

Could Complete Blood Count Sub-Parameters to Albumin Ratios be Biomarkers of Disease Activity in Rheumatoid Arthritis?

Mestan Sahin¹, Mevlut Uzumcu², Meltem Alkan Melikoglu³

¹Ataturk University, School of Medicine, Internal Medicine, Rheumatology, Erzurum, Türkiye

²Ataturk University, School of Medicine, Physical Medicine and Rehabilitation, Erzurum, Türkiye

³Ataturk University, School of Medicine, Physical Medicine and Rehabilitation, Rheumatology, Erzurum, Türkiye

Abstract

Introduction: Our aim was to explore the potential of monocyte to albumin ratio (MAR), neutrophil to albumin ratio (NAR), platelet to albumin ratio (PAR), C-reactive protein to albumin ratio (CAR), neutrophil/lymphocyte ratio (NLR), monocyte/lymphocyte ratio (MLR), platelet/lymphocyte ratio (PLR) and platelet/monocyte ratio (PMR) as biomarkers of disease activity in rheumatoid arthritis (RA).

Material and Methods: Cases with RA and healthy controls were included in this cross-sectional study. Demographic features, disease duration current medications, RA disease activity scores (DAS28-ESR, DAS28-CRP, Simplified Disease Activity Index; SDAI, Clinical Disease Activity Index; CDAI) were recorded. Complete blood count (CBC), erythrocyte sedimentation rate (ESR), C-reactive protein (CRP) and serum albumin levels were obtained. MAR, NAR, PAR, CAR, NLR, MLR, PLR, PMR were calculated. These parameters were compared between patient and controls, correlations between these parameters and disease activity and their discriminatory ability were analyzed.

Results: Fifty-four RA patients and 57 controls were included. Significantly higher levels of CAR, MAR, NAR, and MLR were observed in patients than controls. There was a significant difference between active and inactive patients in terms of ESR, CRP and CAR according to DAS28-ESR while no difference was detected between other parameters. CAR and NAR exhibited notable correlations with clinical disease activity scores.

Conclusion: Higher CAR, MAR, NAR, and MLR in patients with RA, and their correlations with disease activity were determined. By demonstrating their discriminatory potential, they could serve as biomarkers to complement clinical assessments in RA.

Key words: Rheumatoid arthritis; disease activity; biomarker; monocyte to albumin ratio; neutrophil to albumin ratio; platelet to albumin ratio.

Introduction

Rheumatoid arthritis (RA) is a chronic inflammatory condition mainly affects the joints and can also involve other body systems (1). Accurately assessing disease activity is crucial for effective management, with guidelines recommending various common parameters such as RA Disease Activity Score-Erythrocyte sedimentation rate (DAS28-ESR), RA Disease Activity Score-CRP (DAS28-CRP), and RA Simplified Disease Activity Index (SDAI) (2). Alongside the count of tender and swollen joints, these assessments commonly incorporate markers like erythrocyte sedimentation rate (ESR) or C-reactive protein (CRP) (3, 4). However, these evaluations may be influenced by factors such as infections, oncological conditions, ischemia, and trauma. Hence, there is a need for biomarkers indicate disease activity in RA. Biomarkers derived

from routine laboratory tests that reflect RA disease activity can be highly valuable in clinical practice. In last years, there has been a growing focus on using albumin levels and ratios of complete blood count (CBC) sub-parameters as a practical and cost-effective method for estimating inflammation. Ratios such as Neutrophil/albumin ratio (NAR), Monocyte/albumin ratio (MAR), and Platelet/albumin ratio (PAR) have been investigated in various conditions including infections, multiple sclerosis, IgA nephropathy, and certain malignancies, showing promise in predicting prognosis (5-8). Furthermore, these ratios have been investigated as potential indicators of disease activity in autoimmune and rheumatologic conditions (9-14). However, there have been limited conflicting data about these parameters in cases with RA (5, 9). The aim of this investigation was to evaluate the potential of

*Corresponding Author: : Meltem Alkan Melikoglu Ataturk University, School of Medicine, Physical Medicine and Rehabilitation, Rheumatology, Erzurum, Turkey E-mail: mamelikoglu@gmail.com

Orcid: Meltem Alkan Melikoglu [0000-0001-7519-9470](https://orcid.org/0000-0001-7519-9470) , Mestan Sahin [0000-0002-4575-4426](https://orcid.org/0000-0002-4575-4426), Mevlut Uzumcu [0009-0002-5514-2543](https://orcid.org/0009-0002-5514-2543)

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various ratios as biomarkers for disease activity in RA, including the MAR, NAR, PAR, C-reactive protein to albumin ratio (CAR), neutrophil to lymphocyte ratio (NLR), monocyte to lymphocyte ratio (MLR), platelet to lymphocyte ratio (PLR), and platelet to monocyte ratio (PMR).

