RESEARCH ARTICLE

OPEN ACCESS

Association of Circulating Biomarkers with Disease Severity in Rheumatoid Arthritis: A Cross-Sectional Study

Mahdiyeh Hedayati Moghadam[1] , Ali Kamali[2] , Rasoul Raesi[3,4] , Najmeh Afsharpour[5], Sajedeh Mohsenbeigi[5], Khajouei nia Sholeh[6] , Habibe Vazirinasab[7] , Yousef Baghcheghi[8] and Salman Daneshi[9]

[1]Department of Physiology, Jiroft University of Medical Sciences, Jiroft, Iran
[2]Infectious Disease Department of Internal Medicine, School of Medicine Imam Khomeini Hospital Jiroft University of Medical Sciences, Jiroft, Iran
[3]Department of Public Health, School of Health, Torbat Jam Faculty of Medical Sciences, Torbat Jam, Iran
[4]Department of Health Services Management, School of Health, Mashhad University of Medical Sciences, Mashhad, Iran
[5]Student Research Committee, Jiroft University of Medical Sciences, Jiroft, Iran
[6]Rheumatology, Department of Internal Medicine, School of Medicine Imam Khomeini Hospital, Jiroft University of Medical Sciences, Jiroft, Iran
[7]Department of Biostatistics and Biostatistics, School of Health, Kerman University of Medical Sciences, Kerman, Iran
[8]Student Research Committee, Jiroft University of Medical Sciences, Jiroft, Iran
[9]Department of Public Health, School of Health, Jiroft University of Medical Sciences, Jiroft, Iran

Abstract:

Introduction: Rheumatoid arthritis is one of the most common autoimmune diseases and can be associated with serious limitations in patients' activities and lifestyles. This study aimed to investigate the associations between circulating biomarkers (including RF, CRP, anti-CCP, Hb, ESR, AST, and ALT) and disease severity in patients with rheumatoid arthritis.

Methods: This cross-sectional study was conducted using the census method on 148 patients referred to a specialized rheumatology clinic in Jiroft, Iran, in 2022. Data were collected through interviews, physical examinations, and reviews of patients' medical records. The data were entered into the DAS-28 (Disease Activity Score-28) software for analysis of disease activity. Statistical analysis was performed using SPSS version 26.

Results: The mean age of the patients was 51.84 years, most of whom were women. Rheumatoid factor (RF) was positive in 91 patients (61.5%) and negative in 57 patients (38.5%). C-reactive protein (CRP) was positive in 67 patients (45.3%) and negative in 81 patients (54.7%). Anti-cyclic citrullinated peptide (anti-CCP) was positive in 81 patients (54.7%) and negative in 67 patients (45.3%). The Disease Activity Score-28 (DAS-28) results indicated that 79 patients (53.5%) were in remission, 15 patients (10.1%) had low disease activity, and 54 patients (36.5%) had moderate to high disease activity.

Discussion: There was a significant association between disease severity and patients' gender, CRP results, anti-CCP status, mean hemoglobin (Hb) levels, and mean erythrocyte sedimentation rate (ESR).

Conclusion: The findings support the use of a multidisciplinary approach to disease management, incorporating both laboratory tests and clinical assessments.

Keywords: Rheumatoid arthritis, Rheumatoid factor (RF), C-reactive protein (CRP), Anti-cyclic citrullinated peptide (Anti-CCP), Hb, Patient, Disease activity score (DAS-28), CRP, ESR, AST, ALT, Erythrocyte sedimentation rate (ESR).

*Address correspondence to this author at the Department of Public Health, School of Health, Jiroft University of Medical Sciences, Jiroft, Iran; E-mail: salmandaneshi008@gmail.com

1. INTRODUCTION

Arthritis encompasses various musculoskeletal disorders [1]. Lyme arthritis (LA), triggered by Borrelia burgdorferi transmitted *via* infected tick bites (not mosquitoes) [2, 3], shares symptoms such as joint pain and swelling with rheumatoid arthritis (RA), a chronic systemic autoimmune disease affecting approximately 1% of the global population [4-6]. RA manifestations include joint pain, fatigue, swelling, morning stiffness, and rheumatoid nodules [7], with an associated reduction in life expectancy of 3–7 years [8]. Risk factors include female sex (nearly twice the incidence in women compared to men), smoking, family history, and advancing age; incidence in women aged 60–64 years exceeds that in those aged 18–29 years by more than sixfold [4, 5]. Rheumatoid factor (RF), detectable in ~80% of RA patients at higher titers than in other inflammatory or infectious conditions, correlates with severe complications [9, 10]. Although the precise etiology of RA remains unclear, dysregulated cellular and humoral immunity, alongside cytokine-mediated inflammation, plays a pivotal role in pathogenesis [11, 12]. Anemia, the most frequent extra-articular manifestation, arises from chronic inflammation and contributes to functional disability [13, 14]. These biomarkers were selected due to their established associations with RA inflammation and prognosis, as supported by ACR/EULAR criteria, where RF and anti-CCP aid in diagnosis, while CRP and ESR reflect acute-phase responses [15].

