

ARTÍCULO ORIGINAL

Serological Evidence of *Leptospira* Infection in Veterinary Students, Bogota, Colombia

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Abstract

Introduction: Leptospirosis is a zoonotic disease caused by *Leptospira interrogans* and is transmitted by direct contact with animal secretions or contaminated water. Veterinary and veterinary students are population at risk of infection. **Objective:** To determine the seroprevalence of *Leptospira interrogans* infection among students in a School of Veterinary Medicine in Bogotá, Colombia. **Methods:** We processed 445 sera by ELISA test; then we analyzed 42 samples by microagglutination (MAT). **Results:** *Leptospira* seroprevalence by ELISA IgM was 7.9% (35 of 445). 34 sera were positive by MAT at least for one serovar. **Conclusion:** This study confirms the usefulness of serological diagnosis in individuals with occupational risk for Leptospirosis and stand out the importance of using protective barriers by all populations who have regular animal contact including veterinary students.

Key words: *Leptospira*, seroepidemiologic studies, agglutination.

Título: Evidencia serológica de infección por *Leptospira* en estudiantes de medicina veterinaria en Bogotá, Colombia

Resumen

Introducción: La leptospirosis es una enfermedad zoonótica causada por *Leptospira interrogans* y es transmitida por contacto directo con secreciones animales o agua contaminada. Veterinarios y estudiantes de medicina veterinaria son población en riesgo

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de adquirir la infección. **Objetivo:** Determinar la seroprevalencia de infección por *Leptospira interrogans* entre estudiantes de una facultad de medicina veterinaria en Bogotá, Colombia. **Métodos:** Se procesaron 445 sueros por ELISA, de los cuales 42 muestras se analizaron mediante microaglutinación (MAT). **Resultados:** La seroprevalencia a *Leptospira* por ELISA IgM fue del 7,9% (35 de 445). De ellos 34 sueros fueron positivos por MAT al menos para un serovar. **Conclusión:** Este estudio confirma la utilidad del diagnóstico serológico en individuos con riesgo ocupacional para adquirir la leptospirosis y resalta la importancia del uso de barreras de protección para todas las poblaciones que tienen contacto regular con animales, incluidos los estudiantes de medicina veterinaria.

Palabras clave: *Leptospira*, estudios seroepidemiológicos, aglutinación.

Introduction

Leptospirosis is a zoonotic reemerging disease causing a major health problem all over the world. The etiological agent is a gram negative spirochete from the genus *Leptospira* with many serovars capable of surviving in water [1,2].

Animal reservoirs are rodents, dogs, cattle, pigs, among others. Transmission occurs by direct or indirect contact with secretions or material contaminated with the urine of these reservoirs. The clinical presentation in humans ranges from asymptomatic, mild (90%) and severe to Weil syndrome with jaundice and renal failure (5-10%) [1,3].

The global estimated annual incidence of this disease is about half a

million people. In humans, infection is related with animal contact in occupational activities such as veterinarians, farmers, slaughterhouse, and rural workers [4,5].

We aimed this descriptive and observational study to determine the seroprevalence of *Leptospira* in students of a School of Veterinary Medicine in Bogota, Colombia.

Methods

A population of 445 students of veterinary medicine was surveyed according to convenience sampling after signing the written informed consent. In order to assess the sera, we tested each sample by IgM-ELISA test (Panbio®) [3,6], and then positive samples were subjected to microagglutination (MAT) on six *Leptospira* serovars (Hardjo, Pomona, Canicola, Tarassovi, Bratislava, Icterohaemorrhagiae) [3,7].

Data were collected and evaluated in Microsoft Excel® and Statistical Package for Social Sciences (SPSS®).

Results

A total of 455 people enrolled, 224 were men (50.3%) and 221 (49.7%) women aged between 16-45 years and a mean of 21.3 years; 72.6% from Bogota. Of these, 441 students (99.1%) reported contact with animal secretions, 429 (96.4%)

with dogs, 355 (79.8%) with cattle, 312 (70.1 %) with horses, 78 (17.5%) with rodents and less than 15% with other species such as pigs, poultry, sheep and goats; 89.4% of subjects reported contact during their training.

Through an epidemiological survey we collected information about symptoms over the last 3 months before sampling: headache (59.6%), muscular pain (30.8%), fever (25.1%), joint pain (20.4%), suffusion (11.3%), burning during urination (7%), retroauricular pain (5%), and jaundice (3.8%) we reported.

Thirty five students (7.7%) were positive for IgM-ELISA, 42 samples (35 IgM positive, 7 IgM negative) were subjected to MAT agglutination test; seropositivity was obtained in 34 of them at least for one serovar (Table 1).

Table 1. *Leptospira* Serovars by Microagglutination among Students of Veterinary Medicine (n=34), Bogota, Colombia

Serovar	No. positive (%)
Hardjo	14 (41.2)
Pomona	24 (70.6)
Canicola	16 (47.1)
Tarassovi	14 (41.2)
Bratislava	15 (44.1)
Icterohaemorrhagiae	8 (23.5)

Discussion

This study confirms the usefulness of serological diagnosis in individuals with occupational risks for Leptospirosis.

According to the last bulletin of Colombian National Institute of Health, during the first semester of 2013, 327 cases of Leptospirosis (33 deceased) had been confirmed, with an incidence of 0.69 per 100,000 inhabitants [8].

As we expected, Canicola, Pomona, and Hardjo were predominant serovars (Table 1), because at least 80% of students had had dog or cattle contact [1].

