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RESEARCH ARTICLE

Neutrophil: Lymphocyte Ratio and Platelet: Lymphocyte Ratio related to Disease Activity among Rheumatoid Arthritis in Erbil governorate/Iraq

Zekra A. Aziz[1], Dashty A. Al-Bustany[2]
[1]*Department of Prosthetic and Orthotics, Erbil Technical Health and Medical College, Erbil Polytechnic University, Kurdistan Region, Iraq.*
[2]*Department of Medicine, College of Medicine, Hawler Medical University, Kurdistan Region, Iraq.*

***Corresponding author:**
Zekra Ali Aziz
Department of Prosthetic and Orthotics, Erbil Technical Health and Medical College, Erbil Polytechnic University, Kurdistan Region, Iraq.
E-mail:
zekra.aziz@epu.edu.iq

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ABSTRACT

Rheumatoid Arthritis (RA) is a systemic autoimmune inflammatory disorder that causes joints destruction the aim here is to investigate the neutrophil - to - lymphocyte ratio (NLR) and platelet - to- lymphocyte ratio (PLR) in RA patients and relation to Disease Activity Score of 28 joints (DAS28-ESR). The study population included (260) participants which consist of (210) RA patients and (50) healthy persons as a control. RA patients were two groups (107) received conventional diseases modified anti-rheumatic drugs (cDMARD) and (103) received biological agents (bDMARD) plus cDMARD each group subdivided into disease active patients (DAS28-ESR ≥ 2.6) and patients in remission (DAS28-ESR < 2.6). NLR in cDMARD, bDMARD plus cDMARD, and control group was 3.1 (±3.4), 2.2 (±1.3), and 2.4 (±1.3) respectively. PLR in cDMARD, bDMARD plus cDMARD, and control group was 193.3 (±355.3), 115.7 (±42.7), and 115.6 (±43.1) respectively. There was statistically significant difference in NLR and PLR among three groups ($p = 0.023$ and $p = 0.030$) respectively also according to DAS28-ESR in patients with active disease ($p = 0.006$) for both NLR and PLR, while for patients in remission NLR and PLR there were no significant differences detected ($p = 0.901$ and $p = 0.638$ respectively). The result of the current study support the idea that NLR and PLR can be considered as inflammatory parameters for evaluating disease activity in RA patients.

Keywords:

Rheumatoid arthritis, Disease activity, Neutrophil, Platelet, and Lymphocyte.

1. INTRODUCTION

Rheumatoid Arthritis (RA) is a complex inflammatory persistent autoimmune systemic disorder with obscure etiology and many organ involvements characterized by different manifestations. It primarily occurs in the synovial tissues, causes irreversible destruction, and a deformity of joints clinically appears as progressive, symmetric polyarthritis. RA if not treated early and properly it could lead to short life expectancy, decrease quality of life, functional disability, morbidity, and increase mortality, which ends with the societal burden (Mohamad et al.,2021; Kugyelka et al.,2016). Environmental factors and genetic predisposition contribution to the inanition and triggering of RA (Assayag et al., 2014). RA is most common autoimmune disorder in the world. It is more common in women in contrast to men (female: male, 3:1), (Karimifar et al., 2012). Its incidence is about 0.5–1% in the general population (Willemze et al., 2012; Sanmart et al., 2013). Inflammation considers as a key component for the pathogenesis of RA. When RA is in the active stage, inflammatory markers are ordinarily elevated and reflect the disorder recreation (Chandrashekara et al., 2017). Therefore, accurate and appropriate assessments of the RA active disease inflammations rely on markers is critical for customizing the cure method and decide the effectiveness of treatment. The therapeutic aim is lower disease activity or a hold remission, which reduce distraction of the joints, a better pleasant of life, and much less co-morbidity (Hobbs and Cohen, 2012). RA is generally assessed at the time of detection and through the course of the cure and follows up by the Disease Activity Score of 28 joints (DAS-28) system, which is assessed by clinic examination including a number of swollen joints counts (SJC) and a number of tender joints count (TJC), also visual analogue scale (VAS) and laboratory findings such as erythrocyte sedimentation rate (ESR) and/ or C-reactive protein (CRP) (Inoue et al.,2007) which are (CRP) and (ESR) most often used markers for assessment of inflammation in everyday practice. Both of pervious mentioned markers have limitations, like quick time inflammatory pastime beside low recognition capacity with different inflammatory diseases (Colglazier et al., 2005). The systemic inflammatory process is related to the variation in components and blood cells counts. Anemia of normochromic type, neutrophilia, lymphopenia, with thrombocytosis shows up in different inflammatory diseases (Kisacik et al., 2008). For that reason, the characteristics of blood cell composition are used to estimate the inflammatory level. In rheumatology search about the biomarkers come from the need to recognize the underlying mechanisms in

