Neutrophil-to-lymphocyte ratio is increased in patients with rheumatic mitral valve stenosis?

Mehmet Kadri Akboğa, Ahmet Akyel[1], Asife Şahinarslan[2], Çağrı Yayla[2], Yakup Alsancak[2], Gökhan Gökalp[2], Serdar Nurkoç[2], Adnan Abacı[2]

Department of Cardiology, Ministry of Health Etimesgut State Hospital; Ankara-*Turkey*
[1]Department of Cardiology, Dışkapı Yıldırım Beyazıt Education and Research Hospital; Ankara-*Turkey*
[2]Department of Cardiology, Faculty of Medicine, Gazi University; Ankara-*Turkey*

Abstract

Objective: The role of systemic and chronic inflammatory processes in the pathophysiology of rheumatic heart valve disease is well known. The neutrophil-to-lymphocyte ratio (NLR) was shown to be an indicator of systemic inflammation. In this study, we aimed to investigate relationship between NLR as a marker of systemic inflammation and rheumatic mitral valve stenosis (RMVS).

Methods: This is a retrospective study. Among patients who underwent transthoracic echocardiography between January 2008-March 2013, 314 patients with RMVS were included retrospectively in the study. The control group included 57 healthy persons who underwent transthoracic echocardiography during the study period. Basal characteristics and NLR were compared between the two groups. Independent predictors of RMVS were determined by logistic regression analysis.

Results: Basal characteristics were similar among the groups (age, 50.2±14.2 vs. 49.2±13.0, p=0.60). The NLR was significantly higher in patients with RMVS [2.9 (0.6-13.0) vs. 2.1 (0.7-5.8), p<0.001]. Besides, C-reactive protein (CRP) was also higher in the RMVS group [5.99 (0.3-23.7) vs. 2.98 (0.6-6.3), p=0.001]. In the regression analysis, NLR (OR: 2.24, p=0.04), CRP (OR: 1.34, p=0.03), and left atrial diameter (OR: 1.21, p=0.001) were independent predictors of RMVS. In the correlation analysis, there was a significant positive correlation between NLR and CRP (r=0.43, p<0.001).

Conclusion: We found that NLR was significantly increased in RMVS. Furthermore, NLR was an independent predictor of the presence of RMVS in our study population. According to these findings, NLR can be used as a predictor of RMVS. Since it is an easily available and cheap method, it can easily be used in daily clinical practice. Increased NLR can also be a sign of ongoing chronic inflammation in patients with RMVS.
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Keywords: rheumatic mitral valve disease, neutrophil-to-lymphocyte ratio, inflammation, C-reactive protein

Introduction

Rheumatic heart valve disease is one of the most serious sequelae of rheumatic fever, and rheumatic mitral valve stenosis (RMVS) is the most frequently seen component of this devastating disease (1). By primary and secondary preventive measures, the incidence and prevalence of RMVS have been greatly decreased in developed countries. However, RMVS still results in significant morbidity and mortality, especially in underdeveloped and developing countries (2). Although the exact mechanism is unknown, RMVS has the characteristics of inflammatory and autoimmune processes. The role of systemic inflammation in the pathophysiology of RMVS is well established (3-5).

White blood cells and their subtypes have been shown to be predictors of a poor prognosis in many diseases related with inflammatory reactions. As a consequence of lymphocytopenia and increased neutrophils, the neutrophil-to-lymphocyte ratio (NLR) is increased in many inflammatory diseases (6). In several studies, it has been shown that NLR is closely related with unfavorable outcomes in many cardiovascular diseases (7-11).

According to our best knowledge, there are no data regarding the relationship between NLR and RMVS. Since NLR is closely linked to inflammatory status and RMVS is associated with chronic and systemic inflammatory status, we aimed to investigate the relationship between NLR and RMVS in this study.

