ARTICLE

Latent Tuberculosis Diagnosis in Children by Using the QuantiFERON-TB Gold In-Tube Test

Jennifer Lighter, MD, Mona Rigaud, MD, MPH, Roger Eduardo, BS, Chia-Hui Peng, MPH, Henry Pollack, MD

Saul Krugman Division of Pediatric Infectious Diseases and Immunology, Department of Pediatrics, New York University School of Medicine, New York, New York

The authors have indicated they have no financial relationships relevant to this article to disclose.

What's Known on This Subject

IGRA performance for adults has been reported.

What This Study Adds

This study provides information on QFT performance for children.

ABSTRACT

BACKGROUND. The QuantiFERON-TB Gold test was the first blood test to be approved for the diagnosis of latent tuberculosis infection. Although it has been shown to be sensitive and specific in adults, limited data on its performance in children are available.

METHODS. This was a prospective study of children receiving health care in New York, New York. Each child was assessed for risk factors for *Mycobacterium tuberculosis* infection, underwent tuberculin skin testing, and had a QuantiFERON-TB Gold In-Tube test performed. The concordance between tuberculin skin test and QuantiFERON-TB Gold In-Tube test results was calculated, and the results were analyzed according to the likelihood of exposure to *M tuberculosis*.

RESULTS. Data for 207 children with valid tuberculin skin test and QuantiFERON-TB Gold In-Tube test results were analyzed. There was excellent correlation between negative tuberculin skin test results and negative QuantiFERON-TB Gold In-Tube test results; however, only 23% of children with positive tuberculin skin test results had positive QuantiFERON-TB Gold In-Tube test results. Positive QuantiFERON-TB Gold In-Tube test results were associated with increased likelihood of M tuberculosis exposure, and interferon γ levels were higher in children with known recent exposure to *M tuberculosis*, compared with children with older exposure histories. Younger children produced lower interferon γ levels in response to the mitogen (phytohemagglutinin) control used in the QuantiFERON-TB Gold In-Tube test, but indeterminant results were low for children of all ages. Performance characteristics were similar across all age groups.

CONCLUSION. The QuantiFERON-TB Gold In-Tube test is a specific test for M tuberculosis exposure in children, with performance characteristics similar to those for adults residing in regions with low levels of endemic disease. Concerns about test sensitivity, especially for children <2 years of age, will require additional prospective long-term evaluation. *Pediatrics* 2009;123:30–37

www.pediatrics.org/cgi/doi/10.1542/peds.2007-3618

doi:10.1542/peds.2007-3618

This work was presented in part at the First Global Symposium on Interferon-γ Assays; February 21-22, 2007; Vancouver, BC, Canada; Keystone Symposia: Tuberculosis: From Lab Research to Field Trials; March 20-25, 2007; Vancouver, BC, Canada; and the Pediatric Academic Societies meeting; May 5–8, 2007; Toronto, ON, Canada.

Key Words

tuberculosis, children, QuantiFERON, interferon γ-releasing assay

Abbreviations

LTBI—latent tuberculosis infection
IGRA—interferon γ-releasing assay
QFT—QuantiFERON-TB Gold In-Tube test
TST—tuberculin skin test
ESAT-6—early secreted antigenic target 6
ELISA—enzyme-linked immunosorbent assay
IFN-γ—interferon γ

Accepted for publication Apr 9, 2008

Address correspondence to Henry Pollack, MD, Department of Pediatrics, New York University School of Medicine, 550 First Ave, New York, NY 10016. E-mail: henry.pollack@med.nyu.edu

MYCOBACTERIUM TUBERCULOSIS IS estimated to infect one third of the world's population and to cause the death of >450 000 children each year.[1] Young children with tuberculosis are more likely than adults to develop serious disease, because of the decreased ability of their immature immune systems to contain infection.[2,3] Forty percent of children <1 year of age who are infected with tuberculosis develop disseminated disease, compared with <1% of immunocompetent adults.[3]

It is well accepted that, to decrease the incidence of active tuberculosis in low-incidence settings, persons with latent tuberculosis infection (LTBI) need to be identified and treated. The cornerstone of LTBI diagnosis for the past 100 years has been the tuberculin skin test (TST), but it is neither very sensitive nor specific.[4] False-positive TST reactions occur because of cross-reactivity to environmental mycobacteria and BCG vaccination. This may result in the treatment of some children who actually have not been infected.[5] In addition, the TST requires 2 patient encounters, and people may not return for the test results to be read. For these reasons, considerable effort has been devoted to the development of new techniques for diagnosing LTBI in children and adults.