rheumatological disorder. Hemogram parameters especially components of the immune system immune play an important role in the evaluation of different disorders. Among the immune system components lymphocytes, neutrophils, also platelets are regarded as important cells for the management of the inflammatory process (Scapini and Cassatella, 2014). Patients with RA are frequently found with thrombocytosis during active disease stages. While the number becomes less during controlling of inflammation. By that, the degree of thrombocytosis may correlate with the range of active synovitis and maybe correlate with extra-articular features (Elena et al., 2019). As it's recognize in the pathogenesis of RA, neutrophils and, B and T lymphocytes have a key role (Yao et al., 2013; Choi, et al., 2014). Neutrophils have a position in increasing disease activity in RA patients through secreting different substances such as (proteases and intermediates reactive oxygen to the synovial house and lead to activating different cells through secretion of tumour necrosis factor-a (TNF-a), B lymphocyte stimulator (BLyS), interleukin-17 (IL-17), with various extra mediators (Kouri et al., 2014). So, it has been suggested that PLR and NLR is a good index inflammatory marker for the systemic disorder (Scapini and Cassatella, 2014). Recently, many reports have recommended the possible benefit of NLR with PLR, as indicator markers for RA (Fu et al., 2015; Zhang et al., 2016; Du et al., 2017; Zengin et al., 2018) and several rheumatological disorders (Yolbas et al., 2016; Yang et al., 2017; Wu et al., 2016; Balkarli et al., 2016). Many previous researchers have found that NLR with PLR are good biomarkers for systemic inflammation measurement in RA ((Kisacik et al., 2008; Yazici et al., 2013; Muddathir and Haj, 2013). A new meta-analysis showed highly significant increase of NLR and PLR in RA compared to healthy individuals (Erre et al., 2018). In addition, NLR and PLR were consistently prophetic of remission in RA patients in a sustained manner in previous studies (Chandrashekara et al.,2015; Tekeog et al., 2016; Maden et al., 2017; Peng et al.,2015), where positive relations of acute-phase reactant levels with disease activity were found. These results support the suggestion that NLR with PLR may serve as excellent inexpensive markers important for the prognosis of RA. Complete blood counts test is performed in patients with RA at baseline to monitor the side effects of the drug and any changes in the disease that occur. However, there are little data on the relationship between most unstudied chronic inflammatory arthritis and NLRs and PLRs. It is still a concern for RA (Mercan et al., 2016) to discover that new biomarkers play a function in several stages of expansion. As is known that RA is a chronic disorder so correction for illnesses for which there is no recovery requires lifelong treatment with anti-rheumatic drugs (DMARDs) (Park et al., 2018). Today, physicians' aim in the treatment of RA is to eliminate inflammation. This means reducing disease activity that controls symptoms and preventing structural damage (Smolen et al., 2016). Biological drugs have been the mainstay of treatment since the beginning of this century for their established effectiveness in the control of disease activity, although biologics have shown superior results in RA over existing synthetic disease-qualifying anti-rheumatic drugs that initiate this drug. For its cost and impact, it is used in case of if patients with RA do not get an appropriate reaction by methotrexate (MTX) and/or other synthetic DMARDs (Kiely et al., 2012; Bruno et al, 2011). This study was conducted to find out the correlation between DAS28-ESR and the estimated NLR and PLR in RA patients.

2. Patients and Methods:

This case-control study was conducted between September 2019 and March 2021 in Erbil Center of Rheumatology, Rizgary Teaching Hospital. The study population was included (260) participants which consist of (210) patients who have rheumatoid arthritis (RA) and (50) healthy individuals with age and gender matching as a control group. The RA patients were grouped according to the type of treatment into groups A (107) received conventional diseases modified anti-rheumatic drugs cDMARD while group B (103) patients who received biological agents bDMARD plus cDMARD and each group further subdivided into two groups based on Disease Activity Score (DAS28): Patient DAS28 score < 2.6 for the patient in remission and for active disease DAS28 score ≥ 2.6. (Bruno et al., 2018) RA patients are defined according to the diagnostic criteria of the American College of Rheumatology/ European League against Rheumatism (ACR/ EULAR) 2010 (Bruno et al., 2018). Patients who were outpatients of the Department of Rheumatology and Rehabilitation were recruited, and the control group that enrolled was normal subjects. Informed consent was taken from both patient and control groups. The study protocol was accepted by the Kurdistan Board of Medical Specialties Committee of Ethics. Patients with diabetes, coronary artery disease, chronic obstructive pulmonary disease, hypertension, pregnant women, other autoimmune diseases, and patients who received corticosteroid treatment for the past 3 months were excluded. Complete blood cell counts CBC, ESR, CRP, VAS, DAS28 with basal clinical features of the patients and control group were obtained. Complete blood cell counts CBC examined neutrophil counts, lymphocyte counts, and platelet counts. NLR with PLR are studied as the ratio of neutrophil and platelet counts in lymphocyte counts for both patients and controls, respectively, and for the group of RA patients, DAS28 checks 28 joints: (swollen and tender joint number), patient Visual Analog Scale (VAS) (0-100), and the ESR test calculated.

3. Statistical Analysis:

The Social Science Statistical Package (SPSS version 25) was used. Proportions were compared using a chi-square test of association, while, two independent samples Student's t-test (unpaired t-test) was used to compare the means of the two samples. Furthermore, three means were compared using one-way analysis of variance (ANOVA) and by two groups using post hoc tests (LSD). The spearman's rho correlation coefficient

was calculated to evaluate the power of the correlation. A p-value equal to or less than 0.05 was regarded as statistically significant.

4. Results:

This study was composed of two groups of patients and one control group. Group A consisted of cDMARDs (n = 107), group B consisted of bDMARDs and cDMARDs (n = 103), and the third group consisted of 50 subjects without rheumatoid arthritis (control group). As shown in Table 1, the mean ages of the three groups were 50.4, 49.8, and 45.5 years, respectively. Although this table shows that 88.1% of the patients were female, there was no significant difference between the three groups (p=0.892). As shown in Table 1, the mean duration of disease was in the cDMARD group (61 months), and in the bDMARD and cDMARD groups (66 months).

