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Prevalence of *Dirofilaria immitis* in stray dogs from Sofia, Bulgaria

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Abstract

Dirofilaria immitis is a nematode parasite which is the causative agent of a serious pulmonary-cardiac disorder called Heartworm disease. It affects mainly dogs and rarely cats. The infection is transmitted by mosquitoes thus stray dogs and wild carnivores are expected to be an important reservoir of the infection. The aim of the study is to update the current knowledge about the distribution of *D. immitis* among stray dogs from Sofia, Bulgaria. The survey included 293 dogs which were examined with rapid blood test for circulating *D. immitis* antigens. The percentage of positive animals was 11.26 % which is comparable with the results of similar studies from different regions of Bulgaria. The epidemiological data received indicates that *D. immitis* infection is endemic in stray dogs in Sofia.

Keywords: prevalence, stray dogs, *Dirofilaria immitis*, Bulgaria

1. Introduction

The progressive spread of vector-borne nematode diseases in the face of climate changes is a real veterinary challenge ^[1]. Heartworm disease (HWD) is a worldwide parasitic disorder caused by *Dirofilaria immitis* (Leidy, 1856), a representative of the *Onchocercidae* family. Despite the name the main residential location of the parasite is the pulmonary artery which leads to pulmonary hypertension and in some cases to right-sided heart failure. However the disease is often asymptomatic. Infection is transmitted by blood sucking arthropods – mosquitos (*Culex* spp., *Aedes* spp. and *Anopheles* spp.) ^[5, 29]. The final hosts are mainly wild and domestic carnivores. *Dirofilaria immitis* possesses some zoonotic potential ^[24] although humans are usually affected by *D. repens* which is localized in the subcutaneous tissue ^[6, 10]. By the time this article is issued human dirofilariasis cases in Bulgaria are caused by *D. repens* only ^[34].

Heartworm disease shows almost worldwide spread. In Europe the long established endemic areas are around the Mediterranean basin (Italy, South France, Spain) and follow the distribution of mosquitos. There is a progressive tendency of HWD spread to north and east Europe. It now became endemic in countries where it used to be a rare find. Underlying factors include the intense movement of infected dogs across Europe, increased attention toward the disease, emergence of new vector species, wild carnivore population dynamics as well as changes in ecosystem due to human activity ^[21]. However the climate changes and the introduction of new vector species are among the leading factors for HWD progression to wider territories ^[12]. The climate change in the past decades is the reason autochthonous heartworm disease to be detected also in central and northern parts of Europe ^[18].

A main reservoir of the nematode can be the domestic dog (*Canis familiaris*, L., 1758)-pet, stray, hunting etc. and different wild carnivores – wolf (*Canis lupus*, L., 1758), red fox (*Vulpes vulpes*, L., 1758), jackal (*Canis aureus*, L., 1758).

Dirofilaria immitis in dogs was detected for the first time in Bulgaria at the end of the XX century and since then there is increasing data for the distribution of the parasite among canines, mainly pet dogs, in different regions of the country.

Bulgaria is located in south-eastern Europe in the southern part of the North Temperate Zone. However it has some subtropical influence. This transitional location between two zones gives its reflection over the climate, soils, flora and fauna ^[9]. Sofia is the capital of the country with

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app. 1.5 million citizens. It is located in the western part of the territory at 550/600 meters above sea level. The city has continental climate characterized by cold winters and warm summers.

Stray dogs are a serious ecological, social, medical and veterinary problem worldwide. In the last decade the number of stray dogs in Sofia is gradually decreased as a result of spay programs and legislation measures and nowadays their count is app. 24 194 in 2017 in the country (unofficial data). In Sofia district the figures point around 3844 [15]. In urban and suburban areas stray dogs can be an important source for parasite distribution to pet dogs and eventually humans [30].

The aim of this study was to update the information about *Dirofilaria immitis* prevalence in stray dogs from Sofia area.

2. Materials and Methods

2.1. Study area and animals

The survey was carried out in Sofia region between January and December 2017. Study included 293 stray dogs from different parts of the city and neighbored metropolitan areas. All animals were mix breed, from both sex (male - 58 %, female - 42 %), the age of all tested dogs was above 8 months. There was no information about prophylactic measures against parasites.

2.2. Clinical materials

Blood samples were collected from *v. saphena lateralis* or *v. cephalica antebrae* through standard technique in tubes with EDTA. The rapid diagnostic test was performed *ex tempore*.

2.3. Diagnostic assay

The used assay was *Anigen Rapid CaniV-4Test Kit* (BioNote Inc., South Korea) - a chromatographic immunoassay which includes the qualitative detection of *Dirofilaria immitis* antigens. Sensitivity for heartworm is 94.4% and specificity - 100%. The tests were performed according to the manufacturer's instructions.

3. Results and Discussion

The prevalence of *D. immitis* in the studied canine population was 11.26 % (33/293). By sex, male dogs showed a higher incidence 66.7 % (22/33) compared to females 33.3 % (11/33).

