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Erythrocytes Parameters in the Course of Brucellosis

Brusellozun Seyrinde Eritrosit Parametreleri

Abdulkadir Küçükbayrak[1], Tekin Taş[2], Mehmet Tosun[3], Gülali Aktaş[4], İsmail Necati Hakyemez[1], Fırat Zafer Mengeloğlu[2], Hayrettin Akdeniz[1]

[1]Abant İzzet Baysal Üniversitesi Tıp Fakültesi Enfeksiyon Hastalıkları Ve Klinik Mikrobiyoloji Anabilim Dalı, Bolu
[2]Abant İzzet Baysal Üniversitesi Tıp Fakültesi Tıbbi Mikrobiyoloji Anabilim Dalı, Bolu
[3]Abant İzzet Baysal Üniversitesi Tıp Fakültesi Tıbbi Biyokimya Anabilim Dalı, Bolu
[4]Abant İzzet Baysal Üniversitesi Tıp Fakültesi İç Hatalıkları Anabilim Dalı, Bolu

Abstract

Background: To evaluate the all erythrocyte parameters in patients with brucellosis and to investigate the correlation between these parameters and inflammatory markers.
Method: A total of 55 patients who had positive brucella tube agglutination tests were included in the study. The computerized laboratory database records were reviewed for the following data and recorded: Age and gender of the patients, brucella tube agglutination tests, erythrocyte sedimentation rate, C- reactive protein, erythrocyte parameters.
Results: Mean age of the patients was 45.4 ± 18.1 years with a female/male ratio of 25/30. Post-treatment hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration values were significantly higher than the pre-treatment values.
Conclusion: Evaluation of the erythrocyte parameters may help to improve understanding of anemia and its nature related with brucellosis.

Keywords: Brucellosis, erythrocytes, anemia.

Özet

Amaç: Bruselloz hastalarında tüm eritrosit parametrelerini değerlendirmek ve bu parametreler ile inflamatuar belirteçler arasındaki ilişkiyi araştırmak.
Yöntem: Çalışmaya brusella tüp aglütinasyon pozitif toplam 55 hasta alındı. Hastaların yaş ve cinsiyetleri, brusella tüp aglütinasyon testi sonuçları, eritrosit sedimantasyon hızları, C-reaktif protein ve eritrosit parametreleri ile ilgili veriler laboratuvar kayıtlarından incelendi.
Bulgular: Hastalarda ortalama yaş 45,4 ± 18,1 ve kadın/erkek oranı 25/30'du. Tedavi sonrası hemoglobin, hematokrit, ortalama eritrosit hacmi, ortalama eritrosit hemoglobin ve ortalama eritrosit hemoglobini konsantrasyon değerleri tedavi öncesi değerlere gore anlamlı olarak daha yüksek bulundu.
Sonuç: Eritrosit parametrelerinin değerlendirilmesi aneminin ve brusellozla ilişkili doğasının anlaşılmasında fayda sağlayabileceği düşünülmüştür.

Anahtar Kelimeler: Bruselloz, eritrosit, anemi.

Introduction

Brucellosis is a zoonotic infectious disease caused by brucella spp. The most common clinical signs of brucellosis are fever, muscle and joint pain, fatigue and back pain. Brucella has a high affinity for the reticuloendothelial system, including the liver, bone marrow, lymph nodes and spleen. (1). Therefore, hematologic abnormalities are common manifestation of brucellosis. Hematologic abnormalities may be presented with anemia, thrombocytopenia, leukopenia, hemolytic anemia, clotting abnormalities and pancytopenia (2, 3).

Incidence rates of anemia in patients with brucellosis ranges between 6% and 74% (2-7). Anemia in patients with brucellosis results from hypersplenism, autoimmune hemolysis, bone marrow suppression, hemophagocytosis and bleeding (2, 8). Anemia is common in the course of human brucellosis (5). However, to our knowledge, there are no studies investigating the change in all erythrocyte parameters in patients with brucellosis before and after treatment in the literature.

The aim of the present study is to evaluate the erythrocyte parameters in brucellosis and to investigate the correlation between these parameters and inflammatory markers.

