Original Article

Purified protein derivative test and its booster phenomenon in patients with rheumatoid arthritis

Mansour Salesi, Mohsen Meidani[1], Shirin Meshkinfar, Hurie Hashemi, Ziba Farajzadegan[2]

Departments of Rheumatology, [1]Infectious Diseases, [2]Community Medicine, Isfahan University of Medical Sciences, Isfahan, Iran

Abstract

Background: Rheumatoid arthritis (RA) is a common disease in the community, with various complications. An appropriate solution is immunosuppressive drugs, which may lead to weakening of the cellular immune system and body unresponsiveness to tuberculosis (TB). As TB sensitivity is determined by the amount of induration created in the Purified Protein Derivative (PPD) test, this study aims to evaluate the immune response to the PPD test and its booster in RA patients.

Materials and Methods: This cross-sectional study was performed on rheumatoid arthritis patients referred to Alzahra Hospital, Isfahan, treated with ‹20 mg glucocorticoid daily or 7.5 mg Methotrexate (MTX) weekly. The sampling method was simple and accessible. The PPD test was performed in patients using the Mantoux method after 72 hours, and seven days later, the results were interpreted in 72 hours after the PPD booster injection. Induration ≥5 mm was considered to be positive. The data was analyzed using the SPSS software.

Results: Nineteen patients had positive results in the initial and reminder tests and 81 patients had negative results in both tests. Six patients (6.9%) with negative results in the initial test changed to positive in the reminder test. There was no positive result in the initial test and negative result in the reminder one. The frequency distribution of the reminder test, based on the initial test was significant ($P < 0.001$). Also, the McNemar test showed that the changes in the reminder test based on the initial test had a significant difference ($P = 0.031$).

Conclusion: It seems that in the endemic and developing areas, the PPD booster is applicable for diagnosing latent tuberculosis in patients with rheumatoid arthritis.

Key Words: Booster, PPD, rheumatoid arthritis, tuberculin test

Address for correspondence:
Dr. Mohsen Meidani, Department of Infectious Diseases, Isfahan University of Medical Sciences, Isfahan, Iran. E-mail: meidani@med.mui.ac.ir
Received: 14.04.2013, **Accepted:** 21.07.2013

Access this article online	
Quick Response Code:	Website: www.advbiores.net
	DOI: 10.4103/2277-9175.156638

INTRODUCTION

Rheumatoid arthritis (RA) is one of the most common autoimmune diseases with a chronic and inflammatory procedure to systemic involvement associated with immune system disorders, particularly cellular immunity. The disease exists in all regions and races of the world, and its prevalence has been reported as 1% on an average. The prevalence of the disease is three times more common in women than men and the onset age in women is in the age range of

How to cite this article: Salesi M, Meidani M, Meshkinfar S, Hashemi H, Farajzadegan Z. Purified protein derivative test and its booster phenomenon in patients with rheumatoid arthritis. Adv Biomed Res 2015;4:80.

30-60s years, and it is slightly higher in men.[1] In addition to creating signs and symptoms in patients, due to involvement of cellular immunity and the corticosteroid drugs taken by the patient, rheumatoid arthritis causes immune system weakness and may provide a ground for the activation of some latent infection or opportunists in the patient, one of which is the activation of TB-latent infection;[2] therefore, the incidence of rheumatoid arthritis may lead to the lack of the ability to appropriately respond to the PPD skin test.[3] The PPD test is the most widely used method for latent infection diagnosis, with a highly dependent response on the cellular immune function in the body.[3] So far, several studies have investigated the relationship and different results have been obtained from these studies. In the study conducted by Koher *et al.*, the prevalence of a positive PPD test has been less than that of the general population, whereas, there is no relationship between positive PPD and the chest radiograph (CXR) and high-resolution computed tomography (HRCT) findings.[2]

