



## An Investigation of the Usability of Complete Blood Count Parameters in the Diagnosis of Acute Appendicitis

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### ABSTRACT

**Introduction and Objective:** Acute appendicitis is one of the most commonly diagnosed diseases in emergency departments, and it is the abdominal pathology that most frequently requires surgery. The purpose of this study was to investigate the value in the diagnosis of acute appendicitis of routinely investigated complete blood count parameters in patients presenting to the emergency department with abdominal pain.

**Material and method:** This study was planned as a retrospective cohort among patients over 18 years who were diagnosed with acute appendicitis in the tertiary emergency department in Türkiye between January 1 and December 31, 2019. Healthy volunteers aged over 18 with no active complaints undergoing routine blood tests for health screening were included in the control group. Both groups' records were retrieved retrospectively from the hospital database. Data consisting of the groups' demographic characteristics at the time of presentation to the emergency department and parameters obtained from complete blood tests were analyzed.

**Results:** A total of 1200 people were included in the study, 600 of which were in the control group. Men constituted 388 (64.7%) of the acute appendicitis patients and 380 (63.3%) of the control group ( $p=0.674$ ). Leukocyte, neutrophil, nucleated red blood cell, neutrophil-lymphocyte ratio, and platelet-lymphocyte ratio values were significantly higher in the acute appendicitis group compared to the control group ( $p<0.001$ ).

**Conclusion:** An increase in leukocyte, neutrophil, nucleated red blood cell values, neutrophil-lymphocyte ratio, and platelet-lymphocyte ratio and a decrease in lymphocyte, red blood cell distribution width and mean platelet volume values in the complete blood count of patients with acute appendicitis is a useful guide for diagnosis.

**Key Words:** Acute appendicitis, complete blood count, diagnosis, emergency medicine

### 1. INTRODUCTION

Abdominal pain is one of the most common complaints on admission to the emergency department (ED) (1). Appendicitis is the condition most frequently requiring hospitalization in patients presenting with abdominal pain, and the most common form of surgery in treatment is appendectomy (2). Acute appendicitis (AA) can be seen in all age groups, but most frequently between the second and fourth decades. The lifetime prevalence in the entire population is 7-8%, with a male/female incidence of 1.4:1 (3).

Laboratory tests and radiological imaging techniques may be required in addition to physical examination in the diagnosis of AA. However, AA can

manifest with different clinical and examination findings in all patients (4). Diagnosis may therefore not always be easy. There is also a risk of encountering a healthy appendix in surgery performed before a diagnosis is confirmed, and the likelihood of complications such as perforation increases when the diagnosis is delayed (5). Morbidity and mortality are adversely affected by both states of affairs, which may also expose physicians to serious malpractice suits. Although several biomarkers have recently been studied in terms of the diagnosis of AA, there is still no laboratory test capable of establishing a definite diagnosis (6).

In this study we aimed to determine the place in the

diagnosis of AA of white blood cell (WBC), lymphocyte, neutrophil, platelet, mean platelet volume (MPV), red cell distribution width (RDW), nucleated red blood cell (NRBC) parameters and platelet/lymphocyte ratio (PLR) and neutrophil/lymphocyte ratio (NLR) values calculated from these to facilitate the diagnosis of AA in EDs with high patient numbers.

## 2. MATERIALS AND METHODS

### 2.1. Study design

The study was performed after obtaining the necessary local and ethical committee approval. The patients who were over 18 years patients that presented to the tertiary ED due to abdominal pain between 1 January and 31 December 2020, taken for surgery with preliminary diagnoses of AA, and with the diagnosis of AA confirmed by pathology reports were examined retrospectively. Six hundred volunteers over 18 with similar demographic characteristics to those of the patient group, undergoing complete blood tests for routine health screening, and with no known hematological disease or active complaints during screening were enrolled as the control group.

#### 2.1.1. Data Collection

G-power 3.1 analysis software was applied to determine the size of the sample. A minimum requirement of 584 individuals was calculated (alpha: 0.05, beta: 0.80). The records of 720 patients operated on with diagnoses of AA between the study dates were scanned. After excluding a total of 120 patients younger than 18, with missing data, with a known hematological disease, or with normal or neoplastic pathology reports, the study was finally conducted with 600 patients. The patients were divided into complicated appendicitis (CA) and uncomplicated appendicitis (UA) subgroups based on pathological examinations.

