

# Association Between the Percentages of Lymphocytes, Monocytes, and Neutrophils and Brucella Epididymo-orchitis: A Multicentric Study

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## What's known on the subject? and What does the study add?

Mononuclear leukocytes (monocyte cells and lymphocyte cells) play a crucial role in granulomatous infections, such as brucellosis. There is no sufficient data in the literature concerning whether cellular changes in tissue are reflected in blood circulation in brucellosis. In our study, the percentages of lymphocytes and monocytes were higher in patients with brucella epididymo-orchitis than in those with non-brucella epididymo-orchitis.

## Abstract

**Objective:** There are insufficient data in the literature concerning whether cellular changes in tissue are reflected in blood circulation in granulomatous infections, such as zoonotic brucellosis. In this study, we compared laboratory parameters between patients with brucella (BEO) and non-brucella epididymo-orchitis (NBEO).

**Materials and Methods:** This retrospective study included 84 patients with BEO and 92 with NBEO who presented to six medical centers between 2017 and 2021.

**Results:** The median age of the patients was 41 (interquartile range: 27-61) years. In the multivariate analysis, the presence of abdominal pain ( $p=0.003$ ), the percentage of lymphocytes ( $p=0.012$ ) and the percentage of monocytes ( $p=0.029$ ) were significantly higher in the BEO group than in the NBEO group. In addition, the percentage of neutrophils ( $p=0.001$ ) was significantly lower in the BEO group than in the NBEO group. In the receiver operating characteristic analysis, the percentage of lymphocytes had an area under the curve (AUC) value of 0.808 at a cut-off point of >22.1%, the percentage of monocytes had an AUC value of 0.745 at a cut-off point of >7.7%, and the percentage of neutrophils had an AUC value of 0.746 at a cut-off point of <66.8%.

**Conclusion:** To the best of our knowledge, there is no other study comparing the percentages of lymphocytes, monocytes, and neutrophils between patients with BEO and NBEO. The percentages of mononuclear leukocytes (monocytes and lymphocytes) and neutrophils may be useful for the pre-diagnosis of BEO in endemic areas.

**Keywords:** Brucella, orchitis, lymphocytes, monocytes, neutrophils

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## Introduction

Human brucellosis is the most common bacterial zoonosis (1), but its diagnosis can be delayed (2). Because it has a chronic and granulomatous nature (3,4), the timing of diagnosis and treatment is critical in terms of focal complications that may develop. If left untreated, brucellosis can lead to organ involvement, such as osteoarthritis, infective endocarditis, hepatitis, meningitis, and epididymo-orchitis (5).

It has been reported that 2.5-18.8% of men with brucellosis have testicular involvement (5-7). It is also difficult to diagnose brucella cases presenting with isolated orchitis (8,9). The local clinical findings of brucella (BEO) and non-brucella epididymo-orchitis (NBEO) are similar, and acute phase reactants may be elevated in all epididymo-orchitis cases (10,11). Mononuclear leukocytes (monocyte cells and lymphocyte cells, especially macrophages) play a crucial role in granulomatous infections, such as brucellosis. When monocytes in the bloodstream reach the tissue, they are called macrophages and are surrounded by lymphocytes. These reactions also represent lymphohistiocytic inflammation, which is a chronic inflammatory response (12). However, there is no sufficient data in the literature concerning whether cellular changes in tissue are reflected in blood circulation in brucellosis. A detailed evaluation of clinical parameters and the distribution of mononuclear leukocytes in the complete blood count may be useful for the differential diagnosis of epididymo-orchitis. In this study, we compared the clinical and laboratory parameters between the BEO and NBEO patients.

## Materials and Methods

### Study Design

The files of patients with NBEO and BEO who presented to six medical centers (urology and infectious diseases clinics) between 2017 and 2021 were retrospectively reviewed. The study included 84 patients with BEO and 92 with NBEO diagnosed with epididymo-orchitis based on physical examination and laboratory and/or ultrasonography findings. Patients with missing laboratory data, hematological disease, malignancy, or immunosuppressive disease, and those using immunosuppressive drugs were excluded from the study. The study was approved by the ethics committee (approval no: 2022/149, date: 22.11.2023 - University of Health Sciences Türkiye, Ankara Gülhane Training and Research Hospital Clinical Research Ethics Committee).

### Follow-up and Data Collection

The patient's demographic characteristics, physical examination findings, and laboratory parameters [complete blood count, C-reactive protein, and erythrocyte sedimentation rate (ESR)]

were noted. The diagnosis of epididymo-orchitis was made according to the patients' scrotal complaints (pain, redness, tenderness, and/or swelling), acute phase reactants, and/or ultrasonography findings. When brucellosis was suspected clinically, patients with a Wright tube agglutination test result of  $\geq 1/160$  or a positive blood culture test were diagnosed with BEO. Additional focal involvements of patients with BEO were recorded.