Table 1 Basic characteristics of the sample studied.

	cDMARD	bDMARD plus cDMARD	Control	Total	
	No.(%)	No.(%)	No.(%)	No.(%)	p
Age (years)					
< 40	13(12.1)	18(17.5)	14(28.0)	45(17.3)	
40-49	39(36.4)	29(28.2)	18(36.0)	86(33.1)	
50-59	35(32.7)	31(30.1)	11(22.0)	77(29.6)	
≥ 60	20(18.7)	25(24.3)	7(14.0)	52(20.0)	
Mean(±SD)	50.4(11.2)	49.8(11.5)	45.5(10.7)	49.2(11.3)	0.149#
Gender					
Male	13(12.1)	13(12.6)	5(10.0)	31(11.9)	
Female	94(87.9)	90(87.4)	45(90.0)	229(88.1)	0.892#
Total	107(100.0)	103(100.0)	50(100.0)	260(100.0)	
Duration of disease by months					
Mean(±SD)	66 (±3.2)	61 (±2.9)			0.409†
Duration of by months patients received bDMARD plus cDMARD					
Mean(±SD)		48 (±2.2)			

By Chi square test. †By unpaired t test

As can be seen from Table 2, the mean ESR (31.3 mm/hr) was significantly (p< 0.001) higher in the cDMARD group, than the bDMARD plus cDMARD (25.3 mm/hr), which was also higher than the control group (11.1 mm/hr). The mean CRP follow the same pattern, except for the difference between the bDMARD plus cDMARD group and the control (p = 0.162). The mean WBC count was significantly higher ($8.1*10^3$/µl) in the first two groups compared with $6.8*10^3$/µl in the control group. The same pattern can be observed for the means of neutrophils. Regarding the mean of lymphocytes, it was the highest ($2.5*10^3$/µl) in the bDMARD plus cDMARD compared with $2.1*10^3$/µl in the cDMARD, and $2.0*10^3$/µl in the control also. The platelets (PLT) means of the cDMARD group ($269.8*10^3$/µl) and that of the bDMARD plus cDMARD group ($258.1*10^3$/µl) were significantly higher ($203.7*10^3$/µl) (p < 0.001) than the mean of control group. The means of Hb of the cDMARD and bDMARD plus cDMARD groups (12.5 g/dl) were significantly less than the mean of the control group (13.2 g/dl). Regarding the neutrophil-to-lymphocyte ratio (NLR), the highest mean was in the cDMARD group (3.1) compared to (2.2) in the bDMARD plus cDMARD (p = 0.008), and 2.4 in control (p = 0.086). The highest mean of platelets-to- lymphocyte ratio (PLR) was also highest in the cDMARD group (193.3), compared with 115.7 in the bDMARD plus cDMARD (p = 0.015) and 115.7 in the control (p= 0.050).

Table 2. The studied variables means

		N	Mean	(±SD)	P**	Groups (LSD)	p (LSD)
ESR mm/hr	A) cDMARD	107	31.3	(±26.4)		AXB	0.034
	B) bDMARD plus cDMARD	**103**	**25.3**	**(±18.5)**	**<0.00**	**AX C**	**<0.001**
	C) Control	50	11.1	(±3.8)		B X C	<0.001
	Total	**260**	**25.1**	**(±21.8)**			
CRP mg/l	A) cDMARD	107	7.7	(±15.7)		AXB	0.028
	B) bDMARD plus cDMARD	**103**	**4.4**	**(±7.2)**	**0.004**	**AX C**	**0.002**
	C) Control	50	1.7	(±1.1)		B X C	0.162
	Total	**260**	**5.2**	**(±11.3)**			
WBC* 10^3/µl	A) cDMARD	107	8.1	(±2.8)		AXB	0.963
	B) bDMARD plus cDMARD	**103**	**8.1**	**(±2.6)**	**0.008**	**AX C**	**0.005**
	C) Control	50	6.8	(±1.4)		B X C	0.004
	Total	**260**	**7.8**	**(±2.5)**			
Neutrophil *10^3/µl	A) cDMARD	107	5.3	(±2.3)		AXB	0.131
	B) bDMARD plus cDMARD	**103**	**4.9**	**(±1.9)**	**0.002**	**AX C**	**<0.001**
	C) Control	50	4.1	(±1.3)		B X C	0.021
	Total	**260**	**4.9**	**(±2.0)**			
Lymphocyte *10^3/µl	A) cDMARD	107	2.1	(±0.9)		AXB	0.004
	B) bDMARD plus cDMARD	**103**	**2.5**	**(±1.3)**	**0.002**	**AX C**	**0.449**
	C) Control	50	2.0	(±0.9)		B X C	0.002
	Total	**260**	**2.3**	**(±1.1)**			
PLT*10^3/µl	A) cDMARD	107	269.8	(±103.0)		AXB	0.288
	B) bDMARD plus cDMARD	**103**	**258.1**	**(±65.1)**	**<0.001**	**AX C**	**<0.001**
	C) Control	50	203.7	(±39.8)		B X C	<0.001
	Total	**260**	**252.5**	**(±83.1)**			
Hb g/dl	A) cDMARD	107	12.5	(±1.8)		AXB	0.858
	B) bDMARD plus cDMARD	**103**	**12.5**	**(±1.3)**	**0.018**	**AX C**	**0.011**
	C) Control	50	13.2	(±1.6)		B X C	0.008
	Total	**260**	**12.6**	**(±1.6)**			
NLR	A) cDMARD	107	3.1	(±3.4)		AXB	0.008
	B) bDMARD plus cDMARD	**103**	**2.2**	**(±1.3)**	**0.023**	**AX C**	**0.086**
	C) Control	50	2.4	(±1.3)		B X C	0.670
	Total	**260**	**2.6**	**(±2.4)**			
PLR	A) cDMARD	107	193.3	(±355.3)		AXB	0.015
	B) bDMARD plus cDMARD	**103**	**115.7**	**(±42.7)**	**0.030**	**AX C**	**0.050**
	C) Control	50	115.6	(±43.1)		B X C	0.998
	Total	**260**	**147.6**	**(±232.8)**			