The method most studied to date has been based on the detection of interferon γ (IFN-γ) released by T cells after in vitro exposure to antigens from *M tuberculosis*. These IFN-γ-releasing assays (IGRAs) measure T cell responses to

the early secreted antigenic target 6 (ESAT-6) and culture filtrate protein 10, which are transcribed from region of difference 1, which is a region on the mycobacterial genome specific for *M tuberculosis* and absent in BCG and most other mycobacteria. These antigens do not cross-react with BCG or with nearly any nontuberculosis mycobacteria. In the QuantiFERON TB Gold assay (Cellestis, Carnegie, Australia), the antigens are incubated with whole blood, and IFN-γ produced by the antigen-specific T cells is measured with an enzyme-linked immunosorbent assay (ELISA). Studies in adults showed that the results of this assay correlated with exposure history and the test was specific for measuring responses to *M tuberculosis*.[6–10] In 2005, the US Food and Drug Administration approved the QuantiFERON-TB Gold assay, and the Centers for Disease Control and Prevention recommended its usage for diagnosing LTBI in "all circumstances in which the TST is currently used," including testing of children, with the caveat that the sensitivity for young children has not been determined.[11] Management decisions concerning LTBI chemoprophylaxis currently are based on either QuantiFERON assay or TST results alone. After those recommendations were published, many groups, including the New York City Department of Health and Mental Hygiene, began using only the QuantiFERON-TB Gold test for diagnosis in adults and children.[12] However, the American Academy of Pediatrics has withheld recommendation of the use of the QuantiFERON assay until more data for children are available.[13] In this study, we examined the performance of a newer version of the assay, the QuantiFERON-TB Gold In-Tube test (QFT), for children seen at a public hospital in New York, New York, an area with low levels of endemic tuberculosis, with an incidence in 2006 of 4.6 cases per 100 000 population.[14]

METHODS

Study Setting and Participants

After written informed consent was obtained, children <18 years of age were enrolled in this prospective study at Bellevue Hospital, a large public hospital serving a mixed urban population and at a private pediatric practice in New York City. Children at Bellevue Hospital were recruited from the well-child clinic, pediatric chest clinic, and pediatric inpatient ward. Clinical history findings, physical examination results, BCG scar appearance in visual inspection, and chest radiographic findings were recorded for each child enrolled in the study. HIV results were recorded if available, but testing was not performed as part of this study. Data on risk factors for possible *M tuberculosis* exposure, such as contact with an index case, were obtained by interviewing the parents or guardians. The TST was performed by using the Mantoux technique, and blood samples for the QFT were drawn directly into QFT tubes. TST results were checked 48 to 72 hours later and were considered positive when the area of induration was ≥10 mm. All children with positive TST results underwent subsequent chest radiography. Cases of active tuberculosis were diagnosed on the basis of culture, clinical, and radiologic findings. The results of the QFT were not provided to clinicians and had no effect on treatment decisions. This study was approved by the institutional review boards of New York University and Bellevue Hospital.

IFN-γ Assay

The QFT was performed according to the manufacturer's instructions. Briefly, 1 mL of blood was drawn directly into 3 separate heparinized tubes. One tube (the nil control) contained only heparin, 1 tube (the mitogen control) contained phytohemagglutinin, and 1 tube contained the *M tuberculosis*-specific antigens ESAT-6, culture filtrate protein 10, and TB7.7 (Rv2654). Each tube was rotated several times to allow the blood to coat the entire wall. Within 2 hours after venipuncture, the tubes were placed in an incubator set at 37°C. After 24 hours of incubation, the tubes were centrifuged and the plasma was collected. If the ELISA was not performed immediately, then the plasma was frozen and stored at −70°C. The amount of IFN-γ in the plasma was measured by ELISA with the reagents included in the test kit. The amount of IFN-γ released in response to *M tuberculosis*-specific antigens or mitogen was calculated after subtraction of the amount of IFN-γ in the nil control tube. The result was considered positive when the amount of IFN-γ was ≥0.35 IU/mL and ≥25% more than the nil control value, as recommended by the manufacturer and based on previous adult studies.[5] Standard testing was conducted for each ELISA, in triplicate. QFT results were calculated by using the software provided by the manufacturer.

Statistical Analyses

QFT results were compared with TST results by using the κ statistic. Because there is no standard method for diagnosis of LTBI, TST and QFT results were compared with a gradient of likely exposure to *M tuberculosis*, based on risk factors such as contact with an index case or living in a disease-endemic region. Univariate analyses of possible risk factors associated with positive QFT results was evaluated with the χ^2 test. The Mann-Whitney *U* test was used to compare IFN-γ levels. All analyses were performed by using SPSS 15.0 (SPSS, Chicago, IL), with an α of .05.

RESULTS

Study Group

A total of 253 children were enrolled in the study, 42% from the well-child clinic, 56% from the pediatric chest clinic, and 2% from the pediatric inpatient ward. Thirty-one children were receiving antimycobacterial medication at the time of enrollment and were excluded from the analysis, as were 14 additional children who were known to be HIV-infected. One child was excluded from the analysis because of a laboratory error that occurred while the assay was being set up. Table 1 shows the demographic and clinical characteristics of the 207 children included in the analysis. The mean age was 9 years (SD: 5.7 years), and 87 subjects (42%) were female. Previous BCG vaccination was reported for 74 subjects;

TABLE 1 **Patient Characteristics ($N = 207$)**

Characteristic	*n* (%)
Female	87 (42)
Age	
1–59 mo	67 (32)
≥60 mo	140 (68)
Born in disease-endemic region	69 (34)
BCG-vaccinated	74 (36)
First TST placed as part of this study	125 (60)
History of TST	82 (40)[a]
History of ingestion of unpasteurized milk	26 (13)
Household member from disease-endemic region	166 (81)
Born in United States but traveled to disease-endemic region	52 (25)
History of close contact with index case	13 (6)

[a] Results remained negative for 42 and converted for 40.

