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Short communication

Intraarticular *Dirofilaria immitis* microfilariae in two dogs

Steven Hodges [a,*], Mark Rishniw [b]

[a] *Michigan Veterinary Specialists, 29080 Inkster Road, Southfield, MI 48034, United States*
[b] *Department of Biomedical Sciences, Box 11 Veterinary Research Tower, College of Veterinary Medicine, Cornell University, Ithaca, NY 14583, United States*

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Abstract

Dirofilaria immitis microfilariae were found in the synovial fluid of two dogs. One dog had clinical and cytological evidence of polyarthritis at the time of presentation. The second dog presented with severe effusion in a single joint and was later diagnosed with synovial sarcoma of the affected joint. These patients were not protected with heartworm prophylaxis and lived in heartworm endemic areas. Though there is documentation of *D. immitis* microfilaria in the synovial fluid of several clinically normal research dogs with cytologically normal synovial fluid, to our knowledge these are the first documented cases of intraarticular microfilaria in a dog with cytologically confirmed polyarthritis. Based on these unique cases, *D. immitis* infection should be considered a differential diagnosis in patients with polyarthropathies. Interpretive caution must be used when intraarticular microfilaria are present, as concurrent etiologies may also be present.

Keywords: Dirofilaria; Microfilaria; Intraarticular; Polyarthritis; Dog

1. Introduction

The clinical presentation of dogs with *Dirofilaria immitis* infection is typically associated with complications due to the anatomic location of adult heartworms in the right pulmonary artery and, to a lesser degree, the right ventricle. Extra-cardiopulmonary affects secondary to *D. immitis* infection have been well documented in dogs with canine heartworm disease. The aberrant migration of adult worms into numerous tissues appears throughout the veterinary literature. In such cases, clinical signs not classically associated with canine heartworm disease have been reported (Otto, 1974; Blass et al., 1989; Hribernik et al., 1989; Elkins and Berkenblit, 1990; Frank et al., 1997; Healey et al., 2003). A thorough search of the literature revealed minimal references to the aberrant migration of *D. immitis* microfilaria, specifically intraarticular migration and associated clinical signs.

Weinberger et al. (1979) studied the effect of trauma on the canine joint in a group of fifteen dogs from the southern United States. Microfilaria was discovered in the synovial fluid of one of the dogs during postmortem examination. The dog appeared clinically well and had grossly normal appearing joints. Synovial fluid analysis revealed a normal cell count with no evidence of inflammation. Systemic microfilaremia was diagnosed with the Knott test and the acid phosphate test was used to identify the microfilaria as *D. immitis* in origin. The

* Corresponding author at: Oklahoma Veterinary Specialists, 515 W Main Street, Jenks, OK 74037, United States. Tel.: +1 918 299 4900; fax: +1 918 299 6366.
E-mail address: okvetspec@sbcglobal.net (S. Hodges).

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significance of the intraarticular microfilaria was unknown.

2. Case report

2.1. Case 1

An approximately 3-year-old, intact male mixed breed dog presented to Michigan Veterinary Specialists as a rescue from the Gulf Coast of the United States following the 2005 Hurricane Katrina. Physical examination did not reveal any significant abnormalities. A complete blood count (CBC), biochemical profile, urinalysis, fecal floatation and smear and heartworm antigen test were performed. Significant findings included hyperglobulinemia (4.1 g/dL, range 1.6–3.6 g/dL), a slight monocytosis (930/μL, range 0–840/μL), hookworm (*Ancylostoma* spp.) eggs on fecal floatation and a positive heartworm antigen test. A Knott's test was performed and was positive for microfilaria.