**By ANOVA; NLR, neutrophil-to-lymphocyte ratio; PLR, platelets-to- lymphocyte ratio.

From Table 3, the mean ESR was significantly higher (p< 0.0001) in both the cDMARD disease active group (48.8 mm/hr) and the bDMARD + cDMARD disease active group (37.4 mm/hr), than the cDMARD in remission group (12.8 mm/hr) and bDMARD plus cDMARD in remission group (12.9 mm/hr) respectively. The mean CRP follows the same pattern, except for the difference between the bDMARD plus cDMARD groups (p = 0.424). The mean of lymphocytes was the lower (1.7 *10^3/µl) in the cDMARD active disease compared to (2.4*10^3/µl) in the cDMARD remission group the difference was significant (p <0.0001) while in the bDMARD plus cDMARD groups, the mean of lymphocytes vice versa is slightly higher in disease active (4.9) than remission group (4.7).

The mean of platelet (PLT) of the cDMARD disease active group (297*10^3/µl) was high significant than the mean of the cDMARD remission group (240*10^3/µl) (p= 0.004) same things were found between the groups on bDMARD plus cDMARD but there was no significant difference (p= 0.098).Among patients on cDMARD, the result shows that the higher the disease activity (DAS ≥ 2.6), the higher the means of NLR (p = 0.020) and PLR (p = 0.015) compared with the means among

patients with DAS < 2.6. No significant associations were detected between the activity of the diseases with NLR (p = 0.602) also PLR (p = 0.755) among patients on bDMARD plus cDMARD.

Table 3. Means of the studied variables by disease activity and type of treatment.

	DAS < 2.6		DAS ≥ 2.6		
cDMARD	**Patient no. 52**		**Patient no. 55**		
	Mean	**(±SD)**	**Mean**	**(±SD)**	**P***
ESR mm/hr	**12.8**	**(±7.2)**	**48.8**	**(±25.9)**	**<0.0001**
CRP mg/l	**2.5**	**(±2.6)**	**12.6**	**(±20.6)**	**0.0007**
WBC* 10^3/µl	**8.3**	**(±2.5)**	**7.7**	**(±2.9)**	**0.285**
Neutrophil*10^3/µl	**5.2**	**(±2.0)**	**5.3**	**(±2.6)**	**0.810**
Lymphocyte *10^3/µl	**2.4**	**(±0.8)**	**1.7**	**(±0.8)**	**<0.0001**
PLT*10^3/µl	**240**	**(±60.9)**	**297**	**(±125.3)**	**0.004**
Hb g/dl	**12.9**	**(±1.6)**	**12**	**(±1.8)**	**0.008**
DAS	**2.1**	**(±0.3)**	**4.9**	**(±0.9)**	**<0.0001**
NLR	**2.3**	**(±1.1)**	**3.8**	**(±4.5)**	**0.020**
PLR	**108.7**	**(±40.2)**	**273.3**	**(±482.6)**	**0.015**
bDMARD plus cDMARD	**Patient no. 51**		**Patient no. 52**		
	Mean	**(±SD)**	**Mean**	**(±SD)**	**P***
ESR mm/hr	**12.9**	**(±6.6)**	**37.4**	**(±18.3)**	**<0.0001**
CRP mg/l	**3.7**	**(±9.1)**	**4.9**	**(±4.4)**	**0.424**
WBC* 10^3/µl	**7.6**	**(±2.0)**	**8.5**	**(±3.0)**	**0.132**
Neutrophil*10^3/µl	**4.7**	**(±1.6)**	**4.9**	**(±2.0)**	**0.630**
Lymphocyte *10^3/µl	**2.3**	**(±0.9)**	**2.7**	**(±1.5)**	**0.335**
PLT*10^3/µl	**247.4**	**(±53.1)**	**268.6**	**(±73.9)**	**0.098**
Hb g/dl	**12.6**	**(±1.2)**	**12.2**	**(±1.3)**	**0.111**
DAS	**1.9**	**(±0.4)**	**4.3**	**(±0.9)**	**<0.0001**
NLR	**2.30**	**(±1.22)**	**2.17**	**(±1.31)**	**0.602**
PLR	**114.37**	**(±34.89)**	**117.02**	**(±49.49)**	**0.755**

*By unpaired t test; NLR, neutrophil-to-lymphocyte ratio; PLR, platelets-to-lymphocyte ratio.