### Statistical Analysis

The Kolmogorov-Smirnov test was used to evaluate the compliance of the data with the normal distribution. The Mann-Whitney U and chi-square tests were used in the statistical analysis of continuous and categorical variables, respectively. Multivariate logistic regression analysis was performed to determine statistically significant independent parameters. The optimal cut-off values of the percentage of lymphocytes, monocytes, and neutrophils for the differentiation of BEO and NBEO were calculated using receiver operating characteristic (ROC) analysis. A p-value of  $<0.05$  was considered statistically significant.

## Result

The median age of all patients was 41 [interquartile range (IQR): 27-61] years. The median ages of the BEO and NBEO groups were 35 (IQR: 24-51) and 54 (IQR: 27-68) years, respectively ( $p<0.0001$ ). Bilateral involvement was present in eight patients in each group (9.5% for BEO and 8.7% for NBEO) ( $p=1$ ). The BEO and NBEO groups significantly differed in terms of ESR ( $p=0.028$ ), presence of abdominal pain ( $p<0.0001$ ), serum white blood cell count ( $p<0.0001$ ), neutrophil count ( $p<0.0001$ ) and percentage ( $p<0.0001$ ), lymphocyte count ( $p<0.0001$ ) and percentage ( $p<0.0001$ ), monocyte count ( $p=0.023$ ) and percentage ( $p<0.0001$ ), neutrophil-to-lymphocyte ratio (NLR) ( $p<0.0001$ ), and neutrophil-to-monocyte ratio (NMR) ( $p<0.0001$ ). The clinical and laboratory data of the patients are detailed in Table 1.

In the multivariate analysis, the presence of abdominal pain ( $p=0.003$ ), percentage of lymphocytes ( $p=0.012$ ), and percentage of monocytes ( $p=0.029$ ) were significantly higher in the BEO group than in the NBEO group. In addition, the percentage of neutrophils ( $p=0.001$ ) was significantly lower in patients with BEO than in those with NBEO (Table 2).

ROC analysis was performed to calculate the optimal cut-off values for the percentage of lymphocytes, monocytes, and neutrophils required to differentiate between the BEO and NBEO groups. The percentage of lymphocytes had an area under the curve (AUC) value of 0.808 (95% confidence interval: 0.745-0.870) at a cut-off point of  $>22.1\%$ , and the

(n)	Total (n=176)	BEO (n=84)	NBEO (n=92)	p
Age (years), median (IQR)	41 (27-61)	35 (24-51)	54 (27-68)	<0.0001 <sup>a</sup>
Presence of abdominal pain n, (%)	85 (48.3)	59 (70.2)	26 (28.3)	<0.0001 <sup>b</sup>
<b>Involvement side n, (%)</b>				
Unilateral	160 (90.9)	76 (90.5)	84 (91.3)	1 <sup>b</sup>
Bilateral	16 (9.1)	8 (9.5)	8 (8.7)	
CRP (mg/L), median (IQR)	13 (0.1-49)	12 (4-28)	21 (0.1-170)	0.351 <sup>a</sup>
ESR (mm/hour), median (IQR)	71 (3-103)	78 (3-118)	68 (32-99)	0.028 <sup>a</sup>
WBC, median (IQR)	8.600 (3.200-13.110)	7.930 (3.200-11.813)	8.860 (5.350-14.930)	<0.0001 <sup>a</sup>
Neutrophil count (cell/mL), median (IQR)	5.800 (2.320-9.815)	4.890 (2.330-8.592)	6.636 (2.800-10.936)	<0.0001 <sup>a</sup>
Neutrophils (%), median (IQR)	66.8 (36.2-90.3)	60.3 (36.2-83.7)	74.9 (49-91.4)	<0.0001 <sup>a</sup>
Lymphocyte count (cell/mL), median (IQR)	1.840 (100-2.780)	2.380 (900-3.680)	1.700 (100-2.350)	<0.0001 <sup>a</sup>
Lymphocyte (%), median (IQR)	22.1 (2.2-39.8)	29.1 (6.8-39.3)	15.8 (2.2-29.5)	<0.0001 <sup>a</sup>
Monocyte count (cell/mL), median (IQR)	720 (280-1320)	795 (280-1.925)	640 (300-1040)	0.023 <sup>a</sup>
Monocyte (%), median (IQR)	7.7 (2.9-11.8)	9.1 (5.1-18.9)	7.1 (2.9-10.2)	<0.0001 <sup>a</sup>
Platelet count (cell/mL), median (IQR)	240.000 (98.000-340.000)	232.000 (98.000-312.000)	245.000 (146.000-404.000)	0.491 <sup>a</sup>
MPV (fL), median (IQR)	8.7 (5.8-9.7)	8.7 (6.1-9.7)	8.5 (5.8-10.9)	0.698 <sup>a</sup>
RDW (%), median (IQR)	14 (8.1-17) 13.9 1.9	13.7 (8.1-16.5) 13.7 2.2	14.3 (11.2-17.4) 14.3 1.7	0.078 <sup>a</sup>
NMR, median (IQR)	8.7 (1.4-15.5)	6.2 (1.4-13.3)	10.2 (4.7-17.1)	<0.0001 <sup>a</sup>
NLR, median (IQR)	3.13 (0.85-6.98)	2.16 (0.93-4.62)	4.61 (0.85-9.36)	<0.0001 <sup>a</sup>

