The Relationship between Standard Tube Agglutination Titers in Brucellosis And Biochemical and Hematologic Parameters

Brusellozda Standart Tüp Agglütinasyon Titrelerinin Biyokimyasal ve Hematolojik Parametrelerle İlişkisi

Brusella Tanı Testleri / Brucella Diagnosis Tests

Ağca Harun[1], Ertugay Gülin[2]
[1]Clinic of Microbiology, Tavşanlı Doc. Dr. M. Kalemli State Hospital, Kütahya, Türkiye.
[2]Clinic of Biochemistry, Tavşanlı Doc. Dr. M. Kalemli State Hospital, Kütahya, Türkiye.

This study was presented as a poster in the 1st KLIMUD Congress, 2011

Özet

Amaç: Çalışmada Tavşanlı Devlet Hastanesi poliklinik ya da servislerinde bruselloz ön tanısı alan hastalardan standart tüp agglütinasyon titreleri 1/160 ve üzerinde olan serum örneklerinde çeşitli biyokimyasal ve hematolojik parametrelerin ilişkisinin araştırılması hedeflenmiştir. Gereç ve Yöntemler: Çalışmaya dahil edilen 91 hasta serumunda ALT, AST, CRP, eritrosit sedimentasyon hızı (ESH), lökosit, eritrosit ve trombosit sayımı sonuçları araştırılmıştır. Bulgular: Standart tüp agglütinasyon testi pozitif bulunan hastaların 74'ünde (%81) CRP pozitifliği, 7'sinde (%8) ALT pozitifliği, 13'ünde (14) AST pozitifliği, 27'sinde (%30) ESH yüksekliği saptandı. Hastalardan 10'unda (%11) lökositoz, 5'inde (%5) lökopeni, 2'sinde (%2) eritrosit sayısında azalma, 5'inde (%5) trombositopeni görüldü. Sonuç: Biyokimyasal testlerden ALT ve AST ile CRP'nin brusella tüp agglütinasyon testi sonucu ile korelasyon gösterdiği tespit edildi. Bu bulgular; bruselloz tanısında CRP'nin gerek hastaların %81'inde pozitif bulunması ve gerekse korelasyon göstermesi nedeniyle bruselloz düşünülen hastalarda bakılması gerektiğini göstermekte, ALT ve AST titrelerinin ise özellikle standart tüp agglütinasyon testi yüksek titrelerde pozitif bulunan hastalarda yüksek çıkması nedeniyle tanıyı desteklemek üzere araştırılması gerekli parametreler olduğunu göstermektedir.

Anahtar Kelimeler

Brusella; Tüp Agglütinasyon; Biyokimyasal Parametre

Abstract

Aim: We aimed to investigate the correlation between standard tube agglutination titers over 1/160 and various biochemical and hematologic parameters in the sera of patients which were pre-diagnosed as brucellosis in the policlinics or the services of Tavsanli State Hospital. Material and Methods: ALT, AST, CRP, erythrocyte sedimentation rate (ESR), leukocyte count, erythrocyte count and thrombocyte count were investigated in the sera of 91 patients included in the study. Results: Of the standard tube agglutination test positive patients, 74 (%81) were CRP positive, 7 (%8) were ALT positive, 13 (%14) were AST positive and 27's (%30) had elevated ESR levels. 10 (%11) of the patients had leukocytosis, 5 (%5) had leukopenia. Discussion: ALT and AST from the biochemical parameters and CRP were found to be correlated with brucella tube agglutination test. These findings show that; in the diagnosis of brucellosis as CRP was found positive in %81 of the patients and as CRP shows correlation, CRP should be investigated in patients who are thought to be brucellosis, while ALT and AST titers which are elevated in the high titers of brucella standard tube agglutination test should be investigated to support the diagnosis

