Review articles

Dirofilaria repens Railliet et Henry, 1911 – a new parasite acclimatized in Poland¹

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ABSTRACT. Dirofilariosis caused by the *Dirofilaria repens* nematodes is widely dispersed in southern Europe, Asia and Africa among dogs, cats, other carnivores and occasionally, humans. The first case of *D. repens* infection, found in Poland in 2007, concerned humans. In 2009, dirofilariosis was first registered in dogs in 3 focuses in central Poland, Warsaw, Pruszków and Żyrardów, whose range grew considerably with the subsequent identification of *D. repens* microfilariae in 119 dogs in Warsaw and 18 districts of the Mazowieckie Province. The microfilariae of *D. repens* were found in blood samples taken from 1588 dogs from all 16 provinces of Poland. *D. repens* was also recently detected in a mixture of *Culex pipiens* and *Aedes vexans* mosquitoes collected in Mazowieckie Province using Real Time PCR. The results of this study confirms the acclimatisation of *D. repens* on the territory of Poland and a possibility for it to close its life cycle in domestic species of mosquito.

Key words: Dirofilaria repens, autochthonous infection, Poland

Dirofilariosis is a parasitic infection caused by nematodes of the Dirofilaria genus, with its most important agent in Europe, Asia and Africa being Dirofilaria repens (Railliet et Henry, 1911). The nematode is widely dispersed in south Europe among dogs, cats and other carnivores, and occasionally in humans. Adult D. repens are located in nodules, in the subcutaneous or intramuscular connective tissue of dogs (Fig. 1). The viviparous females release microfilariae, which migrate to the blood and are ingested by the intermediate hosts: female mosquitoes of the Anopheles, Aedes and Culex genera (Fig. 2). Invasive D. repens larvae are transmitted by more than 60 mosquito species. Development to the invasive stage III larva takes place in the mosquito and the final host is infected when the mosquito feeds. In dogs, the III stage larvae migrate to the subcutaneous tissue and undergo two moults over the next few months. The preparent period is 6–9 months. The mature nematodes live 2–4 years [1–3].

The following symptoms are observed during the course of infection in dogs: nodular multifocal



Fig. 1. Female D. repens in connective tissue of dog

dermatitis, the presence of itching exanthemas in the form of papules, alopeciae, erythemae, hyperpigmentation of the skin and hyperkeratosis, as well

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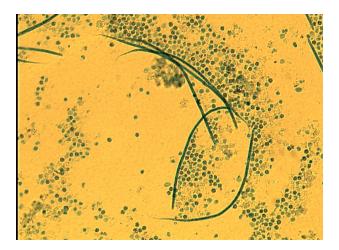


Fig. 2. Microfilariae of D. repens

as occasional purulent inflammatory changes in the skin. Local pathological changes in skin are observed, and these are associated either with the presence of matured nematodes underneath it, or of microfilariae within. Sometimes the invasion of the parasite has an asymptomatic course. Generalized cardio-hepato-renal insufficiency is also noted sometimes, possibly also caused by microfilariae forms circulating in the blood [4–7].

Dirofilariosis is also dangerous for humans. Until 1995, 397 cases of human dirofilariosis had been described in 30 countries all over the world [8], and over 270 cases of dirofilariosis in human had already been diagnosed within the European Union, most of which were caused by D. repens, with 10 cases being attributed to D. immitis [9]. In addition, a total of 1533 cases of human dirofilariosis had been registered in Ukraine before December 2012, with 1465 cases occurring within the last 16 years [10]. Nematodes were located in the subcutaneous connective tissue in different regions of the body: in the lungs, omentum, scrotum, epididymis and spermatic chord, palpebra and under conjunctiva, and also in lymphatic nodules. Tumors associated with the parasites were identified, whose diameter varied from 10 to 25 mm. Subcutaneous and pulmonary tumors evoked by D. repens have sometimes been incorrectly recognized as being of neoplastic origin. In humans, dirofilariosis is most commonly diagnosed during postoperative histological examination [8–11].

The first case of *D. repens* found in Poland in 2007 concerned humans, when histopathological examination revealed the presence of a female nematode in a subcutaneous tumor surgically removed from the abdominal area of a man from Warsaw. However, as the patient was in Greece four

years before the appearance of the tumor, it is not certain whether this was a native invasion [12]. In 2008, four more cases of D. repens were registered in Polish patients, two of which were probably acquired in Poland while the others probably from abroad [13]. Between 2007 and 2012, a total of 18 cases of human dirofilariosis caused by D. repens were reported in Poland. Three of these were confirmed to be autochthonous: the first was found in 2010 in Grójec, and the next two in 2011 in Białobrzegi and Warsaw. In fifteen cases, single nematodes were localised in nodules located in different body parts, while the other three were located subconjunctivally. The nodules were filled with a mass composed of necrotically affected tissue in which the nematode was located. During the last 3 years, since 2011, 12 new cases of human dirofilariosis have been observed in Poland [14,15].

