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Original Research Article

A Study of Neutrophil-Lymphocyte Ratio and Platelet-to-Lymphocyte Ratio and their Impact on Disease Activity in Rheumatoid Arthritis

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Abstract

Background: Rheumatoid arthritis (RA) affects up to 0.5-1% of the adult population worldwide. Inflammation is the clue determinant and primary mechanism resulting in disability and increased mortality in RA patients. Neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) have increasingly become indicators of subclinical systemic inflammation, which can be used to assess disease activity and prognosticate for a variety of infective, inflammatory, neoplastic, and autoimmune conditions, including RA.

Aims and Objectives: To assess the relationship of NLR and PLR with disease activity in RA.

Materials and Methods: A hundred patients of both sex and age 18–75 years diagnosed with RA were studied at the Department of General Medicine from April 2021 to September 2022. All patients were subjected to a detailed history. A thorough clinical examination and investigations were performed, including complete blood count, erythrocyte sedimentation rate, C-reactive protein, RA factor, and DAS score. DAS28 score was calculated and patients were grouped into moderate disease activity (MDA) (3.2 <DAS28 ≤5.1) and high disease activity (HAD) (DAS28 >5.1).

Results: Rheumatoid arthritis was more prevalent in the age groups of 51-60 years and among males. Mean NLR was significantly higher in patients with HDA (4.860±1.083) than in patients with MDA (3.076±2.613). Mean PLR was significantly elevated in patients with HDA (228.52±89.185) compared to MDA (184.90±78.976). The area under the curve (AUC) for predicting the HDA of NLR was 0.627 (p= 0.030), whereas, for PLR, it was 0.669 (p=0.004). Based on the interpretation both NLR and PLR are fair instruments for predicting HDA in patients with rheumatoid arthritis.

Conclusion: NLR and PLR are two emerging inflammatory biomarkers that could be used to evaluate disease activity in active RA patients. Both NLR and PLR values may be potential indices for RA disease-activity assessment.

Keywords: disease activity, biomarkers, rheumatoid arthritis, autoimmune disease, inflammation

Introduction

Rheumatoid arthritis (RA) is a common autoimmune disease characterized by chronic and progressive inflammation of different organs, especially the synovia of the joints leading to joint damage, a shorter life expectancy, and decreased quality of life (QoL). [1]

Inflammation is the clue determinant and primary basic mechanism resulting in disability and increased mortality in RA patients. Therefore, assessment of inflammation in RA with trusted markers is important to detect a long-term outcome. In daily practice, the most commonly used markers for this aim are the erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP). However, both markers have some limitations, such as reflection of short-term inflammatory activity and low discrimination ability with other superimposed inflammatory conditions. [2]

The parameters of a hemogram, particularly those including immune system elements, are important in assessing different diseases and signs. Immune system elements involve the neutrophils, lymphocytes, and platelets that control inflammation while also undergoing changes secondary to inflammation. [3]

Platelet count, mean platelet volume (MPV), and platelet distribution width (PDW) are three useful indices of platelet function reflecting the platelet production rate. Also, neutrophil-lymphocyte ratio (NLR), MPV, and PDW can be determined from routine complete blood counts (CBC) but are usually neglected by clinicians. A relationship between the MPV and PDW with RA has been reported. [4]

NLR is the proportion of absolute neutrophil to lymphocyte counts retrieved from the CBC test. It has become widely agreed that NLR is useful for developing activity in chronic inflammatory diseases like ulcerative colitis (UC) and familial Mediterranean fever. [5] Hence the present study assesses NLR and PLR in RA patients according to the disease activity.

Materials and Methods

A cross-sectional observational Study was performed on 100 patients at the Department of General Medicine, Sri Aurobindo Medical College and Post Graduate Institute Indore, Madhya Pradesh, from April 2021 to September 2022.

Inpatients and Outpatients-both male and female patients having RA, according to the American College of Rheumatology-European League Against Rheumatism 2010 criteria in the age group of 18–75 years, and those who provided written informed consent were included.

Patients with malnutrition, hepatic dysfunction, renal dysfunction or diabetes mellitus, hyperthyroidism, hematologic diseases, dengue fever, septic shock, and COVID-19 infection were excluded.

All patients were subjected to detailed history and thorough clinical examination and investigations, including CBC, ESR, CRP, RA Factor, and DAS scores performed.