72 received a single vaccination. Fifty-nine children had visible BCG scars. Sixty-nine children (34%) were born in tuberculosis-endemic regions (rates of infection of >20 cases per 100 000), and 166 children (81%) lived in homes where a household member was from a disease-endemic region. Thirteen children had documented close contact with an adult diagnosed as having active tuberculosis (index case). For 12 children, the contact with the index case occurred within a few weeks to a few months before study entry. For 1 child, the contact occurred >6 years before study entry.

Figure 1 shows the overall results of the study. Of the 207 children included in the analysis, 3 children (1.4%) had indeterminate results, all because of a mitogen response that was below the allowable cutoff value. For 7 children, the exact dimensions of induration of positive TST results were not recorded.

TABLE 2 **Agreement Between TST and QFT Results as a Function of TST Size and BCG Exposure**

Category	*N*	κ	Agreement, %
All children	204	0.17	55
Children not vaccinated with BCG	130	0.31	72
All children with ≥5-mm induration in TST	201	0.16	53
Unvaccinated children with ≥5-mm induration in TST	130	0.31	72
All children with ≥10-mm induration in TST	200	0.19	56
Unvaccinated children with ≥10-mm induration in TST	128	0.36	66
All children with ≥15-mm induration in TST	199	0.31	79
Unvaccinated children with ≥15-mm induration in TST	128	0.60	91
All children with ≥20-mm induration in TST	199	0.30	86
Unvaccinated children with ≥20-mm induration in TST	128	0.42	91

Comparison of TST and QFT Results

Ninety-one children (44%) had negative TST results, and 116 (56%) had positive TST results. In the TST-negative group of children, 85 (93%) had negative QFT results. The overall agreement between TST and QFT results was 55% for all children and 72% for children who had never been vaccinated with BCG (Table 2). This corresponded to κ values of 0.17 and 0.31, respectively. The agreement and κ values increased when more-stringent TST cutoff criteria were used. The strongest agreement between TST and QFT results (91%) occurred in the subgroup of children who had never been vaccinated with BCG and had TST responses of ≥15 mm; this corresponded to κ of 0.60.

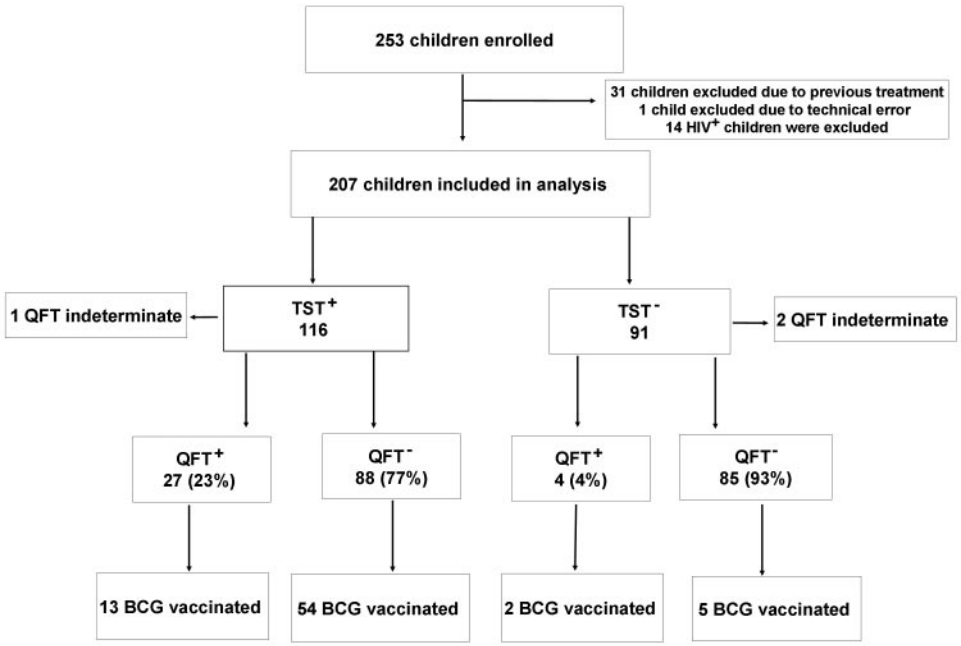

FIGURE 1
Overall results for study participants. There were 3 indeterminate responses attributable to low mitogen levels; 2 subjects had immunodeficiencies and the third subject was a 3-day-old infant.

TABLE 3 **Assay Responses for Groups of Children With Different Likelihoods/Risks of Exposure**

Exposure Risk	Exposure and Test Results	*N*	Proportion With Positive QFT Results, %
Minimal	No known risk, TST-negative	22	0
	No known risk, TST-positive	8	0
Low/moderate	Risk factors, TST-negative	62	6
	Risk factors, TST-positive	99	19
High	Known direct contact with tuberculosis index case, TST-negative	5	0
	Known direct contact with tuberculosis index case, TST-positive	8	100
Definite tuberculosis	Active tuberculosis, TST-negative	1	100
	Active tuberculosis, TST-positive	2	100

Minimal risk indicates no known risk factors for exposure to *M tuberculosis*. For low/moderate risk, the risk factors included birth in or travel to a disease-endemic region and/or living with a household member with specific risks. Specific risks for a household member included emigrating from a disease-endemic region, having HIV infection, or having a history of imprisonment, homelessness, or intravenous drug use. High risk indicates close exposure to an adult with active tuberculosis. Definite tuberculosis was confirmed with culture and/or radiologic findings.