Materials and Methods

Study design and participants: This cross-sectional, descriptive study was carried out at the Rheumatology Clinic of Ataturk University School of Medicine from April to September 2023, in compliance with the Declaration of Helsinki. Its protocol was approved by the local Ethics Committee (Approval Number: B.30.2.ATA.0.01.00/184, date: 30.03.2023), and all participants provided informed consent. The sample size was calculated to achieve 90% statistical power and 95% confidence, resulting in the inclusion of 45 healthy controls and 45 consecutive cases with RA. Our inclusion criteria consisted of consecutive individuals aged ≥ 18 years who met the RA classification criteria based on the 2012 ACR/EULAR criteria (15). The control group included healthy participants, demographically similar to the RA group, with no known diseases. Exclusion criteria included extra-articular involvement, pregnancy or breastfeeding, concurrent inflammatory or autoimmune disorders, infectious diseases, hematologic disorders, or malignancies.

Evaluations: Demographic characteristics (age, gender) of the participants and disease duration and medications of the cases were recorded. Pain was measured by visual analogue scale (VAS), and Patient's Global Assessment (PtGA) and Physician's Global Assessment (GhGA) were noted. Disease activity in cases with RA was scored by assessing four indices: DAS28-ESR, DAS28-CRP, Simplified Disease Activity Index (SDAI), and RA Clinical Disease Activity Index (CDAI). The DAS28-CRP index incorporates the number of tender and swollen joints in 28 joint regions, PtGA, PhGA, and CRP levels, whereas the DAS28-ESR index uses the same components but with ESR instead of CRP (15). According to the EULAR definition (4), individuals with a DAS28 score of ≤ 2.6 were considered to be in remission, those with a DAS28 > 2.6 and ≤ 3.2 were classified as having low disease activity, DAS28 > 3.2 and ≤ 5.1 indicated moderate disease activity, and DAS28 > 5.1 represented high disease activity (4, 16, 17). For the SDAI, scores of ≤ 3.3 signified remission, ≤ 11 indicated low disease activity, ≤ 26 represented moderate disease activity, and > 26 corresponded to high

disease activity. Similarly, for the CDAI, scores of ≤ 2.8 denoted remission, ≤ 10 indicated low disease activity, ≤ 22 represented moderate disease activity, and > 22 corresponded to high disease activity (8). Based on these thresholds, patients were categorized as inactive (remission + low disease activity) or active (moderate + high disease activity). Laboratory evaluations included measurements of CBC, albumin (g/L), CRP (mg/L), and ESR (mm/h). These parameters were assessed by using routine laboratory methodology. CBC sub-parameters were proportioned to each other and to albumin. The calculated ratios included NAR, MAR, PAR, and CAR, along with NLR, MLR, PLR, and PMR.

Statistical analyses: Statistical analyses were conducted using SPSS software, version 22.0 for Windows (SPSS Inc., Chicago, IL, USA). The Shapiro-Wilk test assessed data normality. Descriptive statistics summarized demographic and clinical variables. Categorical variables were examined by the chi-square test, and continuous variables were analyzed with the Mann-Whitney U test or Student's t-test, depending on data distribution. Spearman's correlation analysis calculated the rho coefficient and significance, categorizing correlations as weak ($r = 0.1-0.3$), moderate ($r = 0.3-0.5$), or strong ($r = 0.5-1.0$). Receiver operating characteristic (ROC) curves identified cut-off points for MAR, NAR, PAR, CAR, NLR, MLR, PLR, and PMR. Statistical significance was defined as $p < 0.05$.