#### 2.1.2. Statistical analysis

Statistical analyses were performed on the Jamovi project (2020), Jamovi (Version 1.6.7.0), and JASP (Version 0.14), p values lower than 0.05 were regarded as statistically significant. Descriptive statistics were expressed in table form as distribution

-dependent mean  $\pm$  SD or median (min-max or IQR) for continuous (numerical) variables. Categorical variables were expressed as numbers and percentages. The Kolmogorov-Smirnov test was used to determine numerical variables to the normal distribution. The Mann-Whitney U test was applied to compare non-normally distributed numerical variables between two independent groups. For group-based categorical variable comparisons, the Pearson Chi-Square test was used in 2x2 tables with five or more expected cells, Fisher's Exact test in tables with fewer than five expected cells, and the Fisher Freeman Halton test in RxC tables with fewer than five expected cells. ROC analysis was performed on MedCalc Statistical Software trial version software (MedCalc 2015). Optimal cut-off values, 95% confidence intervals, and area under the curve (AUC) were calculated with Youden's index using the DeLong method.

## 3. RESULT

The 600-member patient group consisted of 388 (64.7%) men and 212 (35.3%) women. The median age of the patient group was 32.0 years. The median age of the CA subgroup was 36.5 years, and the UA group was 31.0 years ( $p=0.001$ ) (Table 1).

Analysis of the complete blood count (CBC) results showed that leukocyte, neutrophil, NRBC, PLR, and NLR values were higher in the patient group, while lymphocyte, MPV, and RDW values were lower. The median leukocyte count in the patient group was higher ( $p<0.001$ ). The median neutrophil count in the patient group was higher ( $p<0.001$ ). The median lymphocyte count in the patient group was  $1.9 \times 10^3 \mu\text{l}$ , and in the control group was  $2.2 \times 10^3 \mu\text{l}$  ( $p<0.001$ ). The median MPV value in the patient group was 9.9 femtoliter(fl) and 10.1 fl in the control group ( $p=0.003$ ). The median RDW in the patient group was 39.5 fl and 39.9 fl in the control group ( $p=0.025$ ). The mean NRBC value in the patient group was  $0.00118 \times 10^3 \mu\text{l}$  and  $0.00070 \times 10^3 \mu\text{l}$  in the control group ( $p=0.005$ ). The PLR value was 131.2 in the patient group and 105.6 in the control group ( $p<0.001$ ). The NLR value in the patient group was 5.7, significantly higher than the 1.7 value in the control group ( $p<0.001$ ) (Table 2).

**Table 1.** Demographic characteristics of groups and subgroups

	<b>Total</b> (n=1200)	<b>Acute Appendicitis Group</b> (n=600)	<b>Control Group</b> (n=600)	<b>p</b>
<b>Gender (%)</b>				
Male	768 (64.0)	388 (64.7)	380 (63.3)	0.674*
Female	432 (36.0)	212 (35.3)	220 (36.7)	
<b>Age</b>	35.0 [18.0 – 88.0]	32.0 [18.0 – 88.0]	36.0 [19.0 – 62.0]	<b>&lt;0.001**</b>
<b>Acute Appendicitis groups according to the histopathology</b>				
	<b>Uncomplicated acute appen- dicitis</b> (n=492)	<b>Complicated acute appendicitis</b> (n=108)		<b>p</b>
<b>Gender (%)</b>				
Male	322 (65.4)	66 (61.1)		0.458*
Female	170 (34.6)	42 (38.9)		
<b>Age</b>	31.00 [18.00 – 83.00]	36.50 [18.00 – 88.00]		<b>&lt;0.001**</b>

Descriptive statistics were given as median, minimum and maximum for numerical variables and as number (%) for categorical variables. \*: Pearson Chi-Square, Fisher's Exact, or Fisher Freeman Halton tests were used. \*\*: Mann-Whitney U tests were used.