In the study done by Sezer *et al.*, the relationship between PPD responses in rheumatoid arthritis patients was investigated. In patients with untreated rheumatoid arthritis rather than those who were treated or patients with AS (Spondyloarthropathies), as well as in the control group, there were significant lower level PPD indurations.[4] In addition, no relationship was found between the disease activity score (DAS28) and PPD in both the treated and untreated groups and similarly there was no relationship between the reactions of the acute phase and PPD. Less PPD response in patients who were not treated with immunosuppressive therapy was caused by the disease activity itself and not by the used drugs. Another important issue is that we need to doubt the response to PPD in patients with rheumatoid arthritis, especially when we are on the verge of administering anti-tumor necrosis factor (TNF) drugs to patients. In the study conducted by Tamporna, it has been shown that high-dose steroids are significant and independent factors for a negative PPD and MTX is a significant and independent factor that causes anergy to the PPD test.[5]

Ponce *et al.* investigated the response to PPD in patients with rheumatoid arthritis and compared it with the control group. The average size of induration in patients with rheumatoid arthritis was significantly lower than that of the control group. They concluded that a PPD skin test was not an appropriate test for diagnosing latent tuberculosis in patients with rheumatoid arthritis.[1,6] The present study aimed to investigate the PPD test booster in the diagnosis of latent TB, in patients with rheumatoid arthritis, in the Al Zahra Medical Center, Isfahan.

MATERIALS AND METHODS

The present study is a cross-sectional study conducted in the Al Zahra Medical Center, Isfahan, in 2012. The studied population consisted of patients having rheumatoid arthritis, receiving a glucocorticoid dose lower than 20 mg, who were referred to the center for treatment.

The inclusion criteria included having rheumatoid arthritis with the American College of Rheumatology (ACR) criteria, lack of active TB, taking corticosteroids less than 20 mg, for less than three weeks, not in touch with patients with active TB, no history of allergic reactions to PPD, and the individual's consent to participate in the study. Also, in cases where the quality of doing the test was not appropriate, such as, subcutaneous injection and the like, patients who did not return to read the test result after 72 hours, those whose response to their PPD test was reported as zero, and patients with other known diseases such as diabetes mellitus (DM), chronic renal failure (CRF), malignancy, active infection, hepatitis, acquired immunodeficiency syndrome (AIDS), and pneumonia were excluded from the study.

In the current study, the sampling method was an easy one and the sample size needed for the study was estimated to be a total number of 100 individuals.

In the study, after obtaining written consent, the patients were injected with the PPD solution made in the Razi Pharmaceutical Company, to the amount of 0.1 cc of 5 units intracutaneously (Mantoux) in the volar forearm, and were not done in places of skin where there were wounds or visible veins, as there could be a possibility of disorder in the test. The injection was given at a minimum distance of 30 mm from these places.

In the interpretation of the next 72 hours, the test was assessed by an experienced person as a single blindness, and seven days later, the patient referred again and the PPD booster test was performed with the previous value intracutaneously, at a distance of 10 cm from the previous injection site, in the same method, and was reviewed and interpreted in terms of induration 72 hours later. Indurations less than 5 mm have been considered negative and indurations greater than or equal to 5 mm have been considered positive in RA patients. The data were analyzed by SPSS software version 20.

RESULTS

In the present study, 106 patients with rheumatoid arthritis were selected and studied. The mean age of the patients was 48.3 ± 15.3 years with the range of 15-85 years.

Among the patients, 14 individuals (13.2%) were male and 92 (86.8%) were female. The mean ages of the studied males and females were 47.8 ± 15.4 and 51.2 ± 14.9 years, respectively, and according to the t-test, no significant difference was observed between the two sexes ($P = 0.46$). In Figure 1, the age distribution of patients has been shown for each sex.

The mean duration of RA was 6.3 ± 1.8 years. In males and females, the mean duration of suffering was 4 ± 1.7 and 6.7 ± 1.4 years, respectively, and according to the t test, no significant difference was observed between the two genders ($P = 0.44$).