Results

Fifty-four cases with RA (43 females and 11 males) and 57 demographically similar healthy controls (40 females and 17 males) were included in our study. Demographic features, disease duration, medications used, disease activity scores and laboratory parameters of the participants were presented in Table 1. There were statistically significant differences in mean \pm standard deviation (mean \pm SD) values of ESR, CRP, albumin, MAR, NAR, CAR and MLR between cases with RA and controls ($p=0.001$, 0.004 , <0.001 , 0.006 , 0.004 , 0.001 , 0.032 , respectively). However, there was no significant difference in PAR, NLR, PLR and PMR between patient and control groups ($p=0.052$, 0.147 , 0.693 , 0.110 , respectively). In the classification of RA disease activity, it was considered as active (moderate+ high disease activity) in 36 cases with RA (66.6%) and inactive (remission+ low disease activity) in 18 cases with RA (34%) according to DAS28-ESR, it was recorded as active in 39 cases with RA (72%) and

Table 1: Demographic, clinical and laboratory characteristics of the participants.

	Patients with RA n=54	Controls n=57	<i>P</i>
Age (years)			
mean \pm SD	54.74 \pm 12.60	47.09 \pm 13.22	0.137
min/max	29/77	21/73	
Gender n(%)			
Male	11(20.4)	17(29.8)	0.252
Female	43(79.6)	40(70.2)	
Disease duration (years)			
mean \pm SD	12.35 \pm 9.89		
min/max	1/44		
Medications; n (%)			
DMARD	50(93)		
cDMARD (mono or combined)	32(59)		
bDMARD veya sDMARD \pm kDMARD	18(34)		
DMARD+steroid	4(7)		
DAS28ESH score Mean \pm SD	2.80 \pm 1.23		
DAS28CRP score Mean \pm SD	2.64 \pm 0.96		
CDAI score Mean \pm SD	7.85 \pm 6.41		
SDAI score Mean \pm SD	8.84 \pm 6.47		
ESR, mm/h Mean \pm SD	20.98 \pm 21.99	10.23 \pm 8.69	0.001
CRP (mg/L) Mean \pm SD	10.88 \pm 14.87	4.31 \pm 3.40	0.004
Albumin (g/L) (mean \pm SD)	3.98 \pm 0.42	4.29 \pm 0.26	0.001
MAR (mean \pm SD)	153.85 \pm 62.49	122.33 \pm 36.69	0.006
NAR (mean \pm SD)	1170.47 \pm 399.52	994.82 \pm 369.15	0.004
PAR (Mean \pm SD)	73054.10 \pm 21151.11	65322.63 \pm 16637.76	0.052
CAR (mean \pm SD)	2.77 \pm 4.00	1.02 \pm 0.84	0.001
NLR (mean \pm SD)	2.36 \pm 1.21	1.99 \pm 0.80	0.147
MLR (mean \pm SD)	0.29 \pm 0.14	0.24 \pm 0.07	0.032
PLR (mean \pm SD)	148.14 \pm 72.34	131.74 \pm 40.03	0.693
PMR (mean \pm SD)	559.07 \pm 331.49	574.70 \pm 208.31	0.110

cDMARD: conventional disease-modifying antirheumatic drug, **bDMARD:** biological disease-modifying antirheumatic drug, **sDMARD:** synthetic disease-modifying antirheumatic drug, **ESR:** Erythrocyte Sedimentation Rate **CRP:** C-reactive protein, **DAS28ESH:** Disease Activity Score-Erythrocyte Sedimentation Rate, **DAS28CRP:** Disease Activity Score-CRP, **CDAI:** RA Clinical Disease Activity Index, **SDAI:** RA Simplified Disease Activity Index, **NAR:** Neutrophil/albumin ratio, **MAR:** monocyte/albumin ratio, **PAR:** platelet/albumin ratio, **CAR:** CRP/albumin ratio, **NLR:** neutrophil/lymphocyte ratio, **MLR:** monocyte/lymphocyte ratio, **PLR:** platelet/lymphocyte ratio, **PMR:** platelet/monocyte ratio

inactive in 15 cases with RA (28%) according to DAS28-CRP. SDAI predicted active disease in 39 cases with RA (72%) and inactive disease in 15 cases with RA (28%), while CDAI predicted active disease in 40 cases with RA (74%) and inactive disease in 14 cases with RA (26%). Statistically

significant differences were determined in ESR, CRP, and CAR ($p=0.004$, 0.018 , 0.014 , respectively) between active and inactive RA cases classified using DAS28-ESH. However, in DAS28-CRP, SDAI and CDAI, none of the parameters were found to be significantly different

Table 2: The comparisons of the laboratory markers between patients with inactive or active disease