Among patients with less active disease (DAS < 2.6), Table 4 shows no significant differences in three studied groups regarding the NLR (p = 0.901) and PLR (p = 0.638). While when patients with active disease are considered (DAS ≥ 2.6), significant differences were detected. The NLR means was 3.8 among patients of the cDMARD group, 2.2 among patients on bDMARD plus cDMARD, and 2.4 in the control. The PLR means of the cDMARD group = 273.3 compared with 117 and 115.6 in the bDMARD plus cDMARD group and the control group respectively.

Table 4. Means of neutrophil -to- lymphocyte ratio and platelet -to - lymphocyte ratio of the three study groups according to disease activity

	Groups	N	Mean	(±SD)	P*	LSD groups	p (LSD)
DAS < 2.6							
NLR	**A) cDMARD**	**52**	**2.3**	**(±1.1)**		**A X B**	**0.851**
	B) bDMARD plus cDMARD	**51**	**2.3**	**(±1.2)**	**0.901**	**A X C**	**0.788**
	C) Control	**50**	**2.4**	**(±1.3)**		**B X C**	**0.650**
	Total	**153**	**2.3**	**(±1.2)**			
PLR	**A) cDMARD**	**52**	**108.7**	**(±40.2)**		**A X B**	**0.464**
	B) bDMARD plus cDMARD	**51**	**114.4**	**(±34.9)**	**0.638**	**A X C**	**0.377**
	C) Control	**50**	**115.6**	**(±43.1)**		**B X C**	**0.877**
	Total	**153**	**112.8**	**(±39.3)**			
DAS ≥ 2.6							
NLR	**A) cDMARD**	**55**	**3.8**	**(±4.5)**		**A X B**	**0.003**
	B) bDMARD plus cDMARD	**52**	**2.2**	**(±1.3)**	**0.006**	**A X C**	**0.012**
	C) Control	**50**	**2.4**	**(±1.3)**		**B X C**	**0.673**
	Total	**157**	**2.8**	**(±2.9)**			
PLR	A) cDMARD	55	273.3	(±482.6)		A X B	0.006
	B) bDMARD plus cDMARD	**52**	**117.0**	**(±49.5)**	**0.006**	**A X C**	**0.006**
	C) Control	**50**	**115.6**	**(±43.1)**		**B X C**	**0.980**
	Total	**157**	**171.3**	**(±296.0)**			

*By ANOVA; NLR, neutrophil-to-lymphocyte ratio; PLR, platelets to- lymphocyte ratio.

There was a positive significant and relatively weak correlation between the DAS score and the NLR (rho = 0.285, p = 0.003) in the cDMARD group. There was also a significant and medium strength correlation between the DAS score and the PLR (rho = 0.548, p<0.001). While among both groups, there was a weak significant correlation between DAS with NLR and PLR as shown in table 5.

Table 5. Correlation between DAS with neutrophil - to - lymphocyte ratio and platelets - to - lymphocyte ratio in the cDMARD and the bDMARD plus cDMARD group

Variable 1	Variable 2	rho	p
cDMARD			
NLR	**DAS**	**0.285**	**0.003**
PLR	**DAS**	**0.548**	**< 0.001**
bDMARD plus cDMARD			
NLR	**DAS**	**-0.020**	**0.840**
PLR	**DAS**	**-0.016**	**0.873**
Both groups			
NLR	**DAS**	**0.161**	**0.019**
PLR	**DAS**	**0.307**	**< 0.001**

NLR, neutrophil-to-lymphocyte ratio; PLR, platelets-to- lymphocyte ratio.

Table 6 shows that, among patients on the cDMARD, around two-thirds of them (65.4%) were on Methotrexate (MTX) only, and 11.2 and 6.5% were taking the MTX with either HCQ or SSZ respectively.

Table 6. Medicine that patient used

	No.	(%)
cDMARD		
MTX (Methotrexate)	**70**	**(65.4)**
Leflunomoid	**4**	**(3.7)**
HCQ (hydroxychloroquine)	**4**	**(3.7)**
SSZ (Sulfasalazine)	**4**	**(3.7)**
MTX + HCQ	**12**	**(11.2)**
MTX + SSZ	**7**	**(6.5)**
SSZ + HCQ	**6**	**(5.6)**
Total	**107**	**(100.0)**
bDMARD which taken by patient group on bDMARD plus cDMARD		
Etanercept	**45**	**(43.7)**
Infliximab	**37**	**(35.9)**
Adalimumab	**13**	**(12.6)**
Rituximab	**8**	**(7.8)**
Total	**103**	**(100.0)**

5. Discussion:

During recent years the neutrophil/ lymphocyte ratio (NLR) and platelet/ lymphocyte ratio (PLR) have been noted as novel effective markers for inflammation (Xuanyu et al., 2017). This evidence is clear in the current study result which is similar to many types of research done previously in this regard such as have been found in the research performed in Egypt which considered the NLR and PLR as two novel inflammatory biomarkers for assessing the activity of the diseases in a patient with RA (Abd-Elazeem and Mohamed, 2018).

Also there is another multicenter retrospective research which conducts in China that agreed RA groups had significantly higher NLRs and PLRs than controls (Jin et al., 2021).