**Table 2.** Comparison of laboratory parameters of acute appendicitis and control groups

	<b>Reference Interval</b>	<b>Total</b> (n=1200)	<b>Acute Appendicitis Group</b> (n=600)	<b>Control Group</b> (n=600)	<b>p*</b>
<b>WBC</b> (x10 <sup>3</sup> µl)	4.5-10	9.5 [3.0 – 26.8]	14.0 [4.9 – 26.8]	7.3 [3.0 – 16.1]	<b>&lt;0.001</b>
<b>Neutrophil</b> (x10 <sup>3</sup> µl)	1.8-7.5	6.0 [0.9 – 24.2]	10.9 [2.2 – 24.2]	4.1 [0.9 – 12.2]	<b>&lt;0.001</b>
<b>Lymphocyte</b> (x10 <sup>3</sup> µl)	0.8-3.2	2.2 [0.4 – 6.2]	1.9 [0.4 – 6.2]	2.4 [0.9 – 5.2]	<b>&lt;0.001</b>
<b>Platelet</b> (x10 <sup>3</sup> µl)	150-450	251.0 [115.0 – 562.0]	248.5 [115.0 – 562.0]	252.0 [120.0 – 526.0]	0.304
<b>MPV (fl)</b>	9-12	10.0 [8.0 – 14.1]	9.9 [8.0 – 13.3]	10.1 [8.1 – 14.1]	<b>0.003</b>
<b>RDW (fl)</b>	37-50	39.8 [33.1 – 61.1]	39.5 [33.1 – 61.1]	39.9 [33.2 – 57.4]	<b>0.025</b>
<b>NRBC</b> (10 <sup>3</sup> µl)	0-0.11	0.00094 [0.0 – 0.02]	0.00118 [0.0 – 0.02]	0.00070 [0.0 – 0.01]	<b>0.005</b>
<b>NLR</b>	0.40-2.34	2.6 [0.6 – 32.7]	5.7 [0.7 – 32.7]	1.7 [0.6 – 9.0]	<b>&lt;0.001</b>
<b>PLR</b>		114.4 [25.8 – 677.1]	131.2 [40.3 – 677.1]	105.6 [25.8 – 381.9]	<b>&lt;0.001</b>

WBC: white blood cell, MPV: mean platelet volume, RDW: red cell distribution width, NRBC: nucleated red blood cell, NLR: neutrophil/lymphocyte ratio, PLR: platelet/lymphocyte ratio.

Descriptive statistics were given as median, minimum, and maximum for numerical variables. \*: Mann-Whitney U tests were used.

No significant difference in mean leukocyte counts was observed between the UA and CA subgroups. The mean lymphocyte count in the CA subgroup

was 1.73x10<sup>3</sup>µl (p=0.005). The NLR was 6.53 in the CA subgroup and 5.54 in the control group (p=0.001). The PLR value in the CA subgroup, 137.6, was also

**Table 3.** Comparison of laboratory parameters of complicated appendicitis and uncomplicated acute appendicitis subgroups

	Acute Appendicitis groups according to the histopathology		p*
	Uncomplicated acute appendicitis (n=492)	Complicated acute appendicitis (n=108)	
WBC (x10 <sup>3</sup> µl)	13.88 [4.92 – 26.80]	14.60 [6.56 – 26.01]	0.127
Neutrophil (x10 <sup>3</sup> µl)	10.71 [2.17 – 24.18]	11.54 [4.07 – 22.10]	0.058
Lymphocyte (x10 <sup>3</sup> µl)	1.96 [0.54 – 6.15]	1.73 [0.41 – 5.02]	<b>0.005</b>
Platelet (x10 <sup>3</sup> µl)	248.00 [133.00 – 562.00]	250.50 [115.00 – 419.00]	0.968
MPV fl	10.00 [8.00 – 13.00]	9.90 [8.10 – 13.30]	0.778
RDW fl	39.55 [33.10 – 53.70]	39.60 [33.90 – 61.10]	0.136
NRBC 10 <sup>3</sup> µl	0.00115 [0.00 – 0.02]	0.00129 [0.00 – 0.01]	0.649
NLR	5.45 [0.68 – 32.68]	6.53 [1.44 – 26.96]	<b>0.001</b>
PLR	128.90 [40.33 – 677.11]	137.62 [54.59 – 554.00]	<b>0.011</b>

WBC: white blood cell, MPV: mean platelet volume, RDW: red cell distribution width, NRBC: nucleated red blood cell, NLR: neutrophil/lymphocyte ratio, PLR: platelet/lymphocyte ratio.

