Original article

Brucellosis seropositivity among adults in al rass city, qassim province, saudi arabia

Reham Alhoshani¹, Sarah Ali¹, Uma M. Irfan¹

1- Department of Medical Laboratories, College of Applied Medical Sciences, Qassim University, Buraydah, Al Qassim, KSA

Corresponding author: Sarah Ali

Email: dr_ sarahrazi@rocketmail.com

Abstract

Introduction: Brucellosis constitutes a major health and economic problem in many parts of the world, including countries Middle East and the Arabian Gulf. Epidemiological investigations conducted in the Middle East, demonstrated a wide spread distribution of brucellosis in the region, with Saudi Arabia having the highest prevalence of brucellosis. Thus the aim of the study was to evaluate the prevalence of brucellosis in Al-Rass city, Qassim region of Saudi Arabia and to determine its associated factors.

Materials and Methods: A cross-sectional survey was carried out in March 2013 with a random consecutive sample of 40 subjects visiting Aljreef dispensary, in Al-Rass city, Qassim region, K.S.A. The blood samples obtained from the study subjects were analyzed in two steps namely: (1) Slide agglutination test; and (2) Standard tube agglutination test.

Results: The prevalence of brucellosis in this study sample was 15% predominantly in the age group of 34 to 46 years. The two predisposing factors associated with brucellosis prevalence were consumption of raw milk and daily direct contact with animals.

Conclusion: It can be concluded that there is a high prevalence of Brucellosis in Al-Rass city, Qassim region. The prevalence of Brucellosis is significantly associated with drinking raw milk and direct contact with animals. An improved healthcare system with outreach programs to educate farmers on Brucellosis preventive measures is needed to minimize the prevalence.

Keywords: : Prevalence, Seropositivity, StandardTube Agglutination Technique (TAT), Slide Agglutination Test, Brucella melitensis,Brucella abortus.

Introduction

Brucellosis constitutes a major health and economic problem in many parts of the world, including countries Middle East and the Arabian Gulf. Brucellosis being primarily a contagious zoonotic disease of domestic livestock's such as goats, sheep, cows and camels, is a constant source of infection to human, and transmitted directly or indirectly, resulting in increased mortality and morbidity among human and animal livestock, leading to huge financial and economic deficit in region. Epidemiological investigations this conducted in the Middle East, demonstrated a wide spread distribution of brucellosis in the region, with Saudi Arabia presenting the highest prevalence of brucellosis.

Brucellosis, commonly known as "undulant fever", "Mediterranean fever" or "Malta fever", constitutes a major health and economic problem in many parts of the world, including countries of Middle East and the Arabian Gulf. It has emerged as a global problem, being, a contagious zoonotic infection^{[1],}primarily among domestic animals such as goats, sheep, cows and camels, transmitted to human directly or indirectly through meat, milk and animal contact. The causative organism is a Gram-negative, rod shaped (coccobacilli), nonmotile. non-spore-forming, facultative intracellularbacteria causing chronic disease. There are four pathogenic species to human:1) Brucella melitensis, found primarily in goats, sheep and camels; 2) Brucella abortus in cows; 3) Brucella suis in pigs; and 4) Brucella canis in dogs. The Brucella species differ in degree of virulence and invasiveness. B. melitensis is the most invasive and produces the most severe disease. B. abortus is the least invasive and causes the mildest illness. In Saudi Arabia, human infection with B. melitensis is

commonly encountered (80%-100%), and B. abortus is less frequent $^{[2, 4, 5, 6]}$.

Important diagnostic methods include; 1) isolation of Brucella from blood, tissue specimens, body fluids and bone marrow; (2) agglutination test (3) ELISA test and 4) polymerase chain reaction (PCR). Saudi Arabia has incidents recorded a critical points when the reported cases reached 8000 cases (22.5%) reported each year to public health authorities which stressed clearly that Saudi Arabia is hyper- endemic for Brucellosis prevalence^[9, 12]

Methods

Across-sectional survey was carried out during March to June 2013, with a random consecutive sampling of 40 subjects attending AlJreef Dispensary, in Alrass city, Qassim region, which has a catchment area of seven localities around the Dispensary. The total residing population of Alrass city is about 92, 501 according to Saudi Census estimates. The study sample included adult males and females who were willing to participate after giving informed consent.

