

Sensitivity to mycobacterial antigens

SALEM F. DAMLUJI¹

In a study carried out in 1971, each of 3 030 persons was tested simultaneously with tuberculin RT 23 and two other PPD sensitins—PPD-S and PPD-A, PPD-B and PPD-Y, or PPD-G and PPD-F. Of the 2 558 persons whose reactions were read, 20.2% had previously been vaccinated with BCG and 79.8% were unvaccinated. RT 23 and PPD-S produced almost identical mean indurations. Both elicited much higher reactions than the sensitins prepared from atypical mycobacteria. Correlation of the sizes of induration obtained with RT 23 and with the other sensitins showed that low-grade sensitivity was due to cross-reactions to human tuberculous infection and that in no case did an "atypical" sensitin produce an induration of wider diameter than RT 23 did. It is concluded that nonspecific tuberculin sensitivity was not demonstrated in this study.

Since 1954, testing with tuberculin (RT 23 with Tween 80) has been carried out in Iraq for epidemiological purposes. Low-grade sensitivity has been observed in persons below the age of 15 years. In a study of 4 227 Iraqis of various age groups, Nyboe (1) found that, in young individuals, most reactions measured between 0 mm and 3 mm in diameter, and few were between 4 mm and 9 mm. So far, no investigation has been made to determine the nature of this finding.

The present study, the purpose of which was to collect information on the frequency of sensitivity to various mycobacterial sensitins, was carried out before the health authorities in Iraq instituted a compulsory programme of mass BCG vaccination of the newborn.

MATERIALS AND METHODS

Study population and tuberculin tests

All persons who attended the outpatient clinic of the Tuberculosis Control Centre, Baghdad, during the period February–September 1971 were included in the study. Most of the persons tested were healthy. The results may therefore be considered as representative of the general population of Baghdad.

Every participant was given three Mantoux tests simultaneously: a group of 1 001 persons received RT 23, PPD-S, and PPD-A; another group (1 024 persons), RT 23, PPD-B, and PPD-Y; and a third

group (1 005 persons), RT 23, PPD-G, and PPD-F. The dose of RT 23 was 2 TU. PPD-S and the "atypical" sensitins (PPD-A, PPD-B, PPD-F, PPD-G, and PPD-Y) were used at a dilution of 0.001 g per litre. The tuberculins were kept under refrigeration until used.

The criterion for allergy to atypical mycobacterial infection was that the reaction to atypical PPD should be 25% greater than that to RT 23 in strong reactors to RT 23 (diameter of induration: ≥ 10 mm), and that the diameter of induration should measure at least 10 mm in nonreactors to RT 23. Cases in which reactions of equal size were produced by "atypical" PPD and by RT 23, or in which the reaction to the former exceeded that to the latter by less than 25%, were considered as suspicious cases in which tuberculous and atypical mycobacterial infection could not be differentiated.

RESULTS

In total, 3 030 persons (1 341 males and 1 689 females) were tested. Their ages ranged from a few months to 80 years. Of those tested, 615 (20.3%) had previously been vaccinated with BCG and 2 415 (79.7%) had not. The test results were read in 2 558 persons (84.4%). X-rays and laboratory tests revealed 65 cases of tuberculosis in this series.

Sensitivity to the 7 PPD products used

Table 1 shows the sizes of reactions to the 7 sensitins used in the unvaccinated group.

¹ Professor and Head of the Department of Medicine, Medical College, Baghdad University, Baghdad, Iraq.

Table 1. Distribution of reactions to 7 PPD sensitins by size of reactions in unvaccinated persons

Diameter of induration (mm)	RT 23		PPD-S		PPD-A		PPD-B		PPD-F		PPD-G		PPD-Y	
	No.	%												
0	524	25.7	168	24.3	283	40.9	376	54.3	437	66.4	256	38.9	265	38.3
1-2	29	1.4	17	2.5	98	14.1	114	16.5	132	20.1	54	8.2	36	5.2
3-4	41	2.0	11	1.6	78	11.2	91	13.2	63	9.6	77	11.7	35	5.1
5-6	17	0.8	7	1.0	58	8.4	52	7.5	14	2.2	40	6.1	43	6.2
7-8	20	1.0	11	1.6	72	10.4	34	4.9	6	0.9	48	7.3	45	6.5
9-10	78	3.8	31	4.5	56	8.1	17	2.5	4	0.6	40	7.3	72	10.4
11-12	118	5.8	51	7.4	26	3.8	7	1.0	1	0.1	70	10.6	78	11.3
13-14	188	9.2	74	10.7	13	1.9	1	0.1	—	—	44	6.7	73	10.5
15-16	255	12.5	91	13.1	6	0.9	—	—	—	—	11	1.7	26	3.7
17-18	255	12.5	100	14.4	2	0.3	—	—	1	0.1	5	0.8	8	1.2
19-20	192	9.4	71	10.3	0	—	—	—	—	—	4	0.6	8	1.2
21-22	148	7.2	29	4.2	0	—	—	—	—	—	—	—	3	0.4
23-24	110	5.4	23	3.3	0	—	—	—	—	—	—	—	—	—
25-26	32	1.6	6	0.9	0	—	—	—	—	—	—	—	—	—
27-28	25	1.2	2	0.3	0	—	—	—	—	—	—	—	—	—
29-30	10	0.5	0	—	—	—	—	—	—	—	1	0.1	—	—
Total	2 042	100	692	100	692	100	692	100	658	100	658	100	692	100
Mean size		12.1		11.5		3.4		1.9		0.9		4.9		6.0

