The effect of neutrophil/lymphocyte ratio and tomography measurements at the time of diagnosis in predicting aneurysm rupture in patients with abdominal aortic aneurysms

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ABSTRACT

Objectives: In this study, we aimed to examine the effect of neutrophil/lymphocyte ratio (NLR) in predicting rupture in patients with abdominal aortic aneurysms (AAA).

Patients and methods: Between March 2014 and September 2017, a total of 42 patients (32 males, 6 females; mean age 66±11.7 years; range, 57 to 81 years) with an AAA were retrospectively analyzed. The patients were divided into two groups as the ruptured (n=13) and non-ruptured groups (n=29), and the effects of baseline NLR and computed tomography results on rupture were evaluated.

Results: The mean follow-up was 2.5 (range, 2 to 4) years. The mean abdominal aortic size, neutrophil percentage, and NLR were significantly higher in the ruptured group, while the mean lymphocyte ratio was significantly lower in the ruptured group. There was a significant correlation between the abdominal aortic size and NLR. Using the receiver operating characteristics, the NLR was found to be more sensitive and specific than the abdominal aortic diameter in predicting rupture.

Conclusion: Our study results indicate that abdominal aortic diameter, smoking, and hypertension are the risk factors for rupture of AAAs. In addition to classical risk factors, NLR is a novel and simple predictor of aortic rupture.

Keywords: Aortic rupture, computed tomography, neutrophil/lymphocyte ratio.

Abdominal aortic aneurysms (AAAs) are a common vascular disease with a predominance of 3% in individuals aged 60 years or older.1 Many studies have consequently been conducted to understand the causal mechanisms which drive aneurysm progress. The AAA progression and rupture follows, when the circulatory stress on the vessel wall exceeds the aortic wall strength. The pathogenesis of this disease is complex, and immune-mediated mechanisms resulting in the activation of matrix metalloproteinases (MMPs) with consequent disruption of the systematic lamellar structure of the aortic media and tissue degradation play essential roles.2,3 Pro-inflammatory CD4+ and CD8+ T cells lacking CD28, the co-stimulatory molecule, are enriched in the peripheral blood and tissue specimens of AAA patients have been reported.4 Therefore, inflammatory porches have an important role in the pathophysiology of AAAs.

Systemic inflammation, usually observed in the form of elevated C-reactive protein (CRP) or neutrophil/lymphocyte ratio (NLR), occurs by various mechanisms involving numerous pro-inflammatory cytokines.5 The NLR defines the balance between the neutrophil and lymphocyte levels and is useful in the determination of systemic inflammation.5 To date, the neutrophil/lymphocyte (N/L) ratio has been used as a systemic index for inflammation by
deciphering the levels of neutrophil and lymphocytes during illness.[6] In the present study, we aimed to examine the effect of NLR in predicting rupture in patients with AAAs.

PATIENTS AND METHODS

In this retrospective study, a total of 156 patients who were admitted with an AAA to our clinic between March 2014 and September 2017 were screened. Patients with bicuspid aorta, severe or symptomatic aortic stenosis, severe aortic regurgitation, significant mitral valve disease (i.e., mitral stenosis or regurgitation), acute or chronic kidney failure and inflammatory diseases such as ankylosing spondylitis, rheumatoid arthritis, scleroderma, or systemic lupus erythematosus were excluded from the study. Finally, a total of 42 patients (32 males, 6 females; mean age 66±11.7 years; range, 57 to 81 years) who were offered surgery or endovascular therapy for AAAs, but refused these treatment options were included in the study. Data were retrieved from medical records considering mortality and patient preferences. A written informed consent was obtained from each patient. The study protocol was approved by the Ethics Committee of Erciyes University, Faculty of Medicine (No. 2020/19). The study was conducted in accordance with the principles of the Declaration of Helsinki.

During follow-up, the patients were divided into two groups as the ruptured (n=13) and non-ruptured groups (n=29) and the effects of baseline biomarkers and computed tomography (CT) results were evaluated. All CT examinations were done with a commercially available CT scanner (Toshiba Co. Ltd., Tokyo, Japan) equipped with 16 detectors with the following scanning parameters: a gantry rotation speed of 0.4 sec per rotation, detector row configuration of 16×1 mm, and helical pitch 23. The images were reconstructed with a 1-mm slice thickness at 0.8 mm intervals. A contrast agent was administered by an infusion pump through an antecubital vein to enhance the aorta and iliac arteries followed by a saline infusion flush. The contrast volume was estimated per protocol and an infusion rate of 4 mL/sec was set. Coordination between the contrast administration and image acquisition was adjusted automatically by the scanner, when 180 HU of contrast concentration was detected at a region of interest located in the ascending aorta.

