ORIGINAL ARTICLE

Comparison of the Diagnostic Values of Leukocytes, Neutrophils, Neutrophil-to-Lymphocyte Ratio and Platelet-to-Lymphocyte Ratio in Distinguishing between Acute Appendicitis and Right Ureterolithiasis

Lixia Chen and Yuezhan Zhang

Department of Emergency, The Fourth Affiliated Hospital Zhejiang University School of Medicine, Zhejiang, China

SUMMARY

Background: **To compare the diagnostic values of leukocytes, neutrophils, neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) in distinguishing between acute appendicitis (AA) and right ureterolithiasis (RU).**
Methods: **In this retrospective study, 106 patients diagnosed with AA (Appendicitis group) and 33 cases with RU (Ureterolithiasis group) were enrolled due to acute right lower abdominal pain. The levels of peripheral blood leukocyte counts (leukocytes), neutrophil counts (neutrophils), lymphocyte counts (lymphocytes), platelet counts (platelets), NLR and PLR were recorded and compared between the two groups. Student's *t*-test for independent samples was adopted for comparing the mean between the two groups. Model discrimination was evaluated using the area under the receiver operating characteristic curve (AUC). Comparison of AUC was performed using the *Z*-test.**
Results: **The levels of leukocytes, neutrophils, NLR, and PLR were significantly increased in AA compared with RU (all p < 0.01), while there were no significant statistical differences of lymphocytes and platelets (all p > 0.05); moreover, AUC in distinguishing AA from RU was 0.797 (95% confidence interval (CI), 0.721 to 0.861) for leukocytes, 0.814 (95% CI, 0.740 to 0.875) for neutrophils, 0.770 (95% CI, 0.691 to 0.837) for NLR, and 0.608 (95% CI, 0.522 to 0.690) for PLR, and significant differences were observed between PLR and any of the three other parameters (all p < 0.01), while there were no significant statistical differences after pairwise comparison between leukocytes, neutrophils and NLR (all p > 0.05). Finally, the cutoff values were 13.1 $\times$ 10^9/L in distinguishing between AA and RU (specificity 87.88%, sensitivity 63.21%, and Youden index 0.511) for leukocytes, 7.4 x 10^9/L (specificity 69.70%, sensitivity 83.02%, and Youden index 0.527) for neutrophils, 5.57 (specificity 81.82%, sensitivity 68.87%, and Youden index 0.507) for NLR, and 182.5 (specificity 84.85%, sensitivity 37.74%, and Youden index 0.226) for PLR.**
Conclusions: **Leukocytes, neutrophils, and NLR can demonstrate more accurate and reliable diagnostic values than PLR, suggesting that they are useful and potential biomarkers in distinguishing between AA and RU.**
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Correspondence:
Dr. Yuezhan Zhang
Department of Emergency
The Fourth Affiliated Hospital Zhejiang
University School of Medicine
322000 Zhejiang
China
E-mail: 8018151@zju.edu.cn

INTRODUCTION

Acute abdominal pain (AAP), accounting for 7 - 10% of the Emergency Department (ED) visits [1,2], is still the most common complaint [3]. Acute appendicitis (AA) is one of the most common abdominal emergencies worldwide [4]. Among all patients with abdominal pains in different areas, evaluation of acute right lower quadrant (RLQ) pain remains a common and challenging clinical scenario for ED physicians due to frequent nonspecific signs, symptoms, and physical examination findings [5]. It is known that the most common cause of acute RLQ pain is AA [6]. The reported lifetime risk of appendicitis in the US is 8.6% in men and 6.7% in women, with an annual incidence of 9.38 per 100,000. In the US, it is estimated that around 326,000 operations for appendicitis were performed in 2007 [7]. However, RLQ pain patients with a suspected diagnosis of AA may have other pathological conditions. The common mimics of AA are related to acute gastrointestinal and genitourinary diseases [8]. As a genitourinary disease, acute right ureterolithiasis (RU) could be challenging to distinguish from AA, especially when the symptoms are often nonspecific. Although computed tomography (CT) and ultrasonography are useful tests in diagnosing AA and RU [9-12], their shortcomings cannot be ignored such as high cost, radiation exposure, and differences in the professional levels of radiologists and ultrasonographers. Therefore, it is necessary for ED physicians to combine medical history, physical examination, laboratory tests, and radiological imaging modalities other than only one diagnostic parameter to decrease the rates of negative laparotomy and morbidity secondary to a delay in diagnosis [13].