Methods

The present study was performed retrospectively in the microbiology laboratory of Abant Izzet Baysal University Hospital. Approval from the local institutional ethics committee was obtained. A search of a computerized patient database was performed to identify all brucellosis patients. A total of 55 patients who had positive brucella tube agglutination tests (BSTAT) at least at a titer of 1/160 were included in the study. The computerized laboratory database records

İletisim Bilgisi / *Correspondence*
Yard. Doç. Dr. Tekin Taş, Abant İzzet Baysal Üniversitesi Tıp Fakültesi Tıbbi Mikrobiyoloji Anabilim Dalı, Bolu **E-posta:** tekints@gmail.com
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were reviewed for the following data and recorded: Age and gender of the patients, BSTAT, erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), red blood cell count (RBC), hemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and red cell distribution width (RDW). Antigens as Brucella abortus s-99 strain were used in BSTAT. CRP and ESR were measured with nephelometric assay in a Siemens ProSpec (Marburg, Germany) device and Westergren method in a Greiner sedimentation device (Frickenhausen Germany), respectively. Erythrocyte parameters were measured with the original kits in an automatic complete blood analyzer (Abbott Cell-Dyn, Wiesbaden-Germany and Beckman Coulter LH780- Fullerton CA-USA). RBC, HGB, HCT, MCV, MCH, MCHC, CRP and ESR values at the time of positive BSTAT at least at a titer of 1/160 and at the time of when BSTAT became negative or when the titers reduced four folds were recorded.

Reference ranges of the measured parameters in our laboratory were as follows: CRP < 3.19 or < 3.41 depends on the reactive, RBC 3.5-5.5 M/uL, HGB 12.2-18.1 g/dL, HCT 37-54 %, MCV 80-97 f/L, MCH 27-34 pg, MCHC 32-36 g/dL, RDW 0.00-99.9 10 (GSD).

Statistical Analyses

Continuous variables were expressed as mean ± standard deviation (SD) and categorical variables were expressed as percentage. A comparison of the categorical and continuous variables between the groups was performed using paired samples t-test. An analysis of normality of the continuous variables was performed with the Kolmogorov–Smirnov test. Pearson test was used for correlation analyses of the parameters measured in the pretreatment period. Logistic regression analysis was used to assess the relationship between as RBC, HGB, HCT, MCV, MCH, MCHC, and RDW as independent variables and BSTAT as dependent variable. P value of less than 0.05 was considered statistically significant. SPSS software program (15.0, Inc., Chicago, Illinois) was used for all statistical analysis.

Results

Mean age of the patients was 45.4 ± 18.1 years. Twenty-five of the patients (45.5%) were female with a mean age of 46.1 ± 17 years and 30 of the patients (54.5%) were male with a mean age of 44.8 ± 19.2 years. BSTAT values of the patients were positive at a titer of 1/160 and 1/1280 on pre-treatment period and were at a range of negative to 1/160 after treatment. BSTAT was positive at a titer of 1/160 in 13 patients, 1/320 in 15 patients, 1/640 in 20 patients and 1/1280 in 3 patients. Four patients were diagnosed by immune capture. Table 1 showed the demographics and BSTAT characteristics of the study patients.

Table 1. Demographics and BSTAT characteristics of the study patients.

	n=55
Mean Age (years)	45.4 ± 18.1
Gender (Female/male)	25/30
Baseline BSTAT titers	
1/160	13
1/320	15
1/640	20
1/1280	3
immun capture	4

** BSTAT: Brucella serum tube agglutination test*

Post-treatment HGB (13.3 ± 1.4 gr/dl), HCT (39.2 ± 4 %), MCV (83.7 ± 5.2 fL), MCH (28.5 ± 2.2 pg) and MCHC (34 ± 0.9 gr/dl) values were significantly higher than pretreatment HGB (12.6 ± 1.6 gr/dl), HCT (37.3 ± 4.9 %), MCV (81.3 ± 6 fL), MCH (27.4 ± 2.5 pg) and MCHC (33.5 ± 1.6 gr/dl) values ($p<0.001$, $p=0.001$, $p<0.001$, $p<0.001$, and $p=0.044$, respectively). Although post-treatment RBC and RDW values were higher than pretreatment values, they did not reach statistically significant level ($p=0.11$ and $p=0.36$, respectively). Mean RBC, HGB, HCT, MCV, MCH, MCHC, and RDW values in pre and post treatment periods and significance levels were given in Table 2.

In our small sample size, we found the frequency of anemia (HGB < 12 gr/dl) in pre- and post-treatment period was 30.9% and 14%, respectively. In pre- and post-treatment period, 18.2% and 4.7% of the brucellosis patients with anemia was hypochrom micrositer, respectively.

Correlation analyses in the pre-treatment period revealed inverse correlations between RBC, HGB, and HCT with ESR (r =-0.225, p<0.001; r =-0.565, p<0.001; r =-0.558, p<0.001, respectively). However, CRP and BSTAT values were not significantly correlated with erythrocyte parameters (p>0.05 for all). Table 3 showed the correlations between the erythrocyte parameters and inflammatory markers in pretreatment period.

Table 2. Mean RBC, HGB, HCT, MCV, MCH, MCHC, and RDW values in pre and post treatment periods in patients with brucellosis.

