

Abstract of proposed student project (1 page limit. This should mirror the aims page of a grant and CLEARLY indicate the student's role.)

Proposal title: **Comparison of heartworm antigen tests pre and post heat treatment in 2 populations of dogs in the same endemic area**

Proposal:

Canine heartworm disease is caused by infection with the parasite *Dirofilaria Immitis*, and is a common disease throughout America and most other countries. It is transmitted via the mosquito, and therefore is more common in tropical and subtropical areas. Heartworm testing is recommended annually by the American Heartworm Society, and current testing is most often performed by a visual test for microfilaria (L1 larval stage) and testing for heartworm antigen in the blood. Because heartworm infection can cause serious illness in the dog and can be transmitted indirectly through the mosquito to other dogs, reliable testing protocols are very important.

Several studies have shown that the antibodies produced in a heartworm positive dog can bind with antigen in the bloodstream, making antigen unavailable for detection and leading to a false negative test.^{1,2,3} These studies have shown that by heat-treating the sample using a standardized protocol, antigen-antibody complexes can be broken down to release the antigen and make it more easily detectable.⁴ Almost all studies of heat-treated samples have been done on shelter dogs that have no history of being on heartworm prevention. No recent data (within 5 years) has been published, and there is no known published data comparing pre and post heat treatment of samples in dogs that are on prevention. Current standard of care does not indicate the use of heat treatment prior to routine testing of dogs on or off heartworm prevention. Heat treatment is currently only recommended by the American Heartworm Society for dogs that test negative on routine testing, but have clinical signs that are consistent with heartworm disease.

In this large scale study, a board-certified veterinary parasitologist will evaluate for microfilaria and perform pre and post heat treated antigen tests on 600 dogs (total) from the same high endemic area. We will test 300 client-owned dogs that have been on heartworm prevention for at least 6 months, and 300 shelter dogs with no known history of prevention. Based on previous studies, we suspect that heat treatment will increase the sensitivity of the antigen test among the shelter dog population. Our goal is to compare the incidence of converted samples (from negative to positive post heat treatment) between the shelter dog population and the population of dogs that are on prevention. If it can be shown that conversion from a negative to positive test happens with regularity in either or both populations of dogs, this data can be used to determine whether heat-treatment of samples should be performed on a routine basis in shelter and/or primary care settings.

The student assigned to our project will be responsible for communication with owners, obtaining study consent, and assistance with collection and submission of samples from both client-owned and shelter dogs. The student will also spend time with the parasitology team performing heat-treatment of samples. At the completion of the study, the student will summarize all data available and present research results (poster format) and a manuscript.

¹ DiGangi B, et al. Impact of heat treatment n *Dirofilaria immitis* antigen detection in shelter dogs; *Parasites and Vectors* 2017; 10(Suppl 2): pp 408; DOI: 10.1186/21307-107-2443-7

² Little S et al. Prime detection of *Dirofilaria immitis*: understanding the influence of blocked antigen on heartworm test performance; *Parasites and Vectors* (2018): 11:186; DOI: 10.1186/s13071-018-2736-5

³ Velasquez L et al. Increased prevalence of *Dirofilaria immitis* antigen in canine samples after heat treatment; *Veterinary Parasitology* (2014): 206: pp 67-70

⁴ Gruntmeir J et al. Canine heartworm and heat treatment: An evaluation using a well based enzyme-linked immunosorbent assay (ELISA) and canine sera with confirmed heartworm infection status; *Veterinary Parasitology* (2020): 283: 109169