Data was collected using a structured questionnaire with 16 questions as an interview schedule. The researcher interviewed all the study subjects to obtain information on demographics, dietary habits including milk handling and consumption such as whether they drank raw milk, consumed milk products, eat raw meat , had contact with livestock, handled parturient animal, had direct contact with animals, history of previous Brucellosis infection, and any current symptoms of Brucellosis. Then a 5 ml venous blood sample was obtained from each participant by a trained nurse at the Dispensary.

Laboratory Tests: Blood samples were collected, and kept at room temperature in the laboratory, and serum was separated from collected blood by centrifugation and stored at -20 ° C, until tested for presence of Brucella antibodies. Serum samples were analyzed in two phases, using rapid slide agglutination kit comprising antigen suspension of B. abortus and B. melitensis. In the first phase, all specimens were screened by the slide agglutination test. Presence of agglutination (visible clumps on slide / white card) was considered as positive for Brucella antibodies (seropositive). In the second phase, seropositive serum samples were analyzed by the standard tube agglutination test (STAT). A positive agglutination in diluted serum samples, with titer of 1:80 or greater was considered as an index of seropositivity.

Data was entered and analyzed by in SPSS (v20) to determine the prevalence of brucellosis. Descriptive statistics were used to examine the distribution of study variables individually with mean and standard deviation for continuous variable and frequency and percent for categorical variables. Comparative measures using quantitative variables were analyzed by independent sample t-test with a level significance set at 0.05

antibodies (**seropositive**). In the second phase, seropositive serum samples were analyzed by the standard tube agglutination test (STAT). A positive agglutination in diluted serum samples, with titer of 1:80 or greater was considered as an index of seropositivity.

Data was entered and analyzed by in SPSS (v20) to determine the prevalence of brucellosis. Descriptive statistics were used to examine the distribution of study variables individually with mean and standard deviation for continuous variable and frequency and percent for categorical variables. Comparative measures using quantitative variables were analyzed by independent sample t-test with a level significance set at 0.05.

Results

A total of 40 blood samples were collected from 45% males and 55% females with a mean age of 37.9 years and standard deviation of 11.12 years (Table 1).From the 40 blood samples, we found 7.5% males had Brucella antibodies at titer of 80, while only 2.5% females had antibodies of these levels at titer of 80.Among the six seropositive cases (1:80), the odds ratio of males having seropositivity was 4.2 times greater than the odds of females who were seropositive, however this association of sex verses seropositivity was statistically not significant (p > 0.05). The odds education and employment versus seropositivity for Brucellosis was found non-significant.

Table 1: Demographic Characteristics of theStudy Sample with the Sero-positive Status ofBrucellosis

Demographic Characteristics	Total Number of Samples	Brucella Sero-positivity Number (%)	P-value	Relative Odds (95% CI)
Age	10			
(20-33) (34-46)	18	0	0.039	1.0*
(47-60)	18	2 (5%)		1.15(0.94-1.40)
	4	2 (5%)		0.53(0.06-4.69)
Sex				
Male	10			1.04
Female	18	3(7.50 %)	0.242	1.0*
	22	1(2.50 %)		4.2 (0.39-44.4)
Education				
No education		2/7 500/)	0.005	1.0*
undergraduate	21	3(7.50%)	0.895	1.0*
Post graduate	6	0		0.85(0.72-1.02)
0	13	1(2.50%)		1.08 (0.92-1.26)
Occupation				
Student	5	0	0.590	1.0*
Unemployed			0.590	
Housewife	14 13	2(5%) 1(2.50%)		0.85(0.64-1.0) 0.50 (0.04-6.2)
Other	8	1(2.50%) 1(2.50%)		0.50 (0.04-6.2) 1.00 (0.92-1.26)
Total	0	1(2.50 /0)		1.00 (0.72-1.20)
10121	40	4(10%)		
	40	4(1070)		

Prevalence of Brucellosis in the Sample: Out of 40 serum samples tested for presence of Brucella antibody by rapid slide agglutination (screening) test 35% were positive. In that, 25% of serum samples were seropositive for Brucella melitensis, while10% samples were seropositive for Brucella abortus. Standard Tube Agglutination Test (STAT) done on 35% seropositive samples (Table 2) revealed 28.6 % with seropositivity for Brucella melitensis at titer of 1:80, while 14.3% samples

were seropositive at titer of 1:80 for Brucella abortus. The overall sero-prevalence of Brucella antibody by Tube agglutination testwith titer 1:80 among thesurveyed individuals was found to be 15%.