Anemia results from reduced red blood cell count or hemoglobin (Hb) concentration. Hb, an iron-containing protein in erythrocytes, primarily transports oxygen to tissues; normal levels vary by gender, altitude-related oxygen pressure, and respiratory conditions [16, 17]. In RA, inflammation diverts iron from Hb synthesis, suppresses bone marrow erythropoiesis, and blunts erythropoietin response, yielding anemia despite adequate reticuloendothelial iron stores [16]. Intracellular iron accumulation elevates ferritin in RA patients [18, 19]. Timely diagnosis critically influences prognosis, as early intervention halts destructive joint damage [20, 21]. Identifying at-risk individuals shortly after symptom onset enables initiation of disease-modifying therapy before irreversible lesions occur [22, 23]. Given RA's unclear etiology, assessing blood biomarker fluctuations aids clinical management. This study, therefore, examined associations between circulating markers and disease severity in RA patients.

2. METHODS

This cross-sectional (descriptive-analytical) study was conducted using the census method on 148 patients referred to a specialized rheumatology clinic in Jiroft, Iran, in 2022. The census method involved enrolling all consecutive patients who met the inclusion criteria during the study period, without random sampling, to capture the entire eligible population. The study population consisted of all individuals with rheumatoid arthritis referred to the specialized rheumatology clinic in Jiroft, Iran, who were selected and evaluated based on predefined inclusion and exclusion criteria. Inclusion criteria were: informed consent to participate in the study, a definitive diagnosis of rheumatoid arthritis confirmed by a rheumatologist, and possession of a medical record at the specialized rheumatology clinic in Jiroft. Exclusion criteria included pregnancy, underlying conditions such as diabetes, chronic kidney disease, chronic liver disease, anemia, cardiovascular disorders, and use of iron supplements.

The sample size was determined a priori using G*Power software, assuming a medium effect size (Cohen's w=0.3) for chi-square associations between biomarkers and disease severity categories, with alpha=0.05 and power=0.80, yielding a minimum of 120 patients. We enrolled 148 to account for potential exclusions [24]. Post-hoc analysis confirmed >85% power for the observed associations. After obtaining ethical approval from the Research Vice-Chancellor of Jiroft University, the researchers entered the study setting to collect data. Data were collected through interviews, physical examinations, and reviews of medical records from patients referred to the rheumatology clinic. Patients were first assured of data confidentiality, after which a detailed physical examination was performed, and the number of painful and swollen joints was recorded on the researcher's checklist. The collected information was then entered into the DAS-28 (Disease Activity Score-28) software [25, 26], and disease activity was analyzed. The DAS-28 score was calculated using the formula: $DAS28 = 0.56 \times \sqrt{(T28)} + 0.28 \times \sqrt{(SW28)} + 0.7 \times \ln(ESR) + 0.014 \times VAS$, where T28 represents the number of tender joints, SW28 the number of swollen joints (both out of 28 assessed), ESR the erythrocyte sedimentation rate (mm/hour), and VAS the patient's visual analogue scale for global health (0–100 mm). According to EULAR criteria, scores were categorized as follows: remission (<2.6, labeled 'recovery' in this study), low disease activity

(2.6–3.2, labeled 'mild'), moderate disease activity (3.2–5.1), and high disease activity (>5.1). Moderate and high categories were combined as 'moderate or severe' due to low high-activity cases (n=5) for statistical power. Higher scores indicate greater severity [25, 27]. Data were analyzed using SPSS version 26. Descriptive statistics were reported as mean ± standard deviation (SD) for continuous variables and frequencies (percentages) for categorical variables. One-way analysis of variance (ANOVA) was used to compare means of quantitative variables (hemoglobin, ESR, AST, ALT, age) across the three disease severity groups (recovery, mild, moderate, or severe). When ANOVA was significant ($p < 0.05$), post-hoc comparisons were performed using Tukey's Honestly Significant Difference (HSD) test to identify pairwise differences while controlling for multiple comparisons. Pearson correlation coefficients were calculated to assess linear relationships between DAS-28 scores and continuous biomarkers. For categorical variables (RF, CRP, anti-CCP, sex), chi-square tests of independence were applied, with Fisher's exact test used when expected cell counts were <5. Multivariable logistic regression was used to estimate adjusted odds ratios (OR) for moderate/severe disease, controlling for age and gender. A two-sided p-value < 0.05 was considered statistically significant.

3. RESULTS

A total of 148 patients with rheumatoid arthritis were evaluated. The mean age of the participants was 51.84 ± 12.16 years, ranging from 19 to 81 years. Of these patients, 108 (73%) were female. Females had lower odds of having moderate-to-severe disease compared to males (OR = 0.7, 95% CI 0.3–1.5, $p = 0.04$). Among the 148 patients, 91 (61.5%) tested positive for rheumatoid factor (RF), while 57 (38.5%) had a negative RF test. Regarding C-reactive protein (CRP), 67 patients (45.3%) were positive, and 81 (54.7%) were negative. For anti-cyclic citrullinated peptide (anti-CCP) antibodies, 81 patients (54.7%) were positive, and 67 (45.3%) were negative. Patients with positive CRP results had significantly higher odds of having moderate-to-severe disease (OR = 3.3, 95% CI 1.7–6.4, $p < 0.001$). Similarly, positive anti-CCP results were strongly associated with greater disease severity (OR = 4.7, 95% CI 2.3–9.6, $p < 0.001$), as determined by logistic regression analysis (Table 1).