BEO: Brucella epididymo-orchitis, NBEO: Non-brucella epididymo-orchitis, CRP: C-reactive protein, ESR: Erythrocyte sedimentation rate, WBC: White blood cells, MPV: Mean platelet volume, RDW: Red cell distribution width, NLR: Neutrophil-to-lymphocyte ratio, NMR: Neutrophil-to-monocyte ratio, MLR: Monocyte-to-lymphocyte ratio, IQR: Interquartile range, <sup>a</sup>: Mann-Whitney U test, <sup>b</sup>: Pearson chi-square test

	Odds ratio	95% Confidence interval		p
		Lower	Upper	
Age	0.247	0.979	1.036	0.619 <sup>a</sup>
ESR	2.231	0.995	1.038	0.135 <sup>a</sup>
Presence of abdominal pain	8.706	0.055	0.558	0.003 <sup>a</sup>
WBC	0.268	0.999	1.000	0.604 <sup>a</sup>
Neutrophil count	0.295	0.999	1.001	0.587 <sup>a</sup>
Neutrophil (%)	10.935	1.172	1.858	0.001 <sup>a</sup>
Lymphocyte count	0.483	0.999	1.003	0.487 <sup>a</sup>
Lymphocyte (%)	6.366	1.089	1.974	0.012 <sup>a</sup>
Monocyte count	0.006	0.992	1.009	0.936 <sup>a</sup>
Monocyte (%)	4.750	1.073	3.748	0.029 <sup>a</sup>
NLR	0.009	0.423	2.187	0.926 <sup>a</sup>
NMR	0.155	0.794	1.415	0.693 <sup>a</sup>

BEO: Brucella epididymo-orchitis, NBEO: Non-brucella epididymo-orchitis, ESR: Erythrocyte sedimentation rate, WBC: White blood cells, NLR: Neutrophil-to-lymphocyte ratio, NMR: Neutrophil-to-monocyte ratio  
<sup>a</sup>: Binary logistic regression analysis

percentage of monocytes had an AUC value of 0.745 (95% confidence interval: 0.670–0.820) at a cut-off point of >7.7%. Finally, the percentage of neutrophils had an AUC value of 0.746 (95% confidence interval: 0.699–0.836) at a cut-off point of <66.8%.

Isolated epididymo-orchitis was present in 27 (32%) patients with BEO, and other focal involvement was observed in 57 (68%). Fifty-six patients (66.7%) with BEO drank or ate unpasteurized dairy products. Occupational exposure was present in 32 patients (38.1%) with BEO. Other focal involvements, including osteoarticular involvement (39.2%), hepatosplenomegaly (29.8%), sacroiliitis (29.8%), spondylitis (16.7%), arthritis (6%), and endocarditis (4.7%) were observed (Table 3). Orchiectomy was performed in one BEO patient (1.2%) because there was no improvement with medical treatment.

Table 3. Clinical presentations of the patients with BEO	
	n (%)
Fever	76 (90.5%)
Sweating	68 (81%)
Scrotal pain and swelling	84 (100%)
Lower urinary tract symptoms	25 (29.8%)
Only epididymo-orchitis (no focal involvement)	20 (23.8%)
<b>Other focal involvements</b>	
Hepatosplenomegaly	25 (29.8%)
Osteoarticular involvement	33 (39.2%)
Spondylitis	14 (16.7%)
Arthritis	5 (6%)
Sacroiliitis	25 (29.8%)
Endocarditis	4 (4.7%)
Positive blood culture	16 (19%)
Positive Wright agglutination test ( $\geq 1/160$ )	84 (100%)
(Some patients had more than one involvement) BEO: Brucella epididymo-orchitis	

## Discussion

In this study, a significant difference was found between the BEO and NBEO groups in terms of the percentages of lymphocytes, monocytes, and neutrophils. The higher percentages of mononuclear cells (lymphocytes and monocytes) in patients with BEO may be due to the effects of chronic and granulomatous changes in tissues on blood circulation.