Calculating the DAS28 score was calculated by counting the number of swollen joints (out of 28). A number of tender joints (out of 28), taking blood to measure the erythrocyte sedimentation rate (ESR) and asking the patient to make a "global assessment of health" (indicated by marking on a 10-point line between very good and very bad). These results were incorporated into a mathematical formula to produce the overall disease activity score:

$$DAS28 = 0.56\sqrt{(28TJC)} + 0.28\sqrt{(28SJC)} + 0.70 \text{ Ln (ESR)} + 0.014VAS$$

(Here TJC = Tender joint count, SJC = Swollen joint count, Ln = log, VAS = Visual analogue scale)

Disease severity was assessed according to the value of the DAS28 score as moderate disease activity (MDA) (3.2 <DAS28 ≤5.1) and high disease activity (HAD) (DAS28 >5.1).

Statistical analysis

Data were recorded in the Microsoft Excel program, and statistical analysis was performed by the SPSS program for Windows, version 25 (SPSS, Chicago, Illinois). Continuous variables were presented as mean ± SD, and categorical variables were presented as absolute numbers and percentages. Data were checked for normality before statistical analysis. A descriptive analysis was performed to obtain the general characteristic of the study population. Categorical variables were analyzed using the chi-square test or Fisher's exact test. Continuous variables were assessed using ANOVA or independent sample t-test. Pearson correlation (r) was performed to establish the correlation between established markers with NLR and PLR. The ROC curve was formed to compare the sensitivity of the test. P<0.05 was considered statistically significant.

Results

Table 1: Comparing different characteristics of the study population with Disease activity according to DAS-28

Characteristics	Disease activity according to DAS-28				P value
	High disease Activity		Moderate disease Activity		
	Mean	SD	Mean	SD	
AGE	47.42	9.806	47.42	9.11	1.000
Sex (M/F)	13 (26)/ 37 (74)		11 (22)/ 39 (78)		0.640
HB	11.44	1.5995	11.400	1.53	0.883
WBC	8684.2	2999.517	8444.0	2540.59	0.667
Neutrophils	70.30	9.148	67.32	10.137	0.126
Lymphocytes	22.88	7.737	24.34	8.208	0.362
ESR	39.42	14.56	26.34	9.921	<0.001(S)
CRP	33.950	55.77	7.22	11.49	0.001 (S)
RA factor	126.4800	135.15	76.90	153.62	0.090

On comparing different characteristics of the study population with Disease activity according to DAS-28, it was found that erythrocyte sedimentation rate (p<0.001) values were significantly higher in patients with HDA (39.42±14.56) as compared to those with MDA (26.34±9.921). Similarly and C - reactive protein (p=0.001) levels were significantly higher in patients with HDA (33.950±55.77) as compared to those with MDA (7.22±11.49). Other parameters such as age (p=1.000), hemoglobin (p=0.883), the white blood cells count (p=0.667), neutrophil count (p=0.126), lymphocytes count (p=0.362) and RA factors (p=0.090) were similar between both HDA and MDA patients.

Table 2: Association between NLR and Disease activity according to DAS-28

Disease activity according to DAS-28	NLR	PLR	ACR-EULAR
High disease Activity	4.860±1.083	228.52±89.185	7.20±1.604
Moderate disease Activity	3.076±2.613	184.90±78.976	6.08±1.307
P value	0.008	0.011	<0.001

The present study observed a significant association between NLR and Disease activity according to DAS-28. Mean NLR was significantly higher in patients with HDA (4.860±1.083) than in patients with MDA (3.076±2.613). This highlights that

NLR is a significant predictor of disease activity in patients with rheumatoid arthritis. The present study observed a significant association between PLR and Disease activity according to DAS-28. Mean PLR was significantly higher in patients with HDA (228.52±89.185) than in patients with MDA (184.90±78.976). This highlight that mean PLR is a significant predictor of disease activity in patients with rheumatoid arthritis.

The present study observed a significant association between ACR-EULAR and Disease activity according to DAS-28. Mean ACR-EULAR was significantly higher in patients with HDA (7.20±1.307) than in patients with MDA (6.08±78.976). This highlights that ACR-EULAR is a significant predictor of disease activity in patients with rheumatoid arthritis.