	DAS28-ESH		DAS28-CRP		SDAI		CDAI	
	Patients with inactive disease (remission+LDA)	Patients with active disease (MDA+HDA)	Patients with inactive disease (remission+LDA)	Patients with active disease (MDA+HDA)	Patients with inactive disease (remission+LDA)	Patients with active disease (MDA+HDA)	Patients with inactive disease (remission+LDA)	Patients with active disease (MDA+HDA)
n	38	16	39	15	39	15	40	14
White blood cell	7440±1734.0	8193±2054.5	7562.3±1722.9	4447.6±1375.5	7603.5±1740.3	7894.0±2151.9	7621.0±1721.4	7865.0±2230.1
Neutrophile	4382.3±1383.6	5139.3±1642.0	4447.6±1375.5	5020.0±1737.9	4500.7±1444.7	4882.0±1622.3	4503.2±1426.1	4902.1±1681.65
Lenfocyte	2225.5±729.4	2138.7±770.3	2220.2±726.7	2146.6±781.1	2211.2±736.1	2170.0±758.9	2221.0±729.2	2139.2±777.8
Monocyte	612.3±214.8	573.7±231.4	617.9±215.6	556.6±226.7	615.3±218.3	563.3±221.3	621.7±219.2	541.4±212.1
Platelet	291105.2±63228.1	278937.5±63228.1	290538.4±77653.0	279600.0±65398.5	288923.0±74006.8	283800.0±76486.4	289075.0±73058.2	283000.0±79308.5
ESH★	15.3±12.3	34.4±32.5	16.9±13.6	31.6±34.0	17.1±13.5	31.0±34.4	17.4±13.5	31.0±35.7
CRP✱	8.8±11.7	15.6±20.1	8.9±11.5	15.8±20.9	9.0±11.5	15.5±21.1	9.2±11.4	15.5±21.9
Albumin	4.0±0.4	3.8±0.4	4.0±0.4	3.8±0.4	4.01±0.42	3.91±0.42	3.99±0.44	3.96±0.39
CAR⊕	2.2±3.1	4.1±5.3	2.2±3.1	4.1±5.6	2.26±3.10	4.10±5.66	2.32±3.08	4.06±5.87
NAR	1102.4±388.5	1331.9±389.8	1123.6±387.8	1292.1±417.2	1140.1±405.6	1249.3±385.2	1147.5±403.1	1235.9±396.0
LAR	558.3±185.9	555.1±200.0	559.3±185.4	552.5±202.0	557.8±187.0	556.1±198.2	564.2±188.9	537.8±192.1
MAR	154.1±59.9	153.1±70.2	156.1±60.2	147.7±69.8	155.7±60.7	148.8±68.9	158.6±62.6	140.0±62.1
PAR	72961.9±22231.0	73272.9±19014.8	73089.5±21887.5	72962.0±19831.4	72939.5±21612.3	73352.0±20631.4	73420.7±21549.4	72006.6±20716.2
NLR	2.2±1.0	2.7±1.4	2.2±1.0	2.67±1.5	2.31±1.20	2.51±1.28	2.29±1.18	2.56±1.31
MLR	0.3±0.14	0.3±0.17	0.3±0.14	0.3±0.18	0.30±0.14	0.30±0.18	0.30±0.14	0.29±0.18
PLR	146.5±71.2	152.0±77.2	146.4±70.4	152.5±79.4	147.2±72.3	150.5±74.7	146.3±71.6	153.1±76.8
PMR	524.4±205.9	641.4±522.8	519.6±206.1	661.5±533.9	523.2±217.6	652.1±524.0	518.6±216.7	674.4±536.3

★ $p=0.004$ ✱ $p=0.018$ ⊕ $p=0.014$

ESR: Erythrocyte Sedimentation Rate **CRP:** C-reactive protein, **PLT:** platelets, **DAS28ESH:** Disease Activity Score-Erythrocyte Sedimentation Rate, **DAS28CRP:** Disease activity score-CRP, **CDAI:** RA Clinical Disease Activity Index, **SDAI:** RA Simplified Disease Activity Index, **LDA:** low disease activity, **MDA:** moderate disease activity, **HDA:** high disease activity **NAR:** Neutrophil/albumin ratio, **MAR:** monocyte/albumin ratio, **PAR:** platelet/albumin ratio, **CAR:** CRP/albumin ratio, **NLR:** neutrophil/lymphocyte ratio, **MLR:** monocyte/lymphocyte ratio, **PLR:** platelet/lymphocyte ratio, **PMR:** platelet/monocyte ratio

