



## Investigation of the Prevalence of *Brucella canis* in Dogs, Veterinarians and Veterinary Faculty Students in Different Part of Turkey

Idil BASTAN<sup>1</sup>, Bulent BAS<sup>2</sup>

<sup>1</sup>Department of Internal Medicine, School of Veterinary Medicine, University of Ankara, Ankara, Turkey

<sup>2</sup>Department of Microbiology, School of Veterinary Medicine, University of Ankara, Ankara, Turkey

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### ABSTRACT

**Objective:** Canine Brucellosis, the caused by *Brucella canis* (*B. canis*), is seen all over the world and the disease is characterized by abortion and infertility. *Brucella canis* is a zoonotic agent and is transmitted to humans by contact with infected dogs or dog secretions. The aim of this study was to determine the presence of antibodies against *Brucella canis* in dog serum collected from different provinces of Turkey and the seroprevalence in veterinarians and veterinary faculty students in some of the provinces using Microagglutination Test (MAT).

**Materials and Methods:** Blood samples collected from 1559 dogs and 225 veterinarians and veterinary faculty students were examined for *B. canis* antibody titers using MAT method.

**Results:** As a result of the study, *B. canis* antibody was found to be positive in 12 (0.8%) of 1559 dog serum and in 13 (5.8%) of 225 human serum.

**Conclusion:** 5.8% of the *B.canis* seropositivity we have identified in the risk groups can give an idea of the state of the infection between the Turkish veterinarians and veterinary faculty students.

**Keywords:** *Brucella canis*, antibody, Microagglutination Test, seroprevalence, seropositivity.

### INTRODUCTION

*Brucella canis* was isolated for the first time in 1966 by Carmichael in the USA as an effective contagious abort of dogs. Dogs are known to be the only animal species that can naturally be infected by *B. canis*. The agent causes abortus in female dogs and epididymitis, testis atrophy and sterility in male dogs. Sometimes the disease can persist with lymphadenitis without any obvious clinical picture. Clinical diagnosis of the infection can be difficult because of not having many clinical symptoms in both disease forms (Aydın N, 2006; Bosu WTK and Prescott JFA, 1980; Corrente M et al., 2010; Flores-Castro R and Segura RA, 1976; Lucero NE et al., 1976; Öncel T, 2005; Wanke MM, 2004).

The agent is infected with dogs and cats in the abortion of abort materials or drinking of infected milk. Thus, vaginal discharge and wastes are important for transmission. Disease symptoms are high in the period when the number of exogenous bacteria to the external environment is higher (during reproduction period) (Aydın N, 2006; Hollett RB, 2006; Öncel T, 2005; Özlem M, 1998).

*B. canis* can be transmitted to humans through direct contact with infected dog or dog secretions and laboratory accidents. The main transmission to humans is by way of contact with dogs that recently aborted or gave birth (Aydın N, 2006; Lucero NE et al., 1976; Yılmaz B, 2010). Clinical signs and symptoms of *B.canis* infection in humans are similar

Corresponding author: Bulent BAS

E-mail: bbas@ankara.edu.tr

to those of brucellosis caused by the other *Brucella* species (Polt SS and Dismukes WE, 1982). Although the disease is seen with different clinical symptoms, deaths due to cardiovascular (infective endocarditis) and central nervous system complications can be seen in untreated cases. Diagnosis of the disease is done either by isolation of the agent or by serological tests. Nowadays, serological tests are preferred more

because of its easy, practical and fast results in the diagnosis the disease (Wanke MM, 2004; Yılmaz B, 2010). This study aims to reveal with the MAT the presence of *B.canis* antibodies in the bloods taken from the dogs from different provinces of Turkey and the veterinarians and veterinary faculty students.

## MATERIALS AND METHODS

In this study, serum samples were collected from 225 veterinarians and veterinary faculty students and 1559 dogs from various provinces of Turkey. The presence of *B. canis* antibodies in serum samples was investigated by MAT method. In the MA test, two-fold dilutions of serum samples were made with PBS in U-base microplates and an equal volume (25 µl) of *B.canis* antigen solution (Kitasato Institute, Tokyo, Japan) stained with Safranin-O was added. The plates

were incubated in a humidified environment for 24 hours at 50 °C after lightly shaking for 20 seconds. As a positive control, hyperimmune serum obtained from the *B. canis* strain in the Public Health Agency of Turkey was used.  $\geq 1/160$  titer was considered positive in the MA test in dogs (George LW, 1979; Monroe PW et al., 1975). However, since the diagnostic titer for *B. canis* infection is not yet definitive in humans, an antibody titer of  $\geq 1/50$  was considered positive in accordance with the previous studies (Sayan M et al., 2011a).