Treatment was administered for intestinal and ectoparasites. Because of suspected very high worm burdens, a treatment protocol for *D. immitis* was recommended by the American Heartworm Society specifically for heartworm positive dogs rescued from Hurricane Katrina (American Heartworm Society, 2007). Microfilaricidal treatment consisted of oral ivermectin (Heartguard®) administered at a dose of 6 μg/kg (preventative dose) on days 1 and 14 and every 30 days thereafter. On day 28, a one-time dose of 50 μg/kg of ivermectin was administered orally. Two months following the initial dose of ivermectin, adulticide therapy was initiated using the two-step, three-dose melarsamine (Immidicide®) protocol.

Four days after the initial treatment with ivermectin, the dog developed a mild fever (39.3 °C) and lameness which initially responded to treatment with a non-steroidal anti-inflammatory (NSAID) (deracoxib (Deramaxx®) 2.2 mg/kg daily). Despite continued NSAID therapy, the dog developed an intermittent, shifting limb lameness and low-grade fever. Joint palpation revealed mild pain, however, no joint effusion was appreciated. Serology for *Ehrlichia canis*, *Borrelia burgdorferi* and *Rickettsia rickettsii* were negative. Discontinuation of NSAID therapy resulted in worsening of fever, progressive lameness and anorexia within 36 h. Because of persistent lameness and low-grade fever, arthrocentesis was performed.

Synovial fluid was obtained from the left radiocarpal joint and left and right stifles. Cytological evaluation revealed low numbers of microfilariae (Fig.1.1) with mild mononuclear inflammation in the right stifle and low numbers of microfilariae with mixed inflammation consisting of macrophages, small lymphocytes and non-degenerate neutrophils in the left stifle. Neither sample had red blood cells or evidence of hemosiderin in macrophages that would be suggestive of contamination or previous intraarticular hemorrhage. Synovial fluid from the left carpus was within normal limits with moderate peripheral blood contamination and no microfilaria seen.

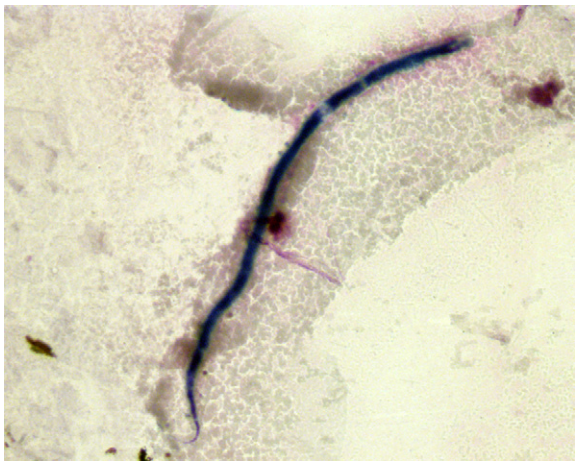

Fig. 1.1. *D. immitis* microfilaria discovered during cytologic examination of synovial fluid obtained from the right stifle.

Molecular identification of the microfilariae observed in the synovial fluid smear was performed as previously described, with minor modifications (Rishniw et al., 2005). Briefly, 75 μL lysis buffer (25 mM NaOH, 0.2 mM disodium EDTA, pH 12) was applied to an air-dried DiffQuick-stained smear of the joint fluid with four microfilariae visible on the slide. After suspending the contents of the smear in the lysis buffer, the solution was aspirated and transferred to a 1.5 mL Eppendorf tube, heated at 95 °C for 20 min and immediately mixed with 75 μL of neutralization buffer (40 mM Tris–HCl, pH 5) on ice (Truett et al., 2000). Five microliters of the resultant solution were used for the PCR genotyping reaction as previously described, using DIDR-F1 and DIDR-R1 primer pairs. Genotyping confirmed the microfilariae to be *D. immitis* in origin.

After the arthrocentesis was performed, NSAID therapy was reinstated and the fever and lameness improved. Five days after the first dose of melarsamine the dog developed a mild cough and hemoptysis. The NSAID was discontinued and treatment initiated with prednisone 1 mg/kg PO q12 h for suspected pneumonitis. The dog was released to a rescue organization and medical care was continued through the primary