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Seroprevalence of *Borrelia burgdorferi* in Dogs From a Mexico-U.S. Border Desert Region: Pilot Study

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Abstract: The aim of this pilot study was to estimate the seroprevalence to *Borrelia burgdorferi* in dogs from a Mexico-U.S. border region. A total of 94 dogs were tested by *Borrelia burgdorferi* ELISA kit. Borreliosis or Lyme desease is a worldwide zoonotic tick-borne disease caused by the espirochete *Borrelia burgdorferi* This disease is characterized by arthritis, lameness, erythema migrans, fatigue, anorexia, general malaise, muscle pain, stiff neck, fever, heart block, kidney failure and neurological changes such as seizures, aggression. In some cases it is cause of death. The resullts show an adjusted prevalence to *Borrelia burgdorferi* of 8.2% (95% I.C. 1.5-13.3%), obtained using Rogan-Gladen estimator. Since *B. burgdorferi* is transmitted by ticks, preventive and control measures to erradicate ticks have to be established in order of minimize the risk of infection.

Key words: Seroprevalence of *Borrelia burgdorferi*, border desert, pilot study, ELISA

INTRODUCTION

Borreliosis or Lyme disease is a worldwide zoonotic disease caused by the espirochete *Borrelia burgdorferi* transmitted by tick bite, primarily *Ixodes scapularis* and *I. pacificus*. This spirochete lenght 8-30 μm and width 0.2-0.3 (Barbour and Hayes, 1986; Greene *et al*., 2000). It is the most frequent tick-borne disease in Europa and the United States in animals and human beings. This disease is characterized by arthritis, lameness, erythema migrans, fatigue, anorexia, general malaise, muscle pain, stiff neck, fever, heart block, kidney failure and neurological changes such as seizures, aggression. In some cases it is cause of death (Burgess, 1986; Faul *et al*., 1999; Straubinger, 2000).

The aim of this pilot study was to estimate the seroprevalence to *Borrelia burgdorferi* in dogs in the urban area of Mexicali Baja California, a Mexico-U.S. border desert region.

MATERIALS AND METHODS

Study design and characteristics of the population: A cross-seccional descriptive study was conduced where 10 veterinary clinics and the Animal Control Center participated. The duration of the study was from September to November 2003. A total of 94 dog serum samples were randomly taken, 54 from veterinary clinics and 40 from the Animal Control Center. Mexicali city is situated along the state's northern border with the U.S. state of California and is the northernmost city in Latin America, located at 32°40'0"N, 115°28'0"W, with 855,962 inhabitants (Wikipedia, 2006). Climate is extreme, desert type and the average annual rainfall is 0.63±.43 cm. Climatic conditions data was collected from the United States National Weather Service of the National Oceanic and Atmospheric Administration (http://www.nws.noaa.gov/).

Data collection: A questionnaire was designed to collect information of the tested dogs and included: General information of dog: gender (female, male), age (≤1 year, >2 years), size (small, medium, big) and Intensity of tick infestation: None, low (1-10 ticks), moderate (11-30 ticks), intense (>30 ticks). The outcomes of most of the questions were dichotomous.

Blood collection: Blood samples were collected by certified personal. Briefly, 3 mL of blood were collected by punction of the cephalic vein after proper antisepsis of

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the area with isopropyl alcohol and placed in tubes Vacutainer®. Each sample was properly labeled and centrifuged at 3500 RPM for 10 min to separate the serum. The serum was transferred into 1mL vials, labeled and stored at-20°C until testing.

Serology: Antibodies against *Borrelia burgdorferi* were measured with the kit *Borrelia burgdorferi* ELISA® Helica Biosystems, Inc. for the detection and semi quantification of canine IgG class which guarantees a 95.8% sensibility and a 94.7% specifity. The Optical Density (OD) at 450 nm was registred, where an OD<1.0 was considered negative and OD = 1.0 as positive, according to the manufacturer.

Statistical analysis: Seroprevalence values were calculated by dividing the number of positive sera obtained by the total number of samples analyzed. The adjusted prevalence and its 95% CI were obtained using Rogan-Gladen estimator (Greiner and Gardner, 2000). The significance of the class variables (gender, age, size and intensity of tick infestation) was determined by Chi-squared test (Walker, 1997). All statistical analysis were performed using the Statistical Analysis System for Windows version 9.1.3 (SAS, 2004).