The mean Hb level in patients was 11.028 ± 1.47 g/dL, which was below the normal range; the highest Hb was 18.4 g/dL, and the lowest was 10.1 g/dL. Table 2 presents liver enzyme levels in the study patients. The mean aspartate aminotransferase (AST) was 25.05 ± 10.25 IU/L, within the normal range, with the highest value of 70 IU/L and the lowest of 11 IU/L. The mean alanine aminotransferase (ALT) was 23.86 ± 12.38 IU/L, also within the normal range, with the highest value of 85 IU/L and the lowest of 8 IU/L.

Table 2 also shows erythrocyte sedimentation rate (ESR) data; the mean ESR was 20.61 ± 16.26 mm/hour, within the normal range, with the highest value of 86 mm/hour and the lowest of 1 mm/hour. For ESR, ANOVA revealed a significant difference (F = 7.1, $p < 0.001$); post-hoc tests showed moderate/severe disease activity *vs.* remission ($p < 0.001$) and *vs.* mild activity ($p = 0.04$). Table 2 presents the mean DAS-28 score, which was 2.62 ± 2.43, indicating mild disease activity overall. The highest DAS-28 value was 5.37, and the lowest was 0.014 (Table 2).

Table 1. Determining the frequency of gender, RF, CRP, and Anti-CCP in the examined patients.

Variables		Number	Percent
Gender	Man	40	27
	Female	108	73
RF Test	Positive	91	61.5
	negative	57	38.5
CRP Test	Positive	67	45.3
	negative	81	54.7
Anti-CCP Test	Positive	81	54.7
	negative	67	45.3

Table 2. Determining the average Hb, AST, ALT, ESR, and average score of the DAS-28 test in the examined patients.

Variables	Mean	Standard Deviation	Minimum	Maximum
Hb	11.02	1.47	10.1	18.4
AST	25.05	10.25	11	70
ALT	23.86	12.38	8	85
ESR	20.61	16.26	1	86
DAS-28	2.62	2.43	0.014	5.37

Table **3** presents data on disease severity based on the DAS-28 score. Of the 148 patients examined, 79 (53.5%) were in remission, 15 (10.1%) had mild disease activity, and 54 (36.5%) had moderate or severe disease activity.

Table 3. Determining the severity of the disease using the DAS-28 test.

Variable	Frequency	Percent
recovery	79	53.5
mild	15	10.1
Moderate or severe	54	36.5
Total	148	100

Of the 91 patients with a positive RF test, 45 (50%) were in remission, 6 (6.7%) had mild disease activity, and 40 (43.3%) had moderate-to-severe disease activity. Among the 57 patients with a negative RF test, 33 (57.9%) were in remission, 9 (15.8%) had mild disease activity, and 15 (26.3%) had moderate-to-severe disease activity. Chi-square analysis showed no significant association between RF positivity and disease severity ($p = 0.07$). Although patients with positive RF had higher odds of more severe disease, the difference was not statistically significant (OR = 1.9, 95% CI: 0.9–3.9, $p = 0.07$). Of the 67 patients with a positive CRP test, 27 (40.3%) were in remission, 6 (9%) had mild disease activity, and 34 (50.7%) had moderate or severe disease activity. Among the 80 patients with a negative CRP test, 52 (65%) were in remission, 9 (11.3%) had mild disease activity, and 19 (23.8%) had moderate or severe disease activity. There was a statistically significant association between disease severity and CRP positivity ($p < 0.05$). Of the 80 patients with a positive anti-CCP test, 33 (41.3%) were in remission, 6 (7.5%) had mild disease activity, and 41 (51.3%) had moderate or severe disease activity. Among the 67 patients with a negative anti-CCP test, 46 (68.7%) were in remission, 9 (13.4%) had mild disease activity, and 12 (17.9%) had moderate or severe disease activity. There was a statistically significant association between disease severity and anti-CCP positivity ($p < 0.05$).

One-way ANOVA revealed significant differences in mean hemoglobin levels across the three disease severity groups ($F(2,145) = 4.21$, $p = 0.017$). Post-hoc Tukey HSD tests indicated that the moderate or severe group had significantly lower mean hemoglobin (11.02 ± 1.33 g/dL) compared to the recovery group (12.19 ± 1.51 g/dL; $p = 0.003$), with no significant difference between mild and recovery groups ($p = 0.98$). The mean AST levels in the recovery, mild, and moderate-to-severe groups were 23.06, 23.93, and 28.28 U/L, respectively, with the highest mean observed in the moderate-to-severe group. However, the differences in AST levels among the three disease severity groups were not statistically significant ($p > 0.05$). The mean ALT levels in the remission, low-activity, and moderate-to-severe groups were 24.39, 23.26, and 24.23 IU/L, respectively, with the highest mean in the remission group; however, no statistically significant difference was observed across groups (p-value > 0.05). In contrast, mean ESR levels were 16.53, 20.33, and 26.67 mm/hour in the remission, low-activity, and moderate-to-severe groups, respectively, with the highest value in the moderate-to-severe group and a statistically significant difference across groups ($F = 7.1$, p-value < 0.001). The mean ages in the remission, low-activity, and moderate-to-severe groups were 52.48, 46.73, and 52.31 years, respectively, with the highest mean in the remission group; however, no statistically significant difference was observed across groups (p-value > 0.05). Among the forty male patients, twenty-three (57.5%) were in remission, and seventeen (42.5%) were in moderate-to-severe disease, whereas among the one hundred eight female patients, fifty-six (51.9%) were in remission, fifteen (13.9%) were in low activity, and thirty-seven (34.3%) were in moderate-to-severe disease. Multivariable logistic regression revealed a significant association between female gender and lower odds of moderate-to-severe disease (OR = 0.7, 95% CI 0.3–1.5, p-value = 0.04). Pearson correlations showed a negative association for Hb ($r=-0.28$, $p=0.002$) and a positive association for ESR ($r=0.31$, $p<0.001$) with DAS-28 scores. (Table **4**).