Methods

The study was approved by the ethics committee of Gazi University Medical Faculty. The echocardiography data (January

Address for Correspondence: Dr. Mehmet Kadri Akboğa, Sağlık Bakanlığı Etimesgut Devlet Hastanesi, Kardiyoloji Kliniği, Ankara-*Türkiye*
Phone: +90 544 698 98 21 Fax: +90 312 243 16 42 E-mail: mkakboga@yahoo.com
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2008-March 2013) of the Gazi University Medical Faculty Department of Cardiology were reviewed retrospectively, and patients with RMVS were included in the study. Among 1267 patients with RMVS, after evaluation according to the exclusion criteria, 314 patients remained for further analysis (our database consisted of 35,806 patients for this time period). Fifty-seven age- and gender-matched individuals who had normal echocardiographic findings were randomly selected from the same echocardiography database as the control group. Significant valvular heart disease except mitral valve disease, decompensated heart failure, presence of acute coronary syndrome, previous cardiac surgery, malignancy, renal or hepatic failure, acute or chronic infectious disease, autoimmune disease, anemia (definition of anemia according to the World Health Organization was a hemoglobin level of <12 g/dL in women or <13 g/dL in men), hematologic disease, and acute or chronic pulmonary disease were the exclusion criteria.

Transthoracic echocardiography (Vivid 7 system, 2.5- to 3.5-MHz transducer, GE-Vingmed Ultrasound AS, Horten, Norway) was performed by standard methods. Ejection fraction was measured by modified Simpson's rule. Left atrial diameter (LAD) and left ventricular end-diastolic diameter (LVEDD) were measured in the parasternal long axis view by M-mode echocardiography. Mitral valve area was calculated by planimetric method in the parasternal short axis. Mitral stenosis (MS) was defined as follows: severe MS was a valve area of <1 cm^2, moderate MS was a valve area of 1.0-1.49 cm^2, and mild MS was a valve area of ≥1.5 cm^2 (12). According to this classification, the study included 70 patients with mild MS, 131 with moderate MS, and 113 with severe MS.

Laboratory analysis

Basal clinical characteristics and laboratory parameters were reviewed from the patients' files. Laboratory parameters, including complete blood count (CBC), routine biochemistry, and cholesterol panel, taken from all study participants, were recorded. For CBC analysis, an automatic blood counter (A Cell-Dyn 3500, Abbot, IL, USA) was used. In our department, CRP levels were routinely studied after at least 8 hours of fasting. The blood samples were routinely centrifuged, and serum samples were collected. The CRP levels were analyzed with a Beckman Coulter Inc (Image 800, California, USA).

Statistical analysis

Statistical analysis was performed with SPSS 17.0 Statistical Package for Windows (SPSS Inc, Chicago, Illinois, USA). Continuous variables were given as the median±standard deviation, and categorical variables were defined as percentages. Data were tested for normal distribution using the Kolmogorov-Smirnov test. Categorical variables were compared with chi-square test. Student t-test or Mann-Whitney U test was used to compare continuous variables. Mean values were compared by ANOVA, followed by Tukey HSD test among different groups. Pearson's correlation analysis was performed to define the correlation between hs-CRP levels and the (NLR). The receiver operating characteristics curve was used to demonstrate the sensitivity and specificity of NLR and the optimal cut-off value for predicting RMVS. In order to define the relationship between RMVS and possible confounding factors, univariate and multiple logistic regression analysis was performed. A p value of <0.05 was accepted as statistically significant.

Results

Basal characteristics are given in Table 1. Basal laboratory parameters are presented in Table 2. Lymphocyte count was lower in the RMVS group as compared to the control group [1.8 (0.4-4.6) vs. 2.2 (0.8-4.0), p<0.001]. NLR was significantly higher in the RMVS group [2.9 (0.6-13.0) vs. 2.1 (0.7-5.8), p<0.001]. The level of CRP was also higher in the RMVS group [5.99 (0.3-23.7) vs. 2.98 (0.6-6.3), p=0.001]. In the correlation analysis, it was seen that there was a significant positive correlation between NLR and CRP (r=0.43, p<0.001). When we excluded patients with atrial fibrillation, NLR in the RMVS group remained higher compared to NLR in the control group [2.91 (0.6-10.6) vs. 2.11 (0.7-5.8), p=0.001]. When the statistical analysis was performed after the exclusion of patients with hypertension, diabetes mellitus, coronary artery disease, and atrial fibrillation, NLR was still significantly higher in the RMVS group [2.89 (0.7-13.0) vs. 2.11 (1.1-5.0), p=0.003].

