RESEARCH PROGRESS REPORT SUMMARY

Grant 02152: Translation of MicroRNA into an Early Diagnostic Test for Chronic Kidney Disease

Principal Investigator: Dr. Mary B Nabity, DVM, PhD

Research Institution: Texas A&M AgriLife Research

Grant Amount: $26,988.00

Start Date: 1/1/2015 **End Date:** 12/31/2016

Progress Report: Mid-Year 2

Report Due: 6/30/2016 **Report Received:** 7/2/2016

Recommended for Approval:

(Content of this report is not confidential. A grant sponsor's CHF Health Liaison may request the confidential scientific report submitted by the investigator by contacting the CHF office. The below Report to Grant Sponsors from Investigator can be used in communications with your club members.)

Original Project Description:

Chronic kidney disease (CKD) is a significant cause of illness and death in dogs and is often due to glomerular diseases. Dogs with glomerular disease often have poor outcomes with standard therapy, and specific treatment recommendations are difficult without performing a kidney biopsy to determine the type of glomerular disease present, since treatment and outcome among these diseases differs substantially. Even then, we lack an understanding of the mechanisms driving these diseases, limiting our ability to optimally treat these dogs. Therefore, tests to non-invasively diagnosis the type of glomerular disease would help veterinarians more appropriately treat these patients and provide insight into the mechanisms that cause the diseases. This could lead to better therapies that slow disease progression and improve quality and length of life in dogs with CKD.

MicroRNAs (miRNAs) are small molecules that can regulate gene expression by up or down regulation of messenger RNA transcripts and proteins in target tissues. Many studies have found that increases or decreases in miRNAs can serve as biomarkers of diseases, including human CKD. They also contribute to the development of diseases.

The main goal of this study is to identify miRNAs in serum and urine of dogs that are specific for the three major causes of glomerular disease in this species. We also aim to identify

miRNAs associated with disease progression for each of these diseases. Successful completion of these goals will help with both non-invasive diagnosis and improved treatment of CKD in dogs.

Grant Objectives:

1. To identify and quantify all miRNAs present in the serum and urine from dogs with each of the 3 glomerular diseases, both early and late in the disease process.

2. To confirm findings in the first objective using a more accurate means of quantifying miRNAs among samples.

Publications:

None at this time.

Report to Grant Sponsor from Investigator:

MicroRNAs (miRNAs) are small, non-coding RNAs that can alter gene expression, and they can serve both as biomarkers of disease and instigators of disease progression. The goal of this project is to identify miRNAs as new biomarkers in the serum and urine of dogs with kidney disease due to primary glomerular diseases. In particular, our goal is to identify miRNAs that can non-invasively (i.e., with a blood or urine sample) distinguish among the 3 most common glomerular kidney diseases in dogs. Currently, such non-invasive identification of these diseases is not possible; yet, their treatment and prognosis vary considerably.

The first objective of this project is to identify and quantify all miRNAs present in the serum and urine from dogs with each of the 3 glomerular diseases, both early and late in the disease process. This will be performed using a technique called small RNA-sequencing. At this point in time, we have identified samples that can be used for each group of diseases and determined reasonable detection and quality control limits for hemolysis in the serum samples. We have also gained experience with10 different urine and serum RNA isolation methods. We are currently in the process of obtaining our first sequencing results that will help us determine which of these methods is best for isolating RNA from our limited samples. While this method comparison has delayed our progress on the objectives of this study, it should confirm the best method for RNA isolation and provide confirmation that all steps in the isolation and sample preparation process will successfully lead to good sequencing results without sacrificing our limited study samples. The graduate student working on this project has recently learned how to perform analysis on sequencing data. Therefore, once we have this first round of

sequencing results, we do not anticipate further delays in this project. Our goal is to complete the first objective toward the end of 2016 or beginning of 2017.

The second objective of this project is to confirm our findings in the first objective using a more accurate means of quantifying specific miRNAs among samples. So far, we have gained experience with techniques that we will use for this objective. We anticipate starting sample analysis for this objective in the first part of 2017.

Thus far, this project has provided research experience to 2 graduate students and is serving as a significant portion of a PhD thesis for one of these students.