

## Low Prevalence of *Brucella* agglutinins in Blood Donors in Central Province of Iran

Masoomeh Sofian<sup>1</sup>, Mehrnoosh Sheikholeslami<sup>2</sup>, Fatemeh- Alsadat Mahdavi<sup>3</sup>, Arezoo Aghakhani<sup>4</sup>,  
Mohammad Banifazl<sup>5</sup>, Ali Eslamifar<sup>4</sup>, Hossein Sarmadian<sup>1</sup>, Ghorban Deiri<sup>3</sup>, Amitis Ramezani<sup>4\*</sup>

<sup>1</sup>Tuberculosis and Pediatric Infectious Research Center, Arak University of Medical Sciences, Arak, Iran. <sup>2</sup>Arak University of Medical Sciences, Arak, Iran. <sup>3</sup>Blood Transfusion Organization Research Center, Advanced Institute for Research and Education in Transfusion Medicine, Tehran, Iran. <sup>4</sup>Clinical Research Dept., Pasteur Institute of Iran, Tehran, Iran. <sup>5</sup>Iranian Society for Support of Patients with Infectious Disease, Tehran, Iran.

Received: October 2012, Accepted: December 2012.

---

### ABSTRACT

**Background and Objective:** Brucellosis is a zoonotic disease of worldwide distribution and has great economic importance. Despite its control in many countries, it remains endemic in Iran. Brucellosis was investigated in many high risk occupational groups; however, few studies on the prevalence of brucellosis among blood donors are available. To determine the seroprevalence of brucellosis antibodies in blood donors, a serological study was carried out in central province of Iran.

**Materials and Methods:** A total of 897 healthy blood donors with mean age  $37.23 \pm 10.9$  years were enrolled in the study. Laboratory tests including Standard Tube Agglutination Test (STA) and 2-mercaptoethanol (2ME) agglutination were checked in all samples. STA dilution  $\geq 1:80$ , and in the presence of 2-mercaptoethanol (2ME) agglutination  $\geq 20$  was considered positive.

**Results:** Out of 897 cases, 11.9% were inhabitants of rural areas. 41.5% had history of consumption of unpasteurized dairy products and 9.3% had history of contact with domestic animals. A very low level of *Brucella* agglutinins was present in 3(0.33%) of the samples and only one sample (0.11%) was found to be truly positive for *Brucella* agglutinins. 2ME was negative in all samples. None of these 4 subjects showed signs and symptoms of brucellosis in 6 months follow-up.

**Conclusion:** On the basis of our data, brucellosis has no epidemiological and clinical importance in our blood donors; therefore, it is not recommended to perform screening tests such as, STA and 2ME to identify brucellosis antibodies in the sera of blood donors.

**Keywords:** *Brucella* agglutinins, blood donors, Brucellosis

---

### INTRODUCTION

Brucellosis is a zoonotic disease that may have a significant veterinarian, public health and economic impact (1, 2). It can be transmitted to humans through

close contact with infected animals or animal products and through the consumptions of unpasteurized milk and dairy products (3). In addition, it is an occupational hazard to persons involved in certain professions such as farming, ranchers, veterinarians and slaughterhouse workers (2, 4).

In humans, brucellosis is an acute febrile illness and behaves as a systemic infection with a very variable clinical spectrum. The pathogen can involve any organ such as the cardiovascular, musculo-skeletal and central nervous systems with sometimes serious complications, and symptoms can vary depending on

---

\* Corresponding author: Amitis Ramezani MD.  
Address: Clinical Research Dept. Pasteur Institute of Iran,  
13164, Pasteur Ave., Tehran, Iran.  
Tel: +98-21-66968852  
Fax: +98-21-66465147  
E-mail: amitisramezani@hotmail.com

the site of infection (1, 2, 5).

Brucellosis is endemic in certain parts of Iran. Recent clinical studies have shown that brucellosis is still a common health problem in Iran and sometimes causes severe clinical illness with complications (2, 6, 7). The prevalence of brucellosis in Iran has been reported to be 0.5% to 10.9% in different provinces. It's highly endemic in certain parts of Iran such as central province of Arak (Arak city) with five-year incidence of about 40.5-48/100,000 people (8).

