The Intensity of QuantiFERON TB-Gold Response does not Differentiate Active from Latent Tuberculosis

Abstract:

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We analyzed positive QuantiFERON (QFT) assays, performed between July 2009 and April 2011 in the Mercy University Hospital, Cork, Ireland, which included 94 patients with latent tuberculosis (LTBI) and 111 patients with active tuberculosis. There was no difference in the intensity of response between patients with LTBI and active tuberculosis (p=0.899). In patients with LTBI, there were no correlations between age (p=0.353), sex (p=0.478), smoking (p=0.323), contact (p=0.612), Mantoux response (p=0.055), Irish nationality (p=0.768), previous BCG vaccination (p=0.504), WCC (p=0.187), lymphocyte count (p=0.786), neutrophil count (p=0.017) and the intensity of QFT response. Similarly in patients with active tuberculosis, none of these variables predicted the intensity of QFT response. Thus, the intensity of QFT response does not help to differentiate active from LTBI. The intensity of QFT response is not influenced by age, sex, smoking, remoteness of contact history, Mantoux response, nationality, CXR abnormalities, BCG vaccination and peripheral lymphocyte count.

Introduction

In 2010, there were an estimated 8.8 million incident cases of active TB globally and 1.65 million deaths. It has been estimated that up to one third of the world’s population has latent TB infection (LTBI). However, this figure is estimated from data obtained from tuberculin skin testing (TST). Using TST to diagnose LTBI may cause false-positive results in individuals who have been vaccinated with bacille Calmette et Guérin (BCG) or exposed to environmental mycobacteria. Furthermore, false-negative results occur in immunosuppressed individuals, suggesting that the estimated global-prevalence of LTBI may be inaccurate. The QuantiFERON-TB Gold test (QFT) is based on a whole-blood ELISA developed in the late 1980s and is approved for in vitro diagnosis of tuberculosis infection by the U.S. Food and Drug Administration. Guidelines from the U.S. Centers for Disease Control and Prevention (CDC) in the use of interferon-gamma release assays (IGRAs), to diagnose Mycobacterium tuberculosis infection have been published and incorporate recent data evaluating both LTBI and active TB. The specificity of the QFT assay is higher than the TST, as it is not affected by previous BCG vaccination. In addition, this test is less influenced by anergy than TST. IGRAs could therefore improve existing information about the global prevalence of LTBI.

The QFT assay is of value in distinguishing true positive TST reactions in patients with LTBI from false positive reactions in those who have been vaccinated with BCG, but is also useful in the diagnosis of active TB, particularly in those without culture confirmation. There are some limitations to the QFT assay. A negative QFT test does not exclude TB disease in immunosuppressed patients. The sensitivity of QFT partly depends on peripheral lymphocyte counts. Therefore in the presence of lymphopenia, the ELISPOT assay is superior to QFT for detecting tuberculosis infection. Age-related and time constraints may cause difficulty in completing QFT. Blood samples must be processed within 8-16 hours. Also, limited data existing on use in children younger than 5 years of age, persons recently exposed to TB, and those who will be tested repeatedly.

It is not clear whether the intensity of QFT response can help to distinguish LTBI from active TB. Several studies have reported varying results in this respect. Metcalfe showed that higher quantitative IFN-γ results were associated with active tuberculosis in a cohort of patients from San Francisco and Kobashi demonstrated significantly higher QFT response in patients with active TB in Japan. In contrast, other studies have suggested that IGRAs cannot distinguish TB infection from disease in children and that concentrations of IFN-γ did not differ in children with LTBI and TB either before or at the end of treatment.

We sought to determine whether the intensity of response in patients with a positive QFT assay was predictive of active over latent tuberculosis, and whether other factors determined the intensity of response in adults with latent tuberculosis.

Methods

A retrospective analysis of 129 immunocompetent patients from Mercy University Hospital, Cork, Ireland with a positive QFT test was conducted. The study population consisted of 35 patients with active tuberculosis and 94 patients with LTBI. LTBI was defined as per CDC guidelines with no symptoms or physical signs suggestive of active TB, normal radiologic findings with positive TST or QFT and negative smear and culture of respiratory specimens. The majority of the patients that were included in the study had been referred to the TB outpatient clinic in the Mercy University Hospital from occupational health departments following occupational screening for TB. QFT assays were taken on the day of reading of the Mantoux test, that is, within three days of Mantoux insertion.

One hundred and eleven patients had positive Mantoux tests (31 in group 1 and 80 in group 2). For the TST test, 0.1 mL of tuberculin (Nippon BCG Manufacturing Tokyo, Japan) equivalent to 2 tuberculin units of purified protein derivative (PPD) was injected intradermally into the volar aspect of the forearm, and the transverse diameter of induration was measured 48 h later. Results were interpreted by hospital staff based on CDC guidelines for tuberculin testing. Each patient in both groups had a heparinized venous blood sample collected for the purpose of performing QFT testing. The QuantiFERON-TB Gold test was performed according to the manufacturer's instructions (Cellestis, Carnegie, Australia). The test results were recorded according to the guidelines of the CDC. Detection of IFN-γ, by ELISA was used to identify in vitro responses to ESAT-6 and CFP-10 that are associated with M. tuberculosis infection. All lots of the reagents and controls for the QFT-Gold test were obtained from Cellestis. The concentrations of IFN-γ were determined within 8-16 hours. Also, limited data existing on use in children younger than 5 years of age, persons recently exposed to TB, and those who will be tested repeatedly.

