

## Original Article

# All CBC parameters in diagnosis of acute appendicitis

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**Abstract:** Objective: To investigate leukocyte count, neutrophil percentage, Neutrophil-Lymphocyte Ratio (NLR), Platelet-Lymphocyte ratio (PLR), mean platelet volume (MPV), red cell distribution width (RDW), platelet distribution width (PDW) and CRP in the diagnosis of acute appendicitis (AA). Materials and methods: A retrospective case-controlled study was designed in two groups. Ninety-seven patients containing AA and 94 patients in control group. Leukocyte count, neutrophil percentage, NLR, PLR, MPV, RDW, PDW and CRP were compared in two groups. Results: The mean PLR values in AA and control were  $166\pm 97$  and  $107\pm 28$  respectively, and there was a significant difference in PLR values between the groups ( $P<0.0001$ ). Best cutoff point for WBC count in the diagnosis of AA was  $8650 \times 10^6/\mu\text{L}$ , which had a sensitivity of 76% and a specificity of 94%. Best cutoff in diagnosis AA for NLR 3.15, which had a sensitivity of 77% and a specificity of 94%. Acute appendicitis group having low levels in MPV and RDW values but there were no significant differences in two groups. Leukocyte levels correlated with NLR, PLR and PDW. Conclusion: Neutrophil-Lymphocyte Ratio and leukocyte count seems to be a better inflammatory marker in AA. Higher PLR levels important parameter in diagnosis of AA. However, ROC analysis showed that MPV and RDW levels is not marker in terms of sensitivity and specificity compared to other markers.

**Keywords:** Acute appendicitis, diagnosis, platelet-lymphocyte ratio

## Introduction

Acute appendicitis (AA) is one of the most frequent cause of emergent surgical intervention due to acute abdomen [1]. The diagnosis of the disease in an emergency setting, however, still presents some challenges [2]. Traditionally, the diagnosis is based on a clinical history of abdominal pain, migration of pain to the right iliac fossa and signs of local peritonitis; diagnostic accuracy based on these symptoms ranges from 72% to 83%. Despite ultrasound and CT scan is useful for diagnosing AA, it adds to the cost of patient care and is often not available in rural hospitals [1-3]. Moreover, the accuracy of ultrasound is dependent upon the operator is a disadvantage and may not help in achieving an accurate diagnosis. Thus, surgeons are still in need of an accurate and easy test to obtain the diagnosis.

Hemogram analyses, generally gets physicians thought as unimportant for the diagnosis of acute appendicitis, except white blood cell (WBC) count and neutrophil predominance. On

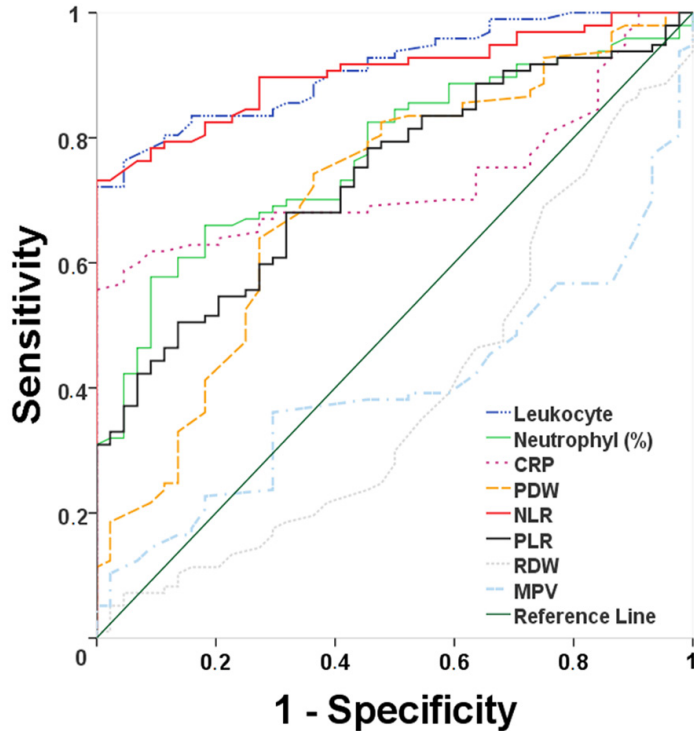
the other hand, recent studies have demonstrated that Neutrophil-to-Lymphocyte Ratio (NLR), mean platelet volume (MPV), red cell distribution width (RDW), platelet distribution width (PDW) are associated with acute appendicitis and inflammatory diseases [4-8]. No studies in literature have examined Platelet-Lymphocyte ratio (PLR) in patients with AA and correlation of CRP with hematological parameter inflammation as NLR for this subject before. In this retrospective case-controlled study, we aimed to seek whether leukocyte count, neutrophil percentage, NLR, PLR, MPV, RDW, PDW and CRP had significant importance in the diagnosis of AA.