There were 4 children who had negative TST results and positive QFT results. One child had active tuberculosis (IFN-γ level: 6.75 IU/mL) and the 3 other children had low to moderate risk factors but were healthy and completely asymptomatic. Two of these children had IFN-γ levels just above the cutoff value (0.36 and 0.39 IU/mL), and the third child had an IFN-γ level of 1.1 IU/mL.

Association Between Likelihood of *M tuberculosis* Exposure and QFT Results

The proportions of positive QFT results for children with increasing gradients of *M tuberculosis* exposure are shown in Table 3. As the likelihood of exposure to *M tuberculosis* increased, the proportion of children with positive QFT results also increased. Among the 13 children with a history of exposure to an adult with active tuberculosis, 8 (61%) had positive results for both TST and QFT and 5 had negative results for both tests. All 3 children with active tuberculosis had positive QFT results.

Among the 30 children with no known risk factors for *M tuberculosis*, none had positive QFT results, whereas 8 of those children had positive TST results (≤12-mm induration for 7 children and 15 mm for 1 child). In this "control group," the estimated specificities of the QFT and TST were 100% and 73%, respectively.

In the group of children who had not received BCG vaccination, the proportion of children with positive QFT results increased with increasing TST size. This correlated with the likelihood of exposure to *M tuberculosis*. In the group of children who had a history of BCG vaccination, there was no difference in the proportions of positive QFT responses with increasing TST induration (Fig 2). Univariate analysis of potential risk factors revealed that only a known index case contact (unadjusted odds ratio: 11.67; 95% confidence interval: 3.52–38.78; $P < .0001$) and travel to a disease-endemic region (limited to children >5 years of age; odds ratio: 5.73; 95% confidence interval: 1.92–17.10; $P = .001$) were significantly associated with positive QFT results.

Potential Effects of Time After Initial *M tuberculosis* Exposure on QFT Results

For the purposes of this study, we defined "recent exposure" as contact with a known index case within 1 year or TST conversion within 2 years. A positive TST result for a child who did not meet the definition of recent exposure or TST conversion was defined as "older exposure history."

IFN-γ levels in QFTs performed for children with recent documented exposure to *M tuberculosis* were much greater than those from children with older exposure histories (Fig 3). For the 8 children who had recent direct contact with an adult with active tuberculosis and who had positive QFT results, the median amount of IFN-γ released was 31.4 IU/mL, compared with 1.1 IU/mL for the 23 children with presumed older infection ($P = .001$). According to the test performance parame-

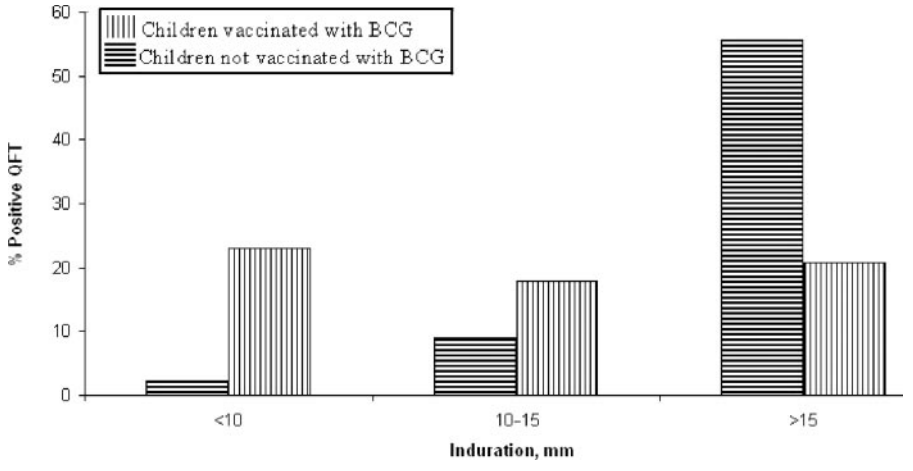

FIGURE 2
Proportions of children with positive QFT results for different TST size categories and BCG vaccination histories.