Five patients (4.8%) took methotrexate only, one patient (1%) had prednisone only, 21 patients (20%) had methotrexate + prednisone, three patients used prednisolone + hydroxychloroquine, and 75 patients (71.4%) received the triple-drug therapy including methotrexate + prednisone + hydroxychloroquine. The mean prednisone dose was 11.1 ± 7.3 mg with a range of 5-25 mg. Also, the mean duration of prednisone was 2.6 ± 1.2 years in the range of 1-6. The mean dose of methotrexate was 7.6 ± 2.3 mg with the range of 2.5-10 and the mean duration of using this drug was 2.26 ± 1.27 years in the range of 1-7. In these patients, the mean dose of hydroxychloroquine was 210.5 ± 1.7.8 mg. The amount and dose distribution of corticosteroids and methotrexate usage has been shown in Figure 2.

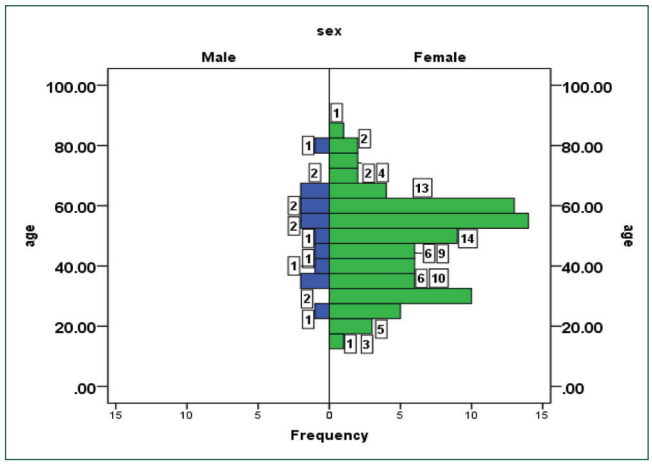

Figure 1: The distribution of age frequency for each sex

In the studied patients, the initial PPD induration mean was 4.2 ± 5.6 in the range of 0-40, and accordingly, the test result was positive for 19 patients (17.9%) and negative for 87 patients (82.1%). Also, the mean of PPD skin induration was 6.35 ± 8 in the range of 0.48 during the seven days after injecting the reminder dose, and accordingly, 25 patients (23.6%) had a positive test result and 81 patients (76.4%) had a negative test result. In Table 1, the frequency distribution of the PPD test result has been indicated in the initial and reminder injections. According to the table, 19 patients had a positive result in the initial and reminder tests and 81 patients had a negative result in both the tests. It should be noted that six patients (6.9%) having a negative result in the initial test had a positive result in reminder test, and there was no case in which the initial test result was positive and the reminder test result was negative. In the mentioned data, doing a chi-square test indicated that the frequency distribution of the reminder test results had a significant difference based on the initial test ($P < 0.001$). Also, performing the McNemar test showed that changes in the reminder test had a significant difference based on the initial test ($P = 0.031$). It should be mentioned that conducting the logistic regression test on the obtained data showed that none of the mentioned variables (including age, sex, type of drug, dosage, and duration of drug use) had any significant effect on the positivity of the PPD test in the patients ($P > 0.05$). In Table 2, the positivity odds ratio of the skin test has been shown in terms of the mentioned variables and in Figure 3, after the initial and reminder injections, the induration distribution of the skin test has been indicated.

DISCUSSION

The main objective of the study was to determine the immunoresponse to the PPD test and the booster effect of the test on patients with rheumatoid arthritis.

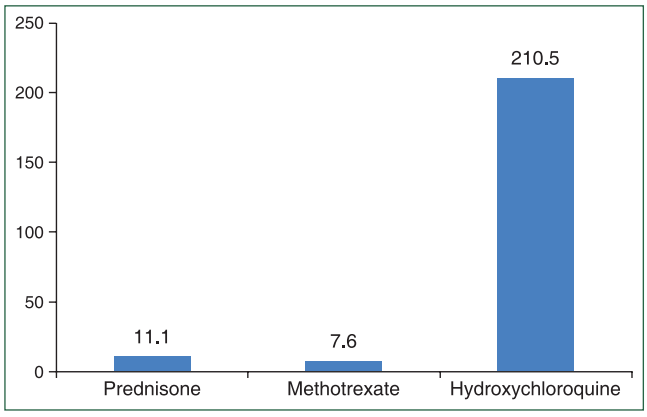

Figure 2: Median, range and percentile of 25% and 75% for amount and duration of corticosteroids and methotrexate usage

