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Gamma Interferon Release Assays for Detection of *Mycobacterium tuberculosis* Infection

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SUMMARY

Identification and treatment of latent tuberculosis infection (LTBI) can substantially reduce the risk of developing active disease. However, there is no diagnostic gold standard for LTBI. Two tests are available for identification of LTBI: the tuberculin skin test (TST) and the gamma interferon (IFN-γ) release assay (IGRA). Evidence suggests that both TST and IGRA are acceptable but imperfect tests. They represent indirect markers of *Mycobacterium tuberculosis* exposure and indicate a cellular immune response to *M. tuberculosis*. Neither test can accurately differentiate between LTBI and active TB, distinguish reactivation from reinfection, or resolve the various stages within the spectrum of *M. tuberculosis* infection. Both TST and IGRA have reduced sensitivity in immunocompromised patients and have low predictive value for progression to active TB. To maximize the positive predictive value of existing tests, LTBI screening should be reserved for those who are at sufficiently high risk of progressing to disease. Such high-risk individuals may be identifiable by using multivariable risk prediction models that incorporate test results with risk factors and using serial testing to resolve underlying phenotypes. In the longer term, basic research is necessary to identify highly predictive biomarkers.

INTRODUCTION

Globally, tuberculosis (TB) continues to be a major public health threat, causing an estimated 8.6 million new cases and 1.3 million deaths from TB in 2012 (1). In most individuals, initial *Mycobacterium tuberculosis* infection is eliminated or contained by host defenses, and infection remains latent. Although latency and active (i.e., symptomatic, infectious) TB disease are likely part of a dynamic spectrum (Fig. 1) (2, 3), persons with latent TB infection (LTBI) are classically considered to be asymptomatic and not infectious. However, latent TB bacilli may remain viable and "reactivate" later to cause active TB disease. Identification and

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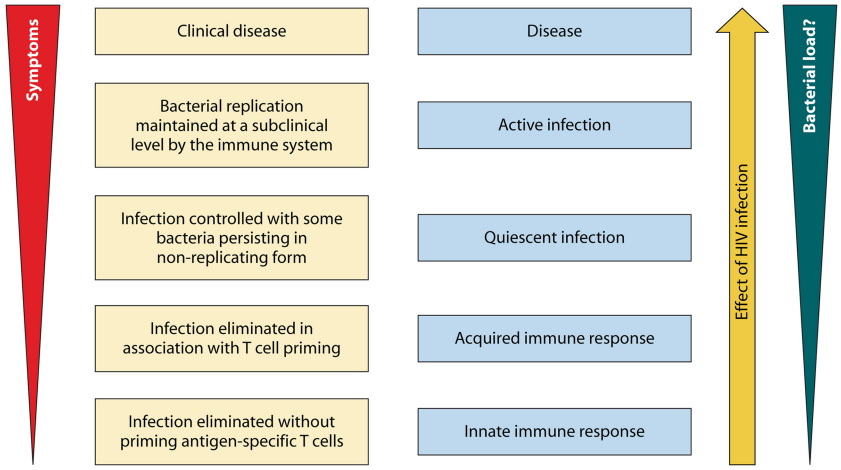

FIG 1 Proposed framework for considering tuberculosis infection as a spectrum. (Reproduced from reference 2 by permission from Macmillan Publishers Ltd.)

treatment of LTBI can substantially reduce the risk of development of disease and are important TB control strategies, especially in settings with a low TB incidence, where reactivation of LTBI often accounts for the majority of nonimported TB disease (4, 5).

TESTING FOR LATENT TUBERCULOSIS INFECTION

The goal of testing for LTBI is to identify individuals who are at increased risk for the development of active TB; these individuals would benefit most from treatment of LTBI (also termed preventive therapy or prophylaxis). Thus, only those who would benefit from treatment should be tested; a decision to test should presuppose a decision to treat if the test is positive (6).

In general, testing for LTBI is indicated when the risk of development of disease from latent infection (if present) is increased; examples include likely recent infection (e.g., close contact of a person with TB) or a decreased capacity to contain latent infection (e.g., because of immunosuppression, as in the case of young children in contact with those with active TB, people living with human immunodeficiency virus [HIV] infection, or otherwise immunosuppressed persons because of medications or conditions such as uncontrolled diabetes). In contrast, screening for LTBI in persons or groups who are healthy and have a low risk of progressing to active disease is not appropriate, since the positive predictive value of LTBI testing is low and the risks of treatment can outweigh the potential benefits (4). The balance of risk and benefit is also different in high-burden settings, where the risk of reinfection may be high and screening for LTBI will have a low negative predictive value. For children, the risk-to-benefit ratio is more favorable than for adults.

There is no diagnostic gold standard for LTBI, and all existing tests are indirect approaches which provide immunological evidence of host sensitization to TB antigens (5). There are two accepted but imperfect tests for identification of LTBI: the tuberculin skin test (TST) and the gamma interferon (IFN-γ) release assay (IGRA). Both tests depend on cell-mediated immunity (memory T-cell response), and neither test can accurately distinguish between LTBI and active TB disease (7, 8).

TUBERCULIN SKIN TESTING: OVERVIEW AND LIMITATIONS

The TST, performed using the Mantoux technique (9), consists of the intradermal injection of 5 tuberculin units (TU) of PPD-S purified protein derivative (PPD) or 2 TU PPD RT23 (these are considered equivalent [6]). In a person who has cell-mediated immunity to these tuberculin antigens, a delayed-type hypersensitivity reaction will occur within 48 to 72 h. The reaction will cause localized induration of the skin at the injection site, and the transverse diameter should be measured (as millimeters of induration) by a trained individual and interpreted using risk-stratified cutoffs (5). It is important to note that cell-mediated immunity to tuberculin antigens can sometimes reflect exposure to similar antigens from environmental mycobacteria or *Mycobacterium bovis* bacillus Calmette-Guérin (BCG) vaccination or a previous infection that has been cleared (through immunological mechanisms or treatment).

In interpreting a positive TST, it is important to consider much more than only the size of the induration (10). Rather, the TST should be considered according to three dimensions: size of induration (for the current test as well as in relation to the induration on a previous test, if done), pretest probability of infection, and risk of disease if the person were truly infected (10). Menzies and colleagues developed a simple, Web-based, interactive algorithm—the Online TST/IGRA Interpreter (version 3.0; www.tstin3d.com)—that incorporates all these dimensions (10) and also computes the risk of serious adverse events due to treatment.

The TST has several known limitations. False-positive and false-negative results can occur. There are two important causes of false-positive results: nontuberculous mycobacterium (NTM) infection and prior BCG vaccination (11). NTMs are not a clinically important cause of false-positive TST results, except in populations with a high prevalence of NTM sensitization and a very low prevalence of TB infection (11). The impact of BCG on TST specificity depends on when BCG is given and on how many doses are administered (11). If BCG is administered at birth (or during infancy) and not repeated, then its impact on TST specificity is minimal and can be ignored while interpreting the results. In contrast, if BCG is given after infancy (e.g., school entry) and/or given multiple times (i.e., booster shots), then TST specificity is compromised (11).