## Materials and methods

### Study design

The study is a case controlled retrospective clinical study. All consecutive individuals undergoing appendectomy at the Department of General Surgery, Kadirli State Hospital, Osmaniye, Turkey, between April 2013 and April

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**Figure 1.** ROC analyze of potential diagnostic hematological parameters (diagonal segments are produced by ties).

2015 considered eligible for the present study. In patients operated with an initial diagnosis of AA, there were supporting findings in history, like right lower abdominal pain, nausea and vomiting and signs of local peritonitis. Histopathologic examination was used as a basis for diagnosis of AA; a normal appendix on histopathologic examination was a reason for exclusion. Additional exclusion criteria included previous abdominal surgery, younger than 18 years old, pregnant women, receiving medical therapy, alcohol consumption, tobacco smoking, diabetes mellitus, heart disease, vascular diseases, hypertension, morbid obesity and patients with severe comorbidities.

According to aforementioned exclusion criteria, a total of 97 patients (58 males and 39 females), were determined as the acute appendicitis (AA) group. Control group were consisted of 94 patients (60 males and 34 females), who were selected from healthy adults of similar age who applied to outpatient check-up of clinic and had no active complaint, chronic disease, or abnormal physical examination. All CBC's were obtained from patients file as well as automation system records. Only CBC's which were taken in a period of 24 hours prior

to surgery were accepted for AA group.

### Laboratory analysis

All tests were performed on blood samples obtained via venous system and collecting into EDTA tubes. All complete Blood Count (CBC) which were taken in a period of 24 hours prior to surgery were accepted for AA group. WBC count, neutrophil ratio, platelet count, MPV, PDW and RDW were analyzed via CBC samples in no time. NLR and PLR were calculated via obtaining ratio to lephocyte value. Hematological parameters were measured by an automated hematology analyzer (Coulter LH 780 Hematology Analyzer, Beckman Coulter Inc, CA, USA). The upper limits of the reference intervals were as follows: Leukocyte counts (WBC)  $4500-10300 \times 10^6/\mu\text{L}$ , Platelet:  $130-400 \times 10^3/\mu\text{L}$ , Neutrophil:  $2-6.9 \times 10^3/\mu\text{L}$  (37%-80%), Lymphocyte:  $0.6-3.4 \times 10^3/\mu\text{L}$  (10%-50%), MPV: 7.2-11 fL, RDW: 11%-16%, PDW: 10%-18% and CRP: 0-10 mg/dl.

### Statistical analysis

Data analysis was performed using the Statistical Package for the Social Sciences for Windows, version 15 (SPSS Inc, Chicago, IL, USA). Shapiro-Wilk test was used for analyzing whether distributions of continuous variables were normal. Data were shown as mean  $\pm$  standard deviation. The Independent Student-T test was used to compare the means of the control and the patient groups for the studied variables because all data had normal distribution. Categorical variables were compared using chi-square significance tests. The Pearson correlation analysis was carried out to examine the linear relationships among the variables. The predictive ability of AA and cutoff values of parameters for discrimination of the groups were determined using the receiver operating characteristic (ROC) analysis (Figure 1). At each value, the sensitivity and specificity for each outcome under study were plotted with Area under Curve (AUC), thus generating an ROC curve. *P* value less than 0.05 was considered statistically significant.