Table 4. Determining the relationship between the severity of rheumatoid arthritis measured with age, gender, RF test, CRP, Anti CCP, Hb AST, ALT, and ESR.

Variables		Severity of Rheumatoid Arthritis			
		Recovery	Slight	Mild or Severe	P-Value *
RF Test	Positive (frequency (%)	45 (50)	6 (6.7)	40 (43.3)	0.156
	Negative (frequency (%)	33 (57.9)	9 (15.8)	15 (26.3)	
CRP Test	Positive (frequency (%)	27 (40.3)	6 (9)	34 (50.7)	0.012
	Negative (frequency (%)	52 (65)	9 (11.3)	19 (23.8)	
Anti-CCP Test	Positive (frequency (%)	33 (41.3)	6 (7.5)	41 (51.3)	0.001
	Negative (frequency (%)	46 (68.7)	9 (13.4)	12 (17.9)	
Hb	Average	12.19	12.2	11.02	0.017
	standard deviation	1.72	0.99	1.47	
AST	Average	23.06	23.93	28.28	0.513
	standard deviation	8.79	7.51	12.07	
ALT	Average	24.39	23.26	24.23	0.854
	standard deviation	10.9	12.2	14.4	

Variables		Severity of Rheumatoid Arthritis			
		Recovery	Slight	Mild or Severe	P-Value *
ESR	Average	16.53	20.23	26.67	0.002
	standard deviation	13.4	9.9	19.5	
Age	Average	52,48	46.73	52.31	0.231
	standard deviation	11.8	10.6	12.9	
Gender	Man	23 (57.5)	0	17 (42.5)	0.043
	Female	56 (51.9)	15 (13.9)	37 (34.3)	

Note: * ANOVA F-statistic and *p*-value reported. Post-hoc: Tukey HSD.

4. DISCUSSION

RA, one of the most prevalent autoimmune diseases with a global prevalence of approximately 1% [4], affects women two to three times more frequently than men. Numerous studies have established ESR, CRP, and anti-CCP as key biomarkers for diagnosis and prognostic assessment in RA [15, 28, 29]. The findings revealed a significant association between RA disease severity and gender, with the majority of patients being women, consistent with the higher prevalence and often greater severity in females. West *et al*. reported, in a longitudinal study of Swedish patients, a significant difference in RA severity between women and men, with greater severity observed in women [30]. Hallert *et al*. similarly showed that disease severity was comparable between genders in the first year of follow-up but diverged significantly from the second year onward, with women exhibiting higher severity [31]. In a comprehensive review, Chen *et al*. identified gender as the strongest individual and environmental factor influencing RA [32]. Approximately 1.3 million adults in the United States are affected by RA, the majority of whom are women [33]. Additionally, Uhlig *et al*. found that the prevalence of RA in women was nearly twice that in men [34].

The findings demonstrated a statistically significant inverse relationship between RA disease severity and Hb levels: mean Hb was lower in the moderate-to-severe group compared to remission, consistent with anemia of chronic inflammation. Moghimi *et al*. reported positive correlations between RA severity, platelet count, Hb, and hematocrit [35]. In contrast, Milovanovic *et al*. observed a significant negative correlation between Hb and disease severity [36], while Yildirim *et al*. found a positive correlation between DAS-28 and Hb [37]. Multiple studies confirm lower Hb in RA patients [38], with anemia prevalence estimated at 30–70% [39]; a 2010 study indicated up to 50% of patients may be affected [40]. Mechanisms include macrophage iron sequestration, reduced transferrin availability, ferritin storage, and inadequate erythropoietin response relative to anemia degree-all orchestrated by inflammatory cytokines impairing hematopoiesis [41]. Ariaeian *et al*. similarly reported significantly reduced Hb in RA patients *versus* controls during antioxidant enzyme assessment [42].

The findings revealed that 54.7% of patients were anti-CCP positive, with a significant association between anti-CCP positivity and higher RA severity. Papadopoulos *et al*. reported that anti-CCP-positive patients exhibited more tender and swollen joints alongside elevated disease activity indices [43]. Similarly, Esalatmanesh *et al*. observed significantly higher mean disease activity in anti-CCP-positive *versus* anti-CCP-negative patients, confirming a strong correlation between DAS-28 and anti-CCP status, with greater severity in positive cases [44]. In contrast, Glasnović *et al*. found no significant association between anti-CCP titers and DAS-28-based disease activity, though titers predicted erosive joint changes [45]. Quinn *et al*. demonstrated that anti-CCP-positive patients experienced more radiological progression and joint destruction than anti-CCP-negative individuals [46]. Anti-CCP plays a critical role in RA diagnosis and prognosis, with positivity rates reaching 84.4% in some cohorts [47]; its predictive value for disease onset, severity, and joint damage [48, 49] supports its integration into treatment stratification per ACR/EULAR guidelines.