The study results could be extrapolated to the resident population of Alrass city as the prevalence of B.melitensis to be 286 per 1000 population and the prevalence of B. abortus to be 142 per 1000 population.







Variable	Total # of samples tested (%)	No. of Sero- positive samples (%)	p-value
Slide AgglutinationTest: Brucella abortus Brucella melitensis	40(100) 40 (100)	4(10) 10 (25)	0.004*
Tube Agglutination Test (STAT): Brucella abortus:1:10 1:20	14 (100)	4 (28.6)	< 0.05*
1:40 1:80	14 (100) 14 (100) 14 (100)	3(21.4) 3(21.4) 2(14.3)	
Brucella melitensis:1:10 1:20 1:40 1:80	14(100) 14(100) 14(100) 14(100)	8(57.2) 7(50.0) 5(35.7) 4(28.6)	< 0.05*
	1 (100)	(200)	

Table 2: Results of the Tests to identif	y Sero-positivity for Brucellosis
--	-----------------------------------

* p< 0.05

The commonest symptoms and signs among the six seropositive cases as reported are summarized in Figure2. Muscle pain was the most common symptom reported. Among the total number of positive cases 75%, 75%, 25%, 25%, and 25% respectively suffered from fever, lethargy, sweating, gastrointestinal symptoms and joint pain.





Discussion

Brucellosis is the most endemic zoonotic disease in Saudi Arabia, with a high prevalence among man and livestock. Along with many studies conducted to assess the situation in many regions of Saudi Arabia, this study estimated the prevalence of brucellosis and the associated risk factors in Alrass city, Qassim region. The results of this study revealed Brucellosis as a major health problem in Alrass city, Saudi Arabia. The overall prevalence rate of Brucellosis based on slide screening test was found to be 15%, while prevalence of seropositivity at 1;80 titer was 42.8%. In our study, the samples were taken randomly from asymptomatic cases, in which the above prevalence rate is comparatively higher than

International journal of Medical Investigation

other studies. This prevalence rate was less than Al-Sekait's finding from theNorthern Region (20%), and Al-Balla's finding from Southern region (18.3%), but higher than and Al-Mofleh's finding in the Central Region (14.6%) of Saudi Arabia ^{[23],[24],[25]}. In view of the fact that this study was conducted among the general asymptomatic population while other above studies included acute cases and high risk population, the mentioned prevalence rate is high in Al-rass city.

Meimish et al, in 2004, reported the prevalence of seropositivity at titer $\geq 1:160$, inpatients of acute brucellosis as 58%, while prevalence of seropositivity at titer >1:160.asymptomatic cases were 8%, while our study showed prevalence of Most other Middle East countries have 5%. reported a lower prevalence rate compared to Saudi Arabia.Dajani ' finding from Jordan (0.03 %), Makarem's finding from Southern Iran(0.01%) and Mousa's finding from Kuwait (1.5%) ^{[26],[27],[28].}The prevalence reported from other developing countries were: Alausa's finding from Western Nigeria (0.8%) and Osman's finding from Kenya [29],[30] (1.3%)and from developed countries:Davos's finding from South Australia (0.7%) and Chomel's finding from California $(0.2\%)^{[31],[32]}$.

Brucellosis is diagnosed by combination of serological tests and clinical findings consistent with brucellosis. Although no single test provides 100% sensitivity and specificity, STAT still remains the test of choice for screening and diagnosis. In the presence of appropriate signs and

symptoms, a presumptive diagnosis of brucellosis is usually defined serologically as a standard tube agglutination titer of 1:80. In this study, it was found, that the prevalence of acute brucellosis was 5 % among individuals between 34 and 46 years of age which is not similar to the rates cited in other reports which mention that risk of brucellosis increases with age ^{[23],[24],[34],[24],[25],[35],[26]}.

The relatively low prevalence found in age group (20-33) compared with age group (34-60) may be the result of raw milk consumption and close contact with livestock. In contrast to other studies, there was no significant difference in the prevalence between male and females in all age groups. This could indicate that both sexes had close contact with animals. Animal shelters are close to human dwellings and the women of this region are just as involved in animal care as men. The widespread habit of drinking raw milk may also diminish any difference in exposure to the disease between the sexes.