Brucellosis was investigated in many high risk occupational groups; however, few studies on the prevalence of brucellosis among blood donors are available. Determination of the seroprevalence of brucellosis in high risk and low risk groups are very important for understanding of the nature of the disease and eradication of brucellosis. This study was carried out to investigate the background prevalence of *Brucella* agglutinins in blood donor population in central province of Iran as an endemic area for brucellosis.

## PATIENTS AND METHODS

A total of 897 blood donors attending the Markazi blood transfusion organization were investigated for *Brucella* agglutinin from April to May 2012. Informed consent was obtained from all cases. The project was approved by Arak University of Medical Sciences ethical committee.

Laboratory tests including Standard Tube Agglutination Test (STA = Wright) and 2-mercaptoethanol (2ME) agglutination were tested in all samples. The STA test was carried out with *Brucella abortus* plain antigen provided by Pasteur Institute of Iran. Serial dilution of the sera in Phosphate buffer saline (PBS) was performed from 1/10 to 1/1280. To each tube 0.5 ml of 10% *Brucella abortus* was added, and incubated at 37°C for 24 h. All the tubes were compared with antigen control tubes for degree of opacity of the supernatant fluid (9). Any serum with titers of 1/80 or above was considered as a positive result.

The 2ME solution was obtained from Pasteur Institute of Iran. The serum treated with 2ME is tested at the same dilutions as STA. To each tube 0.5 ml of 10% *Brucella abortus* was added, and incubated at 37°C for 24 h. The presence of 2ME agglutination  $\geq 20$  was considered positive.

All positive samples were following up for 6 months

for brucellosis signs and symptoms.

A clinical diagnosis of brucellosis was made on the basis of the symptoms, compatible clinical findings, STA test dilution  $\geq 1: 80$  and in presence of 2ME agglutination  $\geq 20$ .

**Statistical analysis.** The SPSS 16 Package program for statistical analysis (Chicago, IL, USA) was used. Data are presented as mean  $\pm$  SD or, when indicated, as an absolute number and percentage.

## RESULTS

Out of 897 cases, with mean age  $37.23 \pm 10.9$  years, 92.1% were male and 7.9% were female. 11.9% of them were habitant in rural area. 41.5% had history of consumption of unpasteurized dairy products and 9.3% had history of contact with domestic animals. Only 4% of cases had occupational risk for brucellosis acquisition. 5% of subjects had infected family members.

A very low level of *Brucella* agglutinin (1: 20) was present in 3 (0.33%) samples from healthy subjects and only one sample (0.11%) was found to be truly positive for *Brucella* agglutinin at 1:80. This sample came from a 26 years old male who lives in Khomein city with history of consumption of unpasteurized milk and milk products and without any history of brucellosis infections and occupational exposure to cattle; but he suffered low back pain. 2ME was negative in all samples. Blood cultures were done in 4 cases and showed negative results. None of these 4 subject revealed signs and symptoms of brucellosis in 6 months follow up.

The *Brucella* agglutinin positive case did not receive any brucella medication because 2ME was negative and his low back pain was mechanical and he did not manifest any brucellosis signs and symptoms in 6 months follow up.

## DISCUSSION

In this study, we investigated the prevalence of *Brucella* agglutinin in blood donor population in central province of Iran and evaluated the epidemiological, clinical, laboratory findings and outcome of serologic positive cases. This survey showed that the prevalence of *Brucella* agglutinin was negligible in blood donors in this region and most cases had insignificant levels of *Brucella* agglutinin,

most likely due to endemicity of the disease. Only one sample was found to be truly positive for *Brucella* agglutinin.

Several studies showed that consumption of unpasteurized dairy products especially in endemic areas is a significant risk factor for brucellosis (2, 10, 11). Brucellosis is also an occupational hazard. Slaughterhouse workers and others involved in animal keeping and handling are at higher risk for disease acquisition (2, 11, 12, 13).

