

SEROLOGICAL DETECTION OF ANTIBODIES TO *EHRlichia CANIS* AND *BORRELIA BURGdorFERI S.L.* IN URBAN HOUSEHOLD DOGS FROM IAȘI

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Abstract

Canine tick-borne diseases are an emerging problem within Romania and also throughout the European Countries. This threat comes not just from Lyme disease which is endemic in our country, but also from other tick-borne diseases as well as ehrlichiosis. The present study consisted in screening of IgG class antibodies to *Borrelia burgdorferi* and *Ehrlichia canis* on 92 urban household dogs (48 females and 44 males) from Iași. The sampling took place during March to June 2021, in three veterinary clinics. The results of the serological testing revealed four positive dogs: one for IgG anti-*E. canis* and three for IgG anti-*Borrelia burgdorferi s.l.* Although the proportion of the sampled dog sex was almost equal, all positive animals were adult females. Our results highlight the silent circulation of the two pathogens in the studied area. These tick-borne pathogens are a significant medical concern to canine health. Changing tick distributions, pet travel and nonspecific clinical signs can make identifying infected pets challenging, so is very important to keep all dogs on appropriate, effective tick prevention year-round.

Key words: *Lyme borreliosis*, *Canine ehrlichiosis*, immunoassay, IgG antibodies

INTRODUCTION

Tick-borne diseases are a global emerging problem for both animals and humans. *Ehrlichia* and *Borrelia* are bacteria genus with some species bearing a zoonotic character (Springer et al, 2020). Lyme borreliosis is probably the most prevalent tick-borne disease in dogs. Its causative agents, Lyme spirochaetes from the *Borrelia burgdorferi sensu lato* complex are transmitted by *Ixodes ricinus* ticks in Europe (Leschnik, 2014). The most common ticks found on Romanian dogs continue to be *Ixodes spp* ticks (Mihalca et al, 2012; Păduraru et al, 2012), *Dermacentor reticulatus* being also present across the Eastern Romania (Brătuleanu et al, 2021). The *Ehrlichia* genus is comprised by six species, *E. canis* being one of them. All species of *Ehrlichia* infect vertebrate hosts, and all of them are transmitted by ticks. Canine monocytic ehrlichiosis is a hemoparasitosis caused by the intracellular bacterium *Ehrlichia canis*, which has a worldwide distribution, although it occurs more frequently in regions with temperate climates due to the high prevalence of its biological vector, the *Rhipicephalus sanguineus* (Carrade et al, 2009).

Animals suffering from TBDs may develop life-threatening conditions if early clinical signs are not recognized and treatment has not been promptly instituted. Most dogs with infections like borreliosis and ehrlichiosis are asymptomatic, and only about 5% to 10% of infected dogs develop

clinical signs (Elsheikha, 2016). During the recent years tick borne diseases have been described more frequently. This is believed to be because of increased pet travel, better animal care from pet owners and increased interest of clinical practitioners to investigate unusual clinical signs.

MATERIAL AND METHODS

A total of 92 samples were collected during March to June 2021 from urban household dogs in Iași city (table 1). The serum samples originate from dogs examined for different pathologies (surgical or internal medicine) in three clinics (two private clinics and in emergency unit of FVM). After collection, the serum samples were stored at -20°C until testing.

All animals were tested for detection of IgG antibodies to *Ehrlichia canis* and *Borrelia burgdorferi s.l* using two available ELISA kits from Euroimmun (Germany). The first immunoassay kit provides a semiquantitative in vitro detection for antibodies of the immunoglobulin class IgG against *Ehrlichia canis*, with a sensitivity of 92% and specificity of 100%. The second ELISA test provides a semiquantitative in vitro assay for canine antibodies of the IgG class against *Borrelia* antigens in serum or plasma. The test kit contains wells coated with a mix of whole antigen extracts of *Borrelia burgdorferi sensu stricto*, *Borrelia afzelii* and *Borrelia garinii*. The testing protocols followed the procedure recommender by the producer. The results were evaluated semiquantitatively by calculation the

ratio of extinction of the sample over the extinction of the calibrator.

Table 1.
Distribution of the tested animals by collection site, sex and age

Consultation place	No. of samples	Sex	Age category		
			0-5 years old	5-10 years old	>10 years old
Private veterinary clinic 1	28	14 females	6	4	4
		14 males	2	5	7
Private veterinary clinic 2	8	5 females	5	-	-
		3 males	3	-	-
Emergency unit of FVM	56	29 females	16	8	4
		27 males	8	14	4

RESULTS AND DISCUSSIONS

Rickettsial organisms are small, obligate intracellular bacteria in the order *Rickettsiales*. These pathogens are transmitted by a variety of tick vectors, maintained in wildlife and domestic reservoirs, and can cause clinical disease in humans, dogs, and other domestic animals.

Our study that consisted in serological testing of 92 household dogs for detection of IgG anti-*E. canis* Ab revealed one positive sample, a 13 years old mixed breed female and a sample with borderline result collected from a 7 years old male. A borderline result does not exclude an infection with *E. canis*. Particularly in the early phase of infection, antibodies may not yet be present or are only present in such small quantities that they are not detectable. Secondly, the results of the serological testing for IgG anti-*Borrelia* antibodies revealed three positive animals: a 13 years Bichon female, a 13 years old and a 4 years old mixed breed female. All four positive animals for rickettsial tick-borne infections were adult females. Similar results were published in 2018 in a study conducted by Galay et al, where more females (37.5%) were found infected with at least one TBP than males (25%). Moreover, adult dogs are more likely to have had attached ticks that went unnoticed in the past, increasing their chances of exposure to tick-borne pathogens.

Our results highlight the silent circulation of the two pathogens in the studied area. All

Rickettsia species known to infect dogs are zoonotic; hence our findings should raise an alarm because of the risk to humans (Chrome, 2011). Dogs have often been reported to serve as effective sentinel animals to assess the risk of human rickettsial tick-borne infection. These tick-borne pathogens are also a significant medical concern to canine health. Changing tick distributions, pet travel and nonspecific clinical signs can make identifying infected pets challenging, so is very important to keep all dogs on appropriate, effective tick prevention year-round.

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