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Review

Prevalence of borreliosis, anaplasmosis, ehrlichiosis and *Dirofilaria immitis* in dogs and vectors in Voronezh Reserve (Russia)

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ABSTRACT

Most of the dogs studied for the prevalence of CVBD have previously received acaricidal and insecticidal treatments. In the present work, a very specific population of dogs (Group 1) that had never been treated against ticks and mosquitoes was studied. Moreover, the territory occupied by this population has also never been treated, because it is a protected area – Voronezh Natural Reserve. Canine patients from veterinary clinics (Group 2) that had been treated against VBD vectors were studied for comparison.

Eighty-two dogs (Group 1) were enrolled in June, 2008. Blood samples were tested using the IDEXX SNAP® 4Dx® test. A specific heartworm antigen was detected in 12.2% samples. The seroprevalence for *Anaplasma phagocytophilum* was found to be 34.1%. The antibodies to *Borrelia* C6 peptide and to *Ehrlichia canis* were detected in 2.4% of the samples. Almost all dogs with infections had no clinical signs. Only 3 mixed-infected dogs showed non-specific clinical signs. During the tick season, 358 *Ixodes ricinus* were collected; the prevalence of *Borrelia burgdorferi s.l.* and *Anaplasma phagocytophilum* was 21.9% and 0.6%, respectively.

Four hundred and forty dogs (Group 2) were studied for comparison. Antibodies to *B. burgdorferi s.l.* were detected only in one dog, seroprevalence for *A. phagocytophilum* represented 1.1%, no *E. canis* seropositive dogs were identified, and 8.2% dogs were found infected with *Dirofilaria immitis*. Fifty-six percent of dogs with dirofilariosis had clinical signs. All dogs with anaplasmosis showed specific clinical signs – fever, anemia, splenitis. Three dogs died within a few days.

Contents

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1. Introduction

Most of the dogs living in Europe and Russia and seen by veterinarians receive acaricidal and insecticidal treatments protecting them from babesiosis, borreliosis, anaplasmosis, dirofilariosis, ehrlichiosis, and are sometimes vaccinated against babesiosis and borreliosis [1].

In the present work, a very specific population of dogs belonging to Voronezh Reserve staff was studied. Voronezh Reserve protected area is located 40 km north from Voronezh City, Russia (Fig. 1). This territory has never been treated with acaricides or insecticides and presents almost the ideal model of the dog–tick interaction in natural conditions.

Ixodes ticks (*Ixodes ricinus* and *Ixodes persulcatus*) transmit *Borrelia burgdorferi s.l.* and *Anaplasma phagocytophilum* [2–4]. *Rhipicephalus sanguineus* ticks transmit *Ehrlichia canis* [5]. Mosquitoes (*Culex*, *Aedes* and *Anopheles*) transmit *Dirofilaria immitis* [6]. All these vectors, except for *R. sanguineus*, were detected in Voronezh, and *R. sanguineus* was detected in Black Sea shore regions south of this area and is known to live in doghouses worldwide [7,8].

Lyme disease in dogs has never been studied in Voronezh Reserve, but a few cases of Lyme disease in humans were reported previously in this area. No cases of anaplasmosis in dogs in this area were reported before, but we did include *A. phagocytophilum* into the scope of the present study as it is transmitted by the same vector as *B. burgdorferi s.l.*

Cases of dirofilariosis in wild animals and cases of Lyme disease in humans were reported previously in this area (unpublished).

Dermacentor reticulatus ticks transmitting *Babesia canis* were detected and cases of babesiosis were reported in Voronezh Reserve, but babesiosis was out of the present research scope.

Eighty-two dogs owned by Voronezh Reserve staff had no travel history and had never been treated against ticks and mosquitoes. Additionally, 440 dogs receiving veterinary care treatment in clinics of six large cities in the European part of Russia and having reported tick bites were selected for comparison study.

IDEXX 4Dx SNAP kits were used to determine *D. immitis* antigens and antibodies to *B. burgdorferi s.l.*, *A. phagocytophilum* and *E. canis* in dog blood samples.

The purposes of this study were: to determine the prevalence of VBDs in dogs living in natural conditions in a protected area and to compare it with the prevalence of VBD in dogs receiving normal acaricidal and insecticidal treatments; to determine the prevalence of VBDs in ticks in natural conditions; to determine the characteristics of the disease in dogs in natural conditions: acute, chronic or subclinical.

2. Materials

2.1. Dogs, Group 1

Group 1 dogs were dogs owned by Voronezh Reserve staff.

All of the 82 dogs (9 breeds and mongrels) had blood collected between June14th and 16th, 2008, 2 months after the peak tick activity in April, to maximize the probability of positive samples. All dogs had no travel history. Depending on the keeping conditions, the dogs were divided into four groups:

- Indoor ("room") – animals lived in the house, the owners examined their dogs after walking, and the ticks were removed;
- Yard – dogs were living outside the house, during the day they walked through the yard without a leash, usually did not leave the village;
- Constraint – dogs were living in open-air cages near the house, the owners taking them hunting or for walks in the forest;
- Free – dogs living outdoors most of the time, visiting the forest without constraints.

Owners were asked to fill in a questionnaire. Blood samples were collected. Each sample was divided into 2 tubes: one with EDTA and one without. Serum were obtained and frozen at −40 °C for future analysis.

Out of the survey, 10 healthy dogs from 1 to 5 years old were selected in order to obtain a reference group for blood parameters. These dogs were negative to Knott's test and IDEXX SNAP® 4Dx® test and have had no history of diseases in the past.