Nikokar et al. (11) reported that the seroprevalence of brucellosis among slaughterhouse workers and the people living in rural areas were 9.8% and 5.5% respectively in North of Iran. The seropositivity rate of 7.8% was reported in high risk groups in South of Iran and they indicated that profession is the main risk factor for seropositivity (14). In a study in Bangladesh, brucellosis seroprevalence determined 11.11% in veterinary personnel, 6.45% in dairy workers and 4.67% in animal farmers (15). In another study on slaughterhouse workers (meat sellers, slaughterers, animal keepers ...) in Pakistan, seroprevalence of *Brucella* antibodies reported 21.7% (16). Swai et al reported that the overall *Brucella* antibodies seroprevalence in abattoir workers of Tanzania was 5.52% (17).

Bhat et al. (18) reported a seroprevalence rate of 8.5% in the general population of Belgaum. In a study by Ajay Kumar et al (19) on the general population and veterinary students, brucellosis prevalence rate of 2.45% and 1.14% were observed respectively.

There is a paucity of literature on *Brucella* agglutinin in blood donors. Torres-Padilla et al. (20) determined the *Brucella* seroprevalence in blood donors of Mexico. They showed brucella seroprevalence in 3.6% of cases. They recommended performing screening tests such as Bengal rose (BR), STA and 2ME to identify brucellosis antibodies in the sera of blood donors. In contrast another study by Vaishnavi et al. (21) in blood donor population of India showed a very low level of *Brucella* agglutinins in 16.8% of the samples and only one sample (0.36%) was positive for *Brucella* agglutinins at 160 IU/mL.

Few studies were carried out on *Brucella* agglutinins prevalence in Iranian blood donors. In a survey conducted by Ghilian et al. (22) in blood donors of Yazd, 6.3% of participants had a STA titer of 1:80 but 2-ME test was positive only in 0.6% of cases.

In contrast, Rabbani Khorasani et al. screened blood donors of Boushehr province (south of

Iran) for serological evidence of brucellosis. *Brucella* agglutinins found in 0.057% of sera samples and 2ME test was positive only in one sample (from 10500 blood donors) with low titer (1/20) without any symptoms of active infection (23).

The young male in our study with positive *Brucella* agglutinin lives in the city and have not provided any past history of brucellosis, cattle contact and high risk occupation for acquisition of the disease; but this seropositivity may be due to consumption of unpasteurized milk and milk products or exposure to an agricultural background.

In conclusion, on the basis of our data, brucellosis has no epidemiological and clinical importance in our blood donors; therefore it is not recommended to perform screening tests such as STA and 2ME to identify brucellosis antibodies in the sera of blood donors.

#### ACKNOWLEDGEMENT

The authors are grateful to Arak University of Medical Sciences for financial support of this study. This study was part of Mehrnoosh Sheikholeslami's MD thesis

#### REFERENCES

1. Gur A, Geyik MF, Dikici B, Nas K, Cevik R, Sarac J, et al. Complications of brucellosis in different age groups: a study of 283 cases in southeastern Anatolia of Turkey. *Yonsei Med J.* 2003; 44: 33-44.
2. Sofian M, Aghakhani A, Velayati AA, Banifazl M, Eslamifar A, Ramezani A. Risk factors for human brucellosis in Iran: a case-control study. *Int J Infect Dis* 2008; 12: 157-161.
3. Bikas C, Jelastopulu E, Leotsinidis M, Kondakis X. Epidemiology of human brucellosis in a rural area of north-western Peloponnese in Greece. *Eur J Epidemiol*, 2003;18: 267-274.
4. Me'ndez Marti'nez C, Pa'ez Jime'nez A, Corte's-Blanco M, Salmoral Chamizo E, Mohedano Mohedano E, Plata C, et al. Brucellosis outbreak due to unpasteurized raw goat cheese in Andalu'cia (Spain), January—March 2002. *Euro Surveill* 2003; 8: 164-8.
5. Colmenero JD, Reguera JM, Martos F, Sa'nchez-De-Mora D, Delgado M, Causse M, et al. Complications associated with *Brucella melitensis* infection: A study of 530 cases. *Medicine (Baltimore)* 1996; 75: 195- 211.
6. Hasanjani MR, Mohrez M, Smailnejad SM, Soleimani MJ, Hajiahmadi M. Epidemiological features and clinical manifestations in 469 adult patients with brucellosis in Babol, northern Iran. *Epidemiol Infect* 2004; 132: 1109-1114.