Other two studies on the relation between RA patient's disease activity and NLRs and PLRs in Turkey supports the result of the current study as the 1st one was concluding that assessment of NLR with PLR levels can be used as additional markers for inflammation in patients who have RA (Senem and Kervansaray, 2019), and 2nd one took into consideration in RA the role of the neutrophils, lymphocytes, and platelets, so they saw that the NLR with PLR which consists of these components as inflammatory markers predict to made changes in the disease, and suggested that they can be used as markers in the disease activity follow up (Uslu et al.,2015). The third study also was done in Turkey which analyzed only the NLR and disease activity and found that it is a useful available low cost and well-correlated marker for disease activity in the evaluation of RA (Mercan et al., 2016).

Regarding the relationship between the DAS score and NLR, the finding of this study were also consistent with the study was done in India that found the worse DAS score positively correlated with NLR and concludes that NLR is a highly significant in patient with RA so could be using as a good indicator for estimate RA active disease patients (Quaiser and

Khan, 2020). Armen et al. were regard the PLR as a novel inflammatory marker together with NLR beside other hematologic parameters, which helps in detect and evaluate the activity and seriousness of much rheumatic disorder, and they suggested that PLR-NLR collectively have more effect on distinguishing RA patients in active stage from a healthy person. A similar thing was detected when RA patients were in remission in comparison with the healthy person (Armen et al., 2019). Also, the meta-analysis which was done by Erre et.al. Supports our result it found that the correlation between RA and peripheral blood NLR and PLR was significant (Erre et al., 2018). Other meta-analysis research was done by Hao et al showed that NLR had strongly related to RA and PLR also correlated to it. So considered NLR with PLR as cheap available diagnostic biomarkers for autoimmune rheumatic diseases (Xuanyu et al., 2017).

It is worth noting that in all the above research they did not address the type of medication used by the patient, so we compared our result of all groups that include in the study together with the previously mentioned studies, while in our research, we put the RA patients according to the type of treatment into two groups for getting more precise results. So when compare each group of the RA patient separately according to the type of treatment in the RA group which received cDMARD we found that the difference was significant statistically among the (patient and control) groups for NLR, PLR when the patients with disease active means DAS28-ESR score of 2.6 and more while no significant differences detected with patients that have a level of less than 2.6 means remission patients, (table4). Also, the correlation between NLR and PLR with DAS28-ESR was significant (table5).

While for the patients who received bDMARD plus cDMARD the difference was not significant in comparison to the control group (table4) and (table5). And since the research that had been conducted previously not identifying patients according to the type of treatment, so the results was found regarding the group that takes cDMARD treatment expected based on the results of the above-mentioned research and compare it with the current study result in term of that the ordinary protocol for the treatment of RA is starting with cDMARD and in case of inadequate response to this treatment shift to use the bDMARD for the treatment so by that most of the patients probably on cDMARD (Smolen et al 2016; Masahito et al 2016). Therefore, it is necessary to conduct more detailed studies in this regard in the future.

Also, with regard to the results of this study of the group receiving treatment bDMARD plus cDMARD, there are no similar finding research, but there are a study conducted in South Korea that evaluated RA patients who started anti-TNFα drug as the first-choice biological therapy for insufficient response to cDMARD treatment and evaluated the response to treatment, and the remission rate of RA patients was performed at 12 and 24 weeks. It found that NLR also PLR levels were correlated positively with a measure of DAS28 disease activity score. This supposed that NLR with PLR are beneficial indicators for monitoring the inflammatory status of RA patients using anti-TNFα agents (Han et al., 2019). This result did not go with current study result but this discrepancy assumed to be because of the differences in the study design. At first, this study analyzed RA patients treated with bDMARD plus cDMARD not only on anti-TNF-α agents. And second, this study investigated the NLR and PLR after 48 (±2.2) months. Nevertheless, even the studies mentioned above also found a significant association at 24 weeks, unlike a significant association at 12 weeks (Han et al., 2019). This concept suggests that NLRs and PLRs can show a quick response in case of inflammatory changes, and the capacity of these biomarkers to portend therapeutic response may be restricted to a short period of time.

Another study from Japan was conducted to detect if NLR could be considered an indicator for the prognosis of the response to biological treatment. Found that active disease and NLR are positively and significantly correlated with DAS28-ESR than in people with low disease activity (Masahito et al., 2016) but can't match the result with the current study because the treatment that the patients received was biological treatment alone and for the duration for 6 months while the participant patients in this study received both bDMARD plus cDMARD and for a longer period so in this case, the effect of both types of the drug will be on the NLR.

6. Conclusion:

The result of the current study supports that NLR and PLR can be regarded as two parameters for inflammation assessment in RA disease activity, especially for patients who receive cDMARD.

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Conflicts of interest:

There are no conflicts of interest to be mentioned.

References:

Abd-Elazeem MI and Mohamed RA, 2018. Neutrophil lymphocyte and platelet lymphocyte ratios in rheumatoid arthritis patients: relation to disease activity. Egypt Rheumatol.; 40:227-231.

Armen Y G, Lilit A, Ulzhan M, Marlen Y, and George D. Kitas, 2019. The Platelet-to-Lymphocyte Ratio as an Inflammatory Marker in Rheumatic Diseases Ann Lab Med;39:345-357 .

Assayag D, Lee JS and King TE Jr, 2014. Rheumatoid arthritis associated interstitial lung disease: a review. Medicina (B Aires); 74 (2), 158–65.