Until recent times, the range of this parasitosis was believed to extend to the 48° parallel [2,16]. However native dirofilariosis was found in dogs in Ukraine in 2000 [7], Germany in 2004 [17], Slovakia in 2005 [18], the Czech Republic in 2006 [19], and in Holland, Austria, and Poland in 2009 [20-22]. Dirofilariosis was first registered in 3 focuses in central Poland, Warsaw, Pruszków and Żyrardów, where 24 of the 64 examined dogs were found to be infected. A high prevalence of between 21.4% and 60% was noted in each focus, which may have been because the dogs were kept in open boxes, which left them susceptible to mosquito bites [22]. In 2010, the presence of D. repens microfilariae was reported in tissue samples from 8 dogs in Poland and infection was confirmed in 2 of



Fig. 3. Nodule of a dog containing female D. repens

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them by PCR, however, the study does not state the origin of the dogs [23]. In the next year, PCR examination confirmed the presence of microfilaremia in another 10 dogs, all within 10 kilometers of the center of Pruszków [24].

Mature *D. repens* nematodes used to be relatively rarely found in dogs in Poland. They have previously been found in the cave on scrotum, in subcutaneous connective tissue, in a nodule on the eyelid (Fig. 3), and inside a cyst in the testicular parenchyma [25–28]. The identification of *D. repens* microfilariae in 119 dogs in Warsaw and in 18 regions of the surrounding Mazowieckie Province have broadened the known range of this parasitosis by a great degree. Taking into consideration that the parasite was registered in the centre of the province, as well as in its outer districts, it is reasonable to assume that dirofilariosis occurs throughout [29].

In the latest examinations, 1588 dogs were examined for dirofilariosis. D. repens microfilariae were found in 11.7% of the blood samples of dogs originating from all 16 provinces of Poland. The intensity, based on the number of microfilariae found in 60 µl of blood, ranged from 1 to 158, and the mean intensity 18 microfilariae. The highest prevalence (25.8%) was found in Mazowieckie Province and the highest mean intensity of invasion (37 microfilariae) was observed in Warmińsko-Mazurskie Province. The prevalence in dogs amounted to between 10% and 16.2% in Wielkopolskie, Podlaskie and Lubelskie. The highest intensity, exceeding 100 microfilariae in 60 ul of blood, was found in Mazowieckie, Lubelskie and Wielkopolskie. The lowest prevalence (1.2%) was registered in Małopolskie Province, and the lowest mean intensity (2 microfilariae) in Zachodniopomorskie and Podkarpackie Provinces. As the infected dogs had never been outside Poland, this appears to be an autochthonous invasion. D. immitis microfilariae were not found in any examined blood samples, and no such circulating antigens were identified by a Canine Heartworm Antigen Test Kit SNAP HTWM. Multiplex PCR results confirmed the presence of D. repens in all cases of microfilaraemic blood samples. The positive controls showed the expected amplification products of approximately 500 bp for the genus Dirofilaria and of 327 bp for D. repens. No cases of D. immitis or mixed invasions were observed [30]. An invasion with a similar profile to the one observed in Mazowieckie Province occurred in

Kiev, Ukraine, where in 2000–2002, D. repens was identified in 30% of stray dogs and 22% of domestic dogs [7]. In eastern Slovakia, the prevalence of dirofilariosis was even higher, reaching a maximal value of 54.38% (mean 34.44%) in the Trebišov district [31]. The identification of dirofilariosis in recent years in countries neighbouring Poland implies that its range is increasing, possibly due to climate change and high summer temperatures. These changes can encourage the proliferation of certain mosquito species and allow for the development of invasive Dirofilaria larvae within them [2,16]. For example, D. repens DNA was detected in samples containing a mixture of Culex pipiens and Aedes vexans mosquitoes collected in Mazowieckie Province during the summer months 2010–2012 [32], which confirms acclimatization of D. repens on the territory of Poland and reveals the possibility of closing its life cycle in domestic mosquito species. In 2012, the first autochthonous case of canine D. immitis infection, detected in Gdynia, northern Poland was an indicator that the range of autochthonous canine D. immitis infection in Europe had extended northwards [33].

Autochthonous dirofilariosis in dogs caused by *D. repens* nematodes is a common occurrence and the parasite has become acclimatized throughout Poland, where the high prevalence of *D. repens* infections in dogs and the presence of the mosquito vectors increase the likelihood of the parasite spreading to the human population. Hence, the number of cases of dirofilariosis in humans can be expected to increase. A nationwide programme to identify the presence of *D. immitis* nematodes in dogs is needed to better determine the epidemiological situation of this new parasitosis.

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