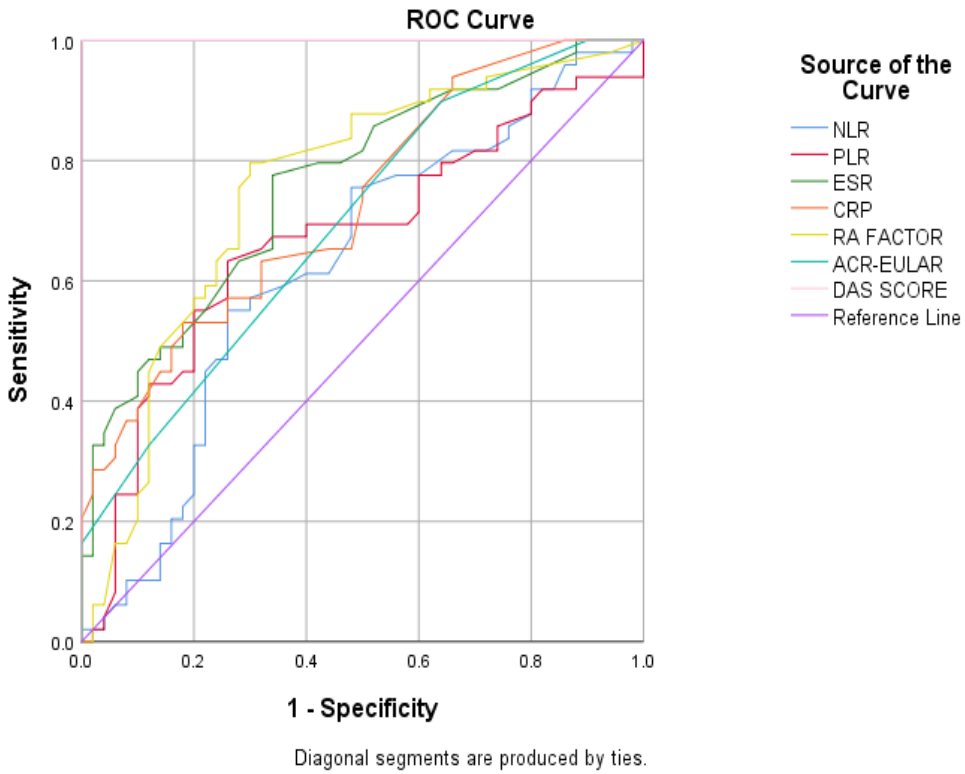

Figure 1: Receptor–operative curve analysis

ROC analysis revealed that the AUC for predicting the higher disease activity of NLR was 0.627 with a p-value of 0.030, whereas AUC for PLR was 0.669 with a p-value of 0.004. Based on the interpretation provided above, it's clear that both NLR and PLR are fair instruments for predicting HDA in patients with rheumatoid arthritis.

Discussion

Rheumatoid arthritis is one of the most common autoimmune diseases characterized by chronic and progressive inflammation of different organs, especially the synovia of the joints leading to joint damage, a shorter life expectancy, and decreased QoL. [6] Over the past decades, white blood count has been used as a traditional inflammatory marker in clinical practice. Inflammation is the clue determinant and primary basic mechanism resulting in disability and increased mortality in RA patients. Therefore, assessment of inflammation in RA with trusted markers is important to detect a long-term outcome. The most commonly used markers for this aim are the ESR and CRP in daily practice. However, both markers have some limitations, such as

reflection of short-term inflammatory activity and low discrimination ability with other superimposed inflammatory conditions. [7]

The inflammatory response can promote the formation of pannus in the joint, which is the major cause of joint damage. [8] Although a deeper understanding of RA has been developed due to more in-depth studies in recent years, assessing the severity of inflammatory activity in patients with RA remains challenging. [9] The current RA evaluation still has some limitations. For example, ESR, CRP, RF, DAS, and the clinical disease activity index are often at cutoff thresholds and are overlooked in patients with low disease activity. However, those patients are at risk of synovial inflammation and progressive joint damage.