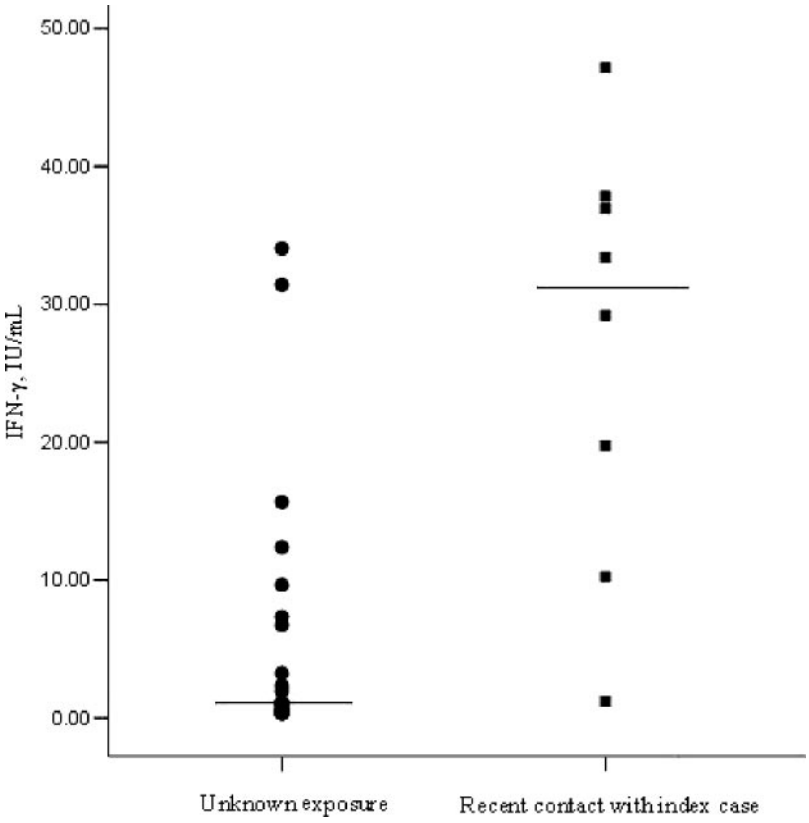

FIGURE 3
Levels of IFN-γ released for QFT-positive children who had known recent contact with an index case ($N = 8$) and children who had unknown exposure histories and therefore might have had older infections ($N = 23$). Reference lines are placed at the median for each group, that is, 1.1 IU/mL for the group with unknown exposure histories and 31.4 IU/mL for the group with recent exposure to an index case ($P = .006$).

ters of the QFT, IFN-γ concentrations of >10 IU/mL may fall beyond the linear range of the QFT software. With the assumption of 10 IU/mL as the maximum, children with recent exposure still had a much higher median IFN-γ level (>10 IU/mL) than did children with presumed older infections (1.1 IU/mL).

Five healthy TST-positive teenagers from tuberculosis-endemic regions, with BCG vaccination, had calcifications on their chest radiographs suggesting old granulomas. None of the subjects had a positive QFT result.

TABLE 4 **IFN-γ Levels From the Mitogen Control Assay ($P = .0001$)**

Age	*N*	IFN-γ Level, Mean ± SD, IU/mL
<24 mo	32	12.8 ± 10.3
24–59 mo	34	19.3 ± 14.6
≥60 mo	138	26.2 ± 19.9

Cellestis reports that IFN-γ levels of >10 IU/mL may fall beyond the linear range of the QFT software. When the maximal control value was limited to 10 IU/mL, children <24 months of age had a mean IFN-γ level of 7.7, whereas children ≥24 months of age had a mean level of 8.9 IU/mL.

QFT Performance Characteristics as a Function of Age

The proportions of indeterminant results were similar for all age groups. There were, however, differences observed in the mean amounts of IFN-γ released after stimulation with the mitogen (phytohemagglutinin) used in the assay. The amount of IFN-γ released after mitogen (phytohemagglutinin) stimulation was correlated directly with age, that is, 12.8 ± 10.3 IU/mL, 19.3 ± 14.6 IU/mL, and 26.2 ± 19.9 IU/mL for children <24 months of age, children 24 to 59 months of age, and children ≥60 months of age, respectively ($P < .0001$) (Table 4).

For this reason, we examined how recalibrating the cutoff value for children might affect results. For the 30 children with no risk factors for *M tuberculosis*, the mean amount of IFN-γ released was 0.045 IU/mL (SD: 0.073 IU/mL). By using the mean for this control group plus 3 times the SD, the cutoff value for children in this study was recalculated to be 0.26 IU/mL. This was less than the 0.35 IU/mL cutoff value recommended by the manufacturer, on the basis of results obtained in studies of adult control subjects.[5] With the application of this adjusted cutoff value to the data in this study, an additional 5 children with IFN-γ levels between 0.26 and 0.35 IU/mL would be considered to have positive results. Four of those 5 children had positive TST results and had risk factors for *M tuberculosis* exposure. The fifth child had negative TST results and had no risk factors for *M tuberculosis* exposure. When the 5 children were included in a reanalysis, an additional 17% of children had positive results for both the TST and the QFT.

Thirty-two children were evaluated at <2 years of age in this study. Only 1 child had a positive QFT result, just above the cutoff value. This child had negative TST results and a risk factor for *M tuberculosis* exposure. Twelve children had TST results of >5-mm induration, 11 of whom had TST results of ≥10 mm. Eleven (91%) of those 12 children had ≥1 risk factor for exposure to *M tuberculosis*, and 3 of those children had received BCG in infancy.