Appropriate diagnostic laboratory tests may be helpful to support other specific diagnoses and thereby play a significant role in warranting more expedited evaluation and accurate treatments. In clinical practice, we often discovered changes in blood routine tests resulted in both AA and renal colic [13-20]. Farooqui et al. [17] found that patients with AA had significantly higher blood levels of white blood cells (WBC) than those without AA, and patients with perforated appendicitis had significantly higher levels of WBCs than those with non-perforated appendicitis. Yardımcı et al. [18] proved that NLR could be useful in predicting severity of AA. Bozkurt et al. [19] drew a conclusion that elevated WBCs and neutrophil counts might be used as diagnostic tests in cases of AA. Paajanen et al. [20] demonstrated that the mean values of C-reactive protein (CRP) (41 mg/L) and blood leukocytes (14 x 10^9/L) were elevated in appendicitis, but not in renal colic (14 mg/L and 10 x 10^9/L, respectively). However, it remains uncertain whether these simple biomarkers could be used to differentiate AA from RU preliminarily. To the best of our knowledge, comparing the diagnostic values of leukocytes, neutrophils, neutrophil-to-lymphocyte ratio (NLR), and platelet-to-lymphocyte ratio (PLR) in distinguishing between AA and RU have not yet been discussed. Our study may provide novel evidence for whether leukocytes, neutrophils, and NLR could demonstrate more accurate and reliable diagnostic values than PLR in distinguishing between AA and RU.

MATERIALS AND METHODS

Study population

A total of 139 patients with acute RLQ pain symptoms from ED admitted to the Fourth Affiliated Hospital Zhejiang University School of Medicine, were consecutively enrolled in this study between January 2018 and May 2019. A total of 106 inpatients were diagnosed with AA (Appendicitis group) which were confirmed later by histopathological analysis of the mass excised during surgery and the remaining 33 cases were outpatients with RU (Ureterolithiasis group). Inclusion criteria were adults ≥ 18 years old, symptoms onset within previous 24 hours. All patients had acute RLQ pains and underwent blood routine tests and CT and/or ultrasound scans. Exclusion criteria were patients with acute or chronic inflammatory diseases, hematological diseases, cancers, autoimmune diseases, thyroid dysfunction, hepatic or renal insufficiency, intracranial hemorrhage, acute ischemic stroke, acute coronary syndrome or other trauma diseases; patients whose symptoms onset were beyond 24 hours; patients who had been treated with antibiotics before laboratory tests; and patients whose clinical data were incomplete. The study was conducted in accordance with the *Declaration of Helsinki* and approved by the Ethics Committee of the Fourth Affiliated Hospital Zhejiang University School of Medicine.

Clinical data

Baseline data were gathered from medical records when patients were admitted to the Fourth Affiliated Hospital Zhejiang University School of Medicine, including age, gender, leukocyte counts (leukocytes), neutrophil counts (neutrophils), lymphocyte counts (lymphocytes), platelet counts (platelets), CT, and ultrasonography results. A Beckman coulter DXH800 hematology analyzer was used to determine leukocytes, neutrophils, lymphocytes, and platelets. NLR and PLR were calculated and recorded. All candidates underwent peripheral blood routine tests and CT and/or ultrasonography scans before any antibiotic treatment.

Statistical analysis

Statistical analysis was performed using SPSS software (version 19.0 for Windows, SPSS Inc., Chicago, IL USA). Descriptive analyses for continuous variables were used to calculate mean values and standard deviations. The qualitative data were compared with chi-square test. Student's *t*-test for independent samples was adopted to compare the mean between the two groups. Prognostic performance was tested by calculation of the receiver operating characteristic (ROC)

Table 1. Comparison of the levels of different variables between two groups.

Variables	Appendicitis group (n = 106)	Ureterolithiasis group (n = 33)	p-value
Age (years)	39.74 ± 14.31	41.45 ± 12.54	0.537
Gender (female/male)	38/68	10/23	0.558 *
Leukocyte (x 10^9/L)	13.72 ± 4.00	9.44 ± 3.25	< 0.001
Neutrophil (x 10^9/L)	11.32 ± 3.85	6.75 ± 3.17	< 0.001
Lymphocyte (x 10^9/L)	1.59 ± 0.87	1.89 ± 0.92	0.083
Platelet (x 10^9/L)	216.79 ± 61.80	215.42 ± 75.99	0.917
NLR	10.05 ± 7.37	4.79 ± 4.19	< 0.001
PLR	181.00 ± 125.52	132.99 ± 59.62	0.003