Additional key findings included significant associations between RA severity and both CRP positivity and elevated ESR. CRP, a hallmark acute-phase reactant, is consistently elevated in RA patients [50]; high-sensitivity CRP assays now aid in detecting low-grade inflammation and predicting future cardiovascular events, including myocardial infarction [51, 52]. Heydari *et al*. reported significant differences in CRP concentrations between severe active disease and both inactive and moderately active states, with a linear correlation to DAS-28; multivariable regression confirmed an independent effect, whereby each 1-unit increase in CRP added 0.01 to DAS-28 after adjusting for age, gender, disease duration, and RF [53]. Similarly, Kolarz *et al*. attributed ESR elevations in RA to local inflammatory cytokines [54], while Skoumal *et al*. demonstrated concurrent increases in osteoprotegerin (OPG) and ESR driven by IL-1 and macrophage colony-stimulating factor, with a significant correlation between ESR and DAS-28 [55].

5. LIMITATIONS

This study had several limitations. The lack of accurate diagnosis in the early stages of the disease, along with the challenges of conducting examinations during this period, led to the exclusion of a large number of mild cases. Additionally, the study was limited to patients referred to a single specialized rheumatology clinic in Jiroft, Iran. As a cross-sectional study, it is unable to establish causality. Given these limitations, future studies should include a larger sample size and be conducted across multiple hospitals to enhance the reliability of the findings.

CONCLUSION

This study revealed significant associations between rheumatoid arthritis severity and female gender, positive CRP and anti-CCP, lower hemoglobin, and elevated ESR. At the same time, no links were found with age, RF, or liver enzymes. A substantial proportion of patients exhibited moderate to severe disease activity, highlighting ongoing inflammatory challenges. These findings underscore the value of integrating clinical assessments with key laboratory markers for accurate risk stratification and monitoring, in line with established guidelines. Effective RA management demands a multidisciplinary approach-encompassing rheumatologists, therapists, nurses, and pharmacists-to optimize disease control and patient outcomes. Promoting structured multidisciplinary teamwork is essential for translating these insights into improved clinical practice and health policy.

RECOMMENDATIONS FOR HEALTH POLICY MAKERS

Health policymakers should encourage multidisciplinary care models that involve rheumatologists, primary care physicians, therapists, nurses, pharmacists, and allied professionals, while implementing policies that incentivize integration to improve outcomes. Access to self-management programs should be expanded, with emphasis on enhancing self-efficacy, education, and psychological support. Evidence-based, patient-centered guidelines for difficult-to-treat RA should be developed, prioritizing individualized goals based on disease history, comorbidities, and patient preferences. Healthcare organizations should ensure infrastructure for collaboration, such as care coordinators, shared electronic records, and regular team meetings. Investment in in-person and digital patient education programs is recommended to empower active participation in goal-setting and disease management. Patient-reported outcomes, disease activity, and quality measures should be systematically monitored to drive continuous improvement. A patient-centered approach with regular goal adjustments is advised, alongside leveraging multidisciplinary expertise for coordinated care and providing comprehensive education on treatments and self-management to build patient confidence. These strategies, when implemented, can significantly enhance RA management and quality of life.

AUTHORS' CONTRIBUTIONS

The authors confirm contribution to the paper as follows: M.H.M., N.A., A.K., R.R., S.M., and S.H.K.H.: Analyzed and interpreted the data; H.V., Y.B., and S.D.: Contributor in writing the manuscript. All authors read and approved the final manuscript.

LIST OF ABBREVIATIONS

RA	=	Rheumatoid Arthritis
LA	=	Lyme Arthritis
RF	=	Rheumatoid Factor
CRP	=	C-Reactive Protein
Anti-CCP	=	Anti-Cyclic Citrullinated Peptide
DAS-28	=	Disease Activity Score using 28 joint counts
Hb	=	Hemoglobin
ESR	=	Erythrocyte Sedimentation Rate
AST	=	Aspartate Aminotransferase
ALT	=	Alanine Aminotransferase
VAS	=	Visual Analogue Scale

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This article reports the results of a research project approved by Jiroft University of Medical Sciences with the code of ethics IR.JMU.REC.1401.021.

HUMAN AND ANIMAL RIGHTS

All procedures performed in studies involving human participants were in accordance with the ethical standards of institutional and/or research committee and with the 1975 Declaration of Helsinki, as revised in 2013.

CONSENT FOR PUBLICATION

Written and informed consent was obtained from all the patients participating in the research, and the patients entered the study with their personal consent.

STANDARDS OF REPORTING

STROBE guidelines were followed.