## RESULTS

The results of 1559 dogs collected from 13 provinces of Turkey are given in Table 1 and the results of the veterinarians and veterinary faculty students are given in Table 2. Antibodies with positive titer ( $\geq 1/160$ ) were detected in 12 (0.8%) of dog sera, while antibodies with  $\geq 1/20$  titer were detected in 512 (32.8%). When the distribution of positivity between provinces was examined, the positivity ranged 0-4% was determined and the highest positivity was found in Diyarbakır province where *B. canis* antibody was detected in 4 of 100 sera (4%). The positives were not

| Province   | MAT Titer |      |      |      |       |      | Total | Positivity % |
|------------|-----------|------|------|------|-------|------|-------|--------------|
|            | Negative  | 1/20 | 1/40 | 1/80 | 1/160 |      |       |              |
| Aydın      | 34        | 5    | 8    | 3    | -     | 50   | %0    |              |
| Amasya     | 13        | 3    | 3    | -    | -     | 19   | %0    |              |
| Kars       | 103       | 38   | 24   | 12   | 1     | 178  | %0.6  |              |
| Diyarbakır | 36        | 38   | 20   | 2    | 4     | 100  | %4    |              |
| Hatay      | 77        | 32   | 7    | -    | -     | 116  | %0    |              |
| Burdur     | 25        | 16   | 5    | 1    | -     | 47   | %0    |              |
| Erzurum    | 47        | 12   | 10   | 1    | -     | 70   | %0    |              |
| İstanbul   | 55        | 14   | 21   | 5    | 2     | 97   | %2    |              |
| Ankara     | 304       | 41   | 13   | 5    | 4     | 367  | %1.1  |              |
| Sivas      | 78        | 18   | 29   | 22   | -     | 147  | %0    |              |
| Şanlıurfa  | 66        | -    | -    | -    | -     | 66   | %0    |              |
| Kırıkkale  | 69        | 8    | 22   | 10   | 1     | 110  | %0.9  |              |
| Bursa      | 140       | 29   | 22   | 1    | -     | 192  | %0    |              |
| Total      | 1047      | 254  | 184  | 62   | 12    | 1559 | %0.8  |              |

**Table 1:** Results of dog sera collected from different provinces of Turkey.

| Provinces    | MAT Titer  |          |          |          |          |
|--------------|------------|----------|----------|----------|----------|
|              | Negative   | 25       | 50       | 100      | 200      |
| Ankara       | 119        | 6        | -        | 2        | 1        |
| Hatay        | 47         | 1        | -        | -        | -        |
| Aydın        | 37         | 1        | -        | 7        | 3        |
| <b>Total</b> | <b>203</b> | <b>9</b> | <b>-</b> | <b>9</b> | <b>4</b> |

**Table 2:** Results of the veterinarians and veterinary faculty students.

found in the samples taken from dogs in provinces of Aydın, Amasya, Hatay, Burdur, Erzurum, Sivas, Sanliurfa and Bursa. Considering the positivity in female and male dogs, positive titer antibodies were detected in 7 (0.85%) of 821 female dogs and 5 (0.67%) of 738 male dogs.

Positivity was determined in 13 (5.8%) of 225 sera collected from the veterinarians and veterinary faculty students. On provincial basis, the highest positivity was found in Aydın (20.8%).