Table 1: The frequency distribution of the reminder test result based on the initial test

Reminder test	Positive		Negative		Total	
	Percentage	Number	Percentage	Number	Percentage	Number
Positive	100	19	6.9	6	23.6	25
Negative	0	0	93.1	81	76.4	81
Total	100	19	100	87	100	106

X2; $P < 0.001$–McNemar; $P = 0.031$

Table 2: The odds ratio, confidence range, and the level of significance in the variables in the positivity of the test

Variable	Regression coefficient	Standard deviation	Odds ratio	Confidence range	*P*
Sex	1.25	1.08	0.25	0.42-29.2	0.25
Age	−0.048	0.04	0.18	0.89-1.02	0.18
Disease duration	0.12	0.14	0.38	0.86-1.5	0.38
Prednisolone dose	0.22	0.2	0.28	1.24-1.8	0.28
MTX dose	0.18	0.26	0.48	0.73-2	0.48
Chloroquine dose	0.02	0.02	0.26	0.99-1.06	0.26
Prednisolone usage duration	-0.74	1.04	0.48	0.06-3.6	0.48
MTX usage duration	0.35	0.93	0.7	0.23-8.7	0.7

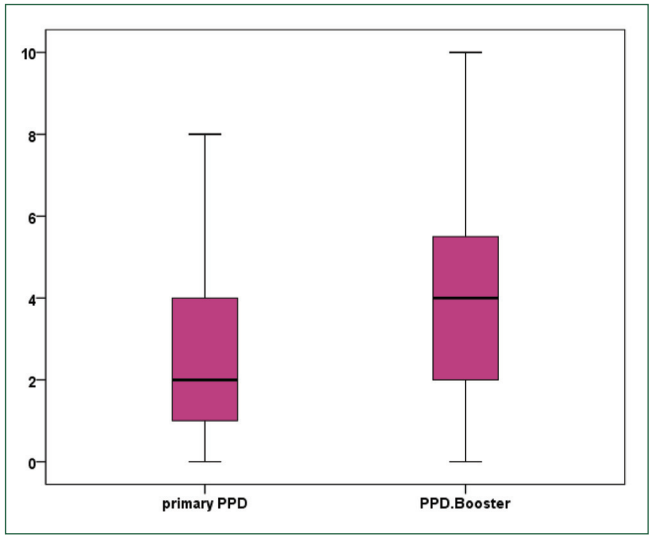

Figure 3: Median, range and percentile of 25% and 75% for the indurations of skin test after initial and reminder injections

According to the results of our study, 25 patients (23.6%) had a positive test result and 81 patients (76.4%) had a negative test result. The results were positive in the initial and reminder tests in 19 patients, 81 patients had a negative result in both tests. It should be mentioned that six patients (6.9%), who had a negative result in the initial test, had a positive result in the reminder test, and the frequency distribution of the reminder test result was significantly different based on the initial test. Also, performing the McNemar test showed that changes in the reminder test had a significant difference based on the initial test, which indicated the booster effect on the positivity of the PPD reminder test in the studied individuals, and although it seemed that due to the initial contact with TB and vaccination there was a Bacilli antibody in the bodies of a high percentage of the community and PPD was positive, in the present study, less than a quarter of the individuals had positive PPD, which could be due to the lack of contact with the community people with TB for a longer time. In a similar study conducted on the Sari Municipal Street Cleaners by Ahanjan *et al.,* in 1996, 91% of the individuals under PPD had an induration diameter between 10 and 15 mm.[5]

