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Case report

Rare case of presence of nematodes *Dirofilaria repens* in a pleural cavity of a dog

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ABSTRACT. Dirofilariosis, caused by nematodes *Dirofilaria repens*, is a parasitosis of wild and domestic carnivores. Nematodes *D. repens* locate in the subcutaneous and intramuscular tissue. The aim of this article is to describe rare case of pleural infection of a dog with nematodes *D. repens*. 12-year-old golden retriever had been treated in the veterinary clinic due to emaciation, weakness and dehydration, observed for 2 weeks. Hematologic and biochemical tests revealed reduction in the number of leukocytes and erythrocytes, significant decrease of hemoglobin, hematocrit and albumins as well as increased ALAT, urea and creatinine levels. Parasitological examination of blood smear revealed the presence of multiple microfilariae. Increased echogenicity of renal parenchyma was visible during ultrasound examination. Necropsy of euthanized animal revealed adult nematodes *D. repens*, located in the pleural cavity. Additionally, enlarged kidneys with subcapsular petechiae and a single cyst in the renal cortex were observed. To our best knowledge, it is the first case of finding nematodes *D. repens* in a pleural cavity of a dog.

Keywords: *Dirofilaria repens*, dog, pleural cavity, renal failure

Introduction

Dirofilariosis is the parasitosis of wild and domestic carnivores all over the world, but mostly observed in southern Europe. The most common species, infecting animals in Europe are: *D. immitis*, locating in the right ventricle and pulmonary arteries and *D. repens*, present in the subcutaneous and intramuscular tissue. Female nematodes produce into the circulation numerous first-stage larvae, called microfilariae. Mosquitos from the genus *Anopheles*, *Aedes* and *Culex* play the role of intermediate hosts and vectors of *Dirofilaria* nematodes. Microfilariae circulate in the blood of the definitive host, and occur in the capillaries of the skin most often in the late evening. It coincides with the increased activity of mosquitoes, which ingest microfilariae in a blood meal. Larve in the mosquito molt twice and develop into infective stage in 10 to 21, depending on environmental conditions. Third-stage larvae are inoculated by mosquitos to the blood of vertebral host during feeding. In the blood of the definitive host, larvae molt twice again, migrate to the final location and mature during 6 to 9 months [1–5].

Case presentation

In April 2015, 12-year-old golden retriever had been treated in the veterinary clinic due to dehydration, weakness and emaciation, lasting for about 2 weeks. Hematologic and biochemical analysis of blood revealed serious decrease in the number of leukocytes and erythrocytes, significant decrease of hemoglobin, hematocrit and albumins as well as increased ALAT, urea and creatinine levels. Blood sample was examined on the presence of microfilariae by the Knott's test [6] and the Kingston and Morton method [7] as well as commercial Canine Heartworm Antigen Test Kit

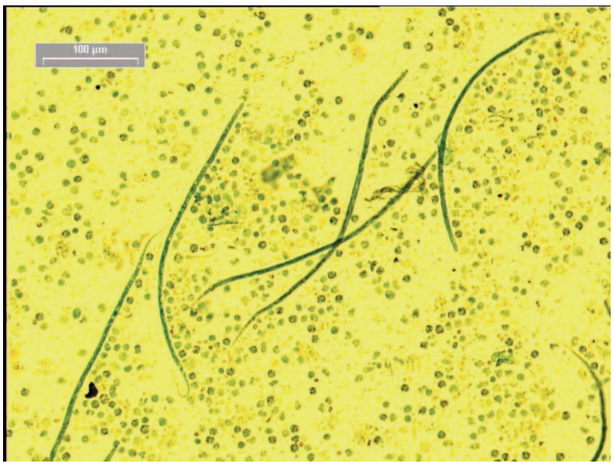

Figure 1. Microfilariae *D. repens* in the Knott's test

(IDEXX, USA) on the presence of *D. immitis* antigens. In the examined sample, numerous microfilariae, belonging to the species *D. repens* were found (Fig. 1) whereas no antigens *D. immitis* were detected. No skin lesions, typically accompanying *D. repens* infection, were observed. Ultrasound examination showed increased echogenicity of renal parenchyma. Treatment was provided by the administration of an antibiotic, antihelmintic, anti-inflammatory and hydrating drugs. Despite the treatment, the animal's condition deteriorated systematically and the dog was euthanized.