RT 23. The mean size of induration was 10.3 mm in the vaccinated group, compared with 12.1 mm in the unvaccinated group. The distribution of reactions between the sexes was almost identical. Older age groups had larger reactions.

PPD-S. Reactions in both vaccinated and unvaccinated groups showed a bimodal distribution. The mean induration in the vaccinated group was 8.1 mm, compared with 11.5 mm in the unvaccinated group.

PPD-A. The mean size of indurations was 2.9 mm in the vaccinated group, as against 3.4 mm in the unvaccinated group. In both groups, most reactions were either negative or weak.

PPD-B. The number of positive reactor was small. There was no appreciable difference between the sexes in the size of reactions. The mean size of indurations was 1.5 mm in the vaccinated group, compared with 1.9 mm in the unvaccinated group.

PPD-F. Most of the persons tested had negative or weak reactions and there was no difference between the sexes in this respect. The mean size of

reactions was 0.4 mm in the vaccinated group and 0.9 mm in the unvaccinated group.

PPD-G. In the vaccinated group, the proportion of males above the age of 20 years who had reactions was higher than that of females in the same age group. However, the proportion of reactors was almost identical for both sexes. In the unvaccinated group, more females than males were tested and a greater number of females reacted with an induration measuring 5 mm or more in diameter, though the difference was not significant. The mean size of reactions was 3.7 mm in the vaccinated group, compared with 4.9 mm in the unvaccinated group.

PPD-Y. In the vaccinated group, larger reactions were seen in females than in males, whereas, in the unvaccinated group, an equal distribution between the sexes was observed. The mean sizes of reactions were 4.4 mm and 6.0 mm in the vaccinated and unvaccinated groups, respectively.

Tuberculin sensitivity in confirmed cases of pulmonary tuberculosis

In all, 666 sputum specimens were examined and

a positive result for acid-fast bacilli by direct microscopy and/or culture was obtained in 65 cases (33 males and 32 females). The mean sizes of indurations resulting from tuberculin testing in this group were as follows: RT 23, 15.7 mm; PPD-S, 16.7 mm; PPD-A, 3.7 mm; PPD-B, 1.9 mm; PPD-F, 0.6 mm; PPD-G, 3.6 mm; and PPD-Y, 6.3 mm.

DISCUSSION

Atypical mycobacteria induce cross-reactions to tuberculin and can interfere with the identification of persons infected with *Mycobacterium tuberculosis*. By simultaneous testing with different mycobacterial sensitins, it has been found that reactions to heterologous PPD sensitins are generally weaker than those to the homologous sensitin (2, 3).

In Iraq, strong reactions to 2 TU of tuberculin RT 23 were always attributed to infection with *M. tuberculosis* (1). Low-grade sensitivity was observed in younger age groups. The object of the present study was to determine the prevalence and nature of low-grade sensitivity by simultaneous testing with various mycobacterial sensitins.

Among the study population, 615 persons (20.3%) had been vaccinated previously with BCG. The highest rates of vaccination were observed in the age groups 10–14 years (47.8%) and 15–19 years (42.1%). Children under the age of 5 years, in whom the risk of exposure and infection is high, showed a vaccination rate of 10.6%.

The prevalence of tuberculous infection was high. Of 2 042 unvaccinated subjects, 1 408 (68.9%) reacted with an induration measuring more than 9 mm in diameter when tested with 2 TU of RT 23. In the older age group (>20 years), this size of reaction was found in 87.5% of the males and 78.5% of the females tested. The higher prevalence rate of tuberculous infection in adults and the low vaccination rate in children made it necessary for the health authorities to implement a programme of BCG vaccination for all infants at birth.

In the older age groups the mean size of reactions was larger, and the number of strong reactors was greater, than in the younger age groups. This was observed in both the vaccinated and the unvaccinated groups. Although one would expect a decrease in the size of reactions with advancing age in vaccinated individuals, this was not the case—probably on account of superinfection with tubercle bacilli.