While this study shows no association with occupation as it was conducted in general population of Alrass, it did not include the highrisk occupations as other studies have done, but indicated that less educated individuals, had a higher prevalence rate than individuals with higher levels of education, because those with low education were more likely to be related with high risk occupations, such as people working with farm animals, shepherds, sheep shearers and goatherds.

The presenting symptoms and the clinical manifestation in our study were similar to those reported in other studies ^{[36].} The rheumatological findings being second only to fever in the clinical symptoms of the cases in this study, the prevalence of muscle pain was 100% which differed significantly from other observational studies. The rate of detection of gastrointestinal symptoms, night sweat and joint pain was lower than that reported in other studies.

The results of this study indicate that the acquisition of Brucella in these individuals may have beenthrough either the contact with infected animals or through the drinking of raw milk as reported in several other studies ^{[23],[33],[34],[24],[25]}.

Conclusion

The prevalence of brucellosis in Saudi Arabia is reported to be 15%, equivalent to the prevalence of brucellosis in Alrass city (15%) as estimated in this study. This significant result could be attributed to two factors namely: the traditional use of raw milk products and having close contact with infected animals particularly goats and sheep.

Although brucellosis had been eradicated from a number of developed countries, it continues to be a major public health and animal health problem in many regions of the world. A well-developed healthcare system and preventive measures would help to reduce potential infection risks and decrease incidence of Brucellosis.

Recommendations: It is recommended to Practice the following control measures:

- 1. Strict adherence to hygienic practices on the farm, and adoption of a policy for disposal of infected animals.
- 2. Proper surveillance of brucellosis in population at risk.
- 3. Surveillance and Quarantine at borders with neighboring countries to control brucellosis.
- 4. Health education programs, and Compulsory Vaccination of Cattle and livestock.

References

1-Ariza J, Bosilkovski M, Cascio A, Colmenero J, Corbel M, et al. (2007) Prospectives for the Treatment of Brucellosis in the 21st Century: The Ioannina Recommendations. PLoS Medicine 4: e317. doi: 10.1371/journal.pmed.0040317.

2. Corbel M (2006) Brucellosis in Humans and Animals: FAO, OIE, WHO. Available: http://www.who.int/csr/resources/publica

tions/Brucellosis.pdf. Accessed 2012 May 7.

3. Roth F, Zinsstag J, Orkhon D, Chimid-Ochir G, Hutton G, et al. (2003) Human health benefits from livestock vaccination for brucellosis: case study. Bulletin of the World Health Organization 81: 867– 876.

4. Perry BR (2002) Ch. 7 - Animal disease impact on the poor: study results. Investing in Animal Research to Alleviate Poverty. Nairobi: International Livestock Research Institute. pp. 67– 78.

5. World Health Organization (2006) The Control of Neglected Diseases: A route to poverty alleviation. Available:

http://www.who.int/zoonoses/Report_Sept06.pdf. Accessed 2012 May 7

6. Pappas G, Papadimitriou P, Akritidis N, Christou L, Tsianos EV (2006) The new global map of human brucellosis. Lancet Infect Dis 6: 91–99. doi: 10.1016/S1473-3099(06)70382-6.

7- Pappas, G. (2006). The new global map of human brucellosis. Lancet Infect Dis.; 6: 91–99.

8- Franco, M. P. (2007). Human brucellosis. Lancent Infect Dis.; 7: 775–86

9-Memish Z. A., (2001). Brucellosis control in Saudi Arabia: prospects and challenges. J Chemother Suppl.; 1:11-7.

10-Al-Eissa, Y A. (1999). Brucellosis in Saudi Arabia: past, present and future. Ann Saudi Med, 19:403-405

11-Ministry of Health Annual Report. (2006.(http://www.moh.gov.sa/statistics/S1427/Chapter%2 01.pdf

12-Elfaki, M. G., Al-Hokail A.A, Nakeeb S. M, Al-Rabiah, F. A. (2005).Evaluation of culture, tube agglutination, and PCR methods for the diagnosis of brucellosis in humans. Med SciMonit; 11: MT69-74

13- Al-Sekait M. Epidemiology of Brucellosis in Al-Medina Region, Saudi Arabia.J. of Family and Community Medicine 1999, (in publication).