The BCG World Atlas (www.bcgatlas.org) provides detailed information on BCG policies and practices in many countries

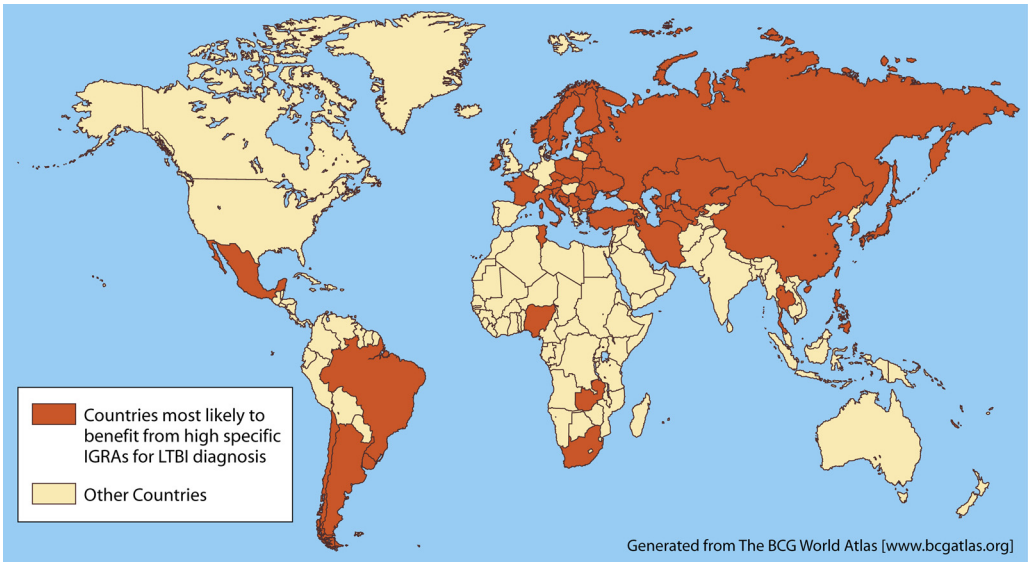

FIG 2 Countries where BCG vaccine is given after infancy or multiple times (at present or in the past). In these settings, IGRAs may be more specific than TST for latent TB infection. (Adapted from reference 12, which was published under a Creative Commons license.)

(12). While most developing countries have a policy of a single BCG vaccine administered at birth, some countries (Fig. 2) give the vaccine later in life and also give booster shots.

False-negative TST results may occur because of limited sensitivity in particular patient subgroups (e.g., immunosuppressed individuals [due to medical conditions such as HIV infection or malnutrition] or those taking immunosuppressive medications) or because of preanalytical or analytical sources of test variability (e.g., improper tuberculin handling or placement or incorrect interpretation of test results) (6). Unfortunately, individuals for whom the TST has limited sensitivity are often the very individuals that are at increased risk of progression to active disease if infected. Anergy induced by active TB itself can cause false-negative TST results (6).

The TST is also known to have problems with reproducibility, with inter- and intrareader variability in measurements of induration (13). Nonspecific variability is expected, and interpretation of repeat testing is complicated by immunologic recall of preexisting hypersensitivity to TB (i.e., boosting), conversions (i.e., new infection), and reversions (of positive results to negative) (13). Cutoffs used for TST conversions are different from the cutoffs used for diagnosis of LTBI (5).

Measurement of the long-term ability of a positive TST to predict development of active TB is difficult, requiring prolonged follow-up of unselected populations. Based on historical studies, there is a modest positive association between tuberculin reactivity and the risk of active TB (14). However, a majority of individuals with positive TST results do not progress to active disease. As a result, many TST-positive individuals need to be treated in order to prevent one disease event (4). Thus, targeted testing of high-risk groups is the common practice.

IGRA: ASSAY PRINCIPLES

IGRAs are *in vitro* blood tests of cell-mediated immune response; they measure T-cell release of IFN-γ following stimulation by antigens specific to the *M. tuberculosis* complex (with the exception of BCG substrains), i.e., early secreted antigenic target 6 (ESAT-6) and culture filtrate protein 10 (CFP-10). These antigens are encoded by genes located within the region of difference 1 (RD1) locus of the *M. tuberculosis* genome (15, 16). They are more specific than PPD for *M. tuberculosis* because they are not encoded in the genomes of any BCG vaccine strains or most species of NTM, other than *M. marinum*, *M. kansasii*, *M. szulgai*, and *M. flavescens* (17). However, not all NTMs have been studied for cross-reactivity. There is some evidence of cross-reactivity between ESAT-6 and CFP-10 of *M. tuberculosis* and *M. leprae* (18, 19), but the clinical significance of this in settings where leprosy and TB are endemic (e.g., India and Brazil) is poorly characterized.

Two commercial IGRAs are available in many countries: the QuantiFERON-TB Gold In-Tube (QFT) assay (Cellestis/Qiagen, Carnegie, Australia) and the T-SPOT.TB assay (Oxford Immunotec, Abingdon, United Kingdom). Both tests are approved by the U.S. Food and Drug Administration (FDA) and Health Canada and are CE (Conformité Européenne) marked for use in Europe.

The QFT assay is an enzyme-linked immunosorbent assay (ELISA)-based, whole-blood test that uses peptides from the RD1 antigens ESAT-6 and CFP-10 as well as peptides from one additional antigen (TB7.7 [Rv2654c], which is not an RD1 antigen) in an in-tube format. The result is reported as quantification of IFN-γ in international units (IU) per milliliter. An individual is considered positive for *M. tuberculosis* infection if the IFN-γ response to TB antigens is above the test cutoff (after subtracting the background IFN-γ response of the negative control).

The T-SPOT.TB assay is an enzyme-linked immunosorbent spot (ELISPOT) assay performed on separated and counted peripheral blood mononuclear cells (PBMCs) that are incubated with ESAT-6 and CFP-10 peptides. The result is reported as the number of IFN-γ-producing T cells (spot-forming cells). An individual is considered positive for *M. tuberculosis* infection if the spot counts in the TB antigen wells exceed a specific threshold relative to the negative-control wells. Indeterminate IGRA results

can occur due to a low IFN-γ response to the positive (mitogen) control or a high background response to the negative control.

TEST CHARACTERISTICS: SENSITIVITY AND SPECIFICITY FOR LTBI

Since there is no gold standard for LTBI, sensitivity and specificity are typically estimated using surrogate reference standards. Sensitivity is estimated among culture-confirmed TB cases, while specificity is estimated among low-risk individuals with no known TB exposure in low-incidence settings (20).

Based on published meta-analyses (7, 8, 21), IGRAs have a specificity for LTBI diagnosis of >95% in settings with a low TB incidence, and specificity is not affected by BCG vaccination. TST specificity is similarly high in populations not vaccinated with BCG (97%). Among populations where BCG is administered, the specificity is much lower (approximately 60%) and variable, depending on when and how often BCG is given. The sensitivity for the T-SPOT.TB assay appears to be higher than that for the QFT assay or TST (approximately 90%, 80%, and 80%, respectively). IGRA sensitivity is diminished by HIV infection and in children (see later discussion) (22, 23).

Because IGRAs are not affected by BCG vaccination status, IGRAs are useful for evaluation of LTBI in BCG-vaccinated individuals, particularly in countries where BCG vaccination is administered after infancy or multiple (booster) BCG vaccinations are given (12, 24). In such countries (Fig. 2), the TST is unlikely to have high specificity.

Although this is based on limited evidence, IGRAs appear to be unaffected by most infections with NTMs which can cause false-positive TSTs (17). However, infection with *M. marinum* or *M. kansasii*, which express ESAT-6 or CFP-10, has been shown to produce positive results in IGRAs, as with the TST (25, 26).

TEST CHARACTERISTICS: REPRODUCIBILITY

While IGRAs have improved specificity over that of TST, concerns have been raised about issues with reproducibility of the test in settings where repeat testing is necessary (27, 28). By nature, functional T-cell assays are highly susceptible to variability by numerous factors at multiple levels, including assay manufacturing, preanalytical processing, analytical testing, and immunomodulation. Therefore, reproducibility is an important consideration (29) that makes it challenging to use a single cutoff value to distinguish between positive and negative results with one-time testing and to define conversion and reversion in individuals undergoing serial testing.

A systematic review on IGRA reproducibility in 2009, based on a small number of studies, showed that variability was substantial, with magnitudes of within-subject IFN-γ responses varying by up to 80% (28). Since then, more research has emerged, providing a better understanding of the sources of variability in IGRAs. A list of potential sources of IGRA variability and their impacts is shown in Table 1. Figure 3 graphically illustrates the sources of variations, with the QFT assay as an example. Although each source can have a positive or negative effect on the assay response, the "total variability" is the net sum of all variability combined.