In the present study, rheumatoid arthritis was more prevalent in the age groups of 51-60. However, on comparing the Disease activity according to DAS-28 with age distribution, no significant difference was observed, as revealed by the insignificant p-value of 0.518. We found the prevalence of Rheumatoid arthritis higher among females than males. However, no significant difference was observed when comparing the Disease activity according to DAS-28 with gender distribution. A study by Nilsson et al. investigated the influence of sex and age of onset on the disease course and treatment of more than 2000 early RA patients over 8 years. [10] The author found that for both sexes, disease activity, function, and pain improved over time significantly more in men than in women in all age groups. In men, those <40 years displayed significantly lower DAS28 than all other groups. [10] In this study, women older than 55 years had a significantly elevated median DAS28 compared with age-matched men at inclusion. [10] During the eight years of follow-up, there was almost no difference in DAS28 between the age groups in women, whereas men younger than 40 consistently had a lower DAS28 than the other age groups. [10] As previously reported [11], the remaining components of DAS28, ESR, and GH-VAS had a greater impact on DAS28 and might not reflect just joint inflammation. However, ESR in both men and women falls dramatically on the commencement of treatment in all age groups. Arnold et al. found that older-onset patients (≥64 years) start and end the first year after diagnosis worse in terms of DAS28 and HAQ than younger patients. [12]

In our study, it was found that ESR ($p<0.001$) values were significantly higher in patients with HDA (39.42±14.56) as compared to those with MDA (26.34±9.921). Similarly, C - reactive protein ($p=0.001$) levels were significantly higher in patients with HDA (33.950±55.77) as compared to those with MDA (7.22±11.49). ESR and CRP are the most widely used markers for measuring acute phase response due to their reliability, reproducibility, and cost-effectiveness. Our results align with the knowledge that ESR increases with age, particularly in women [10], which could mean that the higher DAS28 in older women was probably attributed to the measures of disease activity rather than to the disease activity per se. Nasir et al. found that the DAS28–ESR and DAS28–CRP positively correlated with the mHAQ. [13] However, none of the patients had an mHAQ score ≤ 0.3, and RA patients with HDA had significantly higher mHAQ scores than those with low or MDA. 13 Our results demonstrate that a composite of DAS28–ESR with an assessment of functional status and quality of life utilizing mHAQ determines disease activity. These findings are comparable to those of a study by Egyptian investigators on 130 patients with RA. 14 Their results also demonstrated a significant correlation of DAS28–ESR/DAS28–CRP with other markers of disease activity. They also compared SDAI and CDAI with DAS28 and noted an apparent correlation between disease

activity indices and the mHAQ, akin to our cohort. [14]

Higher CRP levels are associated with greater RA disease activity based on the core components of the 28-joint Disease Activity Score (DAS28). [6] Individual aspects of disease activity, such as swollen joint count, and patient-reported measures, including functional status (Health Assessment Questionnaire score), morning stiffness, fatigue, and pain, have also been associated with CRP. Indeed, CRP levels are widely used for monitoring RA's systemic inflammation and disease activity. CRP level is a component of several composite disease activity measures: DAS28-CRP, SDAI, American College of Rheumatology (ACR), and European League Against Rheumatism (EULAR) definitions of remission.[15] Yet, the usefulness of CRP testing as a routine measure of RA disease activity is not universal due to the substantial proportion of treated patients who experience flares in their RA but still have normal CRP levels.

The present study observed a significant association between NLR and Disease activity according to DAS-28. Mean NLR was significantly higher in patients with HDA (4.860±1.083) than in patients with MDA (3.076±2.613). This highlights that NLR is a significant predictor of disease activity in patients with rheumatoid arthritis.

NLR is a routinely available marker derived from a CBC's absolute neutrophil and lymphocyte counts. This simple method's utility in assessing systemic inflammation has been clearly demonstrated in various disorders. Our results are in accordance with two recently published reports that showed increased NLR in RA. [16] NLR also correlated well with the disease activity of RA in one of these reports. [16]

We reported a significant association between PLR and Disease activity according to DAS-28 in the present study. The mean PLR was significantly higher in patients with HDA (228.52±89.185) than in patients with MDA (184.90±78.976). This highlight that mean PLR is a significant predictor of disease activity in patients with rheumatoid arthritis. Stenvinkel et al. [17] reported that PLR is better than NLR for the predictive value of end-stage renal disease. There is increasing evidence that activated platelets, especially for patients during chronic inflammation, could be an important part of increased atherogenesis. [18]

Although the mechanism between PLR and RA remains unknown, the mechanism to explain the increased levels of PLR in RA is the presence of a chronic inflammatory state that affects the progressive damage of joints. An earlier trial demonstrated the systemic and chronic inflammation associated with thrombocytosis. Several pro-inflammatory cytokines promote megakaryocyte proliferation, such as IL-1 and IL-6, leading to a higher platelet count in patients with RA. It is noteworthy that thrombocytosis induced by pro-inflammatory cytokines such as IL 6, IL 1, GM-CSF, and G-CSF was frequently observed in patients with malignant tumours. [19] Pro-inflammatory cytokines represent a potential cause for megakaryocyte proliferation in patients with RA, contributing to increases in platelet levels. Further research is necessary to clarify the mechanism associated with PLR in RA patients. Further, based on the present results, PLR may be superior to CRP in estimating chronic subclinical inflammation in patients with RA.