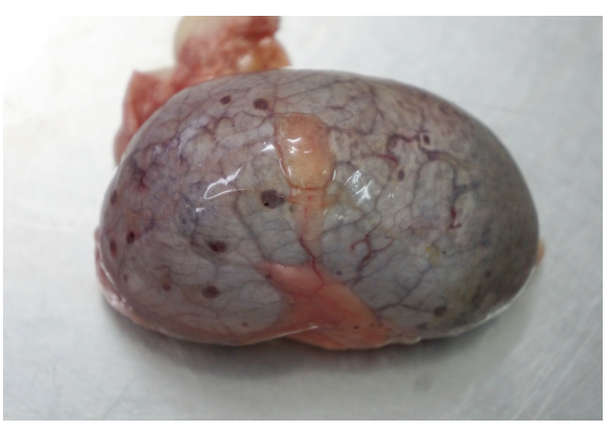

Figure 2. Kidney of the dog with subcapsular petechiae

Necropsy of the dog was performed according to standard necropsy techniques [8]. Pathological changes concerned mostly kidneys, inluding their enlargment, severe renal congestion and subcapsular petechiae with a diameter of 1 to 2 mm (Fig. 2). In the cortex of right kidney, a 6 by 10 mm cyst was found. The heart was moderately enlarged, with distension of the right ventricle and thinning of its wall. No nematodes were found in chambers of the heart, nor in pulmonary arteries. Two adult nematodes – male and female – from the genus *Dirofilaria* were isolated from the surface of lungs (Fig. 3,4); however no pathological changes were observed in infected organ. After isolation, nematodes were preserved in 70% ethanol and 5% glycerol for further identification under light microscope JENAVAL. Morphometrical features of nematodes allowed to identify them as *D. repens* [9]. Examined dog never left Mazowieckie Voivodenship, Central Poland, which suggest that the infection was probably native.

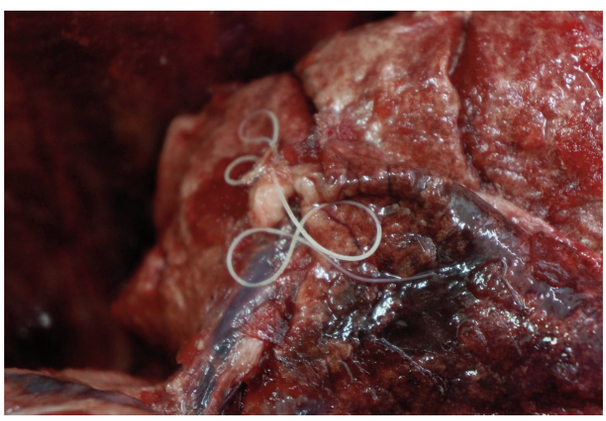

Figure 3. Nematodes *D. repens* in the pleural cavity of a dog

Figure 4. Male and female *D. repens* after isolation

Discussion

There is no recent data about the presence of nematodes *D. repens* in pleural cavity of definitive host. Nematodes *D. repens* typically locate in the subcutaneous and intramuscular tissue of spine or limbs. Subcutaneous dirofilariosis can be

asymptomatic, but nodular multifocal dermatitis or the presence of itchy lumps as well as multifocal alopecia and erythema, hyperpigmentation and hyperkeratosis are often observed. Described skin lesions are accompanied by the presence of mature nematodes under the skin or microfilariae in the skin. The infection may also include general symptoms such as lack of appetite, weakness and weight loss [1,10,11]. Subcutaneous dirofilariosis was observed in many European countries in years 2005–2009, including Slovakia, Czech Republic, Netherlands, Germany, Austria and Poland [5,12–16]. The parasitosis was also diagnosed in Ukraine and Russia [17,18]. During recent years, the range of dirofilariosis shifts to the north, probably due to global warming [3].

Parasitological examination of 1588 dogs in Poland revealed that *D. repens* is a parasite spread all over the country [19]. It was diagnosed in 25.8% of dogs in Central Poland [20]. Later studies revaled an increase of infected dogs to 38.3% and even 89.7% of animals simultaneously infected with *Babesia* spp. [21]. The infection of *D. repens* was also diagnosed in a cat [22].

Nematodes *D. repens* were occasionally found in an atypical locations, including scrotal cavity [16] and testicular parenchyma [22]. Mature nematodes were also observed in the pelvic cavity and mesenterium, and under the conjunctiva in a dog co-infected with *D. repens* and *D. immitis* in Romania. The animal suffered from chronic renal failure. Histopathological examination of kidney biopsy showed a multifocal inflammatory process in the renal glomeruli and in the interstitial space accompanied by atrophy and fibrosis of the renal parenchyma. Fragments of microfilariae, identified by PCR method as *D. repens*, were observed in the glomerular capillaries. These larvae may contain bacteria *Wolbachia pipientis*, which causes immune response of the host [23]. Increase of some biochemical blood parametres, including ALAT and hypoalbuminemia as well as inflammatory changes in kidneys and numerous microfilariae in histopathological examination were also observed in a dog, infected with *D. repens* from Lubelskie Voivodship, East Poland [24]. Cases of renal failure and microfilaremia in dogs infected with nematodes from the genus *Dirofilaria* have also been observed in India [25].

Therefore, renal failure, described in a dog from Central Poland, accompanying atypical *D. repens* infection is not suprising and may be a result of numeral microfilariae in a renal circulation.

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