TABLE 1 Potential sources of variability and their impact on results in IGRAs[c]

Source of variability	Impact on assay	
	QFT	T-SPOT.TB
Manufacturing source		
Between-lot variability	↑ ↓	↑ ↓
Preanalytical sources		
Time of blood draw (a.m. vs p.m.)	↑ p.m.	?
Skin disinfection	?	?
Traumatic blood draw	?	?
Blood vol (0.8–1.2 ml)	↓	NA
Shaking of tubes (gentle to vigorous)	↑	NA
T-cell and APC counts	?	?[a]
Transportation temp	?	↓
Delay in incubation (0–16 h)	↓	↓
Incubation time (16–24 h)	Possible effect	?
Plasma separation delays (seconds to hours)	?[b]	NA
Plasma storage (+4–−80°C)	No effect	NA
Analytical sources		
Within-run imprecision	↑ ↓	↑ ↓
Between-run imprecision	↑ ↓	↑ ↓
Between-operator imprecision	↑ ↓	↑ ↓
Between-laboratory imprecision	↑ ↓	↑ ↓
Immunological sources		
Boosting by PPD	↑	↑
Modulation by PAMP	↑ ↓	?

[a] The mononuclear cell input is normalized.
[b] According to the manufacturer, delay after plasma separation contributes to ELISA variability.
[c] NA, not applicable; APC, antigen-presenting cell; PPD, purified protein derivative; PAMP, pathogen-associated molecular pattern; ↑ ↓, may go in either direction.

Variability Due to Manufacturing Issues

Like all diagnostic tests, IGRAs may be susceptible to manufacturing quality issues, with some lots or reagents affected by issues such as temperature during shipping. This was described for the QFT assay by Slater and colleagues, who investigated a sudden increase in the rate of positive QFT results, from 10% to 31%, at an academic institution in the United States (30). The reason for the sudden increase in the false-positive rate during this incident could not be identified, although a similar issue, attributed to contamination of a specific lot of tubes, led to its withdrawal from the market by the manufacturer in 2012 (31). By monitoring positivity and indeterminate rates, clinical laboratories can rapidly detect and halt utilization of potentially faulty lots, alert the manufacturer to investigate, and prevent reporting of inaccurate test results.

Preanalytical Sources of Variability

Preanalytical sources of variability are several and likely represent a large component of "total variability." Among the list of potential sources shown in Table 1, delay between blood collection and incubation of cells at 37°C has been studied extensively. The manufacturer of the QFT assay allows a 0- to 16-h range of delay before tubes can be incubated. However, Doberne and colleagues showed a significant decline in TB response with a delay in incubation within the recommended range (32). Compared to immediate incubation, 6- and 12-h delays resulted in positive-to-negative reversion rates of 19% (5/26 samples) and 22% (5/23 samples), respectively, for individuals with a high risk for LTBI (32). Indi-

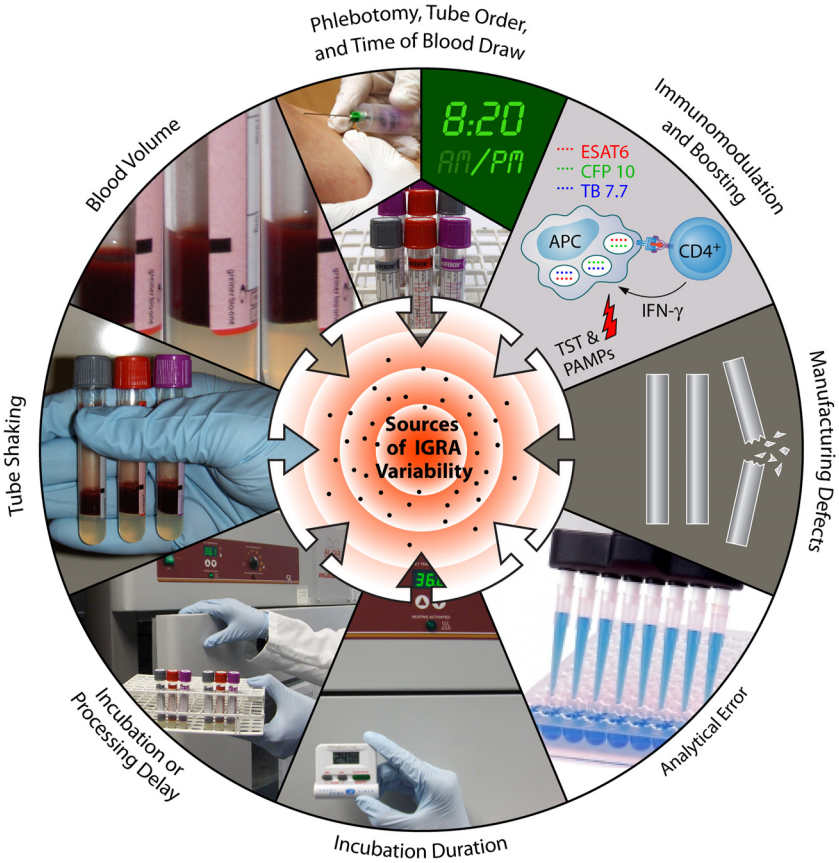

FIG 3 Sources of variability in the QuantiFERON-TB Gold In-Tube assay.

viduals with reversion had a lower TB response, closer to the assay cutoff, than individuals whose results remained positive with incubation delay.

Incubation delay also has a negative effect on test results through reducing the mitogen response in the QFT assay and increasing the rate of indeterminate results (29, 32, 33). Other preanalytical variables shown to impact QFT results include blood volume and tube shaking. Gaur and colleagues showed an inverse relationship between blood volume in the TB antigen tube, within the recommended range, and IFN-γ response (34). Compared to 0.8 ml blood, 1.0 and 1.2 ml blood resulted in significant declines in TB-specific IFN-γ responses in the infected subjects, and 1.2 ml resulted in a significant decrease in the proportion of positive results. Vigorous shaking also caused a significant increase in IFN-γ response in the nil and TB antigen tubes and caused a significant elevation in TB response when vigorously shaken TB antigen tubes were paired with gently shaken nil tubes (34). In the same study, duration of incubation within the recommended range was not shown to be a source of variability in the infected group (34), but this may not be consistent with a similar study that did show that 24 h of incubation led to a higher TB-specific IFN-γ response than that with 16 h of incubation (35). Variation in the timing of blood collection (evening versus morning) may also introduce variability into test results, but the mechanism is unclear (36). While the reproducibility of the QFT assay is reasonably well studied, many of the above considerations also apply to the T-SPOT.TB assay.

Analytical Sources of Variability

The analytical sources of variability refer to fluctuations in measurements due to random errors caused by interference of uncontrolled factors in biological fluids (matrix effects), imprecision of pipetting, manipulation errors in centrifugation, decantation, and washing, and the imprecision of measurement of the final signal. Unlike preanalytical sources of variability, which are mostly systematic and therefore predictable, the analytical sources are mostly random and persist despite extensive efforts to improve the analytical reproducibility.

Indeed, studies such as that of Metcalfe and colleagues (37) have shown considerable within-run and between-run variabilities in the quantitative results, producing discordant results when TB responses are close to the assay cutoff (37). Whitworth and colleagues showed variability in QFT results from the same subjects when ELISAs were performed in different laboratories (38). Analytical error originating from interreader variability has also been investigated, though it appears to be a problem primarily with the T-SPOT.TB assay, not the QFT assay (39).

Immunological Sources of Variability

The two immunological sources of variability described to date include immune boosting and immunomodulation. Van Zyl-Smit and colleagues showed that a significant increase in TB response occurs when QFT and T-SPOT.TB testing is performed more than 3 days after PPD placement, through immunological

recall of preexisting memory T cells to TB antigens (40). Similar findings have been reported in other studies (41–44), although it is not clear how long the IGRA boosting persists and whether the PPD formulation and amount used in TST contribute to boosting. The underlying mechanism of TST boosting is thought to be an anamnestic response of preexisting memory T cells to RD1 antigens, which are contained within PPD (45, 46). In contrast, a previous IGRA will not boost the results of the subsequent IGRA result, as the test itself is performed *ex vivo*.