## DISCUSSION

Although there is considerable information and evidence of *Brucella* infections in the world, studies on *B. canis* infections in dogs and in humans are extremely limited. The seroprevalence of *B. canis* has been reported to vary between 1.1% and 60.6% in serological studies on dogs conducted in different countries. In a study conducted in Italy, 25 (1.1%) of 2328 sera (Ebani VV et al., 2003) were found to be positive and 20 (60.6%) of 33 sera were found to be positive in a study conducted in Canada (Brennan SJ et al., 2008). *B. canis* antibodies were found to be positive in 12 of 485 sera (2.5%) in Japan (Kimura M et al., 2008); in 5 of 102 (4.9%) samples in Iran (Mosallanejad B et al., 2009); in 12 of 113 (10.6%) blood sera in another study conducted in Iran (Behzadi MA and Mohheiseh A, 2012); in 100 of 2000 (5%) sera in another study conducted in Canada (Bosu WTK and Prescott JFA, 1980); in 16 of 219 (7.3%) sera in Argentina (Boeri E et al., 2008); in 33 of 224 (14.7%) sera in another study conducted in Argentina (Lopez G et al., 2009); in 72 of 280 (25.7%) sera in Brazil (Barrouin-melo SM et al., 2007); in 85 of 317 (26.8%) sera in the USA (Brower A, 2007); and in 181 of 463 (39.1%) samples in Korea (Kim JW et al., 2007).

It has been reported that the seroprevalence in dogs changed between 5.4 and 7.7 in studies conducted in our country. In their study on 40 military dogs in Van, Ceylan et al. (2006) did not find *Brucella Canis* antibodies in the serum samples of dogs. *B. canis*

antibodies were found positive in 6 of 111 (5.4%) sera by Yilmaz and Gumussoy (2010); in 14 of 222 (6.3%) samples by Diker et al. (1987); in 9 of 134 (6.7%) sera by İstanbulluoğlu and Diker (1983); and in 28 of 362 (7.7%) samples by Oncel et al. (2005). In these studies, mercaptoethanol tube agglutination test was applied (Diker KS et al., 1987; İstanbulluoğlu E and Diker KS, 1983; Yılmaz B and Gümüşsoy KS, 2010). In the present study, 1559 canine sera collected from different provinces of Turkey were examined using MAT and a positive rate of 0.8% was determined for *B. canis* antibody. When the results of the present study were compared with those of previous studies, *B. canis* antibodies were found to be positive in a smaller number of samples. Rate difference in the positivity sample size is dependent on the difference of the strains used for antigen preparation. Wanke (2004) reported that 62% false positivity reactions were observed in studies using antigen from *B. canis* (RM6/66) strain, and less false positives were observed in studies using antigen from *B. Canis* (M-) strain, a less mucoid strain. Less false positivity and therefore seropositivity at lower rates may be determined due to the antigen prepared from commercial (M-) *B. canis* strain used in the present study.

Although *B. canis* infection is seen worldwide, the actual prevalence of the disease in humans is not fully known in many countries (Carmicheael LE, 1990; Polt SS and Dismukes WE, 1982). In our country, the studies on *B. canis* infection in human are very limited and there is not enough data to reveal the current state of the disease in our country. Determining the seroprevalence of *B. canis* infection in healthy individuals may be important to show the presence and source of the infection in the community. In the studies conducted in cases with the suspect of brucellosis in our country, *B. canis* antibody positivity generally varies between 1.6% and 9.2%. Diker et al. (1987) reported *B. canis* seropositivity in 2 (1.6%) of 123 brucellosis-suspected patients in Bursa region using 2-ME TAT method. Köksal et al. (1988) determined *B. canis* seropositivity as 8.3% (43/514) in patients who were suspected to have brucellosis in Adana using 2-ME TAT method.

Koylu et al. (2009) reported a positivity rate of 9.2% in a study in which seroprevalence of *B. canis* infection were investigated in 76 persons at risk in the province of Konya. Sayan et al. (2011a) determined *B. canis* antibody positivity as 3.7% in 1746 patients with brucellosis-like symptoms but RBPT-negative in all regions of our country with MA test. Sayan et al. (2011b) found *B. canis* seroprevalence as 1.6% (31/1930) in blood donors in Kocaeli region by blood agglutination test. Due to the differences in the study groups, serological diagnostic methods used, and the

accepted diagnostic titers, it is very difficult to compare the data of these studies with our data and to reveal the current status of *B. canis* infections in humans in our country. In our study, 1/50 *B. canis* antibody was detected in 13 (5.8%) of the 225 veterinarians and veterinary faculty students.

As a result; 5.8% of the *B. canis* seropositivity we have identified in the risk groups can give an idea of the state of the infection between the Turkish veterinarians and veterinary faculty students. Therefore, investigation for *B. canis* antibodies with particularly relevant epidemiological information and in cases with the suspect of brucellosis will enable studies on the epidemiology of infection in human to increase and to obtain healthier data on the disease at risk groups.

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