The signals developed in BEO increase the number of monocytes in the blood. An increase in the percentage of monocytes in the blood, which is one of the most important precursors of a granulomatous infection, may be an essential clue in the differential diagnosis. An increase in the percentage of

lymphocytes is also one of the main parameters of chronic infections (12). The percentage of lymphocytes and monocytes in BEO were stronger parameters in differential diagnosis than NLR and NMR. This may be because neutrophils are inflammatory cells with less of a role in chronic infections.

The higher percentage of neutrophils in patients with NBEO may be due to NBEO being an acute infection. In the differential diagnosis of NBEO, neutrophil percentage was a stronger indicator than NLR and NMR. This may be because lymphocytes and monocytes are not essential inflammatory cells in acute infections.

The NLR is useful in the differential diagnosis of inflammatory diseases (13). Cift and Yucel (14) reported that NLR was lower in patients with BEO than in those with NBEO (14). In our study, there was a significant difference between the BEO and NBEO groups in terms of NLR in the univariate analysis, but no significant difference was detected in the multivariate analysis. This difference may be because the independent variables determined for differential diagnosis were not investigated in the previous study. Percentages of lymphocytes, monocytes, and neutrophils can provide a more comprehensive assessment. At the same time, because the number of patients with BEO in the study of Cift and Yucel (14) was lower than that in our study (n=22 vs. 84), the power of their statistical analysis may be lower.

It has been reported that the mean platelet volume (MPV) and red cell distribution width (RDW) are affected in many inflammatory diseases (15,16). Cift and Yucel (14) reported that MPV was lower in patients with BEO than in those with NBEO. They also noted that MPV was higher in pediatric patients with brucella arthritis than in the healthy control group (14). In Aydin et al. (17), RDW was similar between the BEO and NBEO groups. In our study, MPV and RDW were similar between patients with BEO and NBEO, which can be attributed to the presence of inflammation in both groups.

BEO may rarely require orchiectomy. In the literature, the rate of patients with BEO who underwent orchiectomy was reported to be 1.5–11.7% (18,19). In some studies, it has been stated that orchiectomy is not required in BEO (5,20). In our study, the rate of orchiectomy among patients with BEO was 1.2%. Performing the Wright tube agglutination test in patients with suspected BEO can prevent delayed treatment and unnecessary orchiectomy.

Brucellosis may present with isolated orchitis without systemic symptoms. Celen et al. (5) reported no other focal involvement in 22.2% of patients with BEO (5), whereas Gozdas and Bal (6) found this rate to be 12%. In our study, 23.8% of patients with BEO did not have any other focal involvement. Therefore, it should be noted that Brucella infections may present only with

epididymo-orchitis in suspected cases of brucellosis in endemic areas.

BEO is more common in young adults (21). One study noted that the mean age of patients with BEO was 25.5 years (22). Another study reported a median age of 34 years among patients with BEO (23). In our study, the median age of the BEO group was 35 years. These findings may be helpful in terms of identifying BEO in young adult patients from endemic areas.

Bilateral involvement is reported in 3.7-20.8% of patients with BEO (24,25). The rate of bilateral involvement among our patients with BEO (9.5%) is consistent with the literature. Baykan et al. (25) reported that bilateral involvement was higher in patients with BEO than in those with NBEO. However, bilateral involvement was similar between the BEO and NBEO groups in our study.

### Study Limitations

Our study was retrospective in nature. In addition, although patients with BEO were routinely evaluated in detail due to the difficulty of the disease treatment, some of the patients with NBEO did not have available laboratory data, which resulted in the inability to include consecutive NBEO cases in the study. Because BEO is a rare disease, the number of patients is limited. However, this study is important because it has the largest BEO cohort in the literature.

### Conclusion

To the best of our knowledge, there is no other study comparing the percentages of lymphocytes, monocytes, and neutrophils between patients with BEO and NBEO. According to our findings, the percentages of lymphocytes and monocytes were higher, and the percentage of neutrophils was lower in patients with BEO than in those with NBEO. In addition, abdominal pain may be an important sign in the differential diagnosis of BEO.

### Ethics

**Ethics Committee Approval:** The study was approved by the ethics committee (approval no: 2022/149, date: 22.11.2023 - University of Health Sciences Türkiye, Ankara Gülhane Training and Research Hospital Clinical Research Ethics Committee).

**Informed Consent:** Retrospective study.

### Authorship Contributions

Concept: E.B., Design: E.B., Data Collection or Processing: O.E., F.Y.İ., E.K.D., F.Ü., A.H.S., S.A., G.F., Analysis or Interpretation: O.E., E.K.D., S.B., Literature Search: E.B., F.Ü., G.F., Writing: F.Y.İ., A.H.S., S.A., S.B.

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