Another source of immunological variability is caused by immunomodulation through conserved microbial products known as pathogen-associated molecular patterns (PAMPs), such as lipopolysaccharide and peptidoglycan (47). PAMPs are recognized by the innate immune cells via several families of pathogen recognition receptors (PRRs), of which the Toll-like receptor (TLR) family is best characterized. Activation of PRRs triggers intracellular signaling pathways culminating in the expression of inflammatory mediators which stimulate the maturation of antigen-presenting cells and initiation of adaptive immune responses, such as the development and proliferation of antigen-specific effector T-cell subsets (47).

Gaur and colleagues showed that *in vitro* immunomodulation in the QFT assay may occur with Toll-like receptor agonists and at low concentrations and that this may enhance antigen-specific IFN-γ responses in individuals with presumed LTBI (48). PAMPs in an IGRA may, for example, be derived from endogenous microbiota (which can be influenced by diet, antibiotics, and personal hygiene) or from exogenous contaminants (mostly from skin during blood draw) and may account for a fraction of the reported within-subject variability.

Overall, IGRA results can be affected by many sources of variation, not all of which are understood at present. While systematic sources of variability can be eliminated or minimized through standardization by the assay manufacturers and users of the test, random sources of variability are unavoidable and must be accounted for when interpreting results. Once total variability in IGRA responses is determined, appropriate cutoffs and borderline zones can be derived for interpreting serial testing results in light of a patient's TB risk factors and local laboratory practices (49).

TEST CHARACTERISTICS OF SUBGROUPS

Individuals with Suspected TB Disease

Three systematic reviews have assessed the ability of IGRAs to diagnose active TB in adults, including extrapulmonary TB, with consistent conclusions that active TB can neither be ruled in nor ruled out with IGRAs (7, 8, 50). Because IGRAs (like the TST) have suboptimal sensitivity for active TB, especially in HIV-infected persons, a negative result cannot reliably rule out active TB. IGRAs also cannot distinguish between LTBI and active TB, and therefore the specificity of TB diagnosis will always be poor in countries with high TB burdens (51, 52).

Metcalfe and colleagues performed a meta-analysis to assess the diagnostic performance of QFT and T-SPOT.TB assays among adults with suspected or confirmed active pulmonary TB in low- and middle-income countries (7). Among HIV-infected patients, pooled sensitivity estimates were 76% (95% confidence interval [CI], 45% to 92%) for T-SPOT.TB assay and 60% (95% CI, 34% to 82%) for QFT assay. Pooled specificity estimates were low for both IGRAs among all participants (61% [95% CI, 40% to 79%] for T-SPOT.TB assay and 52% [95% CI, 41% to 62%] for QFT assay) and among HIV-infected persons (52% [95% CI, 40% to 63%] for T-SPOT.TB assay and 50% [95% CI, 35% to 65%] for QFT assay). There was no consistent evidence that either IGRA was more sensitive than the TST for active TB diagnosis in low- and middle-income countries. This review informed a 2011 WHO policy on the use of IGRAs in low- and middle-income countries (53). The policy states that neither IGRAs nor the TST should be used for the diagnosis of active TB (53).

Fan and colleagues summarized the performance of IGRAs for extrapulmonary TB diagnosis and found that both IGRAs and the TST had poor specificity in distinguishing extrapulmonary TB from LTBI, especially in low-income countries (50).

A few studies have assessed the incremental value of IGRAs within diagnostic algorithms for active TB (54–56). In other words, considered in light of conventional risk factors, including signs, symptoms, and findings on chest radiograph, do IGRAs improve risk stratification of individual TB suspects? As with the TST, these studies determined that there is limited added value for adults in settings with either low (55) or high (54, 56) TB incidence. A recent study showed similar results for children (57).

Children

Children are at high risk of developing TB disease, if infected (58). Furthermore, diagnosis of TB is a persistent challenge with young children, who are often unable to produce sputum and for whom conventional microbiological tests have low sensitivity (58). Two systematic reviews have assessed the performance of IGRAs for children (23, 59). Available data from these systematic reviews suggest that TST and IGRAs have similar accuracies for the detection of TB infection or the diagnosis of disease in children. Subgroup analysis suggested a lower sensitivity for all tests in young (<5 years of age) or HIV-infected children. Both TST and IGRAs had similar correlations with the exposure gradient in children. However, the ability of either TST or IGRA alone was suboptimal to rule in or rule out active TB. Thus, in children with suspected active TB, every effort should be made to collect appropriate clinical specimens for microbiological and molecular testing, and IGRAs should be used with other clinical data (e.g., TST results, chest X-ray findings, and history of contact) to support a diagnosis of active TB (60).

HIV-Infected Persons

Three systematic reviews have summarized the performance of IGRAs in HIV-infected populations (22, 61, 62), with fairly consistent conclusions. Cattamanchi and colleagues (61) showed that for HIV-infected persons with active TB (a surrogate reference standard for LTBI), pooled sensitivity estimates were heterogeneous but higher for T-SPOT.TB assay (72%; 95% CI, 62 to 81%) than for QFT assay (61%; 95% CI, 47 to 75%) in low- and middle-income countries. However, neither IGRA was consistently more sensitive than the TST in head-to-head comparisons. IGRAs, in particular the T-SPOT.TB assay, may be less affected by the degree of immunosuppression, but results differed across geographical settings.

In another meta-analysis, Santin and colleagues analyzed the impact of HIV on rates of indeterminate IGRA results (22). They estimated the pooled indeterminate proportion to be 8.2% for QFT assay and 5.9% for the T-SPOT.TB assay for HIV-infected persons. Indeterminate proportions were higher in high-burden

settings (12.0% for QFT assay and 7.7% for T-SPOT.TB assay) than in low- or intermediate-burden settings (3.9% for QFT assay and 4.3% for T-SPOT.TB assay). Proportions were also higher for patients with $CD4^+$ T-cell counts of <200 (11.6% for QFT assay and 11.4% for T-SPOT.TB assay) than for those with $CD4^+$ T-cell counts of >200 (3.1% for QFT assay and 7.9% for T-SPOT.TB assay).

Thus, current evidence suggests that IGRAs perform similarly to the TST in identifying HIV-infected individuals with presumed LTBI. Both TST and IGRAs have suboptimal sensitivity for active TB, suggesting a potential role for using both tests, especially in severely immunocompromised individuals.

IMIDs

TB screening before therapy with immunomodulating biologic agents (e.g., tumor necrosis factor alpha [TNF-α] inhibitors) in patients with immune-mediated inflammatory diseases (IMIDs) is an established and growing practice. However, the exact screening approach and algorithm remain controversial. Winthrop and colleagues (63) and Smith and colleagues (64) reviewed the evidence on performance of IGRAs for patients with IMIDs. A summary assessment was limited, as most studies were small and varied considerably with respect to the use of immunosuppressive medications and types of patients with IMIDs. Current evidence does not clearly suggest that IGRAs are better than TST in identifying patients with IMIDs who could benefit from LTBI treatment (63). To date, no studies have been done on the predictive value of IGRAs for patients with IMIDs.

Shahidi and colleagues summarized the evidence for patients with inflammatory bowel disease and evaluated the impact of immunosuppressive therapy on the proportion of indeterminate results and IGRA and TST positivity (65). The pooled percentage of indeterminate results was 5% for QFT assay, with the T-SPOT.TB assay showing similar results. Both positive QFT results (pooled odds ratio [OR] = 0.37; 95% CI, 0.16 to 0.87) and positive TST results (pooled OR = 0.28; 95% CI, 0.10 to 0.80) were significantly influenced by immunosuppressive therapy ($P = 0.02$ for both).