Four students were positive for all 6 serovars tested, two for 5 serovars and 7 for Pomona serovar (Table 2). These findings are similar to described by Sanchez et al. in 69 MAT-positive patients where 97% had antibodies to more than two serovars, predominantly Icterohaemorrhagiae and Gryptophosa [9].

In a different study, Herrmann-Storck concludes that infection with *Leptospira* serovar Icterohaemorrhagiae could be a prognostic factor for severe outcomes despite of Susuki and Ariyoshi concerns about that conclusion [10]. In our study, only 23.5% of students were positive for MAT against that serovar, but in contrast to the study of Herrmann-Storck, similar symptoms were displayed by subjects with infection by other serovars.

Table 2. Co-agglutination by Microagglutination Among 34 Students of a School of Veterinary Medicine, Bogota, Colombia

Serovar	No. of Positive Individuals	Serovar	No. of Positive Individuals
HPCTBI	4	CT	1
HPCTB	2	CTB	1
P	7	PT	1
TBI	1	HPTI	1
B	2	HP	1
PCB	1	PC	1
H	1	HC	1
HPCB	1	PB	1
HPCT	1	C	2
HPC	1	HPBI	1
PTB	1	T	1

H: Hardo; P: Pomona; C: Canicola; T: Tarassovi; B: Bratislaviae; I: Icterohaemorrhagiae.

Previous studies have been conducted in Colombia on Human Leptospirosis, one of them in Sucre (northern state of Colombia) wherein 24.4% of rural workers were positive for ELISA test. This prevalence is almost twice that of our study which might be explained by differences in the type of population surveyed. Rios et al. referred lower prevalence in rural workers with activities that do not involve animal contact or workers who use barriers during animal contact. This finding is consistent with our results because veterinary students are more susceptible to infection during their training [11].

A seroprevalence of 17% was found in veterinarians and veterinary students

by using ELISA IgM test in a study by Gongora et al. In the same report, some risk factors such as having contact with a dog or rodents were strongly associated to *Leptospira* antibodies, and positives from 4% to 61% in different human populations were described [12]. In contrast, the prevalence in our study is 2.5 times lower; a potential explanation for this is that Gongora study included only students from the last year of veterinary medicine while we surveyed students of all years.

Sixty nine patients with symptoms consistent with Leptospirosis were confirmed by MAT in a two-year study (2005-2006) conducted in Quindío (Central state of Colombia). The most frequent symp-

toms were fever, muscular pain, cephalaea, and jaundice [9]. Our work showed similar results except for jaundice.

Echeverri et al. evaluated 14 hospitalized patients with suggestive symptoms of Leptospirosis; 100% were positive for IgM ELISA test, the major symptoms being fever (100%) and jaundice (64.3%) [6]. In our work, up to 60% of people surveyed developed symptoms of Leptospirosis yet just 25.1% had fever and 3.8% experienced jaundice.

Seroprevalence by MAT in workers from Colombian animal sacrifice centers showed that 42% were positive for Hardjo, 38.9% for Bratislava, 8.33% for Icterohaemorrhagiae, 5.6% for Canicola, and 2.8% for Pomona [7]. We found similar results for Hardjo (41.2%) although, in contrast, we found high seroprevalence of other serovars. This discrepancy might be explained by wide animal contact of veterinary students. In the same study, the most common symptoms were conjunctivitis (90%), cephalaea (52%) and jaundice (96.3%), in contrast to our findings of cephalaea (59.6%), fever (25.1%), joint pain (20.4%) and jaundice (3.8%); it also might be due to other infection with similar symptoms to those of Leptospirosis. We reported that students had contact with animal species such as dogs, cattle, horses, rodents, pigs, poultry, sheep and goats. This finding is con-

sistent with other studies where animal contact was higher than 70% with rodents, cattle, horses and dogs [7,9,12].

In our study, all seropositive subjects reported having had any contact with animals or secretions, and 90% of 455 subjects had contact with animals during their school training.

Regarding animal Leptospirosis, some studies have shown that several *Leptospira* serovars are present in animals. Rodríguez et al. reported almost half of stray dogs as infected by Icterohaemorrhagiae serovar (55.6%), followed by Hardjo (54.3%), Gryppotyphosa (45.7%), and Canicola serovar (38.3%) [13]. Likewise, one study of swine Leptospirosis seroprevalence found 43% of prevalence by MAT, where Pomona was the major *Leptospira* serovar (34%) [14].

A study in a Grocery Marketplace in Medellín, Colombia, found 25% of rodents tested positive for MAT; this finding is important if we consider that infection is unusual in the urban context [15]. We found that 17.5% of students reported rodent contact. As Agudelo remarks, rodents and others animals in cities could be creating a new environment for *Leptospira* transmission and thus a new worrisome scenario for Public Health.

Results from this and other serological studies in humans and animals in

our country suggest that ELISA test can be used in screening populations at risk, and that MAT is suitable for confirming the infection. Finally, they stand out the importance of using protective barriers by all populations who have regular animal contact including veterinary students [6].

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Ethical approval: Ethical approval was obtained from the Universidad de La Salle and Universidad Militar, Bogota, Colombia. All subjects in the study signed the informed consent.

Conflict of interest: None of the researchers have conflict of interest. This work was presented in poster session 22.089 at International Meeting on Emerging Diseases and Surveillance IMED 2013, Viena, Austria, February 15-18th 2013.

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