

COMPARISON OF ACUTE PHASE REACTANTS IN RHEUMATOID ARTHRITIS PATIENTS AND HEALTHY ADULTS

Farzana Hakim, Amir Rashid, Amer Fakhr*

Army Medical College/National University of Medical Sciences (NUMS) Rawalpindi Pakistan, *Combined Military Hospital Pano Aqil/National University of Medical Sciences (NUMS) Pakistan

ABSTRACT

Objective: To analyze the acute phase reactants in patients suffering from rheumatoid arthritis (RA) and compare them to healthy controls.

Study Design: Cross-sectional comparative study.

Place and Duration of Study: This study was conducted at the department of Biochemistry & Molecular Biology Army Medical College, Rawalpindi in collaboration with Rheumatology department Military Hospital (MH) Rawalpindi and Military Hospital Laboratories, from Jan 2016 to Jun 2016.

Material and Methods: Approval of this study was given by Ethical Review Committee, Army Medical College (AMC) Rawalpindi. Two groups i.e., group I: normal healthy subjects (n=100) and group II: rheumatoid arthritis patients (n=100) were included in the study. Demographic data: age, gender, ethnicity and marital status of the healthy subjects as well as RA patients was recorded on proforma. Blood sample was collected and patients were monitored for acute-phase response i.e. by evaluating the levels of C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR).

Results: In this study 100 RA patients with 29 males and 71 females were enrolled. Hundred healthy controls included 50 males and 50 females. The mean CRP value of RA patients was 8.44 ± 4.28 mg/l i.e. higher as compared to that of controls whose mean CRP value was 6.2 ± 4.6 mg/l ($p < 0.001$). Among RA patients, the mean ESR value was 29.0 ± 16.9 mm/hr and among controls, the mean ESR value was 20.0 ± 9.42 mm/hr ($p < 0.001$).

Conclusion: Both CRP and ESR are important disease assessment biomarkers. They were significantly elevated among RA patients as compared to healthy subjects in our study. Moreover due to cost effectiveness these (CRP and ESR) are preferred tests for routine assessment. CRP and ESR provide the much desired information about RA activity and severity that cannot be provided by either of them alone.

Keywords: Acute phase reactants, Inflammation, Rheumatoid arthritis.

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INTRODUCTION

Rheumatoid arthritis (RA) is a chronically progressive autoimmune inflammatory systemic disorder¹. RA develops as a result of interaction between genetic and environmental factors that spark immune signaling cascade that finally leads to synovitis, destruction of joints and structural bone deformities. Extra articular manifestations of RA and co morbidities end up with increased mortality².

RA has a variable natural history with a minimum of three likely disease courses i.e. monocyclic, polycyclic and progressive courses.

Monocyclic course comprises of a single episode that finishes within 2-5 years of early diagnosis that could be due to early diagnosis/aggressive treatment. In polycyclic course, disease activity levels waxes and wanes. In progressive course, severity of RA increases and persists³ resulting in complications of the disease.

RA has affected about 1% of population world over². The data for RA prevalence is not available for most parts of the world⁴. In developing countries the RA prevalence is variable. In southern Pakistan, the prevalence of RA in urban population is reported to be 0.142%⁵ while its prevalence is 0.55% in northern Pakistan. As stated in media reports, arthritis has affected 14 million Pakistanis⁶ but no definite figures for RA are available at national level.

Correspondence: Dr Amir Rashid, Biochemistry Dept Army Medical College Pakistan (Email: dramiram@yahoo.com)

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American College of Rheumatology (ACR) and European League against Rheumatism together developed the new criteria for RA classification. According to new criteria, definite RA is classified based on confirmation of synovitis existence in at least one joint, lack of suitable alternate diagnosis explaining the synovitis, getting 6 or greater (with possible 10) total score from four different domains i.e. number of joints and site involved (range 0-5), serological testing for rheumatoid factor (RF) and anti citrullinated protein antibody (ACPA)

The purpose of this study is to analyze the acute phase reactants in RA patients. Both CRP and ESR are cost effective, easily available in all hospitals and are useful biomarkers for diagnosis and prognosis of RA.