In the only prospective, longitudinal study, Chang and colleagues reported a comparison of TST and the QFT assay for LTBI screening in 107 Korean patients with rheumatoid arthritis ($n = 61$) or ankylosing spondylitis ($n = 46$) who were initiating treatment with TNF-α antagonists (66). QFT results were indeterminate for 7 (6.5%) patients. Among the remaining 100 patients, QFT and TST results were discordant for 33 (33%), including 16 with negative QFT and positive TST results and 17 with positive QFT and negative TST results. No patients developed active TB during a median of 18 months of treatment with TNF antagonists, including the 16 patients with positive TST but negative QFT results who were not treated for LTBI. Although Chang and colleagues concluded that screening with the QFT assay is sufficient prior to treatment with TNF antagonists, in regions of moderate or high TB prevalence, or in patients with TB risk factors, there is some evidence that a dual testing strategy of TST and IGRA improves sensitivity (63, 67). Winthrop and colleagues have proposed an algorithm for this purpose (63).

HCWs and Serial Testing

Serial (repeated) testing for LTBI is indicated for specific populations, such as health care workers (HCWs) in high-risk settings and prison inmates and staff. The goal of serial testing is to identify recent TB infections and to target newly infected individuals (who are at increased risk of disease progression) for preventive therapy.

Several studies have evaluated the use of IGRAs in HCWs, and these have been summarized in systematic reviews (68, 69). Zwerling and colleagues found that in settings with low and moderate TB incidences, the cross-sectional prevalence of a positive IGRA in HCWs was significantly lower than that for a positive TST (68). However, in high-incidence settings, there were no consistent differences in the prevalences of positive tests. IGRAs showed good correlation with occupational risk factors for TB exposure in low- and moderate-incidence settings in only some studies (69, 70). Thus, the use of IGRAs instead of TST for one-time screening may result in a lower prevalence of positive tests and fewer HCWs who require LTBI treatment, particularly in settings with a low TB incidence (69).

However, when simple negative/positive cutoffs are used for serial testing, issues may arise from high rates of conversions and reversions, and the higher specificity of IGRAs than of TST must be balanced against the higher probability of false-positive conversions following an initially negative test. This is evident from recent experiences of North American hospitals that began implementing IGRAs for employee screening after the 2005 Centers for Disease Control and Prevention (CDC) guidelines, which recommended that a change from a negative to a positive IGRA result (using the diagnostic IFN-γ cutoff of ≥0.35 IU/ml) can be treated as a "conversion" (71–74). These studies have reported high rates of IGRA conversions and reversions, a phenomenon noted in other studies in settings of low TB incidence as well (75–85) (Table 2).

A recent, large HCW study was conducted by the U.S. CDC's TB Epidemiologic Studies Consortium (TBESC) (86). This study of 2,563 HCWs undergoing occupational TB screening in 4 U.S. hospitals performed testing every 6 months, using TST, QFT, and T-SPOT.TB assays. Proportions of participants with test conversion during the study period were 138/2,263 (6.1%) participants for QFT assay, 177/2,137 (8.3%) participants for T-SPOT.TB assay, and 21/2,293 (0.9%) participants for TST (86). This study also found very high reversion rates among HCWs with positive QFT and T-SPOT.TB results.

In a study of over 9,000 HCWs at Stanford University Medical Center, 4.4% of those with initial negative QFT results had a conversion over 2 years, which is substantially higher than the historic TST conversion rate of 0.4% at this hospital (87). Similarly, a QFT conversion rate of 5.3% was reported from Canadian hospitals (70), with no TST conversions in the same cohort. At the Central Arkansas Veterans Healthcare System, the QFT conversion rate was found to be 30-fold higher than the baseline TST conversion rates in the years preceding the use of the QFT assay (85).

These high IGRA conversion rates are not compatible with the current low rates of TB incidence in the United States and Canada, as indicated by TST conversion rates of well below 1% in many hospitals (86). To overcome these problems, health care institutions have begun using more stringent cutoffs or retesting strategies to eliminate false-positive conversions (71, 74), and some have switched back to serial TST (85).

IGRAs also had high rates of reversions in most studies, ranging from about 20 to 60% (Table 2), and these occurred even without LTBI treatment. In general, IGRA reversions are much more likely to occur among those with IFN-γ values (or spot counts) just above the diagnostic threshold (i.e., borderline zone), indepen-

TABLE 2 Serial testing studies of IGRAs in health care workers in countries with low and intermediate incidences[c,d]

Study, yr (reference)	Country	Duration between tests	No. of conversions or reversions/total no. of participants (%)		
			TST conversions	IGRA conversions[a]	IGRA reversions[a]
Slater et al. (87)	USA	2 yr	0.4% (historical)	361/8,227 (4.4)	613/1,584 (38.7)
Dorman et al. 2013 (86)	USA	6 mo	21/2,293 (0.9)	For QFT, 138/2,263 (6.1); for T-SPOT, 177/2,137 (8.3)	For QFT, 81/106 (76); for T-SPOT, 91/118 (77)
Zwerling et al., 2013 (70)	Canada	1 yr	0/241	13/245 (5.3)	8/13 (62)
Joshi et al., 2012 (85)	USA	1 yr	0.1% (historical)	71/2,232 (3.2)	31/69 (45)
Park et al., 2012 (84)	South Korea	Once-monthly testing for 1 yr	NA	25/48 (52) had ≥1 conversion over 1 yr	Not reported
Joshi et al., 2012 (73)	USA	2–30 days	NA	NA	18/45 (40)
Rafiza and Rampal, 2012 (75)	Malaysia	1 yr	NA	69/703 (9.8)	14/59 (23.7)
Fong et al., 2012 (71)	USA	1 yr or 1–6 mo for repeat of positive IGRA	NA	52/1,857 (2.8)	8/10 (80)[b]
Torres Costa et al., 2011 (76)	Portugal	1 yr	61/199 (30.7); reversion rate = 4/188 (2.1)	51/462 (11)	46/208 (22.1)
Schablon et al., 2010 (77)	Germany	High-risk HCWs tested annually, all others evaluated every other year	NA	15/245 (6.1)	13/42 (32.6)
Ringshausen et al., 2010 (78)	Germany	18 wk	NA	3/162 (1.9)	6/18 (33.3)
Park et al., 2010 (79)	South Korea	1 yr	NA	14/244 (5.7)	NA
Lee et al., 2009 (80)	South Korea	1 yr	16/75 (21.3)	21/146 (14.4)	NA
Chee et al., 2009 (81)	Singapore	1 yr	0/18 (note that the denominator includes only baseline concordant positive results)	9/182 (4.9)	NA
Yoshiyama et al., 2009 (82)	Japan	2 and 4 yr	NA	5/277 (1.8)	13/32 (41)
Pollock et al., 2008 (83)	USA	1–7 mo	NA	2/43 (4.6) selected HCWs at "increased risk" and negative at baseline	NA

[a] All conversions/reversions, using simple negative/positive results.
[b] Note that repeat testing was done among those with positive QFT results close to the cutoff point.
[c] HCW, health care worker; IGRA, gamma interferon release assays; NA, data not available; TST, tuberculin skin test.
[d] Adapted from reference 90 with permission of the publisher.

dent of treatment for LTBI. However, reversions have also been observed in situations with strongly positive initial IGRA responses.

When tests are repeated more frequently on the same individuals, more complex patterns or phenotypes are seen (Fig. 4), including stable and unstable (transient) conversions, persistently positive (long-term positive results) and negative (long-term negative results) results, and other more complex trajectories (88, 89).

