

ORIGINAL ARTICLE

Determination of Rheumatoid Factor by Latex Agglutination Test Among People of Different Age Groups in Pokhara Valley

Kusum Subedi,^{1*} Sushma Gautam²

ABSTRACT

Background: Rheumatoid factor (RF) is an antibody directed against the Fc region of immunoglobulin G (IgG), which is found mostly in the patient with an autoimmune condition. It is an important serological marker to detect autoimmune diseases like rheumatoid arthritis (RA). In this study, we aimed to determine the rheumatoid factor by latex agglutination test among people of different age groups.

Methods: A community-level cross-sectional study was conducted in different areas of Pokhara valley from January 2019 to June 2019. Blood samples collected were processed at the Department of Microbiology, Prithvi Narayan Campus, Pokhara. The presence of rheumatoid factor was detected using the latex kit test and then the concentration of RF was determined in RF positive samples.

Result: Out of 125 participants, 7 (5.6%) of them were detected positive with rheumatoid factor. RF concentration was between the range of 0.5 IU/ml to 4.0 IU/ml. More females (6.8%) tested positive for RF than males. Likewise, 56.73% of people with positive tests were elderly people of age above 60.

Conclusion: Rheumatoid factor suggests the risk of having various autoimmune conditions. Females and elderly people are high-risk groups. Therefore, its use in clinical practice may contribute to making wise treatment decisions at the early stage of autoimmune diseases.

Keywords: Rheumatoid Factor, Autoimmune disease, Serology, Agglutination test

INTRODUCTION

Rheumatoid factors (RFs) are antibodies directed against the Fc region of Immunoglobulin G (IgG), which were initially detected more than 7 decades back in a patient with rheumatoid arthritis. However, RF can also be found in other autoimmune and nonautoimmune conditions as well as in healthy individuals.¹ Rheumatoid factor plays an important role in the differential diagnosis of polyarthritis with

rheumatoid arthritis. RF testing in RA patients has a sensitivity of 60% to 90% and specificity of 85%.² . RF is currently based on only serological diagnostic criteria. For the presence of RA, radiographic changes and routine blood tests for erythrocyte sedimentation rate (ESR) and c-reactive protein (CRP) should be determined along with RF according to the American College of Rheumatology.³ Measurement of anticyclic citrullinated peptide antibody (ACPA) and RF together is recommended in clinical practice as the combination

¹Department of Microbiology, Tri-Chandra Multiple Campus, Ghantaghar, Kathmandu, Nepal.

²Department of Microbiology, Prithvi Narayan Campus, Pokhara, Nepal.

*Corresponding author:

Kusum Subedi
Department of Microbiology, Tri-Chandra
Multiple Campus, Ghantaghar, Kathmandu, Nepal
SubediKushum@gmail.com

of the two markers improves diagnostic accuracy, especially in the case of early rheumatoid arthritis.¹

RF test measures the amount of RF present in the blood.⁴ RF latex kit is a rapid agglutination procedure for the detection of rheumatoid factors in serum. This test is easy to perform, reliable, feasible, and rapid.⁵ Although the RF test alone only cannot confirm the presence of a particular disease, it could aid to determine the level of risk of having a disease. Moreover, in Nepal, only a few studies have been carried out for the determination of RF at the community level. Therefore, this study is intended to study the prevalence of rheumatoid factors among different age groups in Pokhara and aware people of the risks and prevention of autoimmune conditions.

MATERIALS AND METHODS

A cross-sectional study was conducted at the Department of Microbiology, Prithvi Narayan Campus, Pokhara. Local people of Pokhara-19, Lamachaur, students of Life Angel School, Lamachaur and Prithvi Narayan Campus, Pokhara have participated in the study. The study was conducted from January 2019 to June 2019. People of any age and gender were included in the study. Written informed consent of the participants or their guardians was taken before starting the study. During the study, an awareness program about the risks and prevention of autoimmune conditions for students was conducted in the school and campus, while for elderly people it was done before the sample collection.

Laboratory Analysis

With a sterile syringe, 3 ml of blood sample was collected aseptically from each of the participants with the help of registered nurses following the standard procedures and immediately transported to the laboratory after proper labeling. Each blood sample was centrifuged at 3,000 rpm for 5 minutes and serum was separated.⁶ The RF was determined qualitatively by using a rapid latex agglutination kit (lot number 18082726, sensitivity 98%, and specificity 98.8%) following the instructions provided by the manufacturer. In the case of a positive RF test, the titer and RF concentration

were determined semi-quantitatively.⁷ Positive control provided by the manufacturer was used for quality control.