The results of the current study were comparable to Radev [28] which reported 15 % positive dogs from 33 tested animals in a shelter in Sofia. At the same time our data were inconsistent with Stoyanova [30] which announced that 31.25 % (25/80) of tested stray dogs in Sofia were carriers of *D.immitis*. By sex, both studies confirmed that male dogs showed a higher incidence.

Similar epidemiological survey for Sofia but for pet dogs resulted in 7.5 % positive animals (12/160) [4]. Despite the variation in *D. immitis* prevalence in different canine populations, it can be inferred from the accumulated epidemiological data that Sofia is an endemic area and both domestic and stray dogs population are constant parasite carriers.

If we follow the results of previous studies we can concluded that at least for the few decades canines in various parts of the country were stably infected with *D. immitis*. According to Georgieva [13] 12.5 % of tested stray dogs and only 1.4 % of pet dogs were carriers. Results of necropsies of Kirkova [16] for Stara Zagora region (south Bulgaria) showed 10.7 % and comparable were the results of Kostadinov [17] which included

56 stray dogs from Burgas (East Bulgaria) - 12.5 % were infested. Another survey from Stara Zagora region indicated that 16.2 % (27/167) of the examined with rapid antigenic test were positive for HWD [26]. The percentage range of all these surveys was 1.4 % - 16.2%. Our result for Sofia (11.26%) falls in the same interval. All projects which included only pet dogs disclosed lower HWD prevalence (1.4 % - 7.5 %). The possible explanations can be that owned dogs in the city live mostly indoors and are treated with repellent substances which decreased the possibility for mosquito biting and transmission of infection.

Wild carnivores are among the main reservoirs of vector-borne nematodes. The necropsy of golden jackals (*Canis aureus*, L.), red foxes (*Vulpes vulpes*, L.) and stray dogs from different parts of Bulgaria demonstrated infection prevalence of 37.54 %, 25.22% and 33.33 % respectively [25]. The distribution of HWD in jackals from Pazardzik region (100 km west of Sofia) was even more pronounced - 73.1 % (19/26) [20].

Bulgaria borders with five countries – Greece to the south, Romania to the north, Serbia and North Macedonia – to the west and Turkey - to the south-east. According to Diacou [8] the prevalence of HWD in Greece was 4.1 % (31/750). In the northern part of the country (Thessaloniki) the spread was 14 % in comparison to 0.7 % in Attica. The geographical mismatch can be explained by the increased humidity and mosquito distribution in the north. The results of Angelou [2] were in confirmity – *D. immitis* prevalence among 1000 pet dogs was 9 %.

In Romania the reported prevalence of HWD was 3.3 % (38/1146) with significant regional variations – between 2.9 % to 31% [19]. The highest percent was located in Tulcea region near Danube delta where more favorable ecological conditions for the vector development are available.

The distribution rate of HWD in Serbia (Vojvodina province) was 7.2% [32] and in wild carnivores - golden jackals, red foxes and wolves - 7.32 %, 1.55 % and 1.43 % respectively [27].

Turkey has quite different ecological and climate regions which can explain the divergence in results. In Sivas Province of Turkey (central Anatolia) Atas [3] reported 3.7 % in stray and 1.7 % in owned dogs and the highest prevalence in Turkey was reported to be 46.2% in Van Province (east Turkey) [1]. The huge difference can be due to the long snowy winter and dry warm summer in Sivas province therefore mosquito activity lasts less than 3 months.

In the European part of Turkey some of the available results showed that in Istanbul the infection affects 1.52% of dogs [22] and 2% in the area of Gemlik in Bursa [7].

The available epidemiological data from the neighboring countries allows the conclusion that HWD is an endemic parasite in domestic and wild canine species in whole south-eastern Europe. The retrospective study of Széll [31] based on necropsy finding of the parasite demonstrated rise of HWD prevalence from 0.5 % to 8.0 % for eleven years. Although the survey concerned central Europe (Hungary), it clearly demonstrated the rapid progression of infection spread within a short period of time resulting from climate-driven ecological changes and also the impact of wild carnivores in HWD epidemiology.

Stray dogs are highly exposed to a range of zoonotic parasites including *D. immitis* and may become important reservoir for human and pet infection [23]. The limitation of stray dog population should be in the focus of competent authorities, health agencies and institutions in the country. According to

the concept of „one health“ the control and prevention of social and economically important infections require a multidisciplinary approach based on coordinated collaboration between governments, citizens, medical, veterinary and ecological authorities to achieve improvement of human and animal health ^[14, 33].

4. Conclusions

The current epidemiological survey demonstrated that Sofia, Bulgaria is an endemic area for *Dirofilaria immitis* and a significant part of stray dogs in the city are carriers. Because of the potentially substantial role in the life cycle of *D. immitis*, stray dog population in urban areas of Bulgaria should be further examined and tested for heartworm infection.

5. Conflict of Interest

The author reports no conflict of interests.

6. Acknowledgments

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