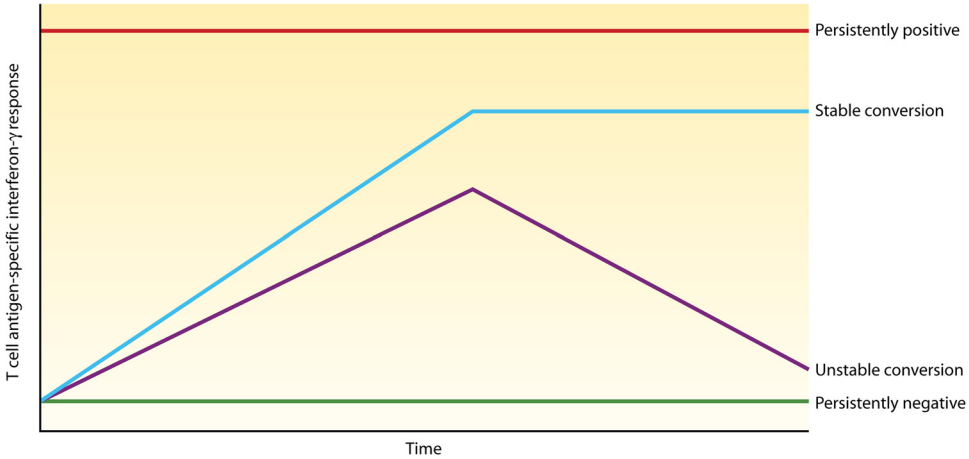

FIG 4 Serial testing with IGRAs reveals underlying phenotypes. The persistently positive pattern is seen in individuals who are repeatedly IGRA positive for a long time. Unstable conversion refers to individuals who convert their IGRA result from negative to positive and then revert again to negativity. Stable conversion refers to individuals who convert their IGRA result and stay converted, at least in the short term. Persistently negative refers to individuals who stay repeatedly IGRA negative for a long time. (Reproduced from reference 88 by permission from Macmillan Publishers Ltd.)

There are limited longitudinal data on the prognosis of such phenotypes, and it is unclear whether any subgroup should be targeted for preventive therapy.

As summarized in recent reviews (49, 90), IGRAs are inherently dynamic in a serial testing context, and this is reflected in the literature, which consistently shows high rates of both conversions and reversions. There are no data to date to suggest that IGRAs are better at identifying the incidence of new TB infection than the TST, and in fact, when manufacturer's dichotomous cutoffs are used for conversions, they will likely result in conversion rates that are incompatible with what is epidemiologically expected for a given setting. While the interpretation of IGRA results is not prone to the subjectivity that adversely affects the reading of TST induration, other factors affect their reproducibility, as reviewed earlier. Occupational testing programs will therefore need to standardize IGRA testing protocols to limit the variability in results, and suggestions for standardization have been proposed (91). It is also clear that simplistic definitions of conversions are no longer valid and that existing guidelines on serial testing need to be updated to reflect the accumulated evidence.

Monitoring of Antituberculosis Therapy

IGRA responses are hypothesized to be related to the bacillary burden and antigenic load present in the body (92). If this is true, then treatment and a decrease of antigenic load should result in a decrease in the IGRA response, which could conceivably be used for treatment monitoring. The clinically important question, therefore, is whether IGRAs can be used to assess treatment responses and to predict failure or relapse in active TB. Smear and culture conversions to negativity are established treatment monitoring parameters (93). Several studies have tried to correlate the changes in IFN-γ responses in IGRAs with these established markers of treatment response (94–98). In one of the largest studies, Denkinger and colleagues found a significant decrease in the IFN-γ response over time but no significant correlation with smear or culture conversion to negativity (98). Other studies have found inconsistent results that are mostly not supportive of the use of IGRAs for active TB treatment monitoring (94–98).

While there are established markers for treatment monitoring in active TB, no biomarker is currently available for the monitoring of LTBI treatment success. Similar to treatment monitoring in active TB, studies have reported conflicting results regarding the impact of treatment of LTBI on IGRA responses (99–102). The highest-quality data thus far come from a randomized trial that assigned patients with LTBI to receive isoniazid (INH) or placebo and measured CFP-10 and ESAT-6 responses by ELISPOT assay at enrollment and months 1, 3, and 6 (103). While there were decreases in responses observed over time, the decreases were similar in both the treatment and placebo groups.

Chiappini and colleagues systematically reviewed the data on use of IGRAs for treatment monitoring in both active and latent TB and concluded that "monitoring IGRA changes over time seems to have only speculative value" (104).

PREDICTIVE VALUE FOR PROGRESSION TO TB DISEASE

Diel and colleagues assessed the positive and negative predictive values of the commercial IGRAs relative to those of the TST for the future development of active TB in untreated individuals (105). Their review suggested that the positive and negative predictive values of commercial IGRAs might be higher than those of the TST, in particular among high-risk populations. A limitation, however, was that the analytic approach did not take the different durations of follow-up into consideration, and therefore, the estimated predictive values were not adjusted for the number of person-years of follow-up.

In a meta-analysis by Rangaka and colleagues (106), the prognostic ability of the IGRAs was summarized in the form of incidence rates and risk ratios for the longitudinal studies included in the review. Fifteen studies with a combined sample size of 26,680 participants were included in this analysis (107–121). The incidence of active TB during a median follow-up of 3 years was 2 to 24 per 1,000 person-years for IGRA-negative individuals (Fig. 5). For IGRA-positive individuals, the TB incidence was 4 to 48 cases per 1,000 person-years (106), suggesting that a majority of IGRA-positive individuals did not progress to TB disease during follow-up. This is similar to the historic data on TST (14).

Compared with negative test results, IGRA-positive and TST-positive results were much the same with regard to risk of TB development (the pooled incidence rate ratio [IRR] in the five studies that used both was 2.11 [95% CI, 1.29 to 3.46] for IGRA versus 1.60 [95% CI, 0.94 to 2.72] for TST at the 10-mm cutoff). However, the proportion of IGRA-positive individuals in 7 of 11 studies that assessed both IGRAs and TST was generally lower than that of TST-positive individuals (106). The authors concluded that neither IGRAs nor the TST have high accuracy for the prediction of active TB, although the use of IGRAs in some populations might reduce the number of people considered for preventive treatment (106).

Since the publication of the aforementioned review, five new longitudinal studies have been published (122–126). Table 3 presents the characteristics of all 20 longitudinal studies. Among IGRA-positive individuals, incidence rates ranged from 3.7 to 84.5 per 1,000 person-years of follow-up, while they ranged from 2.0 to 32.0 per 1,000 person-years for IGRA-negative individuals. The highest incidence rates, among both IGRA-positive and IGRA-negative individuals, were found in studies that followed immunocompromised subjects, such as HIV-infected mothers, HIV-exposed infants, or men with silicosis.

Three studies specifically assessed the prognostic value of the IGRAs in an exclusively HIV-positive cohort (110, 118, 125). Two studies without possible incorporation bias (where IGRAs were not used to make a final diagnosis of active TB) and differential work-up bias (where IGRA-positive individuals were not investigated more intensively for active TB than IGRA-negative individuals) (118, 125) found risk ratios of 2.69 (95% CI, 0.69 to 10.52) and 3.32 (95% CI, 1.09 to 10.08), respectively, meaning that individuals with a positive IGRA result had around a 3-fold-increased risk of progression to TB disease during the follow-up period of the study compared to individuals with a negative IGRA result. Although the rate of disease progression after a presumed TB infection is increased in HIV-infected individuals, there are currently no data that suggest that the predictive value of the IGRAs is better or worse in this subpopulation than in others.