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**Table 1.** Demographics and laboratory findings of patients with significance values

| Variables                 | Control (n:94) | Appendicitis (n:97) | P       |
|---------------------------|----------------|---------------------|---------|
| Gender (m/f)              | 60/34          | 58/39               | ns      |
| Age (year)                | 34±15          | 39±18               | ns      |
| WBC (10 <sup>6</sup> /μL) | 6157±1611      | 11964±3795          | <0.0001 |
| NEU (%)                   | 63.6±8.2       | 73±10.6             | <0.0001 |
| CRP (mg/L)                | 3.3±2.3        | 27.6±43             | <0.0001 |
| PDW                       | 13±2           | 15±2                | <0.0001 |
| NLR                       | 1.9±0.8        | 5.9±4.1             | <0.0001 |
| PLR                       | 107±28         | 166±97              | <0.0001 |
| RDW (%)                   | 14.5±1.5       | 14.1±1.7            | 0.141   |
| MPV (fL)                  | 8.5±0.8        | 8.2±1.2             | 0.168   |

Abbreviations: CRP: C-reactive protein, NLR: Neutrophil to lymphocyte ratio, PLR: Platelet to lymphocyte ratio, WBC: White blood cell, MPV: Mean platelet volume, RDW: Red Cell Distribution of Width, PDW: Platelet distribution width, NEU: Neutrophil.

**Table 2.** Diagnostic parameter comparison of hematological variables

| Variables                 | Cutoff Levels | AUC (95% CI)      | SN (%) | SP (%) |
|---------------------------|---------------|-------------------|--------|--------|
| WBC (10 <sup>6</sup> /μL) | 8650          | 0.911 (0.86-0.95) | 76     | 94     |
| NEU (%)                   | 70.4          | 0.772 (0.69-0.84) | 70     | 82     |
| CRP (mg/L)                | 5.1           | 0.734 (0.65-0.81) | 67     | 80     |
| PDW                       | 14.5          | 0.706 (0.61-0.79) | 64     | 73     |
| NLR                       | 3.15          | 0.903 (0.85-0.95) | 77     | 94     |
| PLR                       | 117           | 0.735 (0.65-0.81) | 66     | 70     |
| RDW (%)                   | 14            | 0.389 (0.29-0.49) | 35     | 45     |
| MPV (fL)                  | 8.7           | 0.401 (0.31-0.49) | 36     | 69     |

Abbreviations: CRP: C-reactive protein, NLR: Neutrophil to lymphocyte ratio, PLR: Platelet to lymphocyte ratio, WBC: White blood cell, MPV: Mean platelet volume, RDW: Red Cell Distribution of Width, PDW: Platelet distribution width, NEU: Neutrophil. AUC: Area Under Curve, SN: Sensitivity, SP: Specificity.

**Table 3.** Pearson correlation of hematological variables

| Variables          | PDW   | NLR     | PLR     | Neutrophil (%) |
|--------------------|-------|---------|---------|----------------|
| Leukocyte count    | 0.274 | 0.715   | 0.318   | 0.651          |
|                    | 0.001 | <0.0001 | <0.0001 | <0.0001        |
| C-Reactive protein | 0.059 | 0.172   | 0.120   | 0.176          |
|                    | 0.487 | 0.041   | 0.157   | 0.036          |
| Neutrophil (%)     | 0.199 | 0.481   | 0.062   | 1              |
|                    | 0.018 | <0.0001 | 0.464   | N.S.           |

First line indicates correlation coefficient (r); Second line indicates correlation significance value.

## Results

The demographic characteristics and laboratory findings of appendicitis and control popula-

tions are shown in **Table 1**. No significant difference was observed between the appendicitis and control groups with respect to age and gender  $P>0.05$  (**Table 1**). White blood cell, Neutrophil ratio, CRP, PDW, NLR, PLR, MPV and RDW results, cut-off, AUC, sensitivity and specificity are demonstrated in **Tables 1** and **2**. Briefly, the mean WBC count in AA group was  $11964\pm3795$   $10^6/\mu\text{L}$  and in control group was  $6157\pm1611$   $10^6/\mu\text{L}$  ( $P<0.0001$ ); neutrophil ratio in AA group and control group were  $73\pm10.6$  and  $63.6\pm8.24$  ( $P<0.0001$ ); and CRP in AA group and control group were  $27.6\pm43$  mg/L and  $3.3\pm2.3$  mg/L ( $P<0.0001$ ). Platelet distribution width values in AA group and control group were  $15\pm2$  and  $13\pm2$  ( $P<0.0001$ ). The mean NLR values in AA and control group were  $5.9\pm4.1$  and  $1.9\pm0.8$ , respectively, and there was a significant difference in NLR values between the groups ( $P<0.0001$ ). The mean PLR values in AA and control were  $166\pm97$  and  $107\pm28$  respectively, and there was a significant difference in PLR values between the groups ( $P<0.0001$ ). AA group having low levels in MPV and RDW values but there were no significant differences in two groups. Mean platelet values in the AA group and control group were  $8.2\pm1.2$  fL and  $8.5\pm0.8$  fL ( $P=0.168$ ). Red cell distribution width values in AA group and control group were  $14.1\pm1.7$  and  $14.5\pm1.5$ , respectively ( $P=0.141$ ).