RESULTS AND DISCUSSION

The results of this study indicate an adjusted seroprevalence to *Borrelia burgdorferi* of 8.2% (95% C.I. 1.5-13.3%) in dogs from Mexicali, calculated by Rogan-Gladen estimator (Greiner and Gardner, 2000). The adjusted prevalence obtained in dogs from veterinary clinics and from the Animal Control Center is presented in Table 1.

The seroprevalence obtained in this study was similar than that from 1988 (6.6%) in the same city (Mexicali). Nevertheless were tested 30 dogs with epistaxis (Romano *et al*., 1998). The prevalence of this study was lower than found in another study done in other region of the Mexican Republic as Monterrey, Nuevo León, where found a prevalencia of 16% (160/850) in dogs tested by an indirect immunofluorescent assay (Salinas-Melendez *et al*., 1999). A possible cause of low seroprevalence to *B. burgdorferi* can be that have not been found the known vectors of this disease in this region as *Ixodes scapularis*, *I. pacificus*, *Dermacentor variabilis* and *Amblyomma americanum* (Magnarelli and Anderson, 1988; Lane, 1996; Adelson *et al*., 2004). The unique tick found it in Mexicali has been *R. sanguineus* (in press),which is not considered as vector of borreliosis in other regions of the world. However, in Sao Paulo, Brasil, where *Ixodes loricatus*, *I. didelphidis* and *Amblyomma cajennense* were found, the prevalence was of 9.7% (23/237) in dogs by ELISA and confirmed by Western blot (Joppert *et al*., 2001).

Table 2 shows the unadjusted prevalence values stratified by origin, gender, age, size and intensity of tick infestation. In general, not differences (p>0.05) were observed in prevalence values according to gender, age and intensity of tick infestation between dogs from veterinary clinics and those from the Animal Control Center. However, dogs of big size from Animal Control Center showed higher (p<0.05) seroprevalence than those dogs of small and medium size.

Although the prevalence found in this pilot study was low, it is necessary to perform a complete study that includes an appropriate sample size, serum sampling all year long and the evaluation of risk factors so that the appropriate preventive and control measures are established. Also, since borreliosis is a zoonotic desease that may require expensive hospitalization and may cause death it is imperative to know the prevalence in humans, particularly in places like Mexicali, where a serologic evidence in dogs has been observed.

Table 1: Adjusted prevalence* to *Borrelia burgdorferi* of dogs from Mexicali, an urban area of Mexico-U.S. border desert region

Origin	n	Positives	Adjusted prevalence* (%)	95%I.C. (%)
General	94	7	8.2	1.5-13.3
Veterinary clinics	54	4	8.1	1.5-13.2
Animal control center	40	3	8.2	1.6-13.3

*Rogan-Gladen estimator (Greiner and Gardner, 2000)

Table 2: Seroprevalence* to *Borrelia burgdorferi* in dogs from Mexicali, stratified by origin, gender, age, size and intensity of infestation

	Origin					
	Veterinary clinics			Animal control center		
Class variable[1/]	n	Positive	(%)	n	Positive	(%)
Gender						
Male	30	3	10.0[a]	15	2	13.3[a]
Female	24	1	4.1[a]	25	1	4.0[a]
Age						
≤1 year old	16	2	12.5[a]	22	1	4.5[a]
>1 year old	38	2	5.2[a]	18	2	11.1[a]
Size						
Small	10	2	20.0[a]	9	0	0.0
Medium	28	1	3.5[a]	23	0	0.0
Big	16	1	6.2[a]	8	3	37.5[a]
Intensity of tick infestation						
None	21	1	4.7[a]	17	0	0.0
Low	26	3	5.5[a]	18	2	11.1[a]
Moderate	5	0	0.0	4	1	25.0[a]
Intense	2	0	0.0	1	0	0.0

* Unadjusted values [1/]Equal letters by class variable within origin, indicate no differences (p>0.05)

CONCLUSION

A low seroprevalence of 8.2% (95% I.C. 1.5-13.3%) to *Borrelia burdorgferi* was observed in this study and because borreliosis is a zoonotic disease, a complete study has to be done, where both dogs and humans are included with an appropriate sample size and the evaluation of risk factors. Since *Borrelia burgdorferi* is transmitted by ticks, preventive and control measures to erradicate ticks have to be established in order of minimize the risk of infection.

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