DISCUSSION

Although the QuantiFERON assay has been studied extensively in adults,[6–10] the sensitivity and specificity of IGRAs in children has not been fully evaluated. Studies of IGRAs in children have been conducted mostly in regions where tuberculosis is endemic.[15–17] One study examined an IGRA for diagnosis of active disease in a region with low levels of endemic disease and found high sensitivity and specificity.[18] Our study is the largest to assess QFT performance in diagnosing LTBI in children in the United States, an area with low levels of endemic tuberculosis. The test was easy to perform and provided interpretable results 98% of the time.

Because there is no gold standard method to measure LTBI, the applicability of any new test for LTBI can only be based on its assessing the likelihood of *M tuberculosis* exposure and correlating it with TST results. Evidence from this and other studies demonstrate that IGRAs are more specific than the TST for *M tuberculosis* detection.[19] It is impossible to exclude all risk of exposure to *M tuberculosis* in New York City but, for children with no known risk, the QFT was more specific than the TST. As the gradient of exposure increased, so did the proportion of positive QFT results. The strongest concordance between TST and QFT was found for children with TST results of ≥15 mm of induration and no BCG vaccination, which also suggests greater specificity of the QFT than the TST.

Seventy-seven percent of children with positive TST results had a negative QFT result. Although this may indicate a lack of specificity of the TST, it is also possible that the QFT is less sensitive than the TST. This may be especially true for infections that occurred much earlier in time, years before the QFT test. Children with a known recent exposure to a tuberculosis index case and a positive QFT response produced much more interferon than QFT-positive children with unknown exposure, and their results correlated well with the TST result. For most of the other TST-positive subjects there was no known exposure to an index case and, therefore, no way of knowing the time of exposure or whether exposure truly occurred. If they were indeed infected, the large majority of these children would likely have been infected outside of the United States, particularly for those children born in countries where the incidence of tuberculosis is high. In most of these cases, the exposure would probably have occurred before emigrating to the United States and long before they were found to have a positive TST. Our data suggest that the QFT test is less likely to be positive when the infection occurred more remotely in time and also for children <2 years of age. Other studies have also found that discordant TST-positive/IGRA-negative results are associated with increased time after exposure,[20,21] suggesting that, as the time after the initial infection increases, IGRA sensitivity decreases. This may explain why 5 asymptomatic teenagers with positive TST results and calcified lesions on their chest roentgenograms, suggestive of *M tuberculosis* infection, had negative QFT results.

It is possible that, for an assay to detect infection from a remote exposure, it may need to measure memory T-cell responses. This is less likely to occur with the 24-hour incubation time used in the QFT, which measures predominantly IFN-γ released from effector T cells. In contrast, the longer 48- to 72-hour incubation times of the TST allow recruitment to the skin of both effector and memory T cells. Two recent reports provided evidence that prolonged in vitro incubation increased IGRA sensitivity and increased considerably the concordance with TST results.[22,23]

Discordant QFT-positive/TST-negative results were much less common and occurred in 4 children. One of those children had active tuberculosis, and the other 3 children had moderate risk factors for *M tuberculosis* exposure. Similar findings were found in another study involving children[24] and in a meta-analysis conducted on IGRAs; discordant positive IGRA results and negative TST results were observed for 6% of all subjects and accounted for 23% of all positive QFT results.[19]

The effect of young age has been another area of concern regarding QFT sensitivity. The cutoff value for the QFT, that is, 0.35 IU/mL, was established for adults in Japan.[6] This cutoff value has not been validated for children, particularly very young children, who produce, on average, less IFN-γ than school-aged children or adults. We found that the amount of IFN-γ released in response to the mitogen phytohemagglutinin was correlated with age. Connell et al[24] also reported lower IFN-γ responses to phytohemagglutinin for younger children. Therefore, the assay cutoff value may need to be adjusted for younger children. When we recalculated the assay cutoff value on the basis of the amount of IFN-γ produced by children in our study with no risk factors for *M tuberculosis*, we found an additional 17% of children had positive results for both the QFT and the TST.

Establishing better cutoff values and increasing the assay incubation time for children may improve the sensitivity of the QFT to identify children exposed to *M tuberculosis* who demonstrate a T helper 1 response to antigenic stimulation. Young children, however, may demonstrate preferentially a T helper 2 response when exposed to *M tuberculosis* antigens, which may predispose them to be more vulnerable to disease progression.[3] One third of the children <2 years of age in this study had positive TST results and negative QFT results. With exclusion of the children who had received BCG vaccination at birth (because there is a higher rate of false-positive TST results secondary to BCG vaccination in the 2 years following vaccination[5]), there were still 9 children with positive TST results and negative QFT results, and 8 of those children had risk factors for *M tuberculosis* exposure. T helper 2 responses are captured by the TST but not by the QFT. The cytokines and chemokines identified in TST reactions include interleukin 4, IFN-γ, tumor necrosis factor α, interleukin 10, interleukin 12, and granulocyte colony-stimulating factor, whereas IGRAs measure only IFN-γ production, a T helper 1 response. Therefore, it remains possible that positive TST results might capture an immune response in very young children truly infected with *M tuberculosis* that is missed by the QFT.