Receiver operating characteristic curve analysis suggested that the best cutoff point for WBC count in the diagnosis of AA was 8650  $10^6/\mu\text{L}$ , which had a sensitivity of 76% and a specificity of 94%, (area under curve [AUC]: 0.911; **Figure 1**). Best cutoff in diagnosis AA for NLR 3, 15, which had a sensitivity of 77% and a specificity of 94% similar to WBC count, (area under curve [AUC]: 0.903; **Figure 1**). Receiver operating characteristic curve analysis suggested that the best cutoff point for neutrophil ratio in the diagnosis of AA was 70.4% which had a sensitivity of 70% and a specificity of

82% (area under curve [AUC]: 0.772; **Figure 1**). Receiver operating characteristic curve analysis show the best cutoff point for CRP level in the diagnosis of AA was 5.1 mg/dL, which had a sensitivity of 67% and a specificity of 80% (area under curve [AUC]: 0.734; **Figure 1**). Platelet distribution width cutoff value 14.5 in the diagnosis of AA which had a sensitivity of 64% and a specificity of 73%, (area under curve [AUC]: 0.706; **Figure 1**). Finally, receiver operating characteristic curve analysis suggested that the best cutoff point for PLR in the diagnosis of AA 117, which had a sensitivity of 66% and a specificity of 70%, (area under curve [AUC]: 0.903; **Figure 1**). Correlation of hematological variables is shown in **Table 3**. Leukocyte levels correlated with NLR, PLR and PDW. NLR correlated with leukocyte levels, CRP and neutrophil ratio. PLR was not correlated with CRP and neutrophil ratio. However, we found a correlation between PLR and leukocyte levels (**Table 3**).

### Discussion

Acute appendicitis is the most common abdominal surgical emergency, but its diagnosis remains a challenge and generally established by a clinician, based on the patient's history, clinical examination and radiological imaging [9, 10]. Despite, the improvements in imaging methods such as ultrasonography and computerized tomography, it adds to the cost of patient care and is often not available in rural hospitals. Due to this reasons and inflammatory process of AA, many authors consider using biomarkers for diagnosis [4-8]. However, there is no single reliable test with satisfactory sensitivity and specificity.

Hemogram and CRP is an inexpensive diagnostic test which can be performed in small laboratories. This is study first study in literature investigated diagnostic all in values of leukocyte count, neutrophil percentage, NLR, PLR, MPV, RDW, PDW and CRP in AA. There is a limited number of studies in literature showing PDW and RDW changes in AA, whereas there is no study on the PLR [3, 7].

While certain traditionally agreed upon hemogram values such as increased leukocyte count, neutrophil percentage and CRP are well known to clinicians as biomarkers in the diagnosis of acute appendicitis. WBC count is most frequently used to laboratory test for diagnosis

acute appendicitis. Many studies support that WBC is the first indicator to be elevated in appendix inflammation [11]. We found that WBC count was significantly higher in AA. Narci found cutoff value  $10.4 \times 10^3/\text{mm}^3$  with a 91% sensitivity and 74% specificity [8]. Dinc found cutoff value  $10.6 \times 10^9/\mu\text{L}$  with a 73.1% sensitivity and 94% specificity [7]. Similar to the literature, the present study found that the sensitivity and specificity of leukocyte level were 76% and 94%, respectively with cutoff level  $8650 \times 10^6/\mu\text{L}$ .

There are very few studies on this subject, but all reported that NLR appears to have greater diagnostic accuracy than traditional diagnostic laboratory tests. It is also reported that NLR on admission to the hospital is an independent predictor of positive appendicitis histology [12]. Kahramanca found NLR value 4.68 for certain diagnosis of AA [6]. According to the results of our study NLR of 3.15 reliable parameter to obtain diagnosis of acute appendicitis with 77% sensitivity and 94% specificity. According to these results, NLR and leukocyte count seems to be a better inflammatory marker in acute appendicitis. Our cutoff value is lower than the value recommended for the diagnosis of AA in leukocyte and NLR.

