TOYO®

INSTRUCTION FOR USE
Tuberculosis (TB) Test

For Tuberculosis Antibody
Detection in Whole Blood / Serum / Plasma

in vitro diagnostic test

Only for professional *in vitro* diagnostic use

Product Code: TTB01

Test for the qualitative detection of TB antibodies (Isotypes IgG, IgM and IgA) in whole blood, serum or plasma specimens

BACKGROUND INFORMATION

Tuberculosis (TB) is spread primarily via airborne transmission of aerosolized droplets developed by coughing, sneezing and talking. Areas of poor ventilation pose the greatest risk of exposure to infection. TB is a major cause of morbidity and mortality worldwide, resulting in the greatest number of deaths due to a single infectious agent. The World Health Organization reports that more than 8 million new cases of active tuberculosis are diagnosed annually. Almost 3 million deaths are attributed to TB as well. Timely diagnosis is crucial to TB control, as it provides early initiation of therapy and limits further spread of infection. Several diagnostic methods for detecting TB have been used over the years including skin test, sputum smear, and sputum culture and chest x-ray. But all these methods have some limitations. Newer tests, such as PCR-DNA amplification or interferongamma assay, have been recently introduced. However, the turn-around time for these tests is long, they require laboratory equipment and skilled personnel, and some are neither cost effective nor easy to use. These tests are also expensive and not practical for developing countries. Serological methods constitute an attractive alternative, since TB serodiagnosis is simple, inexpensive, relatively non-invasive, and it does not depend on detection of mycobacteria.

INTENDED USE

Tuberculosis Test is a rapid chromatographic immunoassay for the qualitative detection of TB antibodies (Isotypes of IgG, IgM and IgA) generated against TB antigens in whole blood, serum or plasma specimens.

REAGENTS

Recombinant TB antigen conjugated with colloidal colored particles and recombinant TB antigens immobilized in the membrane.

METHOD

Tuberculosis Test uses solid-phase immunochromatographic technology for the qualitative detection of anti-TB in human whole blood / serum / plasma. The test is a two-site immunometric assay which selectively detects anti-TB in samples with a high degree of sensitivity. Recombinant TB antigens were immobilized on the test area "T" of the nitrocellulose membrane. Recombinant TB antigens conjugated with colloidal colored particles, were dried on a conjugate pad. Sample is introduced from sampling pad. If there is anti-TB at detectable level in the sample, anti-TB binds to the mobile recombinant TB antigens conjugated with colloidal colored particles. Together they move to the test area "T". Anti-TB molecules bind to the immobilized recombinant TB antigens and as a result of this, anti-TB molecules that have already bound to mobile recombinant TB antigens (conjugated with colloidal colored particles) become immobilized in the test area "T" thus creating a visible colored signal due to the accumulation of colloidal colored particles in the test area "T" (a colored test line), indicating positive test result. If there is no anti-TB at detectable level in the sample then sample moves to the test area "T" together with unbound (free) recombinant TB antigens conjugated with colloidal colored particles.
Immobilized recombinant TB antigens cannot bind to mobilized recombinant TB antigens conjugated with colloidal colored particles, therefore no visible colored signal in test area "T" (no colored test line) can be obtained, indicating negative test result. Regardless of anti-TB content of the liquid sample, accumulation of colloidal colored particles produces a visible colored signal in the control area "C" (a colored control line), indicating a valid test result. Colored line should be visible in the control area "C" in every case; if no visible colored line in control area "C", test result should be indicated as invalid.

PRECAUTIONS AND LIMITATIONS

1. For professional and *in vitro* diagnostic use only.
2. Read this insert completely and carefully prior to use of the test. Test must be performed in strict accordance with these instructions to obtain accurate results.
3. Do not use test kit beyond expiry date. The test device is single use. Do not reuse.
4. The test device should remain in its original sealed pouch until usage. Do not use the test if the seal is broken or the pouch is damaged.
5. Wear disposable gloves while performing the test.
6. Use a new pipette for each sample.
7. This test kit should be handled only by adequately qualified personnel trained in laboratory procedures and familiar with their potential hazards. Wear appropriate protective clothing, gloves and eye/face protection and handle appropriately with the requisite Good Laboratory Practices.
8. Do not use potassium oxalate as anticoagulant to collect plasma and whole blood samples.
9. All patient samples should be handled as taking capable of transmitting disease into consideration. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of samples.
10. This test will indicate only the selectively total anti-TB in the sample, and should not be used as the only basis for the diagnosis of Tuberculosis.

As with all diagnostic tests, it should be kept in mind that an identification diagnosis can't be based on a single test result. Diagnosis can only be reached by an expert after the evaluation of all clinical and laboratory findings.

STORAGE

Test device should be kept away from direct sunlight, moisture, heat and radiation sources. Store at 4 - 30°C (39 - 86°F). Do not freeze.
The test in the original packaging retains stable until expiry date at storage conditions. The test device should be used in maximum one hour after the foil is opened.
Kit components : Test cassettes, pipettes, diluent (for whole blood samples only) and instructions for use.
Additional materials required but not provided : Sample collection tube, centrifuge and timer.
Additional materials recommended but not provided : Micropipettes to deliver mentioned amount of sample in the test procedure, negative and positive control materials.