The NLR ratios were increasing in parallel to the severity of mitral stenosis; however, the difference did not reach statistical

Table 1. Basal characteristics of groups

Parameters	Control n=57	RMVS n=314	†P
Age, years	50.2±14.2	49.2±13.0	0.60¥
Female, n (%)	46 (80.7)	235 (74.8)	0.34
Hypertension, n (%)	25 (43.8)	123 (39.1)	0.59
Diabetes mellitus, n (%)	9 (15.7)	43 (13.7)	0.74
Smoking, n (%)	12 (21.6)	43 (13.7)	0.15
CAD, n (%)	11 (19.2)	49 (15.5)	0.46
RAS blocker, n (%)	24 (42.1)	99 (31.6)	0.15
Diuretic, n (%)	10 (17.5)	90 (28.6)	0.12
CCB, n (%)	8 (14.0)	61 (19.5)	0.36
β-blocker, n (%)	14 (24.5)	112 (35.6)	0.11
Statin, n (%)	14 (24.5)	54 (17.3)	0.23
Aspirin, n (%)	18 (31.5)	112 (35.6)	0.51
OAD, n (%)	7 (12.2)	40 (12.8)	0.88
Warfarin, n (%)	0 (0)	101 (32.2)	-
LVEF, %	65.1±2.0	64.5±5.2	0.74
LAD, cm	33.9 (26-41)	42.9 (30-62)	<0.001#
LVEDD, cm	46.3 (40-54)	46.7 (34-59)	0.34#

Data are given as mean±SD or %. CAD - coronary artery disease; CCB - calcium channel blocker; LAD - left atrial diameter; LVEDD - left ventricular end-diastolic diameter; LVEF - left ventricular ejection fraction; OAD - oral antidiabetic drug; RAS - renin-angiotensin system; RMVS - rheumatic mitral valve stenosis. †Chi-square test, ¥Student t-test, #Mann-Whitney U test

Table 2. Laboratory parameters of the study groups

Parameters	Control n=57	RMVS n=314	†P
Hemoglobin, g/dL	13.6 (12.0-16.5)	13.4 (12.0-16.5)	0.38#
Platelets, 10^3/mm^3	246.7 (136-487)	233.4 (110-503)	0.20#
Mean platelet volume, fL	8.2±2.0	8.7±1.4	0.117
White blood cell, 10^3/mm^3	7.5 (3.1-11.4)	7.6 (2.5-12.9)	0.64#
Neutrophils, 10^3/mm^3	4.4 (1.4-7.7)	4.7 (0.8-10.3)	0.49#
Lymphocytes, 10^3/mm^3	2.2 (0.8-4.0)	1.8 (0.4-4.6)	<0.001#
Monocytes, 10^3/mm^3	0.5±0.2	0.6±0.2	0.86
NLR	2.1 (0.7-5.8)	2.9 (0.6-13.0)	<0.001#
Creatinine, mg/dL	0.77±0.2	0.79±0.2	0.29
CRP, mg/dL	2.98 (0.6-6.3)	5.99 (0.3-23.7)	0.001#
Total cholesterol, mg/dL	196.2±38.2	188.0±36.1	0.16
HDL-cholesterol, mg/dL	47.9±11.3	45.4±10.5	0.12
LDL-cholesterol, mg/dL	122.9±32.8	116.3±28.0	0.17
Triglyceride, mg/dL	140.5±80.0	132.0±63.1	0.76

CRP- C-reactive protein; HDL - high-density lipoprotein; LDL - low-density lipoprotein; NLR - neutrophil-to- lymphocyte ratio; RMVS - rheumatic mitral valve stenosis.
†Student t-test, #Mann-Whitney U test