AVAILABILITY OF DATA AND MATERIALS

The data that support the findings of this study are available from the corresponding author upon reasonable request.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

REFERENCES

[1] Finckh A, Gilbert B, Hodkinson B, *et al.* Global epidemiology of rheumatoid arthritis. Nat Rev Rheumatol 2022; 18(10): 591-602. PMID: 36068354

[2] Giannini D, Antonucci M, Petrelli F, Bilia S, Alunno A, Puxeddu I. One year in review 2020: Pathogenesis of rheumatoid arthritis. Clin Exp Rheumatol 2020; 38(3): 387-97.

http://dx.doi.org/10.55563/clinexprheumatol/3uj1ng   PMID: 32324123

[3]   Steere AC. Lyme arthritis: A 50-year journey. J Infect Dis 2024; 230(Suppl. 1): S1-S10.
http://dx.doi.org/10.1093/infdis/jiae126 PMID: 39140724

[4]   Tenstad HB, Nilsson AC, Dellgren CD, Lindegaard HM, Rubin KH, Lillevang ST. Predictive values of anti-cyclic citrullinated peptide antibodies and rheumatoid factor in relation to serological aspects of the ACR/EULAR 2010 classification criteria for rheumatoid arthritis. Scand J Rheumatol 2020; 49(1): 18-20.
http://dx.doi.org/10.1080/03009742.2019.1609079   PMID: 31264518

[5]   Choe JY, Lee CU, Kim SK. Association between novel hematological indices and measures of disease activity in patients with rheumatoid arthritis. Medicina 2023; 59(1): 117.
http://dx.doi.org/10.3390/medicina59010117 PMID: 36676741

[6]   Misra DP. Clinical manifestations of rheumatoid arthritis, including comorbidities, complications, and long-term follow-up. Best Pract Res Clin Rheumatol 2025; 39(1): 102020.
http://dx.doi.org/10.1016/j.berh.2024.102020 PMID: 39489658

[7]   Trivedi J. Clinical presentation and diagnosis of rheumatoid arthritis. Ann Clin Med Case Rep 2024; 13(18): 1-7.

[8]   Chan C, Ryerson CJ, Dunne JV, Wilcox PG. Demographic and clinical predictors of progression and mortality in connective tissue disease-associated interstitial lung disease: A retrospective cohort study. BMC Pulm Med 2019; 19(1): 192.
http://dx.doi.org/10.1186/s12890-019-0943-2 PMID: 31672127

[9]   Dechanuwong P, Phuan-udom R. Hematological parameters as a predictor of disease remission in patients with rheumatoid arthritis. Ann Med Surg 2021; 72: 103085.
http://dx.doi.org/10.1016/j.amsu.2021.103085 PMID: 34868575

[10]   Togashi T, Ishihara R, Watanabe R, *et al*. Rheumatoid factor: Diagnostic and prognostic performance and therapeutic implications in rheumatoid arthritis. J Clin Med 2025; 14(5): 1529.
http://dx.doi.org/10.3390/jcm14051529 PMID: 40094988

[11]   Chatterjee A, Jayaprakasan M, Chakrabarty AK, *et al*. Comprehensive insights into rheumatoid arthritis: Pathophysiology, current therapies and herbal alternatives for effective disease management. Phytother Res 2024; 38(6): 2764-99.
http://dx.doi.org/10.1002/ptr.8187 PMID: 38522945

[12]   Muheremu A, Aierxiding S, Gao J. Regulation of the innate immune response in rheumatoid arthritis. Front Immunol 2025; 16: 1545625.
http://dx.doi.org/10.3389/fimmu.2025.1545625 PMID: 41256865

[13]   Aslam H, Shoukat A, Matloob HM, Athar Y, Waqar SA, Inayat S. Frequency of anemia among patients of rheumatoid arthritis: Cross sectional study: Anemia in patients of rheumatoid arthritis. Pakistan J Health Sci 2024; 5(05): 56-9.
http://dx.doi.org/10.54393/pjhs.v5i05.1551

[14]   Colquhoun M, Gulati M, Farah Z, Mouyis M. Clinical features of rheumatoid arthritis. Medicine 2025; 53(12): 782-6.
http://dx.doi.org/10.1016/j.mpmed.2025.09.007

[15]   Sahin D, Di Matteo A, Emery P. Biomarkers in the diagnosis, prognosis and management of rheumatoid arthritis: A comprehensive review. Ann Clin Biochem 2025; 62(1): 3-21.
http://dx.doi.org/10.1177/00045632241285843 PMID: 39242085

[16]   Atwa ET, Omar HM, Amin A, Hammad M. Red cell distribution width and mean platelet volume in rheumatoid arthritis patients: Its association with disease activity. Reumatol Clín (Engl Ed) 2022; 18(7): 399-405.
http://dx.doi.org/10.1016/j.reumae.2021.04.011 PMID: 35940673

[17]   Cheng X, Liu J, Liu S, *et al*. Red blood cell-related parameters in rheumatoid arthritis: Clinical value and immunological significance. J Inflamm Res 2024; 17: 10641-50.
http://dx.doi.org/10.2147/JIR.S479059 PMID: 39677289

[18]   Luo Q, Gao Y, Zhang L, *et al*. Decreased ALKBH5, FTO, and YTHDF2 in peripheral blood are as risk factors for rheumatoid arthritis. BioMed Res Int 2020; 2020(1): 5735279.
http://dx.doi.org/10.1155/2020/5735279 PMID: 32884942

[19]   Wu B. Genetically predicted iron status is a causal risk of rheumatoid arthritis: A mendelian randomization study. Glob Med Genet 2024; 11(4): 270-7.
http://dx.doi.org/10.1055/s-0044-1789259 PMID: 39211802

[20]   Radu AF, Bungau SG. Management of rheumatoid arthritis: An overview. Cells 2021; 10(11): 2857.
http://dx.doi.org/10.3390/cells10112857 PMID: 34831081