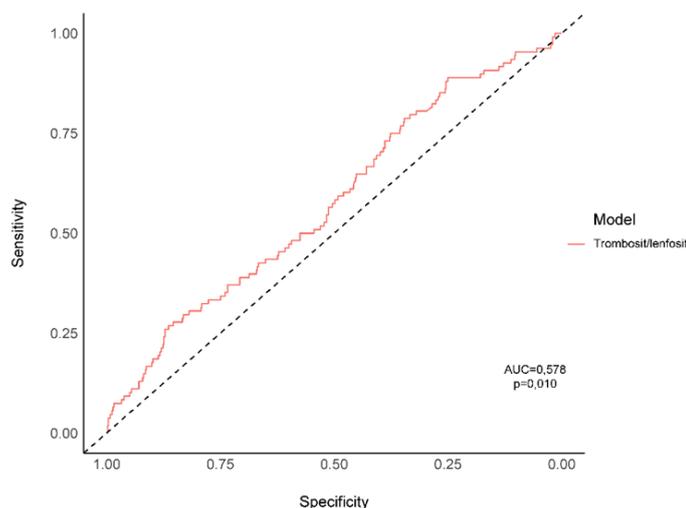
Descriptive statistics were given as median, minimum, and maximum for numerical variables. \*. Mann-Whitney U tests were used.

significantly higher than the 128.9 value in the UA subgroup (p=0.011) (Table 3).

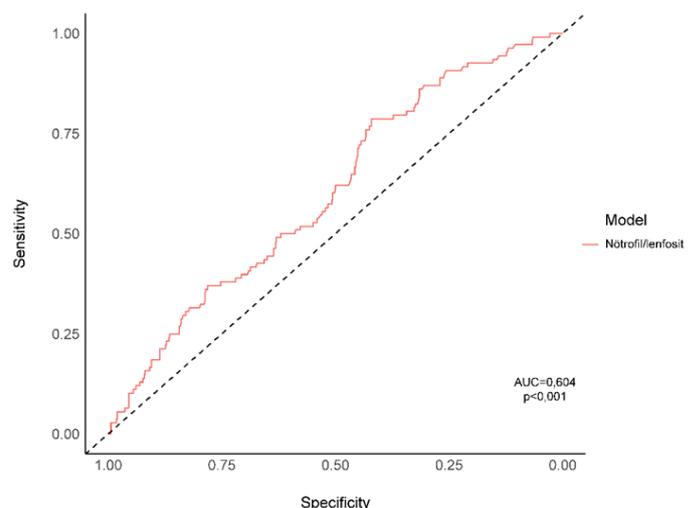
In the analysis performed to determine the role of laboratory values in the diagnosis of AA, the threshold value was 132.43, with the area under the ROC analysis for PLR 0.662 and the confidence interval 0.634-0.688 (p<0.001). In the analysis performed to determine the role of laboratory values in the diagnosis of AA, the threshold value was 2.79, with the area under the ROC analysis for NLR 0.932 and the confidence interval 0.917-0.946 (p<0.001). Analyses also revealed significantly higher PLR and

NLR values, and lymphocyte values significantly lower in the CA subgroup. In the analysis performed to determine the place of laboratory values in the diagnosis of CA, the threshold value was 95.74, with the area under the ROC curve for PLR 0.578 and the confidence interval 0.537-0.618 (p=0.010) (Fig. 1).

In the analysis performed to determine the role of laboratory values in the diagnosis of CA, the threshold value was 4.57, with the area under the ROC curve for NLR 0.604 and the confidence interval 0.563-0.643 (p<0.001) (Fig. 2).



**Figure 1.** Analysis of the threshold value of platelet/lymphocyte ratio with ROC curve in the prediction of complicated acute appendicitis



**Figure 2.** Analysis of the threshold value of neutrophil/lymphocyte ratio with ROC curve in the prediction of complicated acute appendicitis

#### 4. DISCUSSION

AA is a clinical manifestation characterized by infection of the appendix vermiformis (3). Abdominal pain is one of the most common complaints in patients presenting to the ED. These patients often require hospitalization and surgery. (2). The lifetime prevalence of AA in the population is 7-8% (3).

