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Short communication

Prevalence and distribution of *Dirofilaria repens* Railliet et Henry, 1911 in dogs in Poland

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Abstract

In 2011-2013 1588 samples of dogs' blood were examined for dirofilariosis using Knott method, as well as the Kingston and Morton method. The species of microfilariae was determined on the basis of morphometric characteristics. Samples were also examined using the Canine Heartworm Antigen Test. Positive samples were examined using a multiplex PCR assay for species confirmation. Microfilariae belonging to the species *D. repens* were found in the blood samples of dogs from all the provinces of Poland. The mean prevalence of this species observed in Poland was 11.7%. The range of intensity of infection was counted using the number of microfilariae found in 60 µl of blood amounted to between 1 and 158, and the mean intensity was 18 microfilariae. Microfilariae and antigens of *D. immitis* were not found in any examined blood samples.

Key words: *Dirofilaria repens*, dogs, autochthonous infection, Poland

Introduction

Dirofilariosis caused by *Dirofilaria repens* nematodes is widely dispersed in southern Europe among dogs, cats, other carnivores and occasionally humans. It also occurs in Asia, America, Africa and Australia. *D. repens* adult nematodes are located in nodules, in subcutaneous or intramuscular connective dog tissue. *Anopheles*, *Aedes* and *Culex* genera mosquitoes are the intermediate hosts of the mentioned parasites. *D. repens* invasive larvae are transmitted by more than 60 mosquito species (Kuzmin et al. 2005, Genchi et al. 2011).

During the course of infection of this parasite in dogs the following were observed: nodular multifocal dermatitis, presence of itching exanthemas in the form of papules, and also alopeciae, erythema, skin hyperpigmentations and skin hyperkeratosis. Sometimes purulent inflammatory changes occur in the skin. Occasionally invasion of the parasite has an asymptomatic course. It is possibly also caused by microfilariae circulating in the blood in the form of generalized cardio-hepato-renal insufficiency (Demiaszkiewicz et al. 2014). The objective of this study was to determine the prevalence and distribution of canine dirofilariosis in Poland.