SAMPLE COLLECTION AND PREPARATION

The test can be performed using whole blood, serum or plasma. To avoid hemolysis, serum or plasma should be separated from blood as soon as possible.
For Whole Blood Samples : Test should be performed immediately with whole blood samples. Otherwise, whole blood samples should be stored at 2 - 8 °C with anticoagulants (EDTA, heparin, citrate should be used) to avoid coagulation until they are being tested in a period of 2 days after collection.
For Serum Samples: Collect blood into a collection tube without anticoagulant, leave to settle for 30 minutes for blood coagulation and then centrifuge the blood. At the end of centrifuge period remaining supernatant is used as serum.
For Plasma Samples: Collect blood into a collection tube with anticoagulants (EDTA, heparin, citrate should be used) to avoid coagulation of blood sample and then centrifuge the blood. At the end of centrifuge period supernatant is used as plasma.

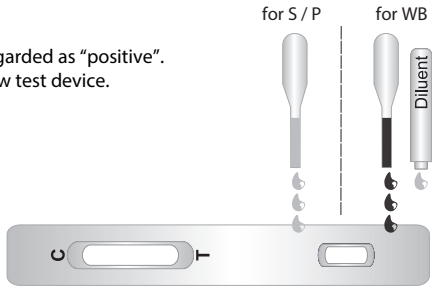
Do not use turbid, hemolyzed samples. If the sample cannot be tested on the day of collection, store the serum, plasma samples in a refrigerator or freezer. Do not freeze and thaw the serum, plasma samples repeatedly. Do not freeze whole blood sample. Bring the samples to room temperature before testing. Frozen samples must be completely thawed and mixed well prior to testing. Turbid test samples should be centrifuged. Using of frozen and thawed samples should be avoided whenever possible, due to the blocking of the membrane by the debris.

TEST PROCEDURE

1. Bring the tests and whole blood / serum / plasma samples to room temperature. Take the test out of its pouch.
2. **For Whole Blood Samples:** Draw whole blood into pipette and put 3 drops (75 µl) into the sample well of the cassette. Immediately after, 1 drop of diluent is added into the sample well and allowed to soak in.
For Serum / Plasma Samples: Draw serum / plasma into pipette and put 3 drops (75 µl) into the sample well of the cassette. Do not use diluent for serum / plasma samples.
Avoid the formation of any air bubbles.
3. Results should be read at 10 minutes as shown below. Do not interpret results beyond 30 minutes, results forming after 30 minutes should be regarded as invalid.

INTERPRETATION OF RESULTS

Negative : Only one colored line is visible in "C" area.
Positive : Two colored lines are visible in "C" and "T" areas.
Low concentration of TB antibody may cause a faint line in "T" area. Even such a faint line in "T" area should be regarded as "positive".
Invalid : No colored line is visible or only one colored line is visible in "T" area; test should be repeated using a new test device.



QUALITY CONTROL

Tests have built in procedural quality control features. When the test is complete, the user will see a colored line in the "C" area of the test on negative samples and a colored line in the "T" and "C" area on positive samples. The appearance of the control "C" line is considered as an internal procedural control. This line indicates that sufficient volume of sample was added as well as valid test result. It is recommended that a negative control and a positive control be used to verify proper test performance as an external control. Users should follow appropriate federal, state and local guidelines concerning the external quality controls.

PERFORMANCE EVALUATION

Clinical Sensitivity and Specificity

TB Tuberculosis Test vs. Smear/Culture

Method		Smear/Culture		Total Results
	Results	Positive	Negative	
Tuberculosis Test	Positive	74	3	77
	Negative	13	282	295
Total Results		87	285	372

Sensitivity : 85 % Specificity : 99 %

+ Predictive V : 96,1 % - Predictive V : 96 %

Intra-Assay:
Within-run precision has been determined by using 10 replicates of four specimens: a negative, a low positive, middle positive and a high positive. The negative, low positive, middle positive, high positive values were correctly identified.

Intra-Assay:
Between-run precision has been determined by using 3 independent assays on the same four specimens: The negative, low positive, middle positive, high positive values. Tuberculosis Test has been tested using negative, low positive, middle positive and high positive specimens. The specimens were correctly identified.

Cross Reactivity:
Tuberculosis Test has been tested with specimens positive for: anti-HIV, pulmonary diseases, anti-CMV, Rheumatoid factor (RF), anti-HCV and specimens from children below 15 years, who have been administered BCG vaccine. No cross-reactivity was observed, indicating that the performance of the Tuberculosis Test is stable in presence of these factors.

INTERFERING SUBSTANCES

Tuberculosis Test has been tested for possible interference from visibly hemolyzed and lipemic specimens, as well as serum specimens containing high bilirubin levels. Results indicate that no interference was observed in specimens containing up to 500 mg/dL hemoglobin; up to 30 mg/dL bilirubin; and up to 2.000 mg/dL human serum albumin.

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TÜRKLAB TIBBİ MALZEMELER SAN. ve TİC. A.Ş.
ITOB 10017 Sokak No: 2 Tekeli Menderes / Izmir / TURKEY
T: +90 232 376 80 81 • F: +90 232 376 80 40 • www.turklab.com.tr • info@turklab.com.tr

CE

Manufacturer
Consult instruction for use
Attention, see instruction for use
In vitro diagnostic medical device
For single use only
Number of test
REF Catalog number
Storage temperature
LOT Lot number
Expiry date