While most longitudinal studies have assessed the predictive value of a single, cross-sectional IGRA result, only a single study has evaluated the predictive value of an IGRA conversion (127). This study found that recent QFT conversion was indicative of an approximately 8-fold higher risk of progression to TB disease (compared to nonconverters) within 2 years of conversion in a cohort of adolescents in South Africa. However, even among QFT

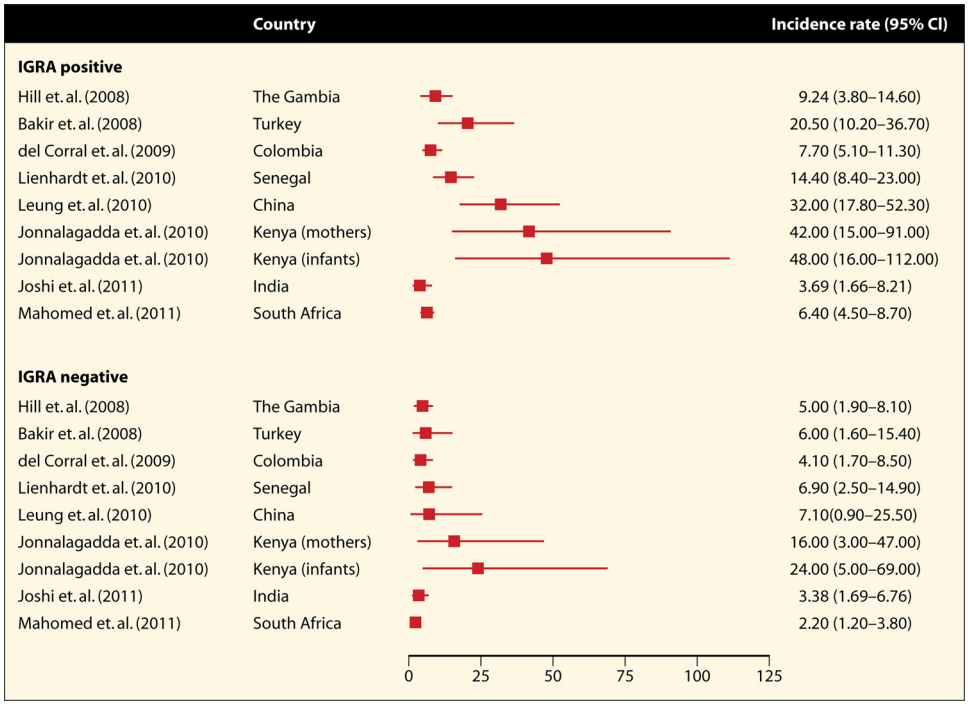

FIG 5 Unadjusted incidence rates for development of active tuberculosis in the short term (median follow-up of 3 years), stratified by IGRA result. Incidence rate estimates are per 1,000 person-years of follow-up, stratified by IGRA result at baseline. Table 3 provides details for each of the studies. (Reproduced from reference 106 with permission from Elsevier.)

converters, the overall risk of TB disease was low (1.46 cases per 100 person-years) (127). Although evidence is limited, this study does suggest that an IGRA conversion (which may indicate recent infection) may be more predictive than a single positive IGRA result.

Overall, the currently available data show that the predictive value of IGRAs for progression to TB disease is low and slightly but not significantly higher than that of the TST. The data suggest that a majority (>95%) of those with positive IGRA or TST results do not progress to TB disease during follow-up.

Why do existing LTBI tests have poor predictive value for active TB? There may be several reasons. First of all, the overall risk of progression from LTBI to active TB—in the absence of recent infection or severe immunosuppression—is low (<5% lifetime risk in healthy populations); thus, even a perfectly accurate test for LTBI would have a low predictive value for progression to active TB. Second, while IGRAs (and TST) are generally evaluated according to their ability to predict future active TB, their true aim is to identify individuals who would benefit from preventive therapy. Since future active TB is a combination of both reinfection events (arguably not amenable to preventive therapy) and reactivation events, and since LTBI may confer some protective immunity against repeat infection (128), the ability of IGRAs to predict future active TB may misrepresent their ability to identify those who would benefit from preventive therapy. Third, IGRAs are immune-mediated tests, and the same immune system is responsible for yielding a positive IGRA result as well as preventing progression to active TB disease; as such, individuals with false-negative IGRAs may be the very individuals (e.g., highly immunosuppressed) at greatest risk of reactivation. Fourth, the sensitivity and specificity of IGRAs are imperfect and dependent on only a few antigens, and antigens expressed by *M. tuberculosis* during latency may not be those expressed during active replication (2, 129).

As a consequence of all the above factors, the IFN-γ response, although important, is probably insufficient to resolve the various phases of the latent TB "spectrum" as illustrated in the framework proposed by Barry and colleagues (reproduced in Fig. 1) (2). Among the stages shown in the figure, both TST and IGRAs are likely to be positive in all stages, with the possible exception of the innate immune response stage (i.e., exposed to TB but negative on both tests) (3, 88).

For all these reasons, both TST and IGRAs are generally unable to select out the phenotypes that are most likely to benefit from LTBI treatment (88, 130). This is underscored by the observed low rates of progression to disease even in IGRA- and TST-positive individuals (106). A more predictive LTBI test or strategy will greatly help to target only those who will benefit from LTBI treatment.

COST-EFFECTIVENESS

A systematic review of cost-effectiveness analyses (CEA) was conducted by Nienhaus and colleagues (131). Cost and cost differences between studies were not fully investigated, as the authors did not adjust or inflate to a common currency to permit comparisons. The study conclusions regarding cost-effectiveness were, however, compared for 7 available CEA studies. The authors concluded that in 6/7 studies, IGRA (as a dual-step strategy following TST or IGRA only) was reported as more cost-effective than TST only. However, the authors also state that comparison of the studies was hampered by several methodologic problems, including differences in assumed costs, test parameters, strategies modeled, and outcomes evaluated. They concluded that until some of these

TABLE 3 Description of published studies on IGRA predictive value[a,b]

Study no.	Author, yr (reference)	Country (income status)	Age group (yr)	Individuals with HIV in cohort	Population	No. of individuals assessed	No. of individuals followed up and included in analysis	Preventive therapy given
1	Doherty et al., 2002 (107)	Ethiopia (LIC)	Adults (15–65)	No; exclusion criterion	Tuberculosis case contacts	38	24	No
2	Hill et al., 2008 (108)	The Gambia (LIC)	Adults and children (0.5–100)	Yes (2%)	Tuberculosis case contacts	2,381	2,348	No
3	Bakir et al., 2008 (109)	Turkey (MIC)	Children (0–16)	Not stated	Tuberculosis case contacts	1,024	908	Yes (76% of 908 individuals)
4	Aichelburg et al., 2009 (110)	Austria (HIC)	Adults (IQR, 31–46)	Yes (100%)	Outpatients with HIV	834	822	No
5	Kik et al., 2010 (111)	Netherlands (HIC)	Adults (16–45+)	No; exclusion criterion	Tuberculosis case contacts	433	339	No; exclusion criterion
6	del Corral et al., 2009 (112)	Colombia (MIC)	Adults and children (IQR, 10–42)	Unknown	Tuberculosis case contacts	2,060	2,060	No
7	Lienhardt et al., 2010 (113)	Senegal (LIC)	Adults and children (18–71)	Unknown	Tuberculosis case contacts	2,762	2,679	Yes (% NS)
8	Yoshiyama et al., 2010 (114)	Japan (HIC)	Adults and children (0–60+)	Unknown	Tuberculosis case contacts (retrospective)	NS	5,676	Yes (20% of 3,102 individuals)
9	Leung et al., 2010 (115)	China (MIC)	Adults (mean, 60)	Unknown	Outpatients with silicosis	331	308	Yes (33% of 203 individuals)
10	Harstad et al., 2010 (116)	Norway (HIC)	Adults (18–50+)	Unknown	Asylum seekers	NS	823	Yes (3%)
11	Diel et al., 2011 (117)	Germany (HIC)	Adults and children (1–62)	No; exclusion criterion	Tuberculosis case contacts	1,417	1,335	Yes (% NS)
12	Jonnalagadda et al., 2010 (118)	Kenya (LIC)	Adults (24–26)	Yes (100%)	HIV cohort with no prior tuberculosis (retrospective)	333	258	No
12	Jonnalagadda et al., 2010 (118)	Kenya (LIC)	Infants (<1)	Unknown	HIV-exposed infants (retrospective)	327	250	No
13	Joshi et al., 2011 (119)	India (MIC)	Adults (18–40)	Unknown	Health care workers with no prior tuberculosis (retrospective)	726	719	Yes (17% of 360 individuals)
14	Mahomed et al., 2011 (120)	South Africa (MIC)	Adolescents (12–18)	Unknown	Individuals with no prior tuberculosis	6,363	5,244	No
15	Torres Costa et al., 2011 (121)	Portugal (HIC)	Adults (<25–50+)	No	Health care workers	2,889	2,876	Yes (2% of 2,876 individuals)
16	Kim et al., 2011 (122)	Korea (HIC)	Adults (16+)	No	Renal transplant recipients	324	296	Yes (13% of 312 individuals)
17	Haldar et al., 2012 (123)	United Kingdom (HIC)	Adults (16–36+)	Unknown	Tuberculosis case contacts	1,769	811	Yes (12% of 851 individuals)
18	Lange et al., 2012 (124)	Germany (HIC)	Adults (18+)	Yes (11%)	Outpatients with diseases of the immunocompromised	460	460	Unknown
19	Kim et al., 2012 (125)	Korea (HIC)	Adult (18+)	Yes (100%)	Outpatients with HIV	124	120	No
20	Bergot et al., 2012 (126)	France (HIC)	Adults and children (0–65+)	No	Tuberculosis case contacts	687	674	Yes (14% of 687 individuals)