Keywords

Brucella; Tube Agglutination; Biochemical Parameter

DOI: 10.4328/JCAM.839 Received: 21.10.2011 Accepted: 28.11.2011 Published Online: 01.10.2012 J Clin Anal Med 2012;3(4):433-435
Corresponding Author: Harun Ağca, Clinic of Microbiology, Tavşanlı Doc. Dr. M. Kalemli State Hospital, Kütahya, Türkiye.
T.: +902726142000 GSM: +905058443004 Fax: +90274 6143798 E-mail: drharunagca@yahoo.com

Introduction

Brucellosis is a zoonosis caused by bacteria of the genus Brucella, which affect both human and animals such as the cow, sheep, goat, camel and pig. Bacteria enters the host through the digestive tract via contaminated dairy products, the respiratory tract via aerosols, or the skin via contact with infected animals on farms in slaughterhouses. Since the symptoms of brucellosis are non-spesific, the clinical diagnosis of the disease is difficult. Therefore the diagnosis must be supported and confirmed by the isolation of the agent, mostly from blood culture or by the detection of antibodies against bacterial antigens.[1,2]

The gold standard in the diagnosis of brucellosis is the isolation of brucella bacteria. The isolation of the bacteria from blood cultures ranges from 47.1% to 94.1% depending on the methods used and the period of incubation.[3,4] In the absence of bacteriologic confirmation, a presumptive diagnosis can be made on the basis of high or rising titers of spesific antibodies. A variety of serologic tests have been applied to brucellosis, of which the standard agglutination test is the most widely used. Routine laboratory findings may support the diagnosis. The purpose of the study was to compare the standard agglutination test titers and some biochemical parameters in the diagnosis of brucellosis.

Materials and Methods

91 patients who were pre-diagnosed as brucellosis in policlinics or services in Tavsanli State Hospital were included in the study. Samples between March 2010 and December 2010 were investigated. The patients were invesitigated for the rose bengal test, the standard agglutination test which were acquired from the Refik Saydam Hygiene Center. Both the tests were done according to the manufacturers instructions. In the standard agglutination test, the serum was diluted between 1/20 to 1/1280 dilutions. Rose bengal test positive sera were investigated for the standard agglutination test. Agglutination over the 1/80 titer were included in the study. The sera were investigated for ALT, AST and CRP by Synchron LX 20 (Beckman Coulter, USA), for erythrocyte sedimentation rate (ESR) by ALIFAX Test I(ALIFAX, Italy), for the white blood cell count, red blood cell count and platelets by Cell Dyn 3700 (Abbott, USA).

Statistics were done according to SPSS 17.0 for Windows.

Results

32 of the patients were male (%35) and, 59 were female (%65). Mean age was 47,9. Standard tube agglutination results positive 74 sera (%81) were found positive for CRP. ALT levels were elevated in 7 sera (%8), AST levels were increased in 13 sera (%14) and there were increase in ESR levels in 27 (%30) sera (Table 1). In higher levels of standard tube agglutinaton tests, CRP positivity and elevation in ALT and AST level's ratio were higher than low levels. White blood cell count was high in 10 sera (%11), while it was found low in 5 sera (%5). Two patients' (%2) erythrocyte count and 5 patients' (%5) thrombocyte count was low (Table 2).

Correlation analysis was made between standard tube agglutination and ALT, AST, CRP, ESR levels, white blood cell count, eryhthrocyte count and thrombocyte count. Correlation was statistically significant between standard tube agglutination and CRP levels (r = 0,32, $P < .05$), standard tube agglutination and ALT levels (r = 0,48, $P < .05$), standard tube agglutination and AST levels (r = 0,45, $P < .05$).

Discussion

In the absence of a positive culture, the diagnosis of brucellosis rests on demonstration of antibodies. A variety of serologic tests have been applied to brucellosis, of which standard tube agglutination is the most widely used.[5,6] Other laboratory findings supporting the diagnosis are, leukopenia, anemia, lymphocytosis, elevation in ESR, ALT and AST levels.

