MedCrave
Step into the World of Research

Journal of Microbiology & Experimentation

Research Article

Open Access

CrossMark

Comparison of standard agglutination test and enzyme-linked immunosorbent assay to detect brucella infection in yemeni pregnant women

Abstract

Introduction: Brucellosis is a zoonotic disease caused by genus Brucella, transmitted to humans by direct or indirect contact with infected domestic animals or their dairy products.

Materials and methods: This study was carried out to compare between standard agglutination test and an enzyme linked immunosorbent assay to detect human brucellosis among pregnant women attending antenatal clinic in some hospitals and health centers in Sana'a city, southern Yemen.

Results: A total of 304 serum samples were tested to detect brucella antibodies of which 87(28.61%) were positive by SAT. However, only 45(15%) were positive by ELISA. Out of 87 participants positive by SAT 58(66.67%) were positive for Brucella abortus and melitensis, 24(27.59%) were positive for brucella abortus, 5(5.75%) were positive for brucella melitensis. Out of 45 subjects positive by ELISA, 17(6%) were positive for IgG antibodies, 42(14%) were positive for IgM antibodies.

Conclusion: A number of seropositivity for brucella antibodies by SAT was more than by ELISA. We recommend confirming the results of SAT by other test such as ELISA because the ELIAS is more sensitive than SAT.

Keywords: brucella, comparison, pregnant women, elisa, sa, yemen

Volume 5 Issue 5 - 2017

Mofeed Al Nowihi, Qais YM Abdullah, Salwa H Alkhyat, Anas A Almahbashi, Assem AlThobahni, Mohammed NQ Al Bana, Saad Al Arnoot

Department of Biology, Sanaa University, Yemen

Correspondence: Saad Al Arnoot, Microbiology Branch, Department of Biology, Faculty of Science, Sana'a University, Sana'a, Yemen , Tel 00967-774667900, Email saad.alarnoot@gmail.com

Received: July 27, 2017 | **Published:** August 21, 2017

Abbreviations: CFT, complement fixation test; CS, cores sectional; ELSIA, enzyme linked immunosorbent assay; RBPT, rose bangel plate test; SAT, serum agglutination test; STA, standard tube agglutination; MRT, milk ring test; IgG, immunoglobulin G, IgM, immunoglobulin M

Introduction

Brucellosis is one of the most common zoonotic diseases worldwide, which caused by genes of brucella.[1] Brucella is a gram-negative, cocobacilli, an aerobic, non-fermenting, facultative intracellular, non-motile, non-spore-forming [2,3]There are four species of *Brucella (B.)* genus (*B. abortus*, *B. melitensis*, *B. suis* and *B. canis*) are causative agents of brucellosis in humans.[4,5] Brucellosis is an endemic disease in Yemen, Middle East, Mediterranean basin, and South America.[6–8] It has been estimated that more than 500,000 human *Brucella spp.* infections occur per year with most reported cases occurring in the Syrian Arab Republic, followed by Mongolia, Kyrgyzstan and Iraq.[9] It is transmitted to humans by direct contact with infected animals or consumption of their raw products such as unpasteurized milk or cheese.[10,11] Milkers, live-stock farmers, abattoir workers, shepherds, veterinarians, meat processing workers and laboratory workers are at high risk of getting infected [12,13]

There are many ways for brucella diagnosis but the most commonly used serological tests which are suitable for screening herds and individual animals are: Rose Bengal test (RBT), standard agglutination test (SAT), complement fixation test (CFT), enzyme linked immunosorbent assay (ELISA) and milk ring test (MRT).[14,15] However, the confirmation diagnosis of human brucellosis used by bacteriological isolation and identification of the causative organism, but this method is complicated and time-consuming.[14,16] Presumptive diagnosis can be made by serological tests. This study amid to compare between SAT and ELISA to detect human brucellosis among pregnant women.

Material and methods

A total of 304 blood samples were collected from pregnant women, who visited the antenatal clinic in some hospitals in Sana'a city, southern Yemen during the period from Jun 2016 to November 2017. Three to five-ml of blood was collected from the pregnant women by venipuncture, transferred into sterile anticoagulant-free sterile bottle, and allowed to clot. The clotted blood sample was centrifuged (3000 rpm, 5 min), and the serum was transferred into cryovials and stored at -10 to -20°C until analysis. All serum samples were tested by ELISA and SAT. SAT was used to determine antibodies for *B. abortus* and *B. meltiensis* according to industrial company. ELISA was used to determine IgG and IgM of brucella antibodies and confirmation to SAT.