between active and inactive disease (Table 2). In the analyzing of the correlations between disease activity scores and laboratory parameters, there were strong positive correlations between DAS28-ESH scores and ESR, CRP, and CAR (for ESR; $r=0.708$, $p<0.001$, for CRP; $r=0.400$, $p=0.003$, for CAR; $r=0.405$, $p=0.002$). However, there was no significant correlation between other parameters and DAS28-ESH scores. Similarly, positive correlations were found between DAS28-CRP and neutrophil count, ESR, CRP, CAR, NAR, and NLR, but no correlation was found with other parameters (for neutrophil; $r=0.282$, $p=0.039$, for ESR; $r=0.359$, $p=0.008$, for CRP; $r=0.455$, $p=0.001$, for CAR; $r=0.459$, $p=0.001$, for NAR; $r=0.459$, $p<0.001$, for NLR; $r=0.288$, $p=0.035$, $r=0.321$, $p=0.018$). There were weak positive correlations between SDAI scores and ESR, CRP, CAR, and NLR, however no correlation was found with other parameters ($r=0.318$, $p=0.019$ for ESR, $r=0.326$, $p=0.016$ for CRP, $r=0.327$, $p=0.016$ for CAR, $r=0.280$, $p=0.040$ for NLR) No correlation was found between CDAI scores and any parameters. On the other hand, we analyzed the discriminative ability of monocyte count, MAR, NAR, PAR, CAR, MLR between cases with RA and healthy controls. AUC for monocyte was 0.620 (95% CI: 0.514-0.726), AUC for CAR was 0.681 (95% CI: 0.580-0.781), AUC for NAR was 0.657 (95% CI: 0.554-0.759), AUC for MAR was 0.653 (95% CI: 0.550-0.756), and AUC for MLR was 0.618 (95% CI: 0.513-0.723) (Figure 1).

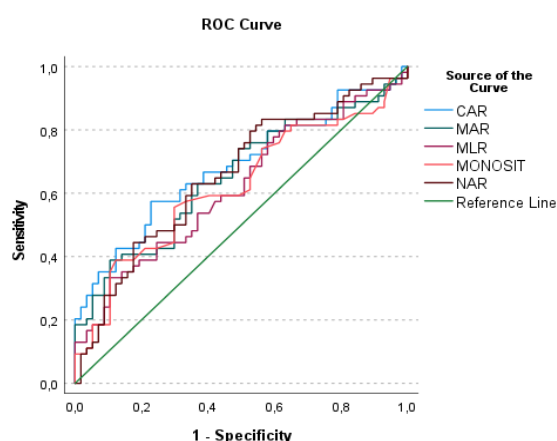


Figure 1: Area under curve of the parameters

Also cut-off points, sensitivity and specificity values were calculated for the variables. The cut-off point for monocytes was 555.00/ μ L

(sensitivity: 59.3%, specificity: 59.6%), for CAR 0.9496 (sensitivity 63.0%, specificity 63.2%), for NAR 1030.58 (sensitivity 63.0% specificity 63.2%), cut-off for MAR was 131.10 (sensitivity 63.0% specificity 63.2%), and cut-off for MLR was 0.2494 (sensitivity 57.4% specificity 57.9%).

Discussion

In the present investigation evaluating the potential of MAR, NAR, PAR, CAR, NLR, MLR, PLR, and PMR as biomarkers for disease activity in cases with RA significantly higher values of MAR, NAR, and CAR were found in cases with RA compared to controls. Notably, among the disease activity scales, ESR, CRP, and CAR values presented significant differences between RA cases with active and inactive disease activity according to DAS28-ESH criteria. Furthermore, there was a strong positive correlation between DAS28-ESH and ESR, CRP, and CAR. Weak to moderate positive correlations were also noted between DAS28-CRP and neutrophil count, ESR, CRP, CAR, NAR, and NLR, as well as between SDAI and ESR, CRP, CAR, and NLR. However, no correlations were found between CDAI and any parameter. Consistent with expectations, ESR and CRP values were higher in cases with RA than controls, and they exhibited positive correlations with scores of disease activity including DAS28-ESH, DAS28-CRP, and SDAI. Conversely, albumin, the negative acute phase reactant, was significantly lower in cases with RA. The MAR, NAR, and CAR were found to be higher in cases with RA than controls in our study. In terms of MAR and NAR, we could not find a previous study evaluating these parameters in cases with RA. However, MAR and NAR were found to be associated with disease activity indicators in cases with Spondyloarthritis (SpA) (14). Numerous studies in the literature investigated the possible association between CAR and disease activity in cases with RA. Sunar et al. showed the presence of a weak correlation between DAS28-ESR and CAR in a study conducted on 121 cases (19). Similarly, another study found a moderate positive correlation between DAS28-ESR disease activity and CAR in cases with RA (20). In our study, in addition to higher values of CAR in RA patients, CAR was also found to be positively correlated not only with DAS28-ESH but also with DAS28-CRP and SDAI. Being the first study evaluating MAR and NAR in disease activity in cases with RA may contribute to literature. There were conflicting data about different CBC parameters and their ratios in cases with RA and control groups in the previous studies. In a study