The quantitative values from the TB antigen (TB) containing tubes are compared to negative (NC) and positive (mitogen tube) controls, which determine the validity of the test. The results can be indeterminate if the NC has inappropriately high levels of IFN-γ. Similarly, a low mitogen indeterminate result can occur due to an inappropriately low IFN-γ response to mitogen in the positive control. This may indicate the mishandled specimens or immunosuppression. The test is interpreted as positive with IFN-γ responses ≥ 0.35 IU/L higher than NC (i.e. TB–NC ≥ 0.35 IU/L is positive and TB–NC < 0.35 IU/L is negative test, in the absence of inappropriate nil or mitogen stimulation).

The sensitivity of QFT partly depends on peripheral lymphocyte consumption, nationality and duration of residence in Ireland if foreign born, previous TST results (in millimeters), contact history with patients with infectious tuberculosis and BCG vaccination. Laboratory and radiologic data, including white cell count (total and differential count), sputum or bronchial washing TB smear and culture results, chest radiograph, and thoracic CT results were collected. Sequential testing with Mantoux, followed by QFT in patients with positive TST results if not performed. QFT assays were taken on the day of reading of the Mantoux test, that is, within three days of Mantoux insertion.

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Information from the collected demographic data, clinical history, laboratory, microbiological and radiological findings was entered into a computerized database. The data were then analyzed using a statistical software package (Graphpad InStat, San Diego, CA) for analysis. Unpaired t-tests were used to determine differences between parametric data and the Mann-Whitney test was used to determine differences between non-parametric data. The significance of the correlation and Spearman's rank correlation coefficient for non-parametric correlation between groups. A p-value<0.05 was considered statistically significant.

Results

35 patients with active TB and 94 with LTBI were analyzed. Of the 35 active TB patients, 12 had extrapulmonary TB and 23 had pulmonary TB. In the latter, 7 patients were sputum culture positive for Mycobacterium tuberculosis (MTB) and the remainder had bronchial washing cultures that were positive for MTB. Of the 12 patients with extrapulmonary TB, 7 had TB lymphadenitis, 3 had tuberculous abscesses (1 epididymal, 1 inguinal and 1 pectoral tuberculous abscess), 1 had TB meningitis and 1 had TB parotitis.

There was no significant difference in intensity of quantiferon TB-Gold response between patients with LTBI and those with active TB (p=0.1589) (Figure 1, Table 2). In patients with latent tuberculosis, there were no correlations between age (p=0.353), sex (p=0.476), smoking status (p=0.323), contact history (p=0.612), Mantoux response (p=0.055), Quantiferon Gold (p=0.768), previous BCG vaccination (p=0.187), peripheral lymphocyte count (p=0.157) and the intensity of QFT response (Table 3). Similarly in the group with active TB group, there were no correlations between these variables and the intensity of QFT response.

Discussion

These data suggest that the intensity of QFT response cannot distinguish latent from active TB. IGRA have revolutionized the management of patients with tuberculosis. Their primary advantage is to differentiate false positive TST reactions in patients who have been vaccinated with BCG (or indeed exposed to environmental mycobacteria) from true positive TST reactions in patients infected with Mycobacterium tuberculosis. In individuals that have not been vaccinated with BCG, there can be a high degree of agreement between IGRA and TST results since the assay is not influenced by previous BCG vaccination. QFT is more reliable than the TST for identifying those who progress from LTBI to active TB. Diel reported progression to active TB in 14.8% of close contacts of active TB cases, compared to 2.3% of cases that were TST positive. Aichelburg reported 8.3% of patients who were QFT positive progressed to active tuberculosis over 19 months among outpatient HIV-positive adults. 17 Rik and colleagues showed lower rates of progression, 2.8% and 3.3%, among QFT and T-SPOT.TB-positive immigrant TB contacts.

IGRAs can also be used to help the diagnosis of active TB. The sensitivity and specificity of the QFT in patients with active TB has been reported at 70.1%-89% and 91.6%-98.1%. In a study in Italy, QFT was positive in 73.8% of pulmonary and 79.2% of extra-pulmonary active cases and indeterminate in 9.3% and 6.2% respectively. Of note, 16.1% had false negative QFT results, occurring more frequently in foreign-born patients (p=0.006). 18 Published studies to date have shown conflicting results as to whether IGRA can help to differentiate LTBI from active TB.8-11 In a way study from Singapore, the authors found no differences in QFT response between persons with LTBI and active TB but significant differences in quantitative T-cell responses as measured by the T-SPOT.TB assay. They concluded however that T-cell response may not be specific and that the intensity of QFT response cannot distinguish latent from active TB. 19, 20 A metaanalysis on the predictive value of QFT and TST for progression from LTBI to active TB disease state concluded that QFT has a higher positive and negative predictive value for progression to active TB compared with those of the TST.

In our group of adults with latent and active tuberculosis, the intensity of QFT response was not influenced by age, sex, smoking, remoteness of contact cases, Mantoux response, nationality, CXR abnormalities, BCG vaccination and WCC, peripheral lymphocyte and neutrophil count. In a study from Hamburg that examined contacts of patients with tuberculosis, QFT, but not TST, results were associated with exposure time (p<0.0001). 21 In a way study from Singapore, the authors found no differences in QFT response between persons with LTBI and active TB but significant differences in quantitative T-cell responses as measured by the T-SPOT.TB assay. They concluded however that T-cell response may not be specific and that the intensity of QFT response cannot distinguish latent from active TB. 19, 20 A metaanalysis on the predictive value of QFT and TST for progression from LTBI to active TB disease state concluded that QFT has a higher positive and negative predictive value for progression to active TB compared with those of the TST.

Recent studies have suggested that other biomarkers, including IL-15 and MCP-1 may help to distinguish LTBI from active TB. 22 Furthermore, biomarkers such as IP-10 may represent novel biomarkers for infection with MTB. 23 Although larger studies might demonstrate a statistically significant QFT response between latent and active TB, we concluded that in routine clinical practice, the intensity of QFT response cannot distinguish latent from active TB.

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References