The QFT detected recent infections effectively, a find-

ing that may be particularly relevant for countries where the prevalence of tuberculosis is low and where the strategy to control tuberculosis revolves around identifying and treating new infections. The fact that there was a statistically significant quantitative difference in the amounts of IFN-γ released by children with known recent infections, compared with children with older exposure histories, may provide additional benefits. It has been argued that very high or increasing IFN-γ levels are associated with greater chances of developing active tuberculosis,[25] and children have a higher risk of progression from infection to disease than do adults. Data from animal models provided strong evidence that there is a correlation between strong ESAT-6 responses and the risk of developing subsequent disease.[26,27] This was validated for human subjects with 2 reports that showed that adults who had very strong ESAT-6 responses shortly after exposure to *M tuberculosis* were those who developed active disease in the subsequent 2 years of follow-up monitoring.[28,29] Moreover, a recent study performed by Diel et al[30] showed that individuals who experienced progression to disease all produced high levels of IFN-γ (> 10 IU/mL) when initially diagnosed as having LTBI up to 2 years earlier. Therefore, QFT results may facilitate the selection of patients who are most likely to benefit from prophylaxis in resource-poor countries, or at least the identification of persons who need close monitoring and follow-up care.

If the QFT were to be used as the diagnostic test for LTBI in children and treatment decisions were based on its results alone, as is the case now for adults and is also the practice for children in some health departments, including that in New York City, many fewer children would require isoniazid prophylaxis. On the basis of the results of this study, up to three fourths of children with positive TST results would not have been treated. Clearly, the public health implications of switching from the TST to the QFT for the management of LTBI in children are significant. Before such a recommendation can be made, it is important to ascertain whether this change will leave a greater number of children, particularly those <2 years old, at risk for developing active tuberculosis.

CONCLUSIONS

The QFT is highly specific, is easy to perform, and requires only 1 visit. For situations such as known recent exposure, it seems to be as sensitive as the TST, at least for children >2 years of age. The situation is less clear for children with unknown exposure histories, because of the large proportion of dicordant TST-positive/QFT-negative results. The clinical significance of this finding merits additional study. Although the immediate risk of developing active tuberculosis seems to be low in this group, only long-term cohort studies will be able to accurately assess risk and determine whether prophylaxis is warranted.

ACKNOWLEDGMENTS

This work was funded in part by grants from the Pott's Memorial Foundation and the Thrasher Research Fund. Dr Lighter is the recipient of the Thrasher Research Fund's Early Career Award. QFT kits were provided at no cost by Cellestis (Carnegie, Australia).

We thank Drs Michael Traister and Satya Narisety for assistance in enrolling subjects, Tiina Moore for excellent technical assistance, Paige Baker for administrative assistance, Dr Ed Fryer for helpful discussions about the study analysis, and Dr William Borkowsky for helpful advice and suggestions.

REFERENCES

1. World Health Organization. *Global Tuberculosis Control: Surveillance, Planning, Financing*. Geneva, Switzerland: World Health Organization; 2002
2. Marais B, Gie R, Schaaf HS, Beyers N, Donald P, Starke J. Childhood pulmonary tuberculosis: old wisdom and new challenges. *Am J Respir Crit Care Med*. 2006;173(10):1078–1090
3. Lewinsohn D, Gennaro M, Scholvinck D. Tuberculosis immunology in children: diagnostic and therapeutic challenges and opportunities. *Int J Tuberc Lung Dis*. 2004;8(5):658–674
4. Huebner RE, Schein MF, Bass JB. The tuberculin skin test. *Clin Infect Dis*. 1993;17(6):968–975
5. Farhat M, Greenwaway C, Pai M, et al. False-positive tuberculin skin tests: what is the absolute effect of BCG and non-tuberculoous mycobacteria. *Int J Tuberc Lung Dis*. 2006;10(11):1192–1204
6. Mori T, Sakatani M, Yamagishi F, Takashima T. Specific detection of tuberculosis infection: an interferon γ based assay using new antigens. *Am J Respir Crit Care Med*. 2004;170(1):59–64
7. Brock I, Munk ME, Kok-Jensen A, Anderson P. Performance of whole blood IFN-γ test for tuberculosis diagnosis based on PPD or the specific antigens ESAT-6 and CRP-10. *Int J Tuberc Lung Dis*. 2001;5(5):462–467
8. Ferrara G, Losi M, Meacci M, et al. Routine hospital use of a new commercial whole blood interferon-γ assay for the diagnosis of tuberculosis infection. *Am J Respir Crit Care Med*. 2005;172(5):631–635
9. Mazurek GH, Zajdowicz MJ, Hankinson AL, et al. Detection of *Mycobacterium tuberculosis* infection in United States Navy recruits using the tuberculin skin test or whole-blood interferon-γ release assays. *Clin Infect Dis*. 2007;45(7):826–836
10. Arend SM, Thijsen SF, Leyten EM, et al. Comparison of two interferon-γ assays and tuberculin skin test for tracing tuberculosis contacts. *Am J Respir Crit Care Med*. 2007;175(6):618–627
11. Mazurek G, Jereb J, LoBue P, et al. Guidelines for using the QuantiFERON-TB Gold test for detecting *Mycobacterium tuberculosis* infection: United States. *MMWR Recomm Rep*. 2005;54(RR-15):49–55
12. Harris TG, Dorsinville M, Munsiff, et al. Piloting of QuantiFERON-TB Gold by the New York City Department of Health and Mental Hygiene. Presented at the First Global Symposium on Interferon-γ Releasing Assays; February 21–22, 2007; Vancouver, Canada
13. Starke JR. Use of new TB test in children should be limited. *AAP News*. 2006;27(3):14–15
14. Centers for Disease Control and Prevention. Trends in tuberculosis incidence: United States, 2006. *MMWR Morb Mortal Wkly Rep*. 2007;56(11):245–250
15. Nakaoka H, Lawson L, Squire SB, et al. Risk for tuberculosis among children. *Emerg Infect Dis*. 2006;12(9):1383–1388
16. Tsiouris S, Austin J, Toro P, et al. Results of a tuberculosis-specific IFN-γ assay in children at high risk of tuberculosis infection. *Int J Tuberc Lung Dis*. 2006;10(8):939–941
17. Dogra S, Narang P, Mendiratta DK, et al. Comparison of a