MATERIAL AND METHODS

The study design was cross-sectional comparative and non-probability purposive sampling technique was employed to collect samples. The study was carried out at department of Biochemistry and Molecular

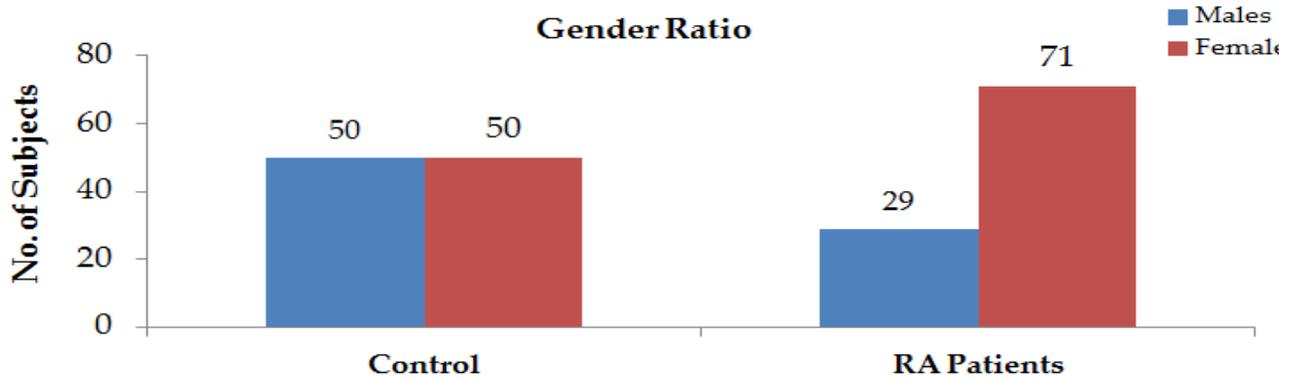


Figure-1: Gender ratio among rheumatoid arthritis patients and healthy controls.

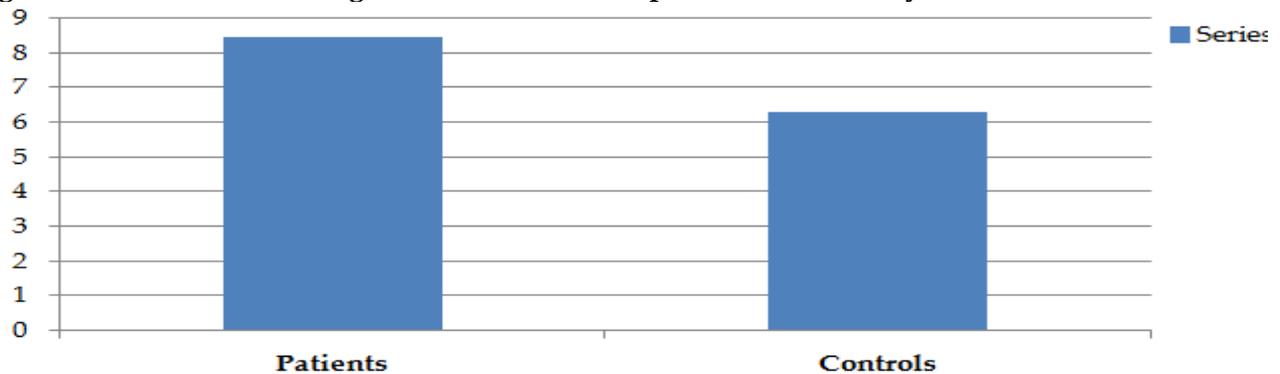


Figure-2: Mean C-reactive protein values among rheumatoid arthritis patients and healthy controls.

abnormality (range 0-3), raised acute-phase reactants i.e. C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) abnormality (range 0-1) and duration of symptoms (two levels; range 0-1)⁷. The acute phase reactants are elevated in all inflammatory disorders whether acute or chronic. There are two types of acute phase reactants i.e. positive and negative. CRP and ESR are both positive acute phase reactants which means their level rises with inflammation⁸.