Neutrophil ratio was also used as a laboratory test for acute appendicitis. In a retrospective study, elevated neutrophil ratio has been detected as a good diagnostic marker in acute appendicitis. The sensitivity of neutrophil ratio has been 60.1% and specificity 76.9% in diagnosing acute appendicitis [13]. In a case-controlled study by Bilici *et al.* [14] in children, the sensitivity was 77% and the specificity was 91%. In the present study, the sensitivity and specificity of neutrophil ratio was found 70% and 82%, respectively.

CRP is a sensitive acute phase protein that lacks specificity due to increased levels in all acute inflammatory processes. Its concentration increases with the duration and extent of the inflammation. In a meta-analysis examining the accuracy of CRP levels in the diagnosis of acute appendicitis, a wide range of sensitivity (40-99%) and specificity (27-90%) was found in literature [15]. Similar to the literature, this study found a sensitivity of 67% and a specificity of 80% for CRP in the diagnosis of acute appendicitis.



MPV, a marker of platelet activation, is being investigated for its correlation with both inflammation and thrombosis. MPV is easily measured in CBC analysis and it presumably reflects the functional and activation status of platelets and their production rate from megakaryocytes as well. Some studies were suggested MPV alteration as a valuable diagnostic marker when it was combined with WBC and neutrophil percentage, but the alteration of MPV was controversial, some of them reported MPV decrease and some of them reported MPV increase in acute appendicitis [14-17]. On the other hand, in one study MPV value was found to have no diagnostic value in pediatric acute appendicitis cases [18]. In the current trial, we detected lower MPV levels in patients with AA. However, we detected not statistical difference between the groups. The values of sensitivity and specificity of MPV in our study, 36% and 69%, respectively, are low compared to the literature.

Platelet distribution width (PDW) are presented in the complete blood cell count, which is routinely used in emergency departments. They are the indicators of platelet activation. In a 202-case study by Aydogan [19], patients were divided into groups as perforated and non-perforated. In this study with no control group, PDW values were significantly higher in the perforated group. Dinc [7] found PDW is higher than WBC and neutrophil percentage diagnostic accuracy for AA with 97.1% sensitivity and 93% specificity. In the present study, the sensitivity and specificity of PDW was found 64% and 73%, respectively is lower from Dinc et al. study.

RDW, which is a measure of heterogeneity in the size of circulating red blood cells, is used in the differential diagnosis of anemia. RDW is commonly used to discriminate between microcytic anemia's due to iron deficiency and those due to thalassemia or hemoglobinopathies [20]. Increased RDW levels are related to impaired erythropoiesis or erythrocyte degradation [21]. RDW is also an independent variable of prognosis in patients with heart failure, myocardial infarction, diabetes mellitus, and pulmonary hypertension [22, 23]. Narci found RDW levels significantly lower in the acute appendicitis group [8]. According to our study we have no significant differences in groups.

Platelets are a source of inflammatory mediators [24]. Increased platelet activation is known to trigger atherosclerosis and plays a major role in its progression [25]. Elevated peripheral blood platelet count is closely related to major adverse cardiovascular outcomes [26-28]. Although preliminary data have shown that the platelet-lymphocyte ratio (PLR) is associated with major adverse cardiovascular outcomes and some cancers. According to our study PLR level was significantly higher in the AA group compared with the control group ( $P < 0.0001$ ) (**Table 1**). Receiver operating characteristic curve analysis suggested that the best cutoff point for PLR in the diagnosis of AA was 117 which had a sensitivity of 66% and a specificity of 70%, (area under curve [AUC]: 0.735; **Figure 1**).

### Conclusion

In conclusion, we have found NLR and leukocyte count seems to be a better inflammatory marker in acute appendicitis. Higher PLR levels important parameter in diagnosis of acute appendicitis. However ROC analysis showed that MPV and RDW levels is not marker in terms of sensitivity and specificity compared to other markers. We believe that further prospective high volume researchs is needed to find more specific and more reliable biomarkers in the diagnosis of acute appendicitis.

### Disclosure of conflict of interest

None.

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