[a] Studies 16 to 20 were published after the previous systematic review by Rangaka et al. (106). HIC, high-income country; MIC, middle-income country; LIC, low-income country. Income classifications are based on the classification system of the World Bank. IQR, interquartile range; NS, not stated.

[b] Adapted from reference 106 by permission from Macmillan Publishers Ltd.

TABLE 4 Comparison of TST and IGRA[a]

Characteristic	Comments	
	TST	IGRA
Potential advantages or benefits	Simple, low-tech test	Requires fewer visits than TST for test completion (follow-up visits will be needed for both tests for IPT initiation)
	Can be done without a laboratory	Potential for boosting test response eliminated
	No equipment necessary	Results can be available within 24 to 48 h (but are likely to take longer if done in batches)
	Can be done by a trained health care worker even in remote locations	Does not have cross-reactivity with BCG
	Effect of BCG on TST results is minimal if vaccination is given at birth and not repeated	Has less cross-reactivity than TST with nontuberculous mycobacteria, though data are limited for low- and middle-income countries
	Longitudinal studies have demonstrated its predictive value, and systematic reviews of randomized trials show that isoniazid preventive therapy (IPT) is highly effective in those who are TST positive	
Risks or undesired effects	May give false-negative reactions due to infections, live virus vaccines, and other factors	Requires a blood draw (which may be challenging in some populations, including young children)
	May give false-positive results because of BCG vaccination and nontuberculous mycobacteria	Risk of exposure to blood-borne pathogens
	Requires an intradermal injection	Risk of adverse events with IGRA may be reduced compared to that with TST
	Can rarely cause adverse reactions (acute reactions, skin blistering, and ulceration)	Interpretation of serial IGRAs is complicated by frequent conversions and reversions and a lack of consensus on optimal thresholds
	Interpretation of serial TST is complicated by boosting, conversions, and reversions	Reproducibility is affected by several preanalytical and analytical factors as well as manufacturing defects
	Interpretation is affected by inter- and intrareader variation	
	Requires 48 to 72 h for a valid result	
Values and preferences	Patients may prefer to avoid visible skin reaction to TST	Patients may prefer to avoid blood draw (for cultural or technical reasons)
	Patients may prefer not to come back for repeat visit for reading the test result	Patients with prior BCG may not trust TST results and prefer IGRA
	Patients with prior BCG may not trust TST results and may be reluctant to accept IPT	
	Patients may self-read their TST results erroneously	
Resource implications	Less expensive than IGRAs (reagent cost is substantially less than IGRA kit costs), but personnel time costs will have to be factored, along with time and cost for 2 patient visits	Need to establish well-equipped laboratory, with electricity, that can perform ELISA or ELISPOT assay
	No laboratory required	Need to procure equipment and supplies for IGRA performance and quality assurance (IGRA reagents cost more than TST reagents)
	Need to establish a program with trained staff to administer and read TST results	Need for staff training, including blood-borne pathogen training
	Staff training is needed to minimize reading errors and variability (underreading, within- and between-reader variability, digit preference, etc.)	Need for cold chain for transport of kits and reagents and for their storage
	PPD must be stored at optimal temperatures	Need for careful handling (e.g., tube shaking) and processing of blood samples (incubation of samples within a specific time window) to ensure reproducibility of tests
	Only standardized PPD must be used	Availability of well-trained staff or staff to be trained
		High likelihood of false-positive conversions during serial testing

[a] Adapted from reference 137 with permission of the publisher (copyright 2012 Karger Publishers, Basel, Switzerland).

issues are addressed, recommendations regarding the cost-effectiveness of IGRAs should be interpreted with caution (131).

Oxlade and colleagues also systematically reviewed the CEA literature (132). They too reported substantial variability in the choice of test characteristics, parameters, and cost estimates used in models. When the IGRA and TST strategies were compared by using a common decision analysis model created by Oxlade and colleagues, predicted costs and effectiveness largely overlapped, emphasizing the difficulty in drawing conclusions about the cost-effectiveness of IGRAs (132). Both systematic reviews ended with recommendations for conducting cost-effectiveness analyses on IGRAs that should improve economic studies to evaluate diagnos-

tic strategies for LTBI and increase their value for informing individual and public health decisions (131, 132).

GUIDELINES AND POLICY STATEMENTS

Recently, Denkinger and colleagues summarized a number of guidelines or recommendations on the use of IGRAs (133). The recommendations in these guidelines were found to vary substantially, particularly for indications where there are limited data (e.g., children younger than 5 years of age and patients on TNF-α inhibitors). The data suggest that IGRAs are increasingly being recommended, primarily in low-incidence settings, as they confer a higher specificity combined with logistical advantages. In contrast, TST is still favored in high-incidence and low-resource settings. In low-incidence countries, especially the United States and Canada, the use of IGRAs has increased substantially over the last 5 years. The most recent, revised U.S. and Canadian IGRA guidelines were published in 2010 and 2013, respectively (134, 135).

CONCLUSIONS

Both TST and IGRAs are acceptable but imperfect LTBI tests, with advantages and disadvantages (Table 4). IGRAs offer some improvements over the TST, but the improvement, as noted by others, is incremental rather than transformational (136). There are situations where neither test is appropriate (e.g., active TB diagnosis in adults) and situations where both tests may be necessary to detect *M. tuberculosis* infection (e.g., immunocompromised populations), and there are situations where one test may be preferable to another. For example, IGRAs may be preferable to the TST in populations where BCG is given after infancy or given multiple times. In contrast, TST may be preferable to the IGRAs for serial testing of health care workers. Both TST and IGRAs have reproducibility challenges, and dichotomous cutoffs are inadequate for interpretation.

The primary goal of IGRAs is to identify those who will benefit from LTBI therapy. Unfortunately, IGRAs (and TST) are limited in this regard, for reasons including the low absolute risk of progression to disease, inability to distinguish reactivation from reinfection, reduced accuracy in immunocompromised patients, and inability to discriminate the various stages within the spectrum of LTBI (2, 88). To maximize the positive predictive value of existing LTBI tests, LTBI screening should be reserved only for those who are at sufficiently high risk of progressing to disease. Such high-risk individuals may be identifiable by using multivariable risk prediction models that incorporate risk factors (e.g., the Online TST/IGRA Interpreter [www.tstin3d.com]) (10) and by using serial testing to resolve underlying phenotypes. In the longer term, highly predictive biomarkers need to be identified. This is an active area of research (93, 129), and future generations of LTBI tests should overcome the limitations of current assays.

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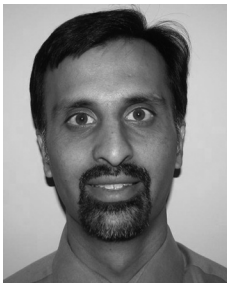

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