Data Analysis

Data were entered in Microsoft Excel 2016 and analyzed through Statistical Package for Social Science (SPSS) version 16. The interpretation was done according to frequency distribution and percentage. Statistical association (chi-square) calculated between age groups and RF was found to be significant. The p-value < 0.05 was considered statistically significant.

RESULTS

In the study, 125 samples from different age groups were collected. Out of the total, 7 (5.6%) blood samples showed RF positive while 118 (94.4%) samples were RF negative. People between the age group of 70 to 80 constituted the highest fraction of positive results, i.e., 3 (42.45%), while there was only 1 (14.28%) positive sample in each age group of 30-40, 40-50, 50-70 and 60-70 (Figure 1). This result was found to be statistically significant with a p-value < 0.05. Of the total positive tests, females were higher at 5 (71.42%) than males 2 (28.57%) (Figure 2).

The semiquantitative test was done for 7 samples that gave a positive RF test to determine their titer and RF concentration values. In total, 3 (42.9%) had RF concentration 16 IU/ml, 2 (28.5%) had 32 IU/ml, remaining 2 (14.3% each) had 128 IU/ml and 256 IU/ml respectively (Table 1).

Table 1: Semi-quantitative test of RF positive samples

S. N.	Titer value	No. of sample	RF Concentration (IU/ml)	Percentage (%)
1	1/2	3	16	42.9%
2	1/4	2	32	28.5%
3	1/16	1	128	14.3%
4	1/32	1	256	14.3%

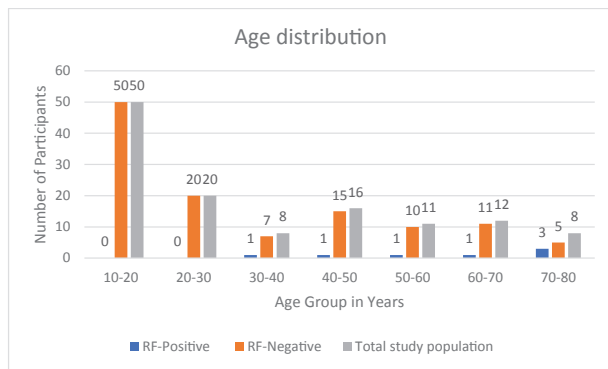


Figure. 1: Age distribution of the participants

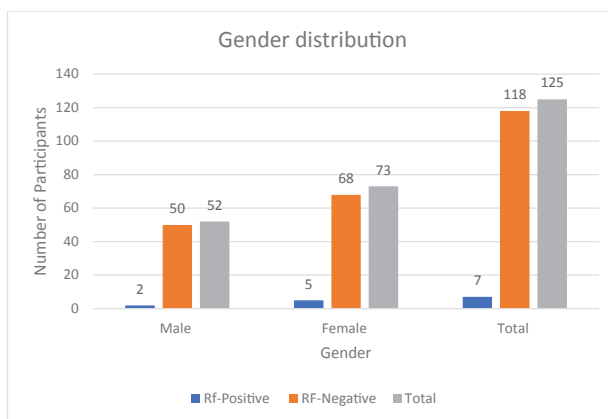


Figure. 2: Gender distribution of the participants

DISCUSSION

Although Rheumatoid factors are useful in predicting the development of autoimmune diseases like Rheumatoid arthritis,⁸ there are very few studies carried out in developing countries like Nepal. This is a small study carried out in a small area with few participants and in a limited period, therefore may not reflect the overall prevalence of the general Nepalese community. Moreover, the use of a latex kit may not confirm the presence of rheumatoid arthritis without any other recommended tests such as ACPA tests.

Our study revealed the prevalence of rheumatoid factor to be 5.6% with a higher number of females than males. The female to male ratio was 5:2. In contrast, a study carried out at Alka hospital had shown 29.35% of positive RF tests out of 184 samples. Similar to our study, females with RF positive tests were more than males with a ratio of 9:1.⁹ The greater occurrence

of RF positive cases in Alka hospital might be due to symptomatic people who reached the hospital for treatment while our study was community-based. The reason behind a large number of females may be due to hormonal fluctuations, the difference in sex hormones, and the presence of two X chromosomes in females.¹⁰