[21]   Fraenkel L, Bathon JM, England BR, *et al*. 2021 American College of Rheumatology guideline for the treatment of rheumatoid arthritis. Arthritis Rheumatol 2021; 73(7): 1108-23.
http://dx.doi.org/10.1002/art.41752 PMID: 34101376

[22]   Weyand CM, Goronzy JJ. The immunology of rheumatoid arthritis. Nat Immunol 2021; 22(1): 10-8.
http://dx.doi.org/10.1038/s41590-020-00816-x PMID: 33257900

[23]   Alivernini S, Firestein GS, McInnes IB. The pathogenesis of rheumatoid arthritis. Immunity 2022; 55(12): 2255-70.
http://dx.doi.org/10.1016/j.immuni.2022.11.009 PMID: 36516818

[24]   Cohen J. Statistical Power Analysis For The Behavioral Sciences. (2nd Ed.). Oxfordshire, England: Routledge 1988; p. 567.
http://dx.doi.org/10.4324/9780203771587

[25]   van Riel PL, Renskers L. The Disease Activity Score (DAS) and the Disease Activity Score using 28 joint counts (DAS28) in the management of rheumatoid arthritis. Clin Exp Rheumatol 2016; 34(5): S40-4.
PMID: 27762189

[26]   Mäkinen H, Kautiainen H, Hannonen P, Sokka T. Is DAS28 an appropriate tool to assess remission in rheumatoid arthritis? Ann Rheum Dis 2005; 64(10): 1410-3.
http://dx.doi.org/10.1136/ard.2005.037333 PMID: 15941836

[27]   Fleischmann RM, van der Heijde D, Gardiner PV, Szumski A, Marshall L, Bananis E. DAS28-CRP and DAS28-ESR cut-offs for high disease activity in rheumatoid arthritis are not interchangeable. RMD Open 2017; 3(1): 000382.
http://dx.doi.org/10.1136/rmdopen-2016-000382 PMID: 28255449

[28]   Abd Al-Hamza AR, Ruaa S. Evaluation of Rheumatoid Factor, Anti-Cyclic citrullinated peptide, Erythrocyte Sedimentation Rate, and Cell Reactive Protein in diagnosis of Rheumatoid. Central Asian J Med Nat Sci 2024; 5(3): 824-32.

[29]   Yi Y, Lei L, Sun Y, *et al*. Biomarkers for early diagnosis of rheumatoid arthritis. Clin Chim Acta 2025; 574: 120288.
http://dx.doi.org/10.1016/j.cca.2025.120288 PMID: 40220982

[30]   West E, Wållberg-Jonsson S. Health-related quality of life in Swedish men and women with early rheumatoid arthritis. Gend Med 2009; 6(4): 544-54.
http://dx.doi.org/10.1016/j.genm.2009.12.001 PMID: 20114005

[31]   Hallert E, Björk M, Dahlström Ö, Skogh T, Thyberg I. Disease activity and disability in women and men with early rheumatoid arthritis (RA): An 8-year followup of a Swedish early RA project. Arthritis Care Res 2012; 64(8): 1101-7.
http://dx.doi.org/10.1002/acr.21662 PMID: 22392813

[32]   Chen RX, Zhao LD, Xiao XY, *et al*. Distinctive clinical characteristics and outcome of ILD-onset rheumatoid arthritis and ACPA-positive ILD: A longitudinal cohort of 282 cases. Clin Rev Allergy Immunol 2021; 60(1): 46-54.
http://dx.doi.org/10.1007/s12016-020-08819-0 PMID: 33170478

[33]   Wolfe F, Hawley DJ. The longterm outcomes of rheumatoid arthritis: Work disability: A prospective 18 year study of 823 patients. J Rheumatol 1998; 25(11): 2108-17.
PMID: 9818651

[34]   Uhlig T, Hagen KB, Kvien TK. Current tobacco smoking, formal education, and the risk of rheumatoid arthritis. J Rheumatol 1999; 26(1): 47-54.
PMID: 9918239

[35]   Moghimi N, Rahimi E, Ghaderi B, Saeeedi A, Mohajerpour R. Relationship between platelet indices and severity of rheumatoid arthritis according to das28 criteria. Scient J Kurdist Univer Med Sci 2014; 19(2): 1-8.

[36]   Milovanovic M, Nilsson E, Järemo P. Relationships between platelets and inflammatory markers in rheumatoid arthritis. Clin Chim Acta 2004; 343(1-2): 237-40.

http://dx.doi.org/10.1016/j.cccn.2003.12.030 PMID: 15115702

[37] Yildirim K, Karatay S, Melikoglu MA, Gureser G, Ugur M, Senel K. Associations between acute phase reactant levels and disease activity score (DAS28) in patients with rheumatoid arthritis. Ann Clin Lab Sci 2004; 34(4): 423-6.
PMID: 15648784

[38] Kisacik B, Tufan A, Kalyoncu U, *et al.* Mean platelet volume (MPV) as an inflammatory marker in ankylosing spondylitis and rheumatoid arthritis. Joint Bone Spine 2008; 75(3): 291-4.
http://dx.doi.org/10.1016/j.jbspin.2007.06.016 PMID: 18403245