comparing female cases with RA and controls, it was found that NLR, PLR and MLR values were higher in the cases with RA than controls and MLR could be used as a diagnostic indicator (21). In another study evaluating these parameters in the diagnostic prediction of osteoporosis and vertebral fractures in postmenopausal women with RA, high NLR, and PLR were determined as associated with osteoporosis; also high baseline NLR, PLR and MLR values were associated with vertebral fractures (22). In another study, PLR was found to be statistically different between RA cases with and without interstitial lung disease, however, no significant difference was found in MLR in this population. In addition, regression analysis in the same study showed a significant positive association between PLR and DAS28 (23). In our present investigation, although there was no significant difference in the PLR, MLR was found to be higher in cases with RA than controls. These parameters were also investigated in a study where disease activity in RA was assessed using ultrasound. In that study it was reported that NLR and PLR were positively correlated with ultrasound joint findings; high NLR and PLR rates could be used as a poor prognosis factor (24). On the other hand, in a study of 547 RA patients in which DAS28 disease activity and parameter ratios were used; although NLR and PLR were found to be statistically different, it was concluded that they would not be used as markers because they showed a weak correlation (25). In our study higher these parameters were similar between patient and control groups except higher MLR levels in cases with RA. Also no correlation was determined between scores of disease activity and these parameters. Considering together with previous studies using different methodologies, it seems that data inconsistencies regarding MLR, NLR, PLR, PMR require further investigation in cases with RA. The CBC sub-parameters, their ratios, and the CRP-to-albumin ratio emerge as informative, straightforward, practical, and cost-effective biomarkers for assessing disease activity in RA. However, they may be affected by factors such as infections and malignancy (26). If these limitations are kept in mind, they can be simple, practical tools reflecting disease activity in RA. The limited number of cases with active RA is the main limitation of the present investigation. This may be due to the inclusion of not only newly diagnosed patients but also patients receiving active treatment. Therefore, study designs including more newly diagnosed cases with RA

seems to be needed. The fact that this is the first study investigating MAR and NAR as potential biomarkers of disease activity in cases with RA can be considered as the main strengths of this study. In conclusion; MAR, NAR, CAR and MLR were found to be higher in cases with RA than controls and CAR presented a correlation with RA disease activity in our investigation. These parameters with their discriminatory potential, might serve as biomarkers to complement clinical assessments in RA.

Key messages:

- In the present investigation evaluating the potential of MAR, NAR, PAR, CAR, NLR, MLR, PLR, and PMR as biomarkers for disease activity in cases with RA, there were statistically significant higher values of MAR, NAR, and CAR in cases with RA than controls.
- Among the disease activity scales, ESR, CRP, and CAR values presented significant differences between cases with active and inactive disease activity according to DAS28-ESH criteria.
- There was a strong positive correlation was identified between DAS28-ESH and ESR, CRP, and CAR. Weak to moderate positive correlations were also noted between DAS28-CRP and neutrophil count, ESR, CRP, CAR, NAR, and NLR, as well as between SDAI and ESR, CRP, CAR, and NLR. However, no correlations were found between CDAI and any parameter.

Study Limitations: The limited number of cases with active RA is the main limitation of the present investigation. This may be due to the inclusion of not only newly diagnosed patients but also patients receiving active treatment. Therefore, study designs including more newly diagnosed cases with RA seems to be needed.

Conclusion

In conclusion; MAR, NAR, CAR and MLR were found to be higher in cases with RA than controls and CAR presented a correlation with RA disease activity in our investigation. These parameters with their discriminatory potential, might serve as biomarkers to complement clinical assessments in RA.

Author contributions: Concept; MS, MU, design; MS, MU, supervision: MAM, data collection: MU, MS, literature review: MAM, writing; MAM, MS.

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