In the present study, the mean ACR-EULAR was significantly higher in patients with HDA (7.20±1.307) compared to patients with MDA (6.08±78.976). According to the 2010 American College of Rheumatology/European League Against Rheumatism (ACR/EULAR) classification criteria, the RA diagnosis also considers rheumatoid factor titers and anti-cyclic citrullinated peptide antibodies (ACPA). [20] Our results found that mean ACR-

EULAR is a significant predictor for disease activity in patients with rheumatoid arthritis.

We found that NLR had a significant positive correlation with RA factors. In contrast, other factors such as ESR, CRP, ACR-EULAR, and DAS scores had no significant correlation with NLR in the present study. However, PLR significantly correlated with ESR, CRP, RA factor, ACR-EULAR, and DAS score. A study by Lijuan et al. showed that the analysis of the correlations of the NLR with CRP, ESR, and DAS28-ESR, the three most extensively used parameters for RA disease activity assessment, showed that all these indices were weakly positively correlated with the NLR. Moreover, the data demonstrated that the PLR was weakly positively correlated with the abovementioned inflammatory markers. [21]

In the present study, ROC analysis revealed that the AUC for predicting the higher disease activity of NLR was 0.627 with a p-value of 0.030, whereas AUC for PLR was 0.669 with a p-value of 0.004. Based on the ROC curves by Lijuan et al., the NLR (sensitivity 31.8%, specificity 77.8%) and PLR (sensitivity 57.3%, specificity 63.9%) were less valuable than the ESR (sensitivity 67.2%, specificity 91.7%) and CRP (sensitivity 76.2%, specificity 91.7%) for differentiating inactive RA patients from active RA patients due to low sensitivity and specificity and combining NLR or PLR also cannot significantly improve the diagnostic value of ESR and CRP. [21] Based on our findings, NLR and PLR are fair instruments for predicting HDA in patients with rheumatoid arthritis. [22]

There may be some limitations in the present study. The variation of PLR, as a cross-sectional study, was not observed dynamically after undergoing effective treatment for patients with RA. Moreover, the association between PLR and the severity of RA should be discussed to clarify whether PLR is an indicator estimating the severity for patients with RA. Finally, our samples were measured from a single laboratory; because PLR values may be slightly different in the various populations and regions. However, our results suggest that PLR is associated with RA, and PLR, but not NLR and CRP, may be an underlying indicator indicating chronic subclinical inflammation in patients with RA.

Conclusion

ESR and CRP are cheap and readily available markers of inflammation that correlate well with other inflammatory markers and disease activity indices in RA. However, NLR and PLR are two emerging inflammatory biomarkers that could be used to evaluate disease activity in active RA patients. Both NLR and PLR values may be potential indices for RA disease-activity assessment. Hence, NLR and PLR could be helpful tools in evaluating disease activity in RA. However, the possible association between NLR and PLR with the outcome of inflammatory arthritides remains to be investigated in prospective long-term studies. A large-scale longitudinal study is recommended to confirm the present results and further demonstrate the relationship to medications received and disease outcome.

References

1. Kugyelka R, Kohl Z, Olasz K, Mikecz K, Rauch TA, Glant TT. Enigma of IL-17 and Th17 cells in rheumatoid arthritis and in autoimmune animal models of arthritis. Mediators Inflamm. 2016 (2016): 6145810.
2. Hobbs F, Cohen MD. Rheumatoid arthritis disease measurement: a new old idea. Rheumatology 2012; 51 (suppl 6): vi21-7.
3. Scapini P, Cassatella MA. Social networking of human neutrophils within the immune system Blood 20 14;124 (5):710-719.