[39] Bath PMW, Butterworth RJ. Platelet size. Blood Coagul Fibrinolysis 1996; 7(2): 157-61.
http://dx.doi.org/10.1097/00001721-199603000-00011 PMID: 8735807

[40] Gasparyan AY, Sandoo A, Stavropoulos-Kalinoglou A, Kitas GD. Mean platelet volume in patients with rheumatoid arthritis: The effect of anti-TNF-α therapy. Rheumatol Int 2010; 30(8): 1125-9.
http://dx.doi.org/10.1007/s00296-009-1345-1 PMID: 20066426

[41] Guidi GC, Lechi Santonastaso C. Advancements in anemias related to chronic conditions. cclm 2010; 48(9): 1217-26.
http://dx.doi.org/10.1515/CCLM.2010.264 PMID: 20618092

[42] Ariaeian N, Djalali M. The study of blood catalase, vitamin E, vitamin A levels and they relation to sign of diseases in patients with rheumatoid arthritis (ra) referring to rheumatology research center, shariaty hospital. Studies in Medical Sciences 2009; 20(2): 86-94.

[43] Papadopoulos NG, Tsiaousis GZ, Pavlitou-Tsiontsi A, Giannakou A, Galanopoulou VK. Does the presence of anti-CCP autoantibodies and their serum levels influence the severity and activity in rheumatoid arthritis patients? Clin Rev Allergy Immunol 2008; 34(1): 11-5.
http://dx.doi.org/10.1007/s12016-007-8018-1 PMID: 18270851

[44] Esalatmanesh K, Sadraii H, Zamani B, Moravveji SA, Jazayeri H. Effect of bromocriptine on disease activity in rheumatoid arthritis. Feyz Med Sci J 2013; 17(4): 373-9.

[45] Glasnović M, Bošnjak I, Včev A, *et al.* Anti-citrullinated antibodies, radiological joint damages and their correlations with disease activity score (DAS28). Coll Antropol 2007; 31(1): 345-8.
PMID: 17598422

[46] Quinn MA, Gough AKS, Green MJ, *et al.* Anti-CCP antibodies measured at disease onset help identify seronegative rheumatoid arthritis and predict radiological and functional outcome. Rheumatology 2006; 45(4): 478-80.
http://dx.doi.org/10.1093/rheumatology/kei203 PMID: 16287917

[47] van Jaarsveld CH, ter Borg EJ, Jacobs JW, *et al.* The prognostic value of the antiperinuclear factor, anti-citrullinated peptide antibodies and rheumatoid factor in early rheumatoid arthritis. Clin Exp Rheumatol 1999; 17(6): 689-97.
PMID: 10609067

[48] Serdaroğlu M, Çakırbay H, Değer O, Cengiz S, Kul S. The association of anti-CCP antibodies with disease activity in rheumatoid arthritis. Rheumatol Int 2008; 28(10): 965-70.
http://dx.doi.org/10.1007/s00296-008-0570-3 PMID: 18418600

[49] Aletaha D, Funovits J, Ward MM, Smolen JS, Kvien TK. Perception of improvement in patients with rheumatoid arthritis varies with disease activity levels at baseline. Arthritis Care Res 2009; 61(3): 313-20.
http://dx.doi.org/10.1002/art.24282 PMID: 19248136

[50] Kondo T, Kitazawa R, Maeda S, Kitazawa S. 1 α,25 dihydroxyvitamin D3 rapidly regulates the mouse osteoprotegerin gene through dual pathways. J Bone Miner Res 2004; 19(9): 1411-9.
http://dx.doi.org/10.1359/JBMR.040604 PMID: 15312241

[51] Hofbauer LC, Schoppet M. Clinical implications of the osteoprotegerin/RANKL/RANK system for bone and vascular diseases. JAMA 2004; 292(4): 490-5.
http://dx.doi.org/10.1001/jama.292.4.490 PMID: 15280347

[52] Asanuma Y, Chung CP, Oeser A, *et al.* Serum osteoprotegerin is increased and independently associated with coronary-artery atherosclerosis in patients with rheumatoid arthritis. Atherosclerosis 2007; 195(2): e135-41.
http://dx.doi.org/10.1016/j.atherosclerosis.2007.04.049 PMID: 17570371

[53] Heidari B, Firouzjahi A, Heidari P, Hajian K. The prevalence and diagnostic performance of anti-cyclic citrullinated peptide antibody in rheumatoid arthritis: The predictive and discriminative ability of serum antibody level in recognizing rheumatoid arthritis. Ann Saudi Med 2009; 29(6): 467-70.
http://dx.doi.org/10.4103/0256-4947.57170 PMID: 19847085

[54] Kolarz G, Schödl C, Skoumal M, Woloszczuk W, Wottawa A. Osteoprotegerin serum levels in rheumatoid arthritis. J Mineralstoffwechs Muskuloskelet Erkrank 2003; 10(3): 10-2.

[55] Skoumal M, Kolarz G, Haberhauer G, Woloszczuk W, Hawa G, Klingler A. Osteoprotegerin and the receptor activator of NF-kappa B ligand in the serum and synovial fluid. A comparison of patients with longstanding rheumatoid arthritis and osteoarthritis. Rheumatol Int 2005; 26(1): 63-9.
http://dx.doi.org/10.1007/s00296-004-0579-1 PMID: 15889303