Using immunosuppressive drugs was another factor affecting the weakness of the immune system and the lack of appropriate cellular immune response to PPD, which was examined in our study, and although in the negative test group, a high percentage of patients had each taken three drugs of Prednisolone, Methotrexate, and hydroxychloroquine, the difference between the two groups was not significant. Also, a similar study done by Ponce de León D, in 2005, had similar conclusions and the result of the PPD skin test showed no significant difference in patients with and without rheumatoid arthritis. The disease duration was also not different in the positive and negative test results.[6] In the Nizam and Emery study performed in 2006, the response of PPD skin test was compared in two groups of healthy subjects and patients with rheumatoid arthritis. In the patients group, the response frequency to the mentioned test was significantly lower than in thehealthy group.[1] In other studies conducted to examine the relationship between tuberculosis and rheumatoid arthritis, it has been shown that in 40% of the people with lupus, the rheumatoid factor was positive.[7] In the study conducted by Kleinert and colleagues in 2012, 1529 patients with rheumatoid arthritis

were examined using the PPD test and 11.3 % of the individuals were positive. According to the results of this study, the prevalence of latent tuberculosis in patients was 8%, while in our study; the incidence was 9/17%.[8] In a study by Dr. Meidani *et al.* on three-stage tuberculin testing in hemodialysis patients, in an area with similar epidemiology, the amount of the induration of the PPD test and PPD booster were significantly different.[9] In our study, which was done on RA patients, the results were also significantly different.

In our study, the dose of drugs and duration of drug use among the test groups did not show significant differences between the positive and negative tests and such results were obtained, while other studies showed decreased levels of cellular immunity in drug users.

In general this study revealed significant differences between the booster PPD and PPD test alone, and also, performing a PPD booster was recommended to identify cases of latent tuberculosis in patients with RA.

ACKNOWLEDGMENT

The authors thank Mr. Ali Mehrabi for doing his best with the data analysis in the present study.

REFERENCES

1. Nizam S, Emery P. Attenuated response to purified protein derivative in patients with rheumatoid arthritis. Ann Rheum Dis 2006;65:980.
2. Köker IH, Pamuk ON, Karlikaya C, Tunçbilek N, Cakir N. A low prevalance of purified protein derivative test positivity in Turkish patients with rheumatoid arthritis. Association with clinical features and HRCT findings. Clin Exp Rheumatol 2007;25:54-9.
3. Hatemi G, Melikoglu M, Fresko I, Masatlioglu S, Tascilar K, Yazici H. Infliximab does not suppress the tuberculin skin test (purified protein derivative). J Rheumatol 2007;34:474-80.
4. Sezer I, Kocabas H, Melikoglu MA, Arman M. Positiveness of purified protein derivatives in rheumatoid arthritis patients who are not receiving immunosuppressive therapy. Clin Rheumatol 2009;28:53-7.
5. Ahanjan M, Nasrollahi M. Evaluation of tuberculin test (ppd) in street cleanser of sari's tawn. J Mazandaran Med Sch 1996;27:40-3.
6. Ponce de LD, Acevedo VE, Sánchez TA, Cucho M, Alfaro J, Perich R, *et al.*. Attenuated response to purified protein derivative in patients with rheumatoid arthritis: Study in a population with a high prevalence of tuberculosis. Ann Rheum Dis 2005;64:1360-1.
7. Modarres B. Review of rheumatologic from laboratory point of view. Available from: http://www.drmodarreslab.ir/index.php [Last accessed on 2013 July 7].
8. Kleinert S, Tony HP, Krueger K, Detert J, Mielke F, Rockwitz K, *et al.*. Screening for latent tuberculosis infection: Performance of tuberculin skin test and interferon-release assays under real-life conditions. Ann Rheum Dis 2012;71:1791-5.
9. Meidani M, Mortazawi Najafabadi M, Emami Naeeni A, Khorvash F, Rezaie N. Booster Phenomenon of 3-Step Tuberculin SkinTesting in Hemodialysis Patients. Isfahan Med Sch 2011;116:1049-55.

Source of Support: Isfahan University of medical sciences, **Conflict of Interest:** None declared.