Likewise, 3 (42.85%) out of 7 samples had RF concentrations of 16 IU/ml while others had higher concentrations. Greater concentration indicates autoimmune disease or a tendency towards aggressive disease. RF does not necessarily be produced by pathological conditions and may present in the normal population, mainly the elderly group. RF may not be routinely detectable in blood without immunogenic stimulus. RF is considered a normal response to a variety of antigenic stimuli like bacterial toxins like lipopolysaccharide or viruses like Epstein-Barr Virus.¹¹

Age groups above 60 years were found to have the most positive RF tests. It is believed that the RF level increases with an increase in age,¹² which was supported by Dequeker et al,¹³ however, a cohort study by Nielsen et al. in 2012 [8] does not confirm this fact. This study showed 7.93% of individuals with symptoms of autoimmune disease tested for RF positive. This is attributed to the fact that rheumatoid factors are frequently detected in patients with systemic autoimmune diseases like rheumatoid arthritis.²

Autoimmune disease is mostly found in elder people, but nowadays, the frequency has increased in earlier age groups and children as a result of changing lifestyles and eating habits. Likewise, RF may even occur due to genetic influence.¹⁴ However, early diagnosis and treatment according to symptoms, regular exercise, therapies, antibiotics, and even prevention from seasonal triggers could help in prevention of the effects of autoimmune diseases; a common problem throughout the world.

CONCLUSION

In conclusion, Rheumatoid Factor was found to be prevalent mostly in elderly people while lower in young ones. Half of the people were above 60 years of age. Likewise, females were higher in number than males with a positive RF test. It suggests that females and elderly individuals are more prone to autoimmune

disease. Hence, early diagnosis and treatment are essential for minimizing the risk of pain or even disability caused by autoimmune diseases.

Conflicts of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

Funding Statement

This research did not receive any funding.

REFERENCES

- Ingegnoli F, Castelli R, Gualtierotti R. Rheumatoid Factors: Clinical Applications. *Disease Markers*. 2013;35:727–34.
- Mahajan Annil, Josrotia Davindder Singh, Manhas A S, Jamwal S S. Prevalence of Major Rheumatic Disorders in Jammu. *JK Science Journal of Medical Education and Research*. 2003;5(2):63–6.
- Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, Bingham CO, et al. 2010 Rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism collaborative initiative. *Annals of the Rheumatic Diseases*. 2010 Sep 1;69(9):1580–8.
- Newkirk MM. Rheumatoid factors: what do they tell us? *J Rheumatol*. 2002 Oct;29(10):2034–40.
- Manandhar S, Sharma S. Practical Approach to Microbiology. In: 3rd ed. Bhotahity, Kathmandu: National Book Centre; 2017. p. 340–1.
- Monica Cheesbrough. District laboratory practice in tropical countries, Part 2. 2nd ed. Cambridge, United Kingdom: Cambridge University Press; 2012.
- Agappe Diagnostics Ltd. *RF*. Agappe Hills, Dist. Ernakulam, Kerala, India. Retrieved from www.agappe.com.
- Nielsen SF, Bojesen SE, Schnohr P, Nordestgaard BG. Elevated rheumatoid factor and long term risk of rheumatoid arthritis: a prospective cohort study. *BMJ*. 2012 Sep 6;345(sep06 2):e5244–e5244.
- Vaidya B, Nakarmi S, Rajbhandari N, Bataju P. Clinical correlation between Anti-CCP antibody, rheumatoid factor and clinical parameters of patients with rheumatoid arthritis. *J Univ Coll Med Sci*. 2014 Jan 12;1(4):13–6.
- Rubtsov AV, Rubtsova K, Kappler JW, Marrack P. Genetic and hormonal factors in female-biased autoimmunity. *Autoimmunity Reviews*. 2010 May;9(7):494–8.
- Slaughter L, Carson DA, Jensen FC, Holbrook TL, Vaughan JH. In vitro effects of Epstein-Barr virus on peripheral blood mononuclear cells from patients with rheumatoid arthritis and normal subjects. *Journal of Experimental Medicine*. 1978 Nov 1;148(5):1429–34.
- Gaston JSH, editor. *Rheumatic diseases: immunological mechanisms and prospects for new therapies*. Cambridge ; New York: Cambridge University Press; 1999. 288 p.
- Dequeker J, Van Noyen R, Vandepitte J. Age-related rheumatoid factors. Incidence and characteristics. *Annals of the Rheumatic Diseases*. 1969 Jul 1;28(4):431–6.
- Simmonds M, Gough S. The HLA Region and Autoimmune Disease: Associations and Mechanisms of Action. *CG*. 2007 Nov 1;8(7):453–65.