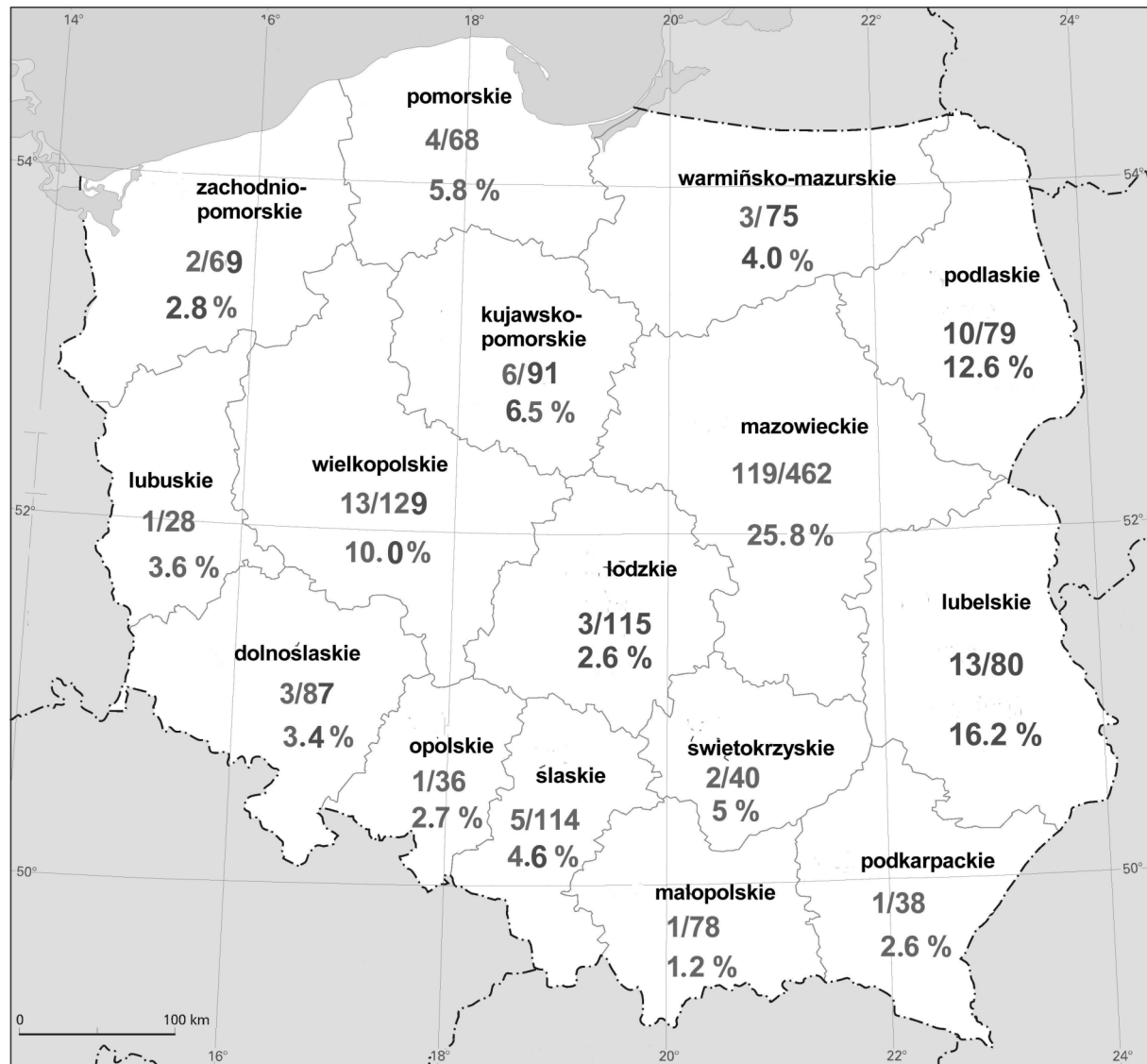


Fig. 1. Prevalence of *Dirofilaria repens* in dogs in Poland (infected/examined).

Materials and Methods

During the period 2011-2013 1588 dogs originated from all the provinces of Poland* were investigated for dirofilariosis. The dogs' age varied from 18 months to 16 years. Examined dogs did not show any pathological symptoms of invasion. Blood samples, taken from cephalic vein to tubes with addition of anticoagulant EDTA, were examined for the presence of microfilariae using the Knott method (1939), as well as the method of Kingston and Morton (1975). The species of microfilariae was determined, after staining, on the basis of morphometric characters (Liotta et al. 2012).

* Parasitological materials from Mazowieckie Province was already used in paper: The prevalence and distribution of *Dirofilaria repens* in dogs in the Mazowieckie Province. Ann Agric Environ Med 2014; 21: in press.

Samples were also examined using a Canine Heartworm Antigen Test Kit SNAP HTWM (IDEXX, USA) which allows circulating antigens of females of *Dirofilaria immitis* to be detected. Adult *D. repens* nematodes, microfilaremic blood samples, as well as negative blood samples were examined using the multiplex PCR assay (Gioia et al. 2010) in order to confirm species adherence of morphological identification of *D. repens* infection.

Results and Discussion

Microfilariae belonging to the species *D. repens* were found in the blood samples of dogs originating from all 16 provinces of Poland (Fig. 1). The mean prevalence of this species observed in Poland was 11.7%. The range of intensity of infection was

counted using the number of microfilariae found in 60 µl of blood amounted to between 1 and 158, and the mean intensity was 18 microfilariae. The highest prevalence, reaching 25.8%, was found in Mazowieckie Province and the highest mean intensity, reaching 37 microfilariae, was observed in Warmińsko-Mazurskie Province. In 3 provinces: Wielkopolskie, Podlaskie and Lubelskie prevalence in dogs amounted to between 10% and 16.2%. In 3 provinces: Mazowieckie, Lubelskie and Wielkopolskie the highest intensity was found, exceeding 100 microfilariae in 60 µl of blood. The lowest prevalence, in Małopolskie Province, was 1.2%, and the lowest mean intensity, in Zachodniopomorskie and Podkarpackie Province, was 2 microfilariae. The infected dogs had never been outside Poland, which means that this is an autochthonous invasion. *D. immitis* microfilariae were not found in any examined blood samples. The Canine Heartworm Antigen Test Kit SNAP HTWM indicated no circulating antigens of that parasite. Multiplex PCR results confirmed the presence of *D. repens* in all cases of microfilaraemic blood samples. No cases of *D. immitis* or mixed invasions were observed.

Until recently there had been an opinion that the northern border for the occurrence of this parasitosis was the 48° parallel. However, in 2000, native dog dirofilariosis were found in Ukraine, in 2004 in Germany, in 2005 in Slovakia, in 2006 in Czech Republic, and in 2009 in the Netherlands, Austria, and Poland (Genchi et al. 2011). In Poland dirofilariosis was first registered in 3 main areas in central Poland: in Warsaw, Pruszków and Żyrardów. 64 dogs were then examined, among whom 24 were infected. The prevalence observed, then reaching between 21.4% and 60%, was caused by the fact that the dogs were examined in 3 kennels where the animals were being held in small areas in open boxes where they were susceptible to mosquito bites (Demiaszkiewicz et al. 2009). In 2010 cases of microfilaremia were again registered in Poland in 8 dogs, in 2 of which the infection was confirmed by the PCR method. The authors, however, did not publish where the infected dogs were from (Sapierzynski et al. 2010). In the subsequent year microfilaremia was confirmed by PCR examination in another 10 dogs. The infected animals originated in a radius of around 10 kilometers from the center of Pruszków (Masny et al. 2011). Subsequent examinations found *D. repens* microfilariae in 119 dogs in

Warsaw and in 18 districts of Mazowieckie Province, significantly widening the already discovered extent of this parasitosis (Demiaszkiewicz et al. 2014). Our results and the published data show that autochthonous dirofilariosis of dogs caused by *D. repens* appears in all of the provinces of Poland.

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