Biology Army Medical College in collaboration with Rheumatology department Military Hospital (MH) Rawalpindi and biochemical markers were analyzed at MH Laboratories. It was approved by Ethical Committee, Army Medical College (AMC), Rawalpindi. Subjects were recruited and their written consent was taken. Two groups i.e. group I: normal healthy subjects (n=100) and group II: rheumatoid arthritis patients (n=100) were included in the

study. RA patients with other chronic diseases e.g anemia, multiple myeloma, tuberculosis, chronic hepatitis, inflammatory bowel diseases, cancer etc were excluded from the study. The age, gender, ethnicity and marital status of the healthy subjects as well as RA patients was documented. Blood sample was collected in specimen tubes under sterile conditions from healthy individuals and RA patients who were visiting Rheumatology department of Military Hospital, Rawalpindi. Patients were monitored for acute-phase response i.e. by evaluating the levels of C-reactive protein (CRP) and erythrocyte sedimentation (ESR). CRP titer was determined

71 (71%) were females. Among controls, 50 (50%) were males and 50 (50%) were females ($p=0.002$) as shown in fig-1. Mean age of RA patients was 49 ± 11.62 years while mean age of controls was 43 ± 9.54 years ($p<0.001$).

Among RA patients, the mean CRP value was 8.44 ± 6.22 mg/l i.e. higher as compared to that of controls whose mean CRP value was 6.2 ± 4.6 mg/l. There was obvious CRP variation between the two groups ($p=0.004$) as shown in fig-2.

The mean ESR values were also found to be higher among RA patients as compared to

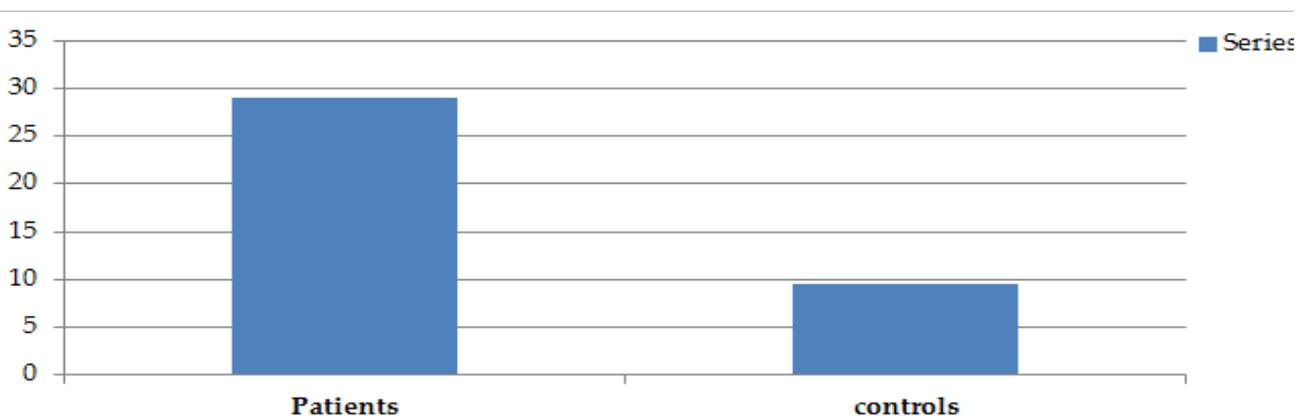


Figure-3: Mean erythrocyte sedimentation rate values among rheumatoid arthritis patients and healthy controls.

through semi-quantitative method mentioned in CRP latex test kit (Cat No. CRP/012). CRP levels in normal adult are <6 mg/l. ESR of the patients was determined by (Westergren method).

Data Analysis

SPSS Version 22 was used to analyze the data. Frequency along with percentages are for categorical data, mean and SD for quantitative data respectively. Chi-square test was applied for gender wise comparison. Independent student's t-test was applied. A p -value less than 0.05 was considered as significant.

RESULTS

A total of 200 subjects were enrolled in the study including 100 patients and 100 controls. Among the RA patients, 29 (29%) were males and

healthy controls. Among RA patients, the mean ESR value was 29 ± 16.6 mm/hr and among controls, the mean ESR value was 20.0 ± 9.4 mm/hr. The ESR values were significantly different between the two groups ($p<0.001$) as shown in fig-3.