4. Briggs C. Quality counts new parameters in blood cell counting. Int J Lab Hematol. 2009; 31 (3):277-297.
5. Ahsen A, Ulu MS, Yuksel S, Demir K, Uysal M, Erdogan M et al. As a new inflammatory marker for familial Mediterranean fever: neutrophil-to-lymphocyte ratio. Inflammation. 2013; 36 (6):1357-1362.
6. Cylwik B, Chrostek L, Gindzienska-Sieskiewicz E, Sierakowski S, Szmitkowski M. Relationship between serum acute-phase proteins and high disease activity in patients with rheumatoid arthritis. Adv Med Sci. 201 0; 55: 80–85.
7. Boyraz İ, Koç B, Boyacı A, Tutoğlu A, Sarman H, Özkan H. Ratio of neutrophil/lymphocyte and platelet/lymphocyte in patient with ankylosing spondylitis that are treating with anti-TNF. Int. J. Clin. Exp. Med. 2014; 7: 2912.
8. Hammer HB, Kvien TK, Terslev L. Ultrasound of the hand is sufficient to detect subclinical inflammation in rheumatoid arthritis remission: a post hoc longitudinal study. Arthritis Res. Ther. 2017;19: 1–7.
9. Coutant F, Miossec P. Evolving concepts of the pathogenesis of rheumatoid arthritis with focus on the early and late stages. Curr. Opin. Rheumatol. 2020; 32: 57–63.
10. Nilsson J, Andersson ML, Hafström I, Svensson B, Forslind K, Ajeganova Set al. Influence of age and sex on disease course and treatment in rheumatoid arthritis. Open Access Rheumatol. Res. Rev. 20121; 13: 123.
11. van Onna M, Boonen A. The challenging interplay between rheumatoid arthritis, ageing, and comorbidities. BMC Musculoskelet. Disord.2016; 17: 1–9.
12. Arnold MB, Bykerk VP, Boire G, Haraoui BP, Hitchon C, Thorne C et al. Are there differences between young- and older-onset early inflammatory arthritis, and do these impact outcomes? An analysis from the CATCH cohort. Rheumatology 2014; 53: 1075–1086.
13. Nasir N, Majid H, Khan A, Awan S, Riaz M. Disease activity correlates and functionality in patients with rheumatoid arthritis–real-world experience from a South Asian country. Reumatologia/Rheumatology 2022; 60: 183–191.
14. Eissa M, El Shafey A, Hammad M. Comparison between different disease activity scores in rheumatoid arthritis: an Egyptian multicenter study. Clin. Rheumatol. 2017; 36: 2217–2224.
15. England BR, Tiong BK, Bergman MJ, Curtis JR, Kazi S, Mikuls TR et al. 2019 update of the American College of Rheumatology recommended rheumatoid arthritis disease activity measures. Arthritis Care Res. 2019; 71: 1540–1555.
16. Uslu AU, Küçük A, Şahin A, Ugan Y, Yılmaz R, Güngör T, et al. Two new inflammatory markers associated with Disease Activity Score-28 in patients with rheumatoid arthritis: neutrophil-lymphocyte ratio and platelet-lymphocyte ratio. Int. J. Rheum. Dis. 2015; 18: 731–735.
17. Stenvinkel P. Malnutrition and chronic inflammation as risk factors for cardiovascular disease in chronic renal failure. Blood Purif. 2001; 19: 143–151.
18. Koyama H, Maeno T, Fukumoto S, Shoji Takuhito, Yamane T, Yokoyama H, et al. Platelet P-selectin expression is associated with atherosclerotic wall thickness in the carotid artery in humans. Circulation 2003; 108: 524–529.
19. Sierko E, Wojtukiewicz MZ. Inhibition of platelet function: does it offer a chance of better cancer progression control? Presented at the Seminars in thrombosis and hemostasis, © Thieme Medical Publishers. 2007: 712–721.
20. Kennish L, Labitigan M, Budoff S, Filopoulos MT, McCracken WA, Swearingen CJ, et al. utility of the new rheumatoid arthritis 2010

ACR/EULAR classification criteria in routine clinical care. BMJ Open 2012; 2: e001117.
21. Lijuan W, Yuting Z, Chaoyang L, Ju Y. Neutrophil-lymphocyte, platelet-lymphocyte, and lymphocyte-monocyte ratios may not be useful markers to assess disease activity in rheumatoid arthritis: A STROBE-compliant article. Medicine (Baltimore). 2021; 100.
22. Khaleel H. Morgagni's diaphragmatic hernia with Large ASD Secondum And Sever PHT mimicking a severe congenital heart disease in a newborn and cause delayed diagnosis of Morgagni's diaphragmatic hernia. a case report. Journal of Medical Research and Health Sciences. 2022; 5(3): 1832–1837.