All blood parameters were taken during hospital admission via an antecubital vein. Blood glucose, creatinine, lipid profile, and all routine biochemical tests were conducted using an autoanalyzer (Roche Diagnostic Modular Systems, Tokyo, Japan). Blood samples for NLR analyses were collected

### Table 1. Baseline characteristics of the patients

|                      | Rupture group (n=13) | Non-rupture group (n=29) | p    |
|----------------------|----------------------|--------------------------|------|
| n                    | 13                   | 29                       |      |
| %                    | 92%                  | 79%                      |      |
| Mean±SD              | 65.9±13.6            | 67±12.8                  | 0.321|
| Age (year)           |                      |                          |      |
| Body mass index (kg/m²) | 22.6±2.4            | 22.5±2.9                 | 0.558|
| Hypertension         | 13                   | 22                       |      |
| Diabetes mellitus    | 5                    | 5                        |      |
| Smoking status       | 10                   | 7                        |      |
| Coronary artery disease | 2                   | 9                        | 0.312|
| Creatine (mg/dL)     | 1.2±0.3              | 1.2±0.4                  | 0.792|
| Glucose (mg/dL)      | 98±23                | 95±19                    | 0.649|
| Total cholesterol (mg/dL) | 177±38              | 181±41                   | 0.470|
| Low-density lipoprotein (mg/dL) | 107±31             | 114±35                   | 0.323|
| High-density lipoprotein (mg/dL) | 45±9              | 47±9                     | 0.459|
| Triglyceride (mg/dL) | 120±82               | 110±47                   | 0.474|
| Hemodynamic parameters |                      |                          |      |
| Systolic blood pressure | 121.8±16.5          | 123.8±19.1               | 0.664|
| Diastolic blood pressure | 79.2±11.6           | 77.7±13.1                | 0.778|

SD: Standard deviation.
in the tripotassium ethylenediaminetetraacetic acid (EDTA)-based anticoagulated tubes. The samples were assessed via a Sysmex K-1000 auto analyzer within 30 min of sampling to avoid swelling of platelets.

**Statistical analysis**

The study power and sample size calculation were performed using the G*Power version 3.1.9.7 (Heinrich-Heine-Universität Düsseldorf, Düsseldorf, Germany). According to a beta value of 0.90 and an alpha value of 0.05, the minimum sample size was calculated as 36.

Statistical analysis was performed using the IBM SPSS version 21.0 (IBM Corp., Armonk, NY, USA) and MedCalc 32 trial version. Descriptive data were presented in mean ± standard deviation (SD), median (min–max) or number and frequency. Univariate analysis (non-metric scales) frequency and chi-square for two or more samples, Mann-Whitney U, and independent simple tests were used. Continuous variables were tested for normal distribution by the Kolmogorov-Smirnov test and compared between the groups using the Student t-test or Mann-Whitney U test. Categorical variables were compared using the chi-square test. The Pearson correlation coefficient was used to estimate the degree of association between the variables. The receiver operating characteristic (ROC) curve was used to calculate the sensitivity and specificity of the rupture, NLR, and aortic aneurysm size and to establish an optimal cut-off value for predicting ruptured AAAs. A \( p \) value of <0.05 was considered statistically significant with 95% confidence interval (CI).

### RESULTS

The mean follow-up was 2.5 (range, 2 to 4) years. Six of the patients who developed rupture during the follow-up were treated with an emergency surgical procedure and seven of them died. Regarding risk factors for rupture, there was a significant difference in the presence of hypertension \( (p=0.04) \) and smoking status \( (p=0.003) \); however, there was no significant difference in diabetes mellitus and coronary artery disease between the groups \( (p=0.120 \) and \( p=0.312, \) respectively). In addition, there was no significant difference between the groups in terms of baseline laboratory test results and medical treatment applied (Table 1).

The mean abdominal aortic size \( (73.31±6.51 \text{ mm vs. } 61.13±7.08 \text{ mm, respectively; } p=0.01) \) was significantly higher in the ruptured group. In addition, the mean neutrophil percentage \( (77.9±10.9\% \text{ vs. } 71.9±9.11\%, \text{ respectively; } p=0.05) \) and NLR \( (8.9±4.3 \text{ vs. } 4.4±2.06, \text{ respectively; } p=0.002) \) were significantly higher, while the mean lymphocyte count \( (13.6±8.6 \times 10^3/\mu\text{L, } \text{respectively; } p=0.035) \) was significantly lower in the ruptured group (Table 2).

The NLR and abdominal aortic size at the time of admission were higher in the patients with AAA rupture \( (p=0.002) \). There was a significant correlation between these two variables \( (r=0.537; p<0.001) \) (Figure 1).
The ROC analysis of the NLR and abdominal aortic size for predicting rupture are shown in Figure 2. An abdominal aortic size of >7.0 mg/L measured at the time of admission had 66% sensitivity and 92% specificity in predicting AAA rupture. An NLR of >6.6 predicted ruptures with 80% sensitivity and 89% specificity.

