Diagnosis of latent tuberculosis in individuals with recent exposure: tuberculin skin test versus interferon-γ release assay

M. REZA SHOKROLLAHI*, S. NOORBAKSH†, L. NASEHI‡ and Z. MOVAHEDI*

Department of Pediatric Infectious Diseases, School of Medicine, Qom University of Medical Sciences and Health Services, Qom; †Research Center of Pediatric Infectious Diseases, School of Medicine, Iran University of Medical Sciences, Tehran, Iran; and ‡Department of Radiology, Ronald Reagan Hospital, David Geffen School of Medicine' University of California Los Angeles, USA

It has been reported that up to a third of the world's population is infected with *Mycobacterium tuberculosis*. The stimulated rate of latent tuberculosis infection (LTBI) is approximately two billion cases worldwide. Identification and treatment of LTBI has an important role in elimination of tuberculosis (TB) as 5–10% of LTBI cases could develop active tuberculosis.[1,2]

Until recently, the tuberculin skin test (TST) has been the only test for identification of LTBI, but it has some technical problems in terms of interpretation due to false-positive and -negative results from cross-reactivity of purified protein derivative (PPD) with other mycobacteria such as Bacillus Calmette-Guerin (BCG) vaccine and *Mycobacterium avium*.[3,4]

The diagnostic value of this test is affected by the booster phenomenon, the quality of substance used and the need for multiple visits.[5] New tests based on detecting interferon-γ (IFNγ) released from T lymphocytes in response to *M. tuberculosis* antigens (e.g., early-secreted antigenic target 6 protein [ESAT-6] and culture filtrate protein 10 [CEP-10]) that are absent from BCG and other *Mycobacterium* strains, would be of great practical use, especially in industrialised countries.[6,7]

Now, two IFNγ-releasing assays (IGRAs), including the QuantiFERON TB Gold in-tube test (QFT) and the Spot TB Test (T-Spot) are available. These tests have proved to be more sensitive and specific than TST and would not be affected by previous BCG vaccination.[5,8,9] The World Health Organization (WHO) suggests that BCG immunisation should be available in Iran and therefore the present study aims to compare TST and QFT for detection of LTBI in family members of patients with confirmed TB in a tertiary centre in Tehran, Iran.

The cross-sectional study was undertaken in 2009–2010 and included patients (n=59) admitted to the Department of Pulmonary Diseases for detection of LTBI. Written informed consent was obtained from each participant.

Tuberculous disease was established from patient history, clinical finding, chest X-ray, positive TST and also positive smear or culture for *M. tuberculosis*. Fifty-nine cases were enrolled in the study, and data including age and gender were obtained by questionnaire.

An intradermal Mantoux test (five units of tuberculin PPD) was performed on each participant by a trained nurse. Investigators injected 0.1 mL PPD solution into the forearm. The result of the test was evaluated 48–72 hours after the injection. Results were classified as negative if TST induration was smaller than 5 mm in diameter, and positive if the reaction was ≥5 mm in diameter.[10]

QuantiFERON-TB Gold (QFT; Cellestis, Carnegie, Victoria, Australia) was performed according to the manufacturer's instructions. Tests were performed <12 hours after collection and aliquots of heparinised whole blood were incubated with the test antigens for 16–24 hours. Phytohaemaglutinin (a mitogen) was used as a positive control and saline was used to measure the background level of IFNγ. After incubation, the concentration of IFNγ in plasma was determined by enzyme-linked immunosorbent assay (ELISA).[11] Based on the manufacturer's information, a positive IFNγ test score was considered to be ≥0.35 iu/mL.

All analyses were performed using SPSS version 16. χ^2 and Student's t-test were used to compare the two methods. $P<0.05$ was considered significant. Accuracy of the IGRA was evaluated by sensitivity and specificity results, with 95% confidence interval (CI).

Mean age of the participants was 21.42±17.84 (range: 1–69) years, 25 (42.4%) were male and 34 (57.6%) female. Mean family size was 3.61±0.929 (range: 1–8 persons). Mean diameter of TST induration was 6.66 mm (range: 0–40 mm). In 12(20.3%) cases, induration was not detected, in 33 (55.9%) it was 5–10 mm, in 10 (16.9%) it was 10–15 mm and in four (6.8%) cases it was >15 mm. If 5-mm induration is regarded as a positive TST, 12 (20%) cases were TST-negative and 47 (80%) were TST-positive. Using the QFT method, negative and positive result was seen in 14 (24%) and 45 (76%) cases, respectively (Table1).

Test sensitivity and specificity are shown in Table 2. Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) were 57%, 87%, 57%, 57%,

Table 1. Comparison of TST and QFT results in participants evaluated for latent TB infection

Results (TST/QFT)	*n*	%
Positive/positive	39	66
Negative/negative	6	10
Negative/positive	6	10
Positive/negative	8	14
Agreement between TST and QFT-GIT	45	76

Table 2. Sensitivity, specificity, PPV and NPV of QFT and TST for diagnosing latent TB infection.

Test performance	%
Sensitivity	57.14
Specificity	86.67
Positive predictive value	57.14
Negative predictive value	57.14
False positive rate	13.33
False negative rate	42.86

Correspondence to: D Zahra Movahedi
Department of Pediatric Infectious Diseases, Qom Children Hospital, Qom University of Medical Sciences and Health Services, Imam Boulevard., Qom, Iran.
Email: movahedizahra@yahoo.com

respectively. Differences between QFT results and TST-positive (2.295±3.97) and TST-negative (0.281±0.85) groups were significant (P=0.025), showing a higher rate of positive QFT in positive cases. There was intermediate overall agreement between the two tests (76%, κ=0.44).

This study compared the results of standard screening using TST with T-cell responses to *M. tuberculosis*-specific and non-specific antigens in family members of patients diagnosed with smear-positive pulmonary TB. The present comparison of QFT and TST revealed a positive QFT in 76% of cases (sensitivity specificity, PPV and NPV: 57%, 87%, 57%, 57%, respectively). The results were consistent with those obtained by Kobashi *et al*.[12] and Dogra *et al*.:[13] however, Mori *et al*.[14] showed that IGRAs have a greater sensitivity (89%) and specificity (98.1%).

In the Moyo *et al*. study, sensitivity and specificity for both tests used to diagnose TB disease were similar, and both had a low PPV, while the NPV was high. The level of agreement (κ=0.44) between TST and QFT in the present study was lower than that observed in studies in India and South Africa, which reported a κ score range of 0.73–0.79.[13–15]

Other studies in children mainly from countries with a low TB incidence have reported lower levels of agreement between QFT and TST (κ range: 0.5–0.56).[16–20] However, lower κ scores have also been reported in studies conducted in TB-endemic countries; for example, The Gambia (0.52)[21] and South Africa (0.56).[22]

Different levels of agreement could be due to the inclusion of children of different ages, different TST cut-offs, TB disease status, variation in BCG vaccination status and differing exposure to *M. tuberculosis* and non-tuberculous mycobacteria. These findings also illustrate the variable performance of IGRAs in different populations and TB incidence settings.[9,22]

In conclusion, the current results show that QFT offers greater specificity than TST and is useful for evaluation of TST-positive cases with low likelihood of LTBI. The QuantiFERON-TB test is a useful tool for TB diagnosis in Iran, where there is only moderate prevalence of TB in a largely vaccinated population. Use of QuantiFERON-TB is also useful in deciding prophylactic treatment for TB. □

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