Results

A total of 304 serum samples which tested to detect brucella antibodies 87(28.61%) were positive by SAT. However, only 45(15%) were positive by ELISA. Out of 87 participants positive by SAT 58(66.67%) were positive for *Brucella abortus* and *melitensis*, 24(27.59%) were positive for *brucella abortus*, 5(5.75%) were positive for *brucella melitensis*. Out of 45 subjects positive by ELISA, 17(6%) were positive for IgG antibodies, 42(14%) were positive for IgM antibodies. The cross-tabulation between SAT and ELISA (IgM and IgG) is shown in Table 1.

Table 1 Comparison between ELISA and SAT for diagnosis of brucella infection among pregnant women

ELISA / SAT	IgG			IgM		
	Yes	No	Total	Yes	No	Total
Brucella abortus	5(20.83%)	19(79.17%)	24(27.59%)	10(41.67%)	14(58.33%)	24(27.59%)
Brucella melitensis	0(0.00%)	5(100%)	5(5.75%)	1(20%)	4(80%)	5(5.75%)
B. abortus and B. melitensis	12(20.69%)	46(79.31%)	58(66.67%)	31(53.45%)	27(46.55%)	58(66.67%)
Total	17(19.54%)	70(80.46%)	87(28.6%)	42(48.28%)	45(51.72%)	87(100%)

Discussion

In this study we compare between SAT and ELISA to detect human brucellosis among Yemeni pregnant women. Out of 304 pregnant women, 28.61% were positive for brucella antibodies by SAT while, only 15% were positive for brucella IgG and IgM antibodies by ELISA. Our findings showed that the number of positive samples was higher by SAT than by ELISA. May be the SAT give false negative at low dilutions of serum, but false negative reactions can be avoided by diluting the serum beyond 1/320. Moreover, false-positive reactions can also be obtained in SAT from cross-reactions with antibodies to *Salmonella spp.*, *Yersinia spp.*, *Vibrio cholera*, *Francisella* and other gram negative bacilli sharing common antigens.[17] In this study, we suggest that the sensitivity and specificity of ELISA was more than the SAT. This agree with other previous studies[18–20] who reported that ELISA typically uses the cytoplasmic proteins as antigens and measures IgM, IgG, and IgA, which allow for better interpretation. On the other hand, some other studies reported that this is not true with some studies who reported that ELISA was more sensitive than the SAT for the diagnosis of human brucellosis.[21, 22]

ELISA as it is the most sensitive and specific serological assay, but ELISA tests are relatively costlier tests in comparison to SAT that require equipment and experience. In a comparative study conducted by Araj et al, it was argued that the ELISA method should be preferred because in chronic and complicated cases, SAT and Rose Bengal tests might miss a serious portion of positive cases.[23] Limitation of study: we had no gold standard test to determine the sensitivity and the specificity of the tests because we did not try to isolate the organism. Also, we were ambitious to use PCR technique, isolation and identification of the bacteria but we could not because of the low facilities.

Conclusion

A number of seropositivity for brucella antibodies by SAT was more than by ELISA. We recommend confirming the results of SAT by other test such as ELISA because the ELIAS is more sensitive than SAT.

Competing interest

There were no financial, personal, or professional competing interests influenced this paper.

Acknowledgments

We would like to pass our gratitude and appreciations to Department of Biology, Faculty of Science, Sana'a University, Sana'a, Yemen for their support. We are also grateful to Al- Thobhani Modern Medical Laboratory, Sana'a, Yemen for their cooperation.

Funding

None.