**DISCUSSION**

In this study, we investigated the effect of systemic inflammation on AAAs and found NLR to be the prognostic factor in rupture. We demonstrated that the NLR, a sign of inflammatory status, was statistically independently associated with AAA rupture. In addition, it was significantly correlated with the abdominal aortic size.

Inflammation has a crucial role in the development and progression of AAAs. In men aged >65 years, the prevalence of AAAs is 5% and ruptures are responsible for 1 to 4% of total mortality in this population, largely because it is asymptomatic before rupture.\(^7\) In this study, we believe that the NLR may be effective as the aortic diameter as a new parameter in predicting rupture. We demonstrated that an NLR of >6.6 could predict the rupture with 80% sensitivity and 89% specificity and the abdominal aortic size was a significant predictor of rupture.

In general, AAAs are characterized by a thinning of the aortic media with rarefaction of vascular smooth muscle cells and extracellular matrix destruction induced by inflammation, oxidative stress, and proteolysis.\(^6\) The aneurysms occurred mainly in the aortic media and adventitia.\(^9\) The cause of the pathophysiology in these plates are oxidative stress, inflammation, matrix degradation, and smooth muscle cells apoptosis.\(^10\) Inflammatory processes play a key role in all stages of aneurysms.\(^5\) Beta cells, and pro-inflammatory CD4+ and CD8+ T cells lacking CD28, the co-stimulatory molecule, are supplemented in blood and tissue specimens of AAA patients, indicating the involvement of adaptive immune responses in the pathogenesis of the disease.\(^6\) In this study, the inflammation induced by inflammation caused cell damage and rupture by expanding the aneurysm due to the cell damage. Previous studies showed that the NLR was a sign of a balance between the neutrophil and lymphocyte levels in the body and an indicator of systemic inflammation.\(^12\) Inflammatory cells such as monocytes, macrophages and T lymphocytes, and a variety of proinflammatory cytokines are participants of a complex pathological pathway and responsible for fibrotic and calcific processes which drive increasing valve stiffness.\(^13\) This increased tension can cause wall expansion and induce rupture. Similarly, in our study, we showed an increase in the NLR, supporting the previous hypothesis.

Furthermore, atherosclerosis is a risk factor of AAA and it has been shown in studies that NLR is as effective as CRP, which is a marker of systemic inflammation, which is considered one of the most important causes of atherosclerosis.\(^1\) Circulatory neutrophils and lymphocytes may have different key roles in the pathophysiological development of atherosclerosis through their respective secretions.\(^14,15\) As in our study, with an increased neutrophil count, neutrophils release superoxide, and chemokines which affect endothelial cells and promote or amplify the recruitment of other inflammatory cells.\(^16\) Neutrophils also support monocyte adhesion and mobilization to the site of inflammation.\(^17\) While the neutrophil count increases, the lymphocyte count decreases with atherosclerosis.\(^18\) In our study, we showed this fall and rise. Therefore, an elevated NLR integrates the predictive risk of these two leukocyte subtypes into a single risk factor. Increased neutrophils and decreased lymphocytes are occasionally observed in patients with systemic inflammation and mortality.
In our study, the neutrophil count was higher and lymphocyte count was lower in the rupture group. These findings suggest that systemic inflammation may affect the rupture. In our study, we showed the effectiveness of the NLR, which is the indicator of inflammation in rupture prediction with high sensitivity and specificity.

Although the pathophysiology of AAA has not been fully elucidated yet, its etiology appears to be multifactorial.\(^{[20-22]}\) Inflammation is not only associated with the clinical presentation of AAAs, but also plays a key role in the AAA pathogenesis.\(^{[23]}\) In our study, similar to this data, smoking, aortic diameter, and hypertension were significantly different in the rupture group. The other important findings in our study were that inflammation alone was an additional factor in the efficacy of rupture. As mentioned in a previous study, rupture was influenced not only by inflammation, but also by smoking and aortic diameter; however, the sample size was low. In our study, on the other hand, an abdominal aortic size of >70 mm predicted rupture with 66% sensitivity and 92% specificity, indicating that the abdominal aortic size is a significant predictor of rupture. According to these findings, the NLR may be used as an important parameter as the width of the abdominal aorta for rupture with a high specificity and sensitivity.

Retrospective and single-center design with a small sample size are the main limitations of this study.

In conclusion, the aortic diameter, smoking, and hypertension are the main risk factors for rupture of AAAs. In addition to classical risk factors, the NLR is a novel and simple predictor of aortic rupture. Further large-scale studies using the combined aortic diameter and NLR may provide more clinically relevant results.

Declaration of conflicting interests

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