Quantitative data were present as mean ± standard deviations, Leukocyte - Leukocyte counts, Neutrophil - Neutrophil counts, Lymphocyte - Lymphocyte counts, Platelet - Platelet counts, NLR - neutrophil-to-lymphocyte ratio, PLR - platelet-to-lymphocyte ratio, Student's *t*-test or chi-square test *.

curve and displayed in the AUC. Comparison of AUC was performed using the *Z*-test of the software MedCalc version 18.2.1 (MedCalc Software bvba, Ostend, Belgium). From ROC coordinates, the cutoff values for the aforementioned scores using the score value with the best Youden index (sensitivity + specificity -1) were identified. p < 0.05 was considered statistically significant.

RESULTS

Characteristics of total population
A total of 106 patients diagnosed with AA (Appendicitis group) and the remaining 33 cases with RU (Ureterolithiasis group) were available for final analysis. The mean age of the study sample was (40.14 ± 13.89) years. Of them, 48 (34.5%) were females and 91 (65.5%) were males. There were no significant statistical differences of age and the rates of gender (all p > 0.05) (Table 1).

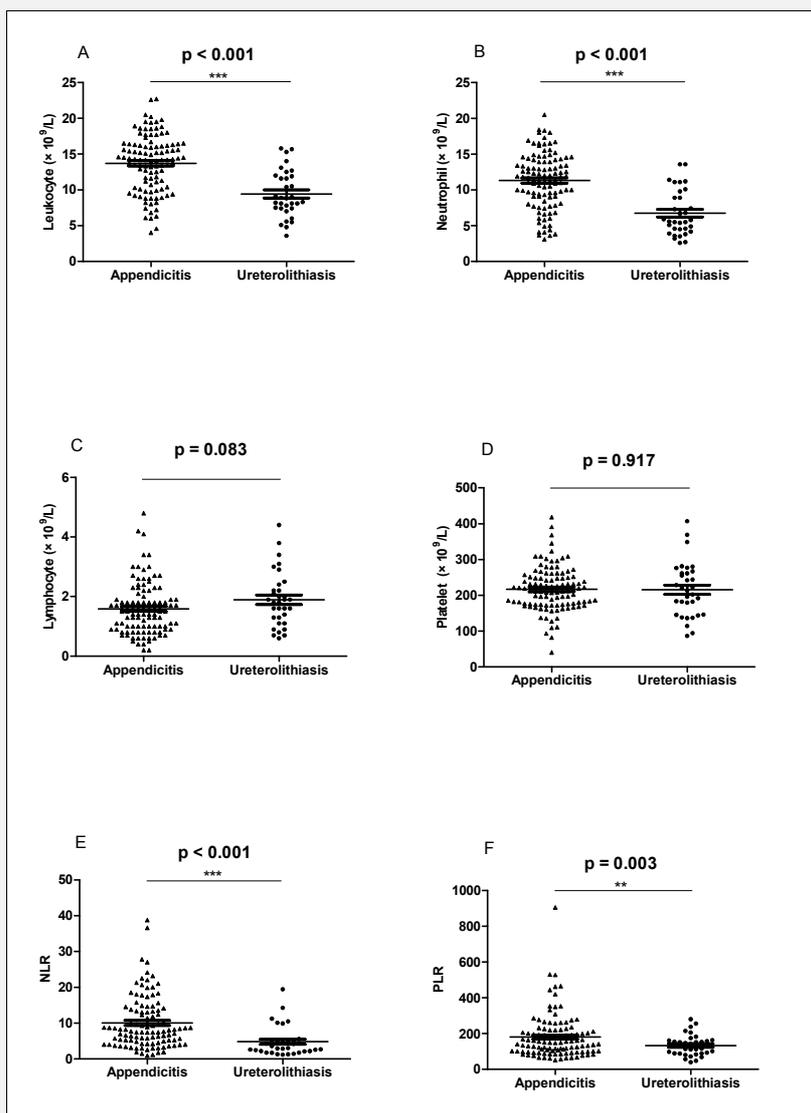**Comparison of leukocytes, neutrophils, lymphocytes, platelets, NLR and PLR between two groups**
As shown in Table 1, Figure 1A, Figure 1B, Figure 1E, and Figure 1F, leukocytes, neutrophils, NLR and PLR were significantly increased in the Appendicitis group compared with the Ureterolithiasis group, and significant differences have been observed (all p < 0.01). While there were no significant differences in lymphocytes and platelets between the two groups (all p > 0.05) (Table 1, Figure 1C, and Figure 1D).