	Pre-treatment	Post-treatment	*P* value
RBC M/uL	4.6 ± 0.6	4.7 ± 0.5	0.11
HGB (gr/dl)	12.6 ± 1.6	13.3 ± 1.4	<0.001*
HCT (%)	37.3 ± 4.9	39.2 ± 4	0.001*
MCV (fL)	81.3 ± 6	83.7 ± 5.2	<0.001*
MCH (pg)	27.4 ± 2.5	28.5 ± 2.2	<0.001*
MCHC (gr/dl)	33.5 ± 1.6	34 ± 0.9	0.044*
RDW (10.GSD)	16.0 ± 2	16.4 ± 2.5	0.362

** HGB: Hemoglobin, HCT: Hematocrit, MCH: Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, MCV: Mean corpuscular volume, RBC: Red blood cell count, RDW: Red cell distribution width.*

Logistic regression analyses were performed to investigate the independent markers of brucellosis in pretreatment period. Erythrocyte parameters (RBC, HGB, HCT, MCV, MCH, and MCHC), inflammatory markers (ESR, CRP) and BSTAT were included in the analysis. We found no independent predictors in logistic regression analysis.

Discussion

In the present study, we found that the frequency of anemia in patients with brucellosis in pre and post-treatment period was 30.9% and 14%, respectively. The frequency of hypochromic microcytic anemia in pre and post-treatment period was 18.2% and 4.7%, respectively. Hemoglobin, hematocrit, MCV, MCH and MCHC values were significantly higher in post-treatment period compared to pre-treatment period. RBC, HGB and HTC levels were inversely correlated with ESR in pre-treatment period.

One previous study by Bulut et al. (4) pointed that anemia prevalence (HGB < 12 gr/dl) in cases of complicated and uncomplicated brucellosis was 46.7% and 36.2% respectively. Prevalence of anemia alone or with leukopenia and thrombocytopenia was reported to be 56% in another study (5). Hemolytic anemia frequency was 0.5% of all anemias in that study population of brucellosis (5). Calık and Gokengin reported a review of the literature between 1990 and 2009 in Turkish brucellosis patients and the authors concluded the percentage of anemia was 17.3 % (6). In those population, 3 patients had microangiopatic hemolytic anemia and another 3 patients had autoimmune hemolytic anemia. Demiroglu et al. reported the anemia prevalence as 51.7% in brucellosis (7). Anemia defined as a hemoglobin level lower than 14gr/dl in men and 12 gr/dl in women in that study. One individual study reported the prevalence of anemia as high as 72% in patients with brucellosis from Turkish ancestry (9). In another study, the prevalence of anemia in brucellosis was reported to be 57.3% in which a hemoglobin level lower than 10 gr/dl considered as anemia (10). High prevalence of anemia in that study explained high frequency of chronic infection (63%) and worse socio-economic conditions (lack of food and hookworm infection) of the patients. Interestingly, in another study by Roushan et al. from the same country reported an anemia prevalence of 15.1% in 469 patients with brucellosis (11).

Basilowski et al (12) claimed that anemia was the most common hematologic sign of brucellosis. Anemia prevalence in patients with and without occupational exposure history were 37.4% and 47.9%, respectively. The authors explained the difference with malnutrition and worse economic conditions. Sari et al (13) reported neutropenia in 30 of 202 brucellosis cases. Five of these 30 patients had hematologic malignity. Mean hemoglobin level in patients with and without hematologic malignancy was 7.5 ± 1.4 gr/dl and 10.6 ± 1.7 gr/dl, respectively. Bone marrow examination of these 30 patients showed hypercellularity in 23 patients and normocellularity in 7 patients. Al Shamahy et al reported anemia

prevalence in childhood and in elderly as high as 20.7% and 72.3%, respectively. In contrast to previous studies, most anemia in that study population were normochromic normocytic type (3). In conclusion, we found high frequency of anemia in brucellosis patients according to results of our small sample size. We think that evaluation of the erythrocyte parameters may help to improve understanding of anemia related to brucellosis and future studies with large numbers of patients are necessary.

Table 3. Pearson correlation analyses in pre-treatment period.

	RBC (M/uL)	HGB (gr/dl)	HCT (%)	MCV (fL)	MCH (pg)	MCHC (gr/dl)	RDW (10.GSD)
CRP (mg/dl)	-0.225	-0.175	-0.21	0.36	0.079	0.103	-0.176
ESR (mm-hour)	-0.515**	-0.565**	-0.558**	-0.021	-0.03	-0.243	-0.129
BSTAT	-0.089	-0.092	-0.103	-0.029	-0.004	0.061	-0.052

*** Significance at p<0.01 level.*

BSTAT: Brucella serum tube agglutination test, CRP: C-reactive protein, ESR: Erythrocyte sedimentation rate, HGB: Hemoglobin, HCT: Hematocrit, MCH: Mean corpuscular, MCHC: Mean corpuscular hemoglobin concentration, MCV: Mean corpuscular volume, RBC: Red blood cell count, RDW: Red cell distribution width.

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