate macrophages via FcγR leading to intracellular killing of pathogens. The purpose of this in vitro summer scholars study will be to molecularly clone, express and determine the biological impact of cloned equine IgG1 on reduction of *R. equi* in infected macrophages.
Project Title: Characterization of post-procedural stress, pain response, and efficacy across four disbudding methodologies (heat cautery, clove oil injection, short-term application of caustic paste, and freezing)

Principal Investigator(s): Kelly Still Brooks

Collaborating Investigator(s): Suzanne Millman

Abstract: (300 words or less): The objective of this project is to determine whether any of three alternate methods of disbudding (injection of clove oil essence, short-term application of caustic paste, or two methods of freezing) are less painful than the industry standard, heat cautery. The clinical phase of the study, which will be conducted during the spring of 2017, includes remote videography of the disbudding process and behavior up to 72 hours after the procedure. Acute pain response during the disbudding process will be assessed through scoring of vocalization and escape behavior. Post-procedural pain will be assessed through application of a standarized ethogram. The summer scholar may have additional opportunities to participate in other small ruminant clinical research or in laboratory analysis of banked serum cortisol and haptoglobin samples from this project.