Comparison of ROC curves of leukocyte, neutrophil, NLR and PLR in distinguishing between AA and RU
AUC in distinguishing between AA and RU was 0.797 (95% confidence interval (CI), 0.721 to 0.861) for leukocytes, 0.814 (95% CI, 0.740 to 0.875) for neutrophils,

0.770 (95% CI, 0.691 to 0.837) for NLR, and 0.608 (95% CI, 0.522 to 0.690) for PLR, and significant differences were observed between PLR and any of the three other parameters (all p < 0.01) (Figure 2), while there were no significant statistical differences after pairwise comparison between leukocytes, neutrophils, and NLR (all p > 0.05) (Figure 2). Finally, the cutoff values were 13.1 x 10^9/L in distinguishing between AA and RU (specificity 87.88%, sensitivity 63.21%, and Youden index 0.511) for leukocytes, 7.4 x 10^9/L (specificity 69.70%, sensitivity 83.02%, and Youden index 0.527) for neutrophils, 5.57 (specificity 81.82%, sensitivity 68.87%, and Youden index 0.507) for NLR, and 182.5 (specificity 84.85%, sensitivity 37.74%, and Youden index 0.226) for PLR.

DISCUSSION

Undeniable evidence was brought that AAP of non-traumatic origin was one of the more frequent complaints leading people to ED. In a retrospective analysis of 5,340 cases, Cervellin et al. [21] found that renal colic (31.18%) was the second most frequent cause, and appendicitis (4.54% vs. 1.47%) and renal colic (34.48% vs. 20.84%) were more frequent in patients aged < 65 years compared with those aged > 65 years, respectively. In addition, renal colic was the most frequent cause of ED admission in men. Due to atypical symptoms, acute RU sometimes mimics AA. Although a mistake of appendicitis for ureteral stone was clinically rare occurring only once or twice per year in the hospital where 700 - 800 emergency appendectomies were performed annually [20]. AA would still be devastating after missed or delayed diagnosis, in that it could lead to perforation, abscess, other serious infections, and death [22], suggesting that it was of importance to differentiate AA from RU early and accurately. If ED physicians made
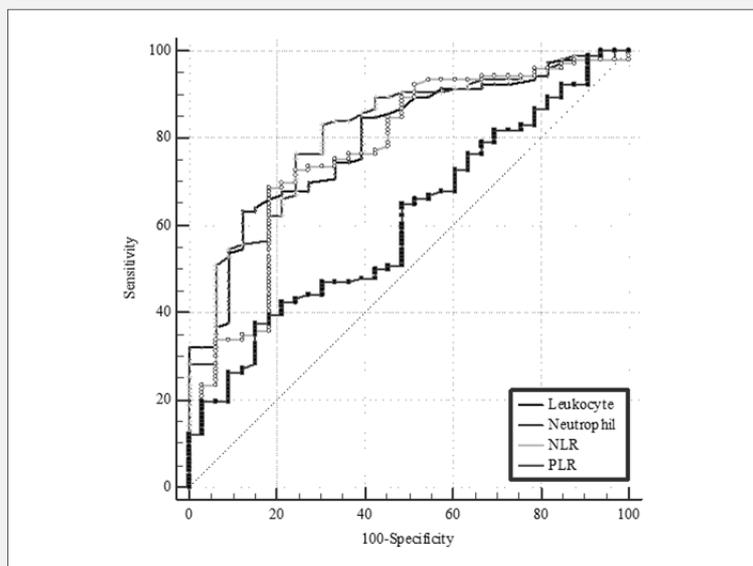
Figure 1. Comparison of leukocytes (A), neutrophils (B), lymphocytes (C), platelets (D), NLR (E), and PLR (F) between two groups.

** p < 0.01, *** p < 0.001, Leukocytes - Leukocyte counts, Neutrophils - Neutrophil counts, Lymphocytes - Lymphocyte counts, Platelets - Platelet counts, NLR - neutrophil-to-lymphocyte ratio, PLR - platelet-to-lymphocyte ratio.

inappropriate differential diagnoses, it was likely to generate both medicolegal litigation and unfavorable outcomes [23,24]. However, how to distinguish between AA and RU by simple means has become a common topic of concern for clinical physicians.