Brucellosis is usually seen in adults. Yuce et al.[7] reported that %50-60 of patients were between 20 and 50 years old, %10-15 were child and %10 were older than 65. Cagatay et al.,[8] reported that the mean age of the brucellosis patients' was 43,3 and %61 of the patients were female. In our study we found that the mean age was 47,9 and %65 of the patients were female.

In the diagnosis of Brucellosis culture and serologic tests are used. Tansel et al. reported that, %30 of the brucellosis patients had leukopenia, %35 had thrombocytopenia, %40 had anemia, %90 had elevated ESR levels, %83 had elevation in CRP levels, %68 had elevated AST, and %55 had elevated ALT levels.[9] Geyik et al.,[10] reported that %25 of brucellosis patients had leukopenia, %7 had leukocytosis, %30 had elevated ESR levels, %39 had elvated ALT and %38 had elevated AST levels and %6 had culture positivity. Gul et al.,[11] reported that ESR had increase in %55 of the brucellosis patients, %34 had leukopenia, %13 had leukocytosis, %14 had anemia, %11 had thrombocytopenia, %32 had elevated ALT levels. Mehli et al.,[12] reported that %62 of the brucellosis patients had elevated CRP levels, %31 had elevated ALT and %32 had AST levels and %69 had elevates ESR levels. There was leukocytosis in %15, leukopenia in %7, anemia in %35, thrombocytopenia in %12 of the patients . In our study there was CRP positivity in %81 of the patients, ALT elevation in %8, AST elevation in %14, ESR elevation in % 30 of the patients. White blood cell count was high in %11, and low in %6 of the patients. Erythrocytopenia was seen in %2 and thrombocytopenia was seen in %6 of the patients. These results show that hematologic parameters are not effected significantly from the

Table 1. Comparison of standard tube agglutination with CRP, ALT, AST, ESR.

Standard tube agglutination		CRP positivity		ALT elevation		AST elevation		ESR elevation	
Titer	n	n	%	n	%	n	%	n	%
1/160	39	29	74	0	0	3	8	7	18
1/320	28	23	82	1	4	3	11	11	39
1/640	17	15	88	4	24	4	24	7	41
1/1280	7	7	100	2	29	3	43	2	29
Total	91	74	81	7	8	13	14	27	30

Table 2. Comparison of standard tube agglutination with hematologic parameters

Standard tube agglutination		WBC						Decrease in erithrocytes		Decrease in thrombocytes	
		High		Normal		Low					
		n	%	n	%	n	%	n	%	n	%
1/160	39	1	3	37	94	1	3	0	0	2	5
1/320	28	5	18	22	78	1	4	0	0	1	4
1/640	17	3	18	13	76	1	6	1	6	1	6
1/1280	7	1	14	4	57	2	29	1	14	1	14
Total	91	10	11	76	84	5	6	2	2	5	6

disease. Hematologic parameters can help the diagnosis but are usually normal in most of the patients. Acute phase reactant CRP is usually found high in most of the patients. CRP, which is synthised by the hepatocytes is important in immune response. CRP serum levels are elevated in infections, rheumatologic diseases, cancers, and chronic diseases.[12-14] CRP levels in our study is similar to the others.[9,12] CRP levels are elevated parallel to the standard tube agglutination titers.

Elevation in ALT and AST levels are usually seen in high standard tube agglutination titers (1/640 and 1/1280). These findings show that hepatocytes are prominently effected in high antibody titers and this leads to elevation in ALT and AST. Hematologic parameters are prominently effected in high (1/1280) titers. This is due to the severity of the underlying disease.[15]

Conclusion

ALT and AST from the biochemical parameters and CRP were found to be correlated with brucella tube agglutination test. These findings show that; in the diagnosis of brucellosis as CRP was found positive in %81 of the patients and as CRP shows correlation, CRP should be investigated in patients who are thought to be brucellosis, while ALT and AST titers which are elevated in the high titers of brucella standard tube agglutination test should be investigated to support the diagnosis

Acknowledgement

This study was presented as a poster in the 1st KLIMUD Congress, 2011.