Bruno F, Suzanne M.M, Vertappen and Annelies B, 2011. Economic consequences and potential benefits. Best Pract Res Clin Rheumatol, 27, pp. 607–624

Balkarli A, Kucuk A, Babur H, and Erbasan F, 2016.Neutrophil/lymphocyte ratio and mean platelet volume in Behçet's disease. Eur Rev Med Pharmacol Sci.; 20:3045-3050.

Bruno F, Stephene M, and Peter T, 2018. Treatment of Rheumatoid arthritis. In: Johannes WJ. and Eric H. (Eds.), Eular Textbook on rheumatic diseases 3rd ed. BMJ Publishing Group;10:279

Chandrashekara S, Rajendran A, Bai Jaganath A, and Krishnamurthy R., 2015. Neutrophil-lymphocyte ratio, pain perception, and disease activity score may serve as important predictive markers for sustained remission in rheumatoid arthritis. Reumatismo; 67(3):109–115.

Chandrashekara, S, Mukhtar Ahmad M, Renuka, P, Anupama, K.R, and Renuka, K, 2017.

Characterization of neutrophil-to-lymphocyte ratio as a measure of inflammation in rheumatoid arthritis. Int. J. Rheum. Dis.; 20, 1457–1467.

Choi JL, Li S and Han JY, 2014. Platelet function tests: a review of progresses in clinical application. Biomed Res Int; 456569.

Colglazier CL, and Sutej PG, 2005. Laboratory testing in the rheumatic diseases: a practical review. South Med J; 98(2):185–91.

Du J, Chen S, Shi J, Zhu X, Ying H, Zhang Y, Chen S, Shen B, and Li J, 2017. The association between the lymphocyte-monocyte ratio and disease activity in rheumatoid arthritis. Clin Rheumatol.; 36:2689-2695.

Elena M., Carl T. and Eric L. Matteson, 2019. Extraarticular features of rheumatoid arthritis. In: Marc C. Hochberg, Ellen M. Gravallese, Alan J. Silman et. al. Rheumatology7thed. Elsevier, Philadelphia; 95:768.

Erre GL, Paliogiannis P, Castagna F, Mangoni AA, Carru C, Passiu G, and Zinellu A, 2018. Meta-analysis of neutrophil-to-lymphocyte and platelet-to-lymphocyte ratio in rheumatoid arthritis. Eur J Clin Investig. ; https ://doi.org/10.1111/eci.13037

Fu H, Qin B, Hu Z, Ma N, Yang M, Wei T, Tang Q, Huang Y, Huang F, Liang Y et al., 2015. Neutrophil and platelet to lymphocyte ratios are correlated with disease activity in rheumatoid arthritis. Clin Lab.; 61:269-273.

Han-N L, Yun-K K, Geun-T K, Eunyoung A, Min W S, Dong H S, and Seung-G L, 2019. Neutrophil-to-lymphocyte and platelet-to-lymphocyte ratio as predictors of 12-week treatment response and drug persistence of anti-tumor necrosis factor-α agent in patients with rheumatoid arthritis. Rheumatol Int. May;39 (5):859-868.

Hobbs KF and Cohen MD, 2012. Rheumatoid arthritis disease measurement: a new old idea. Rheumatology;51(suppl 6). vi21-7.

Inoue E, Yamanaka H, Hara M, Tomatsu T, and Kamatani N, 2007 .Comparison of Disease Activity Score (DAS) 28-erythrocyte sedimentation rate and DAS28- C-reactive protein threshold values. Ann Rheum Dis; 66, 407–9.

Jin Z, Cai G, Zhang P, Li X, Yao S, Zhuang L, Ren M, Wang Q, and Yu X, 2021. The value of the neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio as complementary diagnostic tools in the diagnosis of rheumatoid arthritis. *J Clin Lab Anal*.;35:e23569.

Karimifar M, Salesi M. and Farajzadegan Z , 2012. The association of anti-CCP1 antibodies with disease activity score 28 (DAS-28) in rheumatoid arthritis. Adv Biomed Res;1(30),6.

Kiely PD, Deighton C, Dixey J, and Ostor AJ, 2012. British Society for Rheumatology Standards, Guidelines and Audit Working Group: Biologic agents for rheumatoid arthritis-negotiating the NICE technology appraisals. Rheumatology (Oxford); 51: 24—31.

Kisacik B, Tufan A, Kalyoncu U, Karadag O, Akdogan A, Ozturk MA, Kiraz S, Ertenli I, and Calguneri M, 2008. Mean platelet volume (MPV) as an inflammatory marker in ankylosing spondylitis and rheumatoid arthritis. Joint Bone Spine.; 75: 291–4.

Kouri VP, Olkkonen J, Ainola M, Li TF, Björkman L, Konttinen YT, and Mandelin J, 2014. Neutrophils produce interleukin-17B in rheumatoid synovial tissue. Rheumatology; 53(1):39–47.

Kugyelka R, Kohl Z, Olasz K, Mikecz K, Rauch TA, Glant TT, and Boldizsar F, 2016. Enigma of IL-17 and Th17 cells in rheumatoid arthritis and in autoimmune animal models of arthritis. Mediators Inflamm; 6145810.

Maden M, Pamuk G. and Pamuk O., 2017. Development of atherosclerotic cardiovascular mortality in gouty arthritis and rheumatoid arthritis patients: are they associated with mean platelet volume and neutrophil lymphocyte ratio?. Arch Rheumatol.;32:39-45.