Small reactions to RT 23 could either be due to a low level of specific sensitivity in persons who have

been infected with *M. tuberculosis* or to low-grade, nonspecific sensitivity in those infected with atypical mycobacteria. To determine the nature of this low-grade sensitivity and whether it was caused by infection with *M. tuberculosis* or with atypical mycobacteria, simultaneous testing with RT 23 (or PPD-S) and other sensitins is necessary. A higher reaction to RT 23 than to other sensitins is more likely to be due to infection with *M. tuberculosis*. Criteria for nonspecific sensitivity have been given at the beginning of this report (see page 279).

Comparative studies of simultaneous testing with RT 23 and with "atypical" sensitins revealed that reactions to RT 23 were much larger in size than those "atypical" sensitins. Table 2 shows the correlation by size of reaction (diameter of induration) to RT 23 and to PPD-A in 42 unvaccinated children. According to our criteria, there was no case in which allergy could be attributed to atypical mycobacterial infection. The mean size of indurations was the largest for PPD-Y, followed by PPD-G, PPD-A, PPD-B, and PPD-F.

Study of the tuberculin reaction in confirmed cases of tuberculosis showed that the mean size of reac-

Table 2. Correlation of reactions to RT 23 and PPD-A by diameter of induration in 42 unvaccinated children 10–14 years old

Diameter of induration (mm)	RT 23	PPD-A
0	20	27
1–2	0	6
3–4	0	4
5–6	0	2
7–8	1	2
9–10	1	—
11–12	1	—
13–14	6	1
15–16	0	—
17–18	1	—
19–20	4	—
21–22	4	—
23–24	2	—
25–26	1	—
27–28	1	—

tions to all "atypical" sensitins was far smaller than that to RT 23 and PPD-S. One would conclude, therefore, that most reactions to "atypical" sensitins represent cross-reactions among infected persons.

Little is known of sensitivity to "atypical" myco-

bacterial sensitins in the Near East and Middle East. We are aware of one similar study made in Turkey by Kocacitak in 1966 (4), in which allergy to atypical mycobacteria was found in 17 cases (2.44%) in an unvaccinated group and in 23 cases (3.73%) in a BCG-vaccinated group.

ACKNOWLEDGEMENTS

I am grateful to Dr Lydia Edwards, US Public Health Service, for supplying the typical mycobacterial sensitins; to Dr Adib Al-Fukaiki, Director, Tuberculosis Control Centre, Baghdad, for his help in conducting the study;

and to Mrs Adiba Dalali and Mrs Amira Dalali for performing the tuberculin testing.

This study was supported by a grant from the University of Baghdad.

RÉSUMÉ

SENSIBILITÉ À DES ANTIGÈNES MYCOBACTÉRIENS

De février à septembre 1971, on a pratiqué sur 3 030 habitants de Bagdad (Irak) des épreuves cutanées (tests de Mantoux) avec divers antigènes mycobactériens. Chaque personne a subi un test avec une tuberculine RT 23 et, simultanément, des tests avec deux sensitines PPD (PPD-S et PPD-A; PPD-B et PPD-Y; ou PPD-G et PPD-F). La lecture des réactions a pu être effectuée chez 2 558 sujets (84,4%) parmi lesquels 516 (20,2%) avaient subi antérieurement une vaccination par le BCG et 2 042 (79,8%) n'avaient jamais été vaccinés.

Le RT 23 et le PPD-S ont provoqué des indurations de diamètre moyen quasi identique et de loin supérieur

à celui des indurations produites par les autres sensitines. L'étude des corrélations entre la taille des indurations dans l'épreuve au RT 23 et celle des indurations suscitées par les sensitines de mycobactéries atypiques montre que la sensibilité de faible niveau est due à des réactions croisées chez des sujets infectés par le bacille tuberculeux. En aucun cas, une sensitine de mycobactérie atypique n'a entraîné une réaction de dimensions supérieures à celles des réactions au RT 23. La conclusion des auteurs est que la présente étude n'a mis en évidence aucune sensibilité tuberculitique non spécifique.

REFERENCES

1. NYBOE, J. *Bulletin of the World Health Organization*, **22**: 5 (1960).
2. EDWARDS, L. B. ET AL. *In: Proceedings of the XVI International Tuberculosis Conference*, Toronto, Canada, 10-14 September 1961, vol. 2, p. 384.
3. LJUBISAVLJEVIC, S. ET AL. *In: Selected papers of the Royal Netherlands Tuberculosis Association*. The Hague, 1973, vol. 15, p. 5.
4. KOCACITAK, D. 1965 Yilinda Yozgat ve Ankarada nonspesifik allerji yonunden cesitli mikobakteriyel sensitinlerle "PPD-RT23, PPD-S, PPD-G, PPD-6, NT-S18, PPD-A" yapılan arastirmalar. Ankara, Ankara Basim ve Ciltevi, 1966 (in Turkish).