References

1. Corbel MJ. Brucellosis an overview. *Emerg Infect Dis*. 1997;3(2):213–221.
2. Grimont F, Verger JM, Cornelis P, et al. Molecular typing of brucella with cloned DNA probes. *Res Microbiol*. 1992;143(1):55–65.
3. Moreno E, Stackebrandt E, Dorsch M, et al. *Brucella abortus* 16S rRNA and lipid A reveal a phylogenetic relationship with members of the alpha–2 subdivision of the class Proteobacteria. *J Bacteriol* . 1990;172(7):3569–3576.
4. Sauret JM, Vilissova N. Human brucellosis. *J Am Board Fam Pract*. 2000;15(5):401–406.
5. Young E. An overreview of human brucellosis. *Clin Infect Dis*. 1995;21(2):283–290.
6. Al Arnoot S, Abdullah QYM, Alkhyat SH, et al. Human and Animal Brucellosis in Yemen. *J Hum Virol Retrovirol*. 2017;5(4):00162.
7. Corbel MJ. Recent advances in brucellosis. *J Med Microbiol*. 1997;46(2):101–103.
8. Young EJ. Brucellosis. In: Connor DH, et al. editors. Pathology of infectious diseases. Stamford, USA, 1997;2:146.
9. Pappas G, Papadimitrou PH, Akritidis N, et al. The new global map of human brucellosis. *Lancent Infect Dis.* 2006;6(2):91–99.
10. Ali S, Ali Q, Neubauer H, et al. Seroprevalence and risk factors associated with brucellosis as a professional hazard in Pakistan. *Foodborne Pathog Dis*. 2013;10(6):500–555.
11. Chen S, Zhang H, Liu X, et al. Increasing threat of brucellosis to low–risk persons in urban settings. *China Emerg Infect Dis.* 2014;20(1):126–130.
12. Olabode HOK, Adah BMJ, Nafarnda WD, et al. Seroprevalence of *Brucella abortus* antibodies in slaughtered cattle and meat by–product handlers in Ilorin abattior, Kwara state–Nigeria. *Prime J Microbiol Res*. 2012;2:109–113.
13. Pathak AD, Dubal ZB, Doijad S, et al. Human brucellosis among pyrexia of unknown origin cases and occupationally exposed individuals in Goa Region, India. *Emerg Health Threats J.* 2014;7:23846.
14. Malone F, Athanassiou A, Nores L, et al. Poor perinatal outcome associated with maternal *Brucella abortus* infection. *Obstet Gynecol.* 1997;90(4):674–676.
15. Mangen MJ, Otte J, Pfeiffer D , et al. *Bovine brucellosis* in Sub–Sahran Africa:Estimation of Sero–Prevalence and Impact on Meat and Milk Offtake Potential. Livestock Information and Policy Branch. 2002. pp. 1–58.
16. Bamaiyi PH, Hassan L, Khairani Bejo L, et al. Updates on Brucellosis in Malaysia and Southeast Asia. *MJVR*. 2014;5(1):71–82.
17. Young EJ. *Brucella* species. In: Mandell GL, et al. editors. Principles and Practice of Infectious Diseases. 5th edn, Churchill Livingstone, Philadelphia, USA, 2000. pp. 2386–2393.

18. Fadeel MA, Wasfy MO, Pimentel G, et al. Rapid enzyme–linked immunosorbent assay for the diagnosis of human brucellosis in surveillance and clinical settings in Egypt. *Saudi Med J* 2006;27(7):975–981.

19. Osoba AO, Balkhy H, Memish Z, et al. Diagnostic Value of Brucella ELISA IgG and IgM in Bacteremic and Non–Bacteremic Patients with Brucellosis. *J Chemother.* 2001;13(suppl 1):4–9.

20. Young EJ. *Brucella* species. In:Mandell GL, et al. (Eds.), Principles and Practice of Infectious Diseases. 6th edn, Churchill Livingstone, Philadelphia, USA, 2005. pp. 2669–2672.

21. Kostoula A, Bobogianni H, Virioni G, et al. Detection of Brucella Ig G, IgM and IgA antibodies with ELISA method in patients with Brucellosis. *Clin Microbiol Infect*. 2001;7:108.

22. Prado A, Gutierres P, Duenas A. Serological Follow–up of Brucella patients using an immunocapture–agglutination test (Brucellacapt), Coombs anti–Brucella and LPS– ELISA tests. *Clin Microbiol Infect.* 2001;7:109–110.

23. Araj GF, Kattar MM, Fattouh LG, et al. Evaluation of The PANBIO Brusella immunglobulin G and IgM enzyme–linked immunosorbent assays for diagnosis of human bruscellosis. *Clin Diagn Lab Immunol*. 2005;12:1334–1335.

Citation: Al Nowihi M, Abdullah QYM, Alkhyat SH, et al. Comparison of standard agglutination test and enzyme-linked immunosorbent assay to detect brucella infection in yemeni pregnant women. *J Microbiol Exp.* 2017;5(5):11–12. DOI: 10.15406/jmen.2017.05.00160