14- Fallatah, S. M., Oduloju A. J, Al-Dusari S. N, Fakunle Y. M. (2005). Human brucellosis in Northern Saudi Arabia. Saudi Med. J. 26:1562-1566

15-Malik G. M. (1997). A clinical study of brucellosis in adults in the Asir region of southern Saudi Arabia. Am J Trop Med Hyg. 56:375-377

16. Alballa, S. R. (1995). Epidemiology of human brucellosis in southern Saudi.Arabia.J Trop Med Hyg. 98: 185-189 17-Cooper, C. W. (1991). The epidemiology of human brucellosis in a well defined urban population in Saudi Arabia. J Trop Med Hyg. 94: 416-422

18 - Al-Sekait M. Epidemiology of Brucellosis in Al-Medina Region, Saudi Arabia.J. of Family and Community Medicine 1999, (in publication).

19-Kiel F. W., Khan M. Y. (1993). Brucellosis among hospital employees in Saudi Arabia. Infect Control HospEpidemiol. 14:268-272.

20 - Elbeltagy KE. An epidemiological profile of brucellosis in Tabuk Province, Saudi Arabia. East Mediterr Health J 2001;7(July—September (4— 5)):791—8

21-Al-Sekait M. A. (1993). Prevalence of brucellosis among abattoir workers in Saudi Arabia. J. R. Soc. Health. 113:230-233

22- Talukdar MAS, Abomelha MS, Higham RH (1984) Brucellosis in a farming community in Saudi Arabia. Dev Biol Stand 56:593–595.

23- Al-Sekait MA. Epidemiology of Brucellosis in Northern Saudi Arabia.Saudi Medical Journal 1992; 6:29-31.

24- Al-Moleh IA, Al-Aska AL, Al-Sekait MA, Al-Balla SR, Al-Nasser AN. Brucellosis in Saudi Arabia: Epidemiology in the central region. Annals of Saudi Medicine 1996;16:349-52.

25.Al-Balla SR. Epidemiology of Human brucellosis in Southern Saudi Arabia. J Trop Med & Hygiene 1995;95:185-9.

26-Dajani Y, Masoud A, Barakat H. Epidemiology and diagnosis of human brucellosis in Jordan. Journal of Tropical Medicine and Hygiene 1989;92:209-14.

27-Makarem E, Karjoo R, Omidi A. Frequency of Brucella Melitensis in Southern Iran. Journal of Tropical Pediatrics 1982;28:97-100.

28-Mousa A, Elhaq K, Khogali M, Marafie A. Nature of Human Brucellosis in Kuwait. Reviews of Infectious Disease 1988;19:211-7.

29-Alausa O, Awoseyi A. Brucellosis: The situation in Western Nigeria. Tropical Geographic Med 1976;28:54-9.

30-Osman AJI.Human Brucellosis in Kenya. Tropical Geographic Medicine 1976;28:45-53.

31-Davos D, Cargill C, Collin F, Kyrkou M, Jamieson J, Rich G. Outbreak of Brucellosis at a South Australian Abattoie. Medical Journal Aust 1981;2:657-60.

32-Chomel BB, De Boss EE, Mangiamele DE, Reilly KF. Changing trends in the Epidemiology of human brucellosis in California from 1973-1992. J Infect Disease 1994;170:1216-23.

33-Kambal A, Mahgoub E, Jamjoom G, Chowdry M. Brucellosis in Riyadh, Saudi Arabia. Trans R Soc Trop Med Hyg 1983;77:820-4.

163

34-Madkour M, Rahman A, Mohammed E, Talukdar M, Kudwah A. Brucellosis in Saudi Arabia. Saudi Medical Journal 1985; 6:324-32.

35 -Hashim N, Galil G, Hulaibi M, Saleem E. The incidence of brucellosis and species of brucella organism isolated from Al Hassa. World Animal 1987;61:32-5.

36 -Norton LW.Brucellosis and rheumatic syndromes in Saudi Arabia. Ann Rheumatology Disease 1984;43:810-5.

37 -Al-SekaitMA .Seroepidemiology survey of brucellosis antibodies in Saudi Arabia . Ann Saudi Med. 1999 May-Jun;19(3):219-22.

38- Lifeso RM, Harder E, McCorkell SJ. Spinal brucellosis. J Bone Joint Surg Br. 1985 May;67(3):345–351