RESEARCH PROGRESS REPORT SUMMARY

Grant 02945-MOU: Understanding the Genetic Basis of Addison's Disease in Portuguese Water Dogs

Principal Investigator: Steven Friedenberg, DVM, PhD

Research Institution: University of Minnesota

Grant Amount: $209,381.00

Start Date: 5/1/2021 **End Date:** 4/30/2025

Progress Report: End-Year 3

Report Due: 4/30/2024 **Report Received:** 4/18/2024

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Original Project Description:

Addison's disease (AD) is a common, life-threatening disorder in dogs characterized by the immune-mediated destruction of portions of the adrenal gland. This damage prevents the adrenal gland from synthesizing hormones that are necessary for normal cell metabolism, kidney function, and maintenance of the immune system. Dogs with AD are also highly predisposed to succumbing to a life-threatening adrenal crisis. AD is most common in Portuguese Water Dogs (PWDs), which have a 29-fold greater risk of developing the disease compared to other dog breeds, indicating a strong genetic component. To date, no genetic variants have been associated with AD in PWDs. This lack of knowledge has prevented the development of a genetic test that would allow for prediction of a dog's disease risk and the development of informed breeding practices related to AD. In this study, investigators will use state-of-the-art scientific tools to understand the genetic basis of AD in PWDs. The data generated here will provide the foundation for the development of a genetic test for AD in PWDs, enabling early diagnosis and treatment, as well as maintenance of genetic diversity within the breed while helping to decrease disease incidence.

Funding for the research is provided through the collaborative efforts and generosity of the Portuguese Water Dog Foundation, Inc. The AKC Canine Health Foundation supports the funding of this effort and will oversee grant administration and scientific progress.

Publications:

We recently published a manuscript related to environmental exposures that might be associated with Addison's disease in Portuguese Water Dogs and Standard Poodles. This is indirectly related our work on this grant. The paper can be found at this link: DOI: 10.1111/jvim.16896.

Presentations:

Dr. Friedenberg gave a 50-minute talk at the Portuguese Water Dog National Specialty on September 29, 2021 in Norfolk, VA. This talk discussed the pathophysiology of Addison's disease, Dr. Friedenberg's research strategy for the funded grant, as well as some preliminary data. There was significant audience engagement via Q&A throughout the talk.

Dr. Friedenberg also participated in a Q&A webinar through the CARE website on September 18, 2021. While this webinar was not solely related to this study, he did discuss it and encourage qualifying dogs to enroll in the study.

We gave a webinar on Addison's disease and our research to a seminar sponsored by the PWDF/PWDCA on 3/28/2023. The seminar was recorded and can be viewed at this link: https://www.pwdfoundation.org/project/understanding-the-genetic-basis-of-addisons-disease-in-pwds/

We will be presenting some initial findings from our work studying the genetics of Addison's disease at the International Conference on Canine and Feline Genetics and Genomics in Helsinki, Finland in early June. We also plan to give a presentation to the Portuguese Water Dog Foundation in October 2024 to provide an update on our results.

Report to Grant Sponsor from Investigator:

Over the first 3 years of this study, we have focused primarily on sample collection and running an initial set of genetics experiments. We collected samples mostly through PWD-related channels working directly with our collaborators in the PWDF/PWDCA. We also attended the PWD National Specialty in September 2021 and August 2022 to promote the study and collect samples, and we established a collaboration with Dr. Anita Oberbauer at UC Davis to share samples that she had already collected. To date, we have in hand 151 samples from affected PWDs and 319 samples from unaffected PWDs. Based upon relatedness, we have performed low-pass whole genome sequencing on 46 affected dogs and 55 unaffected dogs, and we have used data from these genotyping efforts to perform a genome-wide association study (GWAS). This initial study using traditional GWAS analysis approaches did not detect any major genetic differences between affected and unaffected PWDs. This is consistent with our thinking that the disease is genetically "complex." We then looked at the same data from 46 affected/55 unaffected dogs using two other analysis approaches which can identify subtler differences between cases and controls as well as regions of the genome that are under active selection in PWDs. These analyses highlighted several regions of the genome that could harbor genes

important in the development of Addison's disease in PWDs, and we are actively analyzing these genes at the moment.

We also performed an updated analysis of the variations in a set of genes called the major histocompatibility complex genes. These genes are very important in the immune system. We found one "version" of this gene set that continues to be associated with Addison's disease in PWDs – which is consistent with what we found in the last progress report.

We look forward to ongoing fruitful collaborations with the PWDF and PWDCA on this work.