

Immunodiagnosis of Filarial Infections

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OBJECTIVE: To determine the incidence of *Dirofilariasis* in humans and dogs with above normal numbers of circulating eosinophils.

BACKGROUND: Eosinophilia frequently appears in patients with certain parasitic infections, allergies and dermatosis. The actual role of the eosinophil leukocyte has not yet been elucidated, but there is evidence that the mechanism of eosinophilia may be mediated by immune processes. Most of the experimental evidence for this is based on experimental infections with *Trichinella spiralis* in rats and mice¹. The clinical syndrome, Tropical Eosinophilia, has been recognized for many years and high titers of antibody against filarial antigens have been detected in active cases. These titers subside after treatment with diethylcarbamazine^{2,3}. This syndrome has been reported from many areas, including a large part of Southeast Asia. The inciting organism is believed to be *Wucheria bancrofti* in many instances⁴.

An increase in circulating eosinophils is often observed in Americans in Thailand without any other symptoms of Tropical Eosinophilia. Exposure to *W. bancrofti* is limited in Bangkok, but exposure to *Dirofilaria immitis*, a parasite of dogs, could be high as the incidence of heartworms in dogs in Bangkok is high. A survey of outpatients seen at the US Army Hospital, Bangkok, with an above normal number of eosinophils was performed to determine if serological titers for *D. immitis* antigens could be demonstrated.

DESCRIPTION: All outpatient records were screened to select patients with eosinophil counts above 8 percent. These individuals were bled and a portion of the serum sample kept while the remainder was processed by the Knott's Concentration Method for circulating microfilaria. The sera were tested against *D. immitis* antigen by the indirect hemagglutination assay (IHA) using antigen prepared from a saline extract of adult worms. Specificity of the assay was determined by using sera of patients with known helminthic infections (i.e. Gnathostomiasis) in assays against the filarial antigen, and by using antigens of other parasites and the sera of patients positive for filaria. All sera will be tested in the Soluble Antigen Fluorescent Antibody Test (SAFA) as an additional verification of the positives obtained with the IHA. Control sera (100) were obtained from the U.S. Army Hospital, Bangkok, from outpatients demonstrating less than 4% eosinophils. When a member of a family had eosinophilia, the remaining members of the family and pets, if any, were included in the study. Dogs which were brought to the Veterinary Clinic were routinely checked for *D. immitis* and those that were positive or demonstrated eosinophilia were included in the study with the owners and families.

PROGRESS: To date 349 human samples and 36 canine samples have been tested in the IHA. Of these samples 57(16.3%) human (Table 1) and 30(83.3%) canine (Table 2) samples were serologically positive for filariasis. Of the 177 human samples with 8% eosinophilia or greater 19 were positive (10.7%).

Of the 29 IHA positive sera from individuals with less than 4 percent eosinophiles, 28 were members of family groups which had been sampled based on the eosinophilia of the index member. There were no IHA positive sera among the 100 control sera obtained from the hospital laboratory.

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Future plans call for the data to be statistically analyzed to determine whether it supports the hypothesis that individuals in close contact with infected dogs will develop eosinophilia and antibody to the microfilaria. No microfilaria were isolated from human samples; however, a number of the canine samples had microfilaria. Family groups will be studied and the canine-human relationship investigated. A number of the sera obtained from military guard dogs were IHA positive and the handlers of these dogs will be included in the study. All sera presently on hand will be tested with the SAFA test and all future collections will be tested with both the IHA and the SAFA.

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Table 1: Results of IHA for Human Samples

Per cent Eosinophils	0-4	5-7	8-11	12-15	16-19	20+	Total
Observations	133	36	97	41	19	23	349
IHA Positive	29 ^a	9 ^b	6	4	4	5	57
Per cent Positive	21.8	25.0	6.2	9.8	21.7	21.7	16.3

- a. 28 individuals in this group were members of family groups sampled after the index individual was found to have eosinophilia.
- b. All positives in this group were members of family groups sampled after the index individual was found to have eosinophilia.

Table 2: Results of IHA for Canine Samples

Per cent Eosinophils	0-4	5-7	8-11	12-15	16-19	20+	Total
Observations	2	3	8	9	7	7	36
IHA Positive	1	3	8	8	3	7	30
Per cent Positive	50.0	100.0	100.0	88.9	42.9	100	83.3