References

1. Young EJ. Brucella species. In: Mandell GL, Bennet JE, Dolin R, editors. Principles and Practice of Infectious Diseases. 5th ed. Philadelphia: Churchill Livingstone; 2000 p. 2386-93.
2. Altuglu I, Zeytinoglu A, Bilgic A, Kamcioglu S, Karakartal G, Smits H. Evaluation of brucella dipstick assay for the diagnosis of acute brucellosis. Diagn Microbiol Infect Dis 2002; 44(3): 241-243.
3. Ruiz J, Lorente I, Perz J, Simarro E, Martinez-Campos L. Diagnosis of brucellosis by using blood cultures. J Clin Microbiol 1997; 35(9): 2417-2418.
4. Ozturk R, Mert A, Kocak F, Ozaras R, Koksal F, Tabak F et al. The diagnosis of brucellosis by use of BACTEC 9240 blood culture system. Diagn Microbiol Infect Dis 2002; 44(3): 133-135.
5. Mernish ZA, Almuneef M, Mah W, Qassem LA, Osobad AO. Comparison of Brucella Standard Agglutination Test with the ELISA IgG and IgM in patients with Brucella bacteremia. Diagn Microbiol Infect Dis 2002; 44(3): 129- 132.
6. Ertek M, Yazgı H, Ozkurt Z, Ayyıldız A, Parlak M. Comparison of the diagnostic value of the Standard Tube Agglutination Test and the ELISA IgG and IgM in patients with Brucellosis. Turk J Med Sci 2006;36(3):159- 163.
7. Yuce A, Cavus SA, Yapar N, Cakır N. Brucellosis. An evaluation of 55 cases. Klimik Derg 2006;19:87-97.
8. Cagatay AA, Kucukoglu S, Berk H, Ozsut H, Eraksoy H, Dilmener M et al. The evaluation of 36 patients with Brucellosis. Klimik Derg 2002;15:19-21.
9. Tansel O, Yavuz M, Kuloglu F, Akata F, Evaluation of 40 Brucellosis cases admitted to the Trakya University Hospital. İnfek Derg 2003;17:1-4.
10. Geyik MF, Kokoglu OF, Hosoglu S, Ayaz C. Evaluation of 184 Brucellosis pateints. Dicle Tıp Derg 2002;29:1-2.
11. Gul HC, Coskun O, Turhan V, Besirbellioglu BA, Bilgeturk A, Erdem H et al. Brucellosis: Retrospective investigation in 140 patients. TAF Prev Med Bull 2007;6:249-252.
12. Mehli M, Karslıgil T, Gayyurhan ED, Akın FE. The relationship between standard tube agglutination titres and the Rose Bengal test results with biochemical parameters in brucellosis. Turk Mikrobiyol Cem Derg, 2008;38:16- 22.
13. Bayraktar M, Bayraktar N, Bayındır Y, Durmaz R. Evaluation of serum levels of C-reactive protein, iron and ferritin in diagnosis and monitoring of Brucellosis. Ankem Derg 2005;19:61-63.
14. Danese I, Haine V, Delrue RM, Tibor A, Lestrate P, Stevaux O et al. The Ton System, an ABC Transporter, and a universally conserved GTPase are involved in iron utilization by Brucella mellitensis 16M. Infect Immun 2004;72:5814-5823.
15. Bellaire BH, Elzer PH, Hagius S, Walker J, Baldwin JL, Roop II RM et al. Genetic organization and iron-responsive regulation of the Brucella abortus 2,3-dihydroxybenzoic acid biosynthesis operon, a cluster of genes required for wild-type virulence in pregnant cattle. Infect Immun 2003;71:1794-1803.