DISCUSSION

Rheumatoid arthritis (RA) is a chronic, inflammatory systemic and prototype autoimmune disorder affecting females more than the males. The prevalence of RA is higher among females than that of males⁹. The studies in northern France showed a higher female: male ratio i.e 2.99:1¹⁰. A study was carried out in Japan to evaluate the RA prevalence. The ratio being 2.6:1¹¹ i.e. higher in females than males. In our

study, the female to male ratio was also high i.e. 2.5:1 thus matching international data.

In order to diagnose RA at early stage and start treatment as soon as possible, different criteria were used including disease activity score and ESR based estimation¹². Earlier, it was observed that CRP and ESR have a positive linear correlation in RA along with high variability but CRP was suggested to be more sensitive marker¹³. Similarly in another study, serum CRP was found to correlate with RA activity and important in objective evaluation of RA. Both ESR and CRP are extensively used for monitoring disease activity in RA. Unlike CRP, ESR is affected by age, gender, red cell defects, abnormal immunoglobulin levels i.e. multiple myeloma, smoking, dietary lipids, medications, phases of menstrual cycle and serum fibrinogen levels. ESR has similar pattern like that of CRP but due to shortfall of ESR because of its dependency on above mentioned factors, CRP levels strongly correlate with subjective as well as semi-objective criteria as compared to ESR¹⁴. In another study, high and significant association was observed between acute phase response, swelling of joints, and radiological progression¹⁵. A study of 774 RA patients concluded that CRP is a superior measure of RA activity as compared to ESR. As evident through simple comparisons, both ESR and CRP estimations are similar, but partial correlation reveals that fraction of association between ESR and clinical variables are resultant from non-acute phase factors which in turn results in discordance between the ESR and CRP outcome. Therefore, CRP seems to be a superior test to measure acute phase. ESR is a better marker to evaluate the general severity as compared to CRP because of its sensitivity to immunoglobulin and RF, but still a poorer marker to evaluate the inflammation. CRP together with ESR provides the much desired information about RA activity and severity that cannot be provided by a single test¹⁶. Based on the findings, CRP being an important acute-phase reactant was included to assess the disease¹². American College of Rheumatology (ACR) and

European League against Rheumatism developed a criteria for RA classification where a patient should be evaluated in 4 domains, one being raised acute-phase response i.e. CRP and ESR abnormality estimation⁷. CRP levels in first year of RA are associated with quick and severe succession of joint damage, and persistently elevated CRP levels are correlated with extensive succession in radiological damage of joints. In RA pathogenicity, CRP is observed to have an inflammatory role and hence likely associated with destruction of bones¹⁷. In another study, acute phase reactants (APR) were estimated among RA patients and mean values ratios of CRP and ESR were similar to our findings with highly significant difference between normal and patients i.e. $p < 0.001$. The study concluded CRP being the most useful measurement among various ARP tests in RA patients¹⁸. In an Iranian study carried on RA patients, RF-T positive patients were found to have longer duration of RA, young age at disease onset and raised levels of ESR and CRP along with increased swollen joints¹⁹. Our findings suggest that CRP and ESR both are important parameters as they are significantly elevated among RA patients as compared to that of healthy subjects. However CRP together with ESR provides the much desired information about RA activity and severity that cannot be provided by either of them alone. As evident by our findings, not all the patients had raised ESR and CRP. Some patients had elevated levels of both ESR and CRP while others were with elevated ESR or CRP. None of the patients had normal values of both ESR or CRP.

CONCLUSION

Both CRP and ESR are important disease assessment biomarkers. They were significantly elevated among RA patients as compared to healthy subjects in our study. Moreover due to cost effectiveness these (CRP and ESR) are preferred tests for routine assessment. CRP and ESR provide the much desired information about RA activity and severity that cannot be provided by either of them alone.

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CONFLICT OF INTEREST

This study has no conflict of interest to declare by any author.

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