AA is generally seen between the second and fourth decades of life (3). Sevinç et al. reported a median age of 32.0 among patients with AA, with mean ages of 33.9±12.2 in the UA subgroup and 38.7±15.8 in the CA subgroup (7). Ugurlu et al. reported a median age of 37.79 in the perforated appendicitis group and 45.97 in the non-perforated appendicitis group (8). The median age of the appendicitis group in the present study was 32.0. The median age in the CA subgroup was significantly higher at 36.5 than the median age of 31.0 in the UA subgroup.

AA is 1.4 times more common in men than women (3). Sevinç et al. reported that men represent 59.2% of AA patients and women 40.8%, while Almström et al. reported figures of 57.1% for men and 42.9% for women (7,9). Consistent with the previous literature, men constituted 64.7% of the patients diagnosed with AA in the present study.

Several studies have investigated the diagnostic value of laboratory tests for AA. The presence of an increase in the leukocyte count is considered one of the early markers of inflammation of the appendix (10). Boshnak et al. reported a mean leukocyte value of 14.36x10<sup>3</sup>µl in their AA group, while Daldal et al. reported a value of 13.27x10<sup>3</sup>µl, significantly higher (11,12). The median leukocyte value in the AA group in the present study was higher than the median value in the control group. A cut-off value of 9.69x10<sup>3</sup>µl was determined for the differentiation of patients with AA from the control group. In the leukocyte values, there was no significant between the UA and CA subgroups.

In terms of the leukocyte formula, an increase is also observed in neutrophil counts and percentages in patients with AA. Akyüz et al. reported a neutrophil count of 10.1±3.9x10<sup>3</sup>µl in their AA group, while Maghsoudi et al. reported a value of 11.03x10<sup>3</sup>µl,

these values being significantly higher than in a negative appendectomy group (13,14). The median neutrophil count in the AA group in the present study was 10.9x10<sup>3</sup>µl, and this was also significantly higher than the median neutrophil count of 4.1x10<sup>3</sup>µl in the control group. Analysis revealed a cut-off value for the neutrophil count of 6.38x10<sup>3</sup>µl in the differentiation of AA patients from the controls, and a value of 9.25x10<sup>3</sup>µl for differentiating patients with CA from the control group.

Neutrophil elevation and a left shift in the leukocyte formula in AA are frequently accompanied by relative lymphopenia. Boshnak et al. reported a median lymphocyte value of 1.65±0.87x10<sup>3</sup>µl in an AA group, while Pehlivanlı et al. reported a value of 1.8x10<sup>3</sup>µl, these values being significantly lower compared to a negative appendectomy group (11,15). The median lymphocyte value among the AA in the present study was 1.9x10<sup>3</sup>µl, significantly lower than the value in the control group. Mean lymphocyte counts in the UA and CA subgroups were 1.96x10<sup>3</sup>µl and 1.73x10<sup>3</sup>µl, respectively, the difference being statistically significant. The cut-off value for the lymphocyte count in differentiating CA was 2.17x10<sup>3</sup>µl.

The response exhibited by leukocytes to stress developing during inflammation takes the form of an increased neutrophil count and a decreased lymphocyte count. Recent studies have shown that the ratio between these two subgroups (NLR) can be used as a marker of the body's inflammatory response (16). Rajalingam et al. reported a cut-off value of 4.75 for the NLR in differentiating patients with AA from the control group, and a cut-off value of 6.96 for differentiating patients with CA (17). Another study showed a significant difference between the complicated AA groups and the histopathologically normal appendix group in terms of NLR (18). The NLR value of the AA group in the present study was 5.7, significantly higher than that in the healthy control group. The cut-off value for the NLR in differentiating patients with AA from the control group was 2.79. NLR values in the patients with UA and CA were 5.45 and 6.53, respectively, the

difference being statistically significant. The cut-off value for the NLR in differentiating CA was 4.57.