7. Afsar H, Baydar I, Sirmatel F. Epididymo-orchitis due to brucellosis. *Br J Urol* 1993; 72: 104-105.
8. Farahani SH, Shah Mohamadi S, Navidi I, Sofian M. An investigation of the epidemiology of brucellosis in Arak City, Iran, (2001-2010). *Arak Medical University Journal* 2012, 14: 49-54. [In Persian].
9. Collee JC, Dugid JP, Fraser AG, Marmion BP. Mackie & McCartney Practical Medical Microbiology, Churchill Livingstone, New York, 1989. p. 529.
10. Gwida M, Al DS, Melzer F, Rosler U, Neubauer H, Tomaso H. Brucellosis - regionally emerging zoonotic disease? *Croat Med J* 2010; 51: 289-295.
11. Nikokar I, Hosseinpour M, Asmar M, Pirmohbatee S, Hakeimeh F, Razavei MT. Seroprevalence of Brucellosis among high risk individuals in Guilan, Iran. *J Res Med Sci* 2011; 16: 1366-1371.
12. Al-Majali AM, Talafha AQ, Ababneh MM, Ababneh MM. Seroprevalence and risk factors for bovine brucellosis in Jordan. *J Vet Sci* 2009; 10: 61-65.
13. Yoo SJ, Choi YS, Lim HS, Lee K, Park MY, Chu C, et al. [Seroprevalence and risk factors of brucellosis among slaughterhouse workers in Korea] *J Prev Med Public Health*. 2009; 42: 237-242.
14. Beheshti S, Rezaian GR, Azad F, Faghiri Z, Taheri F. Seroprevalence of brucellosis and risk factors related to high risk occupational groups in kazeroon, South of Iran. *Int J Occup Environ Med* 2010; 1: 62-68.
15. Muhammad N, Hossain MA, Musa AK, Mahmud MC, Paul SK, Rahman MA, et al. Seroprevalence of human brucellosis among the population at risk in rural area. *Mymensingh Med J* 2010; 19: 1-4.
16. Mukhtar F, Kokab F. Brucella serology in abattoir workers. *J Ayub Med Coll Abbottabad* 2008; 20: 57-61.
17. Swai ES, Schoonman L. Human brucellosis: seroprevalence and risk factors related to high risk occupational groups in Tanga Municipality, Tanzania. *Zoonoses Public Health* 2009; 56: 183-187.
18. Bhat K, Hemashettar, BM, Jain R, Anrade AT, Patil CS. Latex agglutination test for antigen detection in human brucellosis. *Indian J Med Microbiol* 1997; 15: 210.
19. Ajay Kumar VJ, Nanu E. Sero-positivity of brucellosis in human beings. *Indian J Public Health* 2005; 49: 22-24.
20. Torres-Padilla JC, López-Merino A, García-Escamilla RM, Gutiérrez-García JN. Anti-Brucella antibody seroprevalence in blood donors for therapeutic ends at three blood banks of the Mexican Institute of Social Security. *Gac Med Mex* 2004; 140: 391-398.
21. Vaishnavi C, Kumar S. Investigation for background prevalence of Brucella agglutinins among blood donors. *Indian J Med Microbiol* 2007; 25: 302-304.
22. Ghilian R, Hekmati Moghaddam SH, Fatemi A, Eslamieh H, Dargahi M. Seroepidemiologic status of brucellosis in blood donors in Yazd, 2009. *Sci J Blood Transfus Organ* 2011; 7 : 196-205.
23. Rabbani Khorasgani M, Esmaeili H, Pourkarim MR, Mankhian AR, Zahraei Salehi T. Anti-brucella antibodies in blood donors in Boushehr, Iran. *Comp Clin Pathol* 2008; 17: 267-269.