whole blood interferon-γ assay with tuberculin skin testing for the detection of tuberculosis infection in hospitalized children in rural India. *J Infect.* 2007;54(3):267–276
18. Detjen A, Keil T, Roll S, et al. Interferon-γ release assays improve the diagnosis of tuberculosis and nontuberculosus mycobacterial disease in children in a country with a low incidence of tuberculosis. *Clin Infect Dis.* 2007;45(3):322–328
19. Menzies D, Pai M, Comstock G. Meta-analysis: new tests for the diagnosis of latent tuberculosis infection: areas of uncertainty and recommendations for research. *Ann Intern Med.* 2007;146(5):340–354
20. Arend S. A supersize contact investigation with comparison of two IGRA. Presented at the First Global Symposium on Interferon-γ Releasing Assays; February 21–22, 2007; Vancouver, Canada
21. Mori T, Harada N, Higuchi K. Waning of the specific interferon-γ response after years of tuberculosis infection. *Int J Tuberc Lung Dis.* 2007;11(9):1021–1025
22. Cehovin A, Cliff JM, Hill PC, Brookes RH, Dockrell HM. Extended culture enhances sensitivity of γ interferon assay for latent *Mycobacterium tuberculosis* infection. *Clin Vaccine Immunol.* 2007;14(6):796–798
23. Leyten EM, Arend SM, Prins C, Cobelens FG, Ottenhoff TH, van Dissel JT. Discrepancy between *Mycobacterium tuberculosis*-specific interferon-γ release assays using short versus prolonged in vitro incubation. *Clin Vaccine Immunol.* 2007;14(7):880–885
24. Connell TB, Curtis N, Ranganathan SC, Buttery JP. Performance of a whole blood interferon γ assay for detecting latent infection with *Mycobacterium tuberculosis* in children. *Thorax.* 2006;61(7):616–620
25. Andersen P, Doherty TM, Pai M, Weldingh K. The prognosis of latent tuberculosis: can disease be predicted? *Trends Mol Med.* 2007;13(5):175–182
26. Vordermeier HM, Chambers MA, Cockle PJ, Whelan AO, Simmons J, Hewinson RG. Correlation of ESAT-6-specific γ interferon production with pathology in cattle following *Mycobacterium bovis* BCG vaccination against experimental bovine tuberculosis. *Infect Immun.* 2002;70(6):3026–3032
27. Buddle BM, Parlane NA, Keen DL, et al. Differentiation between *Mycobacterium bovis* BCG-vaccinated and *M bovis*-infected cattle by using recombinant mycobacterial antigens. *Clin Diagn Lab Immunol.* 1999;6(1):1–5
28. Doherty TM, Demissie A, Olobo J, et al. Immune responses to the *Mycobacterium tuberculosis*-specific antigen ESAT-6 signal subclinical infection among contacts of tuberculosis patients. *J Clin Microbiol.* 2002;40(2):704–706
29. Demissie A, Abebe M, Aseffa A, et al. Healthy individuals that control a latent infection with *Mycobacterium tuberculosis* express high levels of Th1 cytokines and the IL-4 antagonist IL-4δ2. *J Immunol.* 2004;172(11):6938–6943
30. Diel R, Loddenkemper R, Meywald-Walter K, Niemann S, Nienhaus A. Predictive value of whole-blood IFN-γ assay for the development of active tuberculosis disease after recent infection with *Mycobacterium tuberculosis*. *Am J Respir Crit Care Med.* 2008;177(10):1164–1170

A CONVERSATION WITH NINA V. FEDEROFF

"If everybody switched to organic farming, we couldn't support the earth's current population—maybe half."

Ed. Note: Dr Federoff, a member of the National Academy of Sciences, is science adviser to the secretary of state and administrator of the Agency for International Development.

Dreifus C. *New York Times*. August 19, 2008
Noted by JFL, MD

Latent Tuberculosis Diagnosis in Children by Using the QuantiFERON-TB Gold In-Tube Test

Jennifer Lighter, Mona Rigaud, Roger Eduardo, Chia-Hui Peng and Henry Pollack

Pediatrics 2009;123;30-37
DOI: 10.1542/peds.2007-3618