The PLR has been linked to poor prognosis in peripheral vascular diseases, coronary artery diseases, and hepatobiliary malignancies (19). It has recently been suggested that PLR values can be used as an inflammatory marker, and research has commenced into its association with several inflammatory diseases, including appendicitis. Pehlivanlı et al. reported PLR values of 100.19 for a negative appendectomy group, 127.75 for a UA group, and 166.74 for a CA group, the differences being statistically significant (15). PLR values in the present study were 105.6 in the healthy control group and 131.2 in the appendicitis group. PLR values in the UA and CA subgroups were 128.90 and 137.62, and the difference was also significant. A PLR cut-off value of 132.43 was calculated for differentiating patients with AA from the healthy control group.

MPV is an important parameter that shows platelet function and activation. In the literature, it has been shown that MPV levels are associated with acute pancreatitis, ulcerative colitis, sepsis, and atherosclerotic heart diseases (20). Since AA is related to inflammation, studies have also investigated the association between this disease and MPV. Haghi et al. reported MPV values of  $9.52 \pm 1.06$  for a healthy control group and  $8.38 \pm 0.96$  for an appendicitis group, the difference being statistically significant (6). MPV values in the present study were 10.1 in the control group and 9.9 in the appendicitis group.

RDW shows the variation in erythrocyte dimensions. Inflammation and oxidative stress lead to RDW elevation by impairing the structure of erythrocytes. The number of studies investigating the relationship between AA and RDW has increased recently. Daldal et al. reported an RDW value of  $13.16 \pm 1.98$  in their AA group, while Haghi et al. determined a value of  $13.09 \pm 0.09$ , the low values in the patients with AA being statistically significant in both studies (12,6). In contrast to previous studies, the RDW values in the present research were expressed as femtoliters (fl). RDW values in the present study were 39.5 fl in the

AA group and 39.9 fl in the control group. The difference was statistically significant. Although RDW values are known to rise with inflammation, in agreement with previous research, we also observed lower values in our AA group. We think that the change in RDW values may differ in acute and chronic inflammatory conditions and that further studies are needed on the subject.

NRBCs, also known as erythroblasts, are immature erythrocyte cells present in the bone marrow in the process of hematopoiesis (21). Circulating NRBCs are not generally present in healthy individuals. Publications have suggested that NRBC elevation in the neonatal period may be associated with asphyxia. The presence of NRBCs in peripheral blood after the neonatal period is generally associated with bone marrow diseases, malignant neoplasms, and severe infections. Recent studies have shown that NRBC values increase in inflammatory conditions and are associated with high mortality in cardiovascular diseases (21,22). There has been an increase in the number of studies investigating the relationship between NRBC values and inflammation, although our scan of the literature revealed no studies regarding its value in the diagnosis of AA. The mean NRBC value in the AA group in this study was  $0.00118 \times 10^3 \mu\text{l}$ , and higher than the control group. The fact that we encountered no studies of the applicability of NRBC values in the diagnosis of appendicitis makes our results particularly noteworthy.

The principal limitation of this study lies in its retrospective nature. However, its particular strengths lie in the large patient number, the fact that numerous complete blood count parameters were evaluated simultaneously, and the absence of any similar research in the literature regarding the diagnostic significance of NRBC values in AA.

## 5. CONCLUSION

Although the PLR and NLR calculated using the CBC parameters of neutrophil, leukocyte, and lymphocyte counts and MPV, RDW, and NRBC values appear to be a useful guide to the diagnosis of AA, they are not by themselves diagnostic. In our

study, leukocyte, neutrophil, and NRBC values were found to be higher and lymphocyte, MPV, and RDW values were found to be lower in AA patients. Also, NLR and PLR values were found to be significantly higher in AA patients. NLR and PLR values were found to be significantly higher and lymphocyte values were found to be significantly lower in CA patients compared to UA patients. Since there are no studies in the literature regarding the applicability of NRBC in the diagnosis of appendicitis, the present research is valuable in terms of investigating this relationship and yielding significant results. We think that further studies on the subject are now needed.

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**Conflicts of Interest:** The authors declared that there is no conflict of interest.

**Ethical Statement:** The study was approved by the Kayseri City Training and Research Hospital Ethics Committee with the date and number 04.02.2021/101.

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