Masahito K, Susumu G, Kenji T, Toshikazu K, Shinro T and Hiroshi N, 2016 Neutrophil/Lymphocyte Ratio in Patients with Rheumatoid Arthritis Treated with Biological Agents J Nippon Med Sch; 83 (3)

Mercan R, Bitik B, Tufan A, Bozbulut UB, Atas N, Ozturk MA, Haznedaroglu S, and Goker B, 2016. The association between neutrophil/ lymphocyte ratio and disease activity in rheumatoid arthritis and ankylosing spondylitis. J Clin Lab Anal.;30:597-601.

Mohamad, A.A., Majid, F.A.A., Assaw, S., Abidin, F.Z., Fadhlina, A., Aluru, S.K., Zainol, S.N. and Ahmed, N.J., (2021). "Anti-inflammatory and oral toxicity assessment of r-38tm: a traditional herbal supplement for arthritis", Jurnal Teknologi, 84(1), pp. 249-262.

Muddathir ARM and Haj FEJ., 2013. Platelet indices in sudanese patients with rheumatoid arthritis. Asian J Biomed Pharm Sci.; 3: 1–3.

Park EY, Lee SG, Park EK, Koo DW, Park JH, Kim GT, Tag HS, Kim HO, and Suh YS, 2018. Drug survival and the associated predictors in South Korean patients with rheumatoid arthritis receiving tacrolimus. Korean J Intern Med; 33(1):193–202.

Peng Y-F, Cao L, Zeng Y-H, Chen D, Zhang Q, and Zhu YS, 2015. Platelet to lymphocyte ratio and neutrophil to lymphocyte ratio in patients with rheumatoid arthritis. Open Med (Wars).;10:249-253.

Quaiser S. and Khan R., 2020. Correlation of neutrophil–lymphocyte ratio with disease activity in rheumatoid arthritis. J Clin Sci;17:46-8.

Sanmart -R, Ruiz-Esquide V. and Hern_andez MV., 2013. Rheumatoid arthritis: a clinical overview of new diagnostic and treatment approaches. Curr Top Med Chem; 13, 698–704.

Scapini P. and Cassatella MA ., 2014. Social networking of human neutrophils within the immune system. Blood; 124, 710–9.

Senem Şaş. and Kervansaray, 2019. Neutrophil/lymphocyte and platelet/lymphocyte ratios are associated with disease activity in rheumatoid arthritis.J Clin Anal Med;10(3):379-83

Smolen JS, Breedveld FC, Burmester GR, Bykerk V, Dougados M, Emery P, Kvien TK, Navarro-Compan MV, Oliver S, Schoels M et al., 2016. Treating rheumatoid arthritis to target: 2014 update of the recommendations of an international task force. Ann Rheum Dis; 75(1):3–15.

Tekeog˘lu I, Gürol G, Harman H, Karakeçe E, and Çiftçi IH, 2016. Overlooked hematological markers of disease activity in rheumatoid arthritis. Int J Rheum Dis; 19 (11):1078–82.

Uslu, A.U.; Küçük, A.; ¸Sahin, A., Ugan, Y.; Yılmaz, R.; Güngör, T.; Ba˘ gcacı, S.; Küçük¸ and sen, S, 2015. 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.; 18, 731–735.

Willemze A, Toes RE, Huizinga TW, and Trouw LA, 2012. New biomarkers in rheumatoid arthritis. Neth J Med; 70, 392–9.

Wu Y, Chen Y, Yang X, Chen L, and Yang Y, 2016. Neutrophil to lymphocyte ratio (NLR) and platelet to lymphocyte ratio (PLR) were associated with disease activity in patients with

systemic lupus erythematosus. Int Immunopharmacol.;36:94-99.

Xuanyu H, Dongyang L, Dan W, and Ning Z, 2017. The Relationship between Hematological Indices and Autoimmune Rheumatic Diseases (ARDs), Published:07 September Scientific Reports; 7: *10833*

Yang Z, Zhang Z, Lin F, Ren Y, Liu D, Zhong R, and Liang Y, 2017. Comparisons of neutrophil, monocyte, eosinophil, and basophil lymphocyte ratios among various systemic autoimmune rheumatic diseases. APMIS.; 125:863-871.

Yao Y, Simard AR, Shi FD, and Hao J, 2013. IL-10-producing lymphocytes in inflammatory disease. Int Rev Immunol; 32, 324–36.

Yazici S, Yazici M, Erer B, Calik Y, Ozhan H, and Ataoglu S, 2010. The platelet indices in patients with rheumatoid arthritis: mean platelet volüme reflects disease activity. Platelets; 21: 122–5.

Yolbas S, Yildirim A, Gozel N, Uz B, and Koca SS., 2016. Hematological indices may be useful in the diagnosis of systemic lupus erythematosus and in determining disease activity in Behçet's disease. Med Princ Pract.; 25:510-516.

Zengin O, Onder ME, Kalem A, Bilici M, Türkbeyler I H, Ozturk Z A, Kisacik B, and Onat A M, 2018. New inflammatory markers in early rheumatoid arthritis. Z Rheumatol.; 77:144-150.

Zhang Y, Yin Y, Kuai S, Shan Z, Pei H, and Wang J., 2016. Combination of neutrophil to lymphocyte ratio and platelet to lymphocyte ratio as diagnostic biomarker for rheumatoid arthritis. Int J Clin ExpMed.; 9:22076-22081.