Table 3. Multiple logistic regression analysis of rheumatic mitral valve stenosis

Variable	†P	Odds ratio (95% CI)
Mean platelet volume	0.29	0.81 (0.55-1.20)
Lymphocytes	0.69	1.21 (0.47-3.12)
Neutrophil-to-lymphocyte ratio	0.04	2.24 (1.03-4.86)
CRP	0.03	1.34 (1.02-1.76)
LAD	0.001	1.21 (1.08-1.36)

CRP - C-reactive protein; LAD - left atrial diameter.
†Multiple logistic regression analysis

significance. On the other hand, in the comparison of mitral stenosis groups with the control group, NLR in all mitral stenosis groups was significantly higher than in the control group [2.66 (0.8-8.2), 2.92 (0.6-13.0), and 3.01 (0.7-10.6) vs. 2.10 (0.7-5.8), respectively, p=0.002] (Fig. 1).

In the univariate logistic regression analysis, mean platelet volume, lymphocyte count, LAD, NLR, and CRP were possible independent predictors of RMVS. In the multiple logistic regression analysis, LAD, NLR, and CRP remained independent predictors of RMVS (Table 3).

In the ROC curve analysis, an NLR level cutoff point of more than 2.3 predicted the presence of RMVS with a sensitivity of 60.8% and specificity of 77.2% (Fig. 2).

Discussion

In this study, we showed that NLR was significantly higher in patients with RMVS. Besides, we also found that CRP levels of

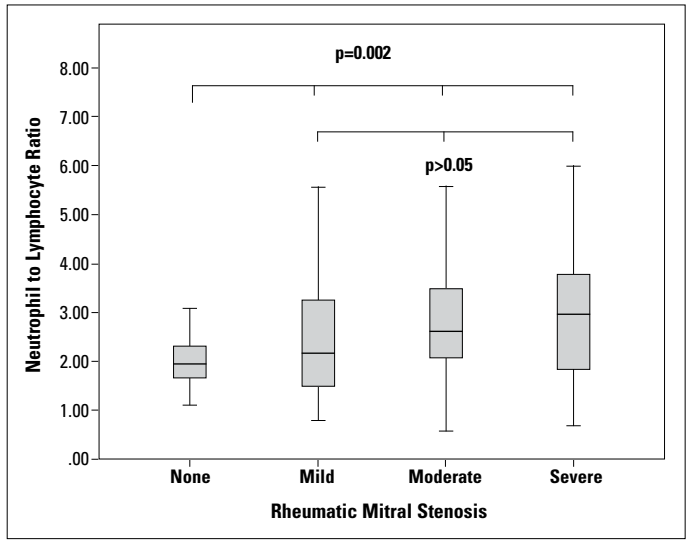

Figure 1. Neutrophil-to-lymphocyte ratio according to presence and severity of rheumatic mitral valve stenosis
†ANOVA followed by Tukey test

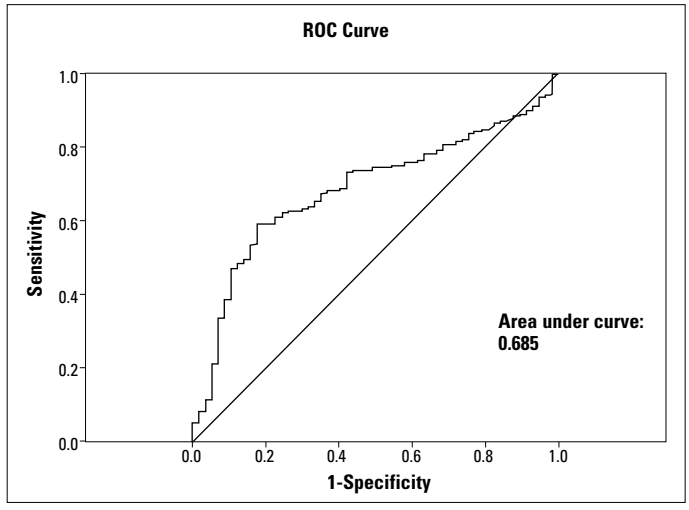

Figure 2. ROC curve analysis for neutrophil-to-lymphocyte ratio as a predictor of rheumatic mitral valve stenosis
†ROC analysis

patients with RMVS were higher than in the control group and that LAD was increased in patients with RMVS. Furthermore, NLR, CRP, and LAD were independent predictors of RMVS.

