

Interferon-Gamma Release Assays (IGRAs)

What are they?

Interferon-Gamma Release Assays (IGRAs) are whole-blood tests that can aid in diagnosing *Mycobacterium tuberculosis* infection. They do not help differentiate latent tuberculosis infection from tuberculosis disease. Two IGRAs that have been approved by the U.S. Food and Drug Administration (FDA) are commercially available in the U.S.:

- QuantiFERON®-TB Gold In-Tube test (QFT-GIT);
- T-SPOT®.TB test (T-Spot)

How do they work?

IGRAs measure a person's immune reactivity to *M. tuberculosis*. White blood cells from most persons that have been infected with *M. tuberculosis* will release interferon-gamma (IFN-g) when mixed with antigens (substances that can produce an immune response) derived from *M. tuberculosis*.

To conduct the tests, fresh blood samples are mixed with antigens and controls. The antigens, testing methods, and interpretation criteria for IGRAs differ (see Table 1).

What are the advantages of IGRAs?

- Requires a single patient visit to conduct the test.
- Results can be available within 24 hours.
- Does not boost responses measured by subsequent tests.
- Prior BCG (bacille Calmette-Guérin) vaccination does not cause a false-positive IGRA test result.

What are the disadvantages and limitations of IGRAs?

- Blood samples must be processed within 8-30 hours after collection while white blood cells are still viable.
- Errors in collecting or transporting blood specimens or in running and interpreting the assay can decrease the accuracy of IGRAs.
- Limited data on the use of IGRAs to predict who will progress to TB disease in the future.
- Limited data on the use of IGRAs for:
 - Children younger than 5 years of age;
 - Persons recently exposed to *M. tuberculosis*;
 - Immunocompromised persons; and
 - Serial testing.

Table 1: Differences in Currently Available IGRAs

	QFT-GIT	T-Spot
Initial Process	Process whole blood within 16 hours	Process peripheral blood mononuclear cells (PBMCs) within 8 hours, or if T-Cell Xtend® is used, within 30 hours.
<i>M. tuberculosis</i> Antigen	Single mixture of synthetic peptides representing ESAT-6, CFP-10 & TB7.7	Separate mixtures of synthetic peptides representing ESAT-6 & CFP-10
Measurement	IFN-g concentration	Number of IFN-g producing cells (spots)
Possible Results	Positive, negative, indeterminate	Positive, negative, indeterminate, borderline

What are the steps in administering an IGRA test?

Confirm arrangements for testing in a qualified laboratory, and arrange for delivery of the blood sample to the laboratory in the time the laboratory specifies to ensure testing of samples with viable blood cells.

- Draw a blood sample from the patient according to the test manufacturer's instructions.
- Schedule a follow-up appointment for the patient to receive test results.
- Based on test results, provide follow-up evaluation and treatment as needed.

How do you interpret IGRA test results?

IGRA interpretations are based on the amount of IFN-g that is released or on the number of cells that release IFN-g. Both the standard qualitative test interpretation (positive, negative, or indeterminate) and the quantitative assay measurements (Nil, TB, and Mitogen concentrations or spot counts) should be reported.

As with the tuberculin skin tests (TSTs), IGRAs should be used as an aid in diagnosing infection with *M. tuberculosis*. A positive test result suggests that *M. tuberculosis* infection is likely; a negative result suggests that infection is unlikely. An indeterminate result indicates an uncertain likelihood of *M. tuberculosis* infection. A borderline test result (T-Spot only) also indicates an uncertain likelihood of *M. tuberculosis* infection.

A diagnosis of latent TB infection (LTBI) requires that TB disease be excluded by medical evaluation. This should include checking for signs and symptoms suggestive of TB disease, a chest radiograph, and, when indicated, examination of sputum or other clinical samples for the presence of *M. tuberculosis*. Decisions about a diagnosis of *M. tuberculosis* infection should also include epidemiological and historical information.

Recommendations on when to use IGRA tests

- IGRAs can be used in place of (but not in addition to) TST in all situations in which CDC recommends TST as an aid in diagnosing *M. tuberculosis* infection, with preferences and special considerations noted below. This includes contact investigations, testing during pregnancy, and screening of health care workers and others undergoing serial evaluation for *M. tuberculosis* infection. Despite the indication of a preference, use of the alternative test (FDA-approved IGRA or TST) is acceptable medical and public health practice. Caution in interpretation should be used when testing certain populations because of limited data on the use of IGRAs (see Updated Guidelines for Using Interferon Gamma Release Assays to Detect *Mycobacterium tuberculosis* Infection, United States).
- Populations in which IGRAs are preferred for testing:
 - Persons who have received BCG (either as a vaccine or for cancer therapy); and
 - Persons from groups that historically have poor rates of return for TST reading.
- TST is preferred over IGRAs for testing children less than 5 years of age.
- As with TST, IGRAs generally should not be used for testing persons who have a low risk of infection and a low risk of disease due to *M. tuberculosis*.
- Each institution and TB control program should evaluate the availability and benefits of IGRAs in prioritizing their use.

Recommendations on when to use IGRA tests (cont.)

- Routine testing with both TST and IGRA is not recommended. However, results from both tests might be useful in the following situations:

- o When the initial test is negative and:
 - The risk for infection, the risk for progression to disease, and the risk for a poor outcome are high (e.g., HIV infected persons or children under 5 years of age who are exposed to a person with infectious TB).
 - There is clinical suspicion for TB disease (e.g., signs, symptoms, and/or radiographic evidence suggestive of TB disease) and confirmation of *M. tuberculosis* infection is desired.
 - Taking a positive result from a second test as evidence of infection increases detection sensitivity.
- o When the initial test is positive and:
 - Additional evidence of infection is required to encourage acceptance and adherence (e.g., foreign-born healthcare workers who believe their positive TST is due to BCG). A positive IGRA might prompt greater acceptance of treatment for LTBI as compared with a positive TST alone.
 - The person has a low risk of both infection and progression from infection to TB disease. Requiring a positive result from the second test as evidence of infection increases the likelihood that the test reflects infection. An alternative is to assume, without additional testing, that the initial result is a false positive or that the risk for disease does not warrant additional evaluation or treatment, regardless of test results.

- o In addition, repeating an IGRA or performing a TST might be useful when the initial IGRA result is indeterminate, borderline, or invalid and a reason for testing persists.

Multiple negative results from any combination of these tests cannot exclude *M. tuberculosis* infection. Steps should be taken to minimize unnecessary and misleading testing of persons at low risk.

Selection of the most suitable test or combination of tests for detection of *M. tuberculosis* infection should be based on the reasons and the context for testing, test availability, and overall cost of testing.

Can IGRAs Be Given To Persons Receiving Vaccinations?

As with TST, live virus vaccines might affect IGRA test results. However, the effect of live virus vaccination on IGRAs has not been studied. Until additional information is available, IGRA testing in the context of live virus vaccine administration should be done as follows:

- Either on the same day as vaccination with live-virus vaccine or 4-6 weeks after the administration of the live-virus vaccine
- At least one month after smallpox vaccination

Additional Information

Centers for Disease Control and Prevention. Updated Guidelines for Using Interferon Gamma Release Assays to Detect *Mycobacterium tuberculosis* Infection, United States. MMWR 2010; 59 (No. RR-5) <http://www.cdc.gov/mmwr/pdf/rr/rr5905.pdf>