AA is an inflammation of appendix vermiformis which is directly associated with inflammatory processes [13]. Therefore, many inflammatory markers (i.e., leukocytes, neutrophils, NLR and PLR) have been studied in AA [13-20]. Chung et al. [25] reported that these in-

flammatory markers had guided many unexperienced physicians and surgeons in their decision-making processes for surgery. Cases of elevated leukocytes due to acute RU are encountered frequently in clinical practice. Reasons for leukocyte elevation other than infection have been discussed in the literature. An interaction between the stone and the ureteral mucosa during acute renal colic might induce an inflammatory reaction, which leads to cytokine release and subsequent WBC elevation [26].

4

Figure 2. Comparison of ROC curves of leukocytes, neutrophils, NLR, and PLR in distinguishing between AA and RU.

Leukocytes - Leukocyte counts, Neutrophils - Neutrophil counts, NLR - neutrophil-to-lymphocyte ratio, PLR - platelet-to-lymphocyte ratio.

In the present study, we have proven that leukocytes were significantly increased in the Appendicitis group compared with Ureterolithiasis group ((13.72 ± 4.0) x 10^9/L vs. (9.44 ± 3.25) x 10^9/L, p < 0.001), suggesting that leukocytes could be used to distinguish between AA and RU. Our study result was also consistent with Acar [13] and his colleagues' conclusion, as they found that WBCs were significantly increased in Appendicitis group compared with Renal Colic group ((13.3 ± 2.8) x 10^9/L vs. (8.05 ± 1.6) x 10^9/L, p < 0.001). Moreover, we also observed that neutrophils were significantly increased ((11.32 ± 3.85) x 10^9/L vs. (6.75 ± 3.17) x 10^9/L, p < 0.001), suggesting that neutrophils could distinguish AA from RU, similarly. Previous clinical evidence [13] also supported this view ((10.60 ± 0.27) x 10^9/L vs. (4.96 ± 0.16) x 10^9/L, p < 0.001). Furthermore, we also observed that the NLR and PLR were significantly increased in the Appendicitis group compared with Ureterolithiasis group (p < 0.001, p = 0.003), consistent with the previous study [13], suggesting that NLR and PLR could distinguish between AA and RU, similarly. Whereas, we found no significant differences in lymphocytes and platelets between the two groups (all p > 0.05). In other words, lymphocytes and platelets could not be used to differentiate AA from RU. However, this result was not completely consistent with the conclusion of the previous study [13], as it demonstrated that lymphocytes other than platelets could distinguish between Appendicitis and Renal Colic, similarly.

The specific reason of the controversial conclusion is unclear and whether it is related to the sample size needs to be confirmed by further studies.

In addition, we also discovered that the AUC in distinguishing between AA and RU was 0.797 for leukocytes, 0.814 for neutrophils, 0.770 for NLR, and 0.608 for PLR, and there were significant statistical differences between PLR and any of the three other parameters (0.797 vs. 0.608, Z = 3.024, p = 0.0025; 0.814 vs. 0.608, Z = 3.756, p < 0.001; 0.770 vs. 0.608, Z = 3.780, p < 0.001), while there were no significant statistical differences after pairwise comparison between leukocytes, neutrophils, and NLR (0.797 vs. 0.814, Z = 1.030, p = 0.303; 0.814 vs. 0.770, Z = 1.410, p = 0.158; 0.797 vs. 0.770, Z = 0.642, p = 0.521) (Figure 2); finally, the Youden indices of leukocytes (0.511), neutrophils (0.527), and NLR (0.507) were superior to PLR (0.226), which further verified that leukocytes, neutrophils, and NLR could demonstrate more accurate and reliable diagnostic values than PLR. Our study result was also consistent with the previous study [13], in that it was reported that the AUC in distinguishing between the Appendicitis group and Renal Colic group was 0.896 for leukocytes, 0.916 for neutrophils, 0.888 for NLR, and 0.725 for PLR. In the present study, we have proposed the more potential diagnostic values of leukocytes, neutrophils, and NLR in distinguishing between AA and RU.

There are several limitations in our study. First, al-

though we have taken special care to avoid sources of bias and confounding, some potential bias (i.e., collecting blood samples at different times and disease severity) may still exist in this retrospective, and it is a single-center study. Second, CRP, as one of the most important inflammation markers, was not observably tested in file screening studies. Finally, we only described the differences of laboratory tests between AA and RU, while we did not investigate the underlying mechanisms. A larger group and more homogeneous samples from a multicenter study are needed to verify the conclusions.

CONCLUSION

Leukocytes, neutrophils, and NLR can demonstrate more accurate and reliable diagnostic values than PLR, suggesting that they are useful and potential biomarkers in distinguishing between AA and RU.

Declaration of Interest:
The authors report no competing financial interests.

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