RMVS is one of the most serious complications of acute rheumatic fever, occurring in approximately 30% of patients (13). The pathogenic mechanism involved in this disease is believed to be an autoimmune process due to "antigen mimicry" (14). This pathophysiological mechanism is mainly based on antigenic similarity that is caused by an inappropriate cross-reaction between heart valves and M protein of group A streptococci. This reaction eventually leads to valvular damage in susceptible individuals (14). Since auto-reactivity has been thought to activate the complement system and starts inflammatory reactions, inflammation takes a crucial role in this cross-reaction (15). CRP is a well-known inflammation marker. In their study, Gölbaşı et al. (4) showed that the CRP level in chronic rheumatic valve

disease (CRVD) was significantly higher than in the control group. They concluded that this could be a sign of ongoing inflammation. In another study, it was found that CRP was also related with severity of CRVD (16). Chiu-Braga et al. (17) investigated advanced oxidation protein products (AOPPs) and CRP levels (as markers of inflammation) in patients with CRVD. They found that AOPPs and CRP levels were significantly higher in patients with CRVD as compared to the control group. According to their results, they concluded that oxidative stress and systemic inflammation are involved in the pathophysiology of CRVD (17).

In recent studies, NLR was shown to be an indicator of systemic inflammation (18-20). It has also been shown that NLR is significantly elevated in many cardiovascular diseases and is related with a poor prognosis (21-23). Turak et al. (24) showed that admission NLR was an independent predictor of poor prognosis in patients with infective endocarditis. According to these data, we thought that there might be a relationship between NLR and the presence of RMVS. According to our results, NLR was significantly higher in patients with RMVS. The leading cause of increased NLR was a decreased lymphocyte counts in the RMVS group. The main cause of lymphopenia was probably the decreased production of lymphocytes as a result of increased steroid levels due to RMVS-induced stress conditions (25, 26). The other probable cause may be the increased apoptosis of lymphocytes triggered by the increased inflammatory status in RMVS (25, 26).

When we subdivided the RMVS patients into mild, moderate, and severe MS groups, we found that NLR levels in each of three groups were higher than in the control group. Furthermore, in the regression analysis, NLR was an independent predictor of RMVS. The increased CRP levels and the close correlation between NLR and CRP levels strengthen our hypothesis. Our results are also in accordance with previous literature (4, 7, 16). These findings suggest that NLR can be used as a predictor for the presence of RMVS. Although the sensitivity and specificity levels were low, NLR still remained an independent predictor of RMVS in the logistic regression analysis. Thus, we thought that although NLR was significantly related with RMVS, there might be possible confounders other than NLR. Further studies are needed to understand this relationship. According to these data, it can be suggested that NLR can not be used as a single marker to rule out RMVS; however, it might be used to predict the presence of RMVS, since NLR is a cheap, fast, and routinely used test in daily clinical practice.

Study limitations

There are some limitations of our study. First, this is a retrospective study. Second, we used a spot NLR value for our analysis, rather than follow-up values. Third, we did not perform an analysis of the prognostic value of NLR in RMVS. Fourth, we also could not study other inflammatory markers. The absence of diastolic parameters can also be a restriction of our study, because these parameters are not studied in detail routinely.

Conclusion

We found that NLR was significantly increased in RMVS and that NLR was an independent predictor of the presence of RMVS in our study population. According to these findings, NLR can be used as a marker of RMVS. Increased NLR can also be a sign of ongoing chronic inflammation in patients with RMVS. In order to understand this relationship in detail, further studies are needed.

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