OnSite® TB IgG/IgM Combo Rapid Test

REF R0053C CE

Instructions for Use

INTENDED USE

The *OnSite* TB IgG/IgM Combo Rapid Test is a lateral flow chromatographic immunoassay for the simultaneous detection and differentiation of anti-*Mycobacterium tuberculosis* (*M. TB*) IgM and IgG in human serum, plasma or whole blood. It is intended to be used by professionals as a preliminary test result to aid in diagnosis of infection with *M. TB*.

Any interpretation or use of this preliminary test result must also rely on other clinical findings as well as on the professional judgment of health care providers. Alternative test method(s) should be considered to confirm the test result obtained by this device.

SUMMARY AND EXPLANATION OF THE TEST

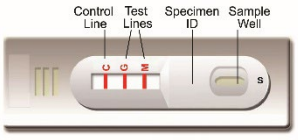
Tuberculosis is a chronic, communicable disease caused principally by *M. TB hominis* (Koch's bacillus), occasionally *by M. TB bovis*. The lungs are the primary target, but any organ may be infected.

The risk of TB infection exponentially declined in the 20th century. However, the recent emergence of drug-resistant strains[1], particularly among patients with AIDS[2], has rekindled interest in TB. The incidence of infection was reported to be around 8 million cases per year with a death rate of 3 million per year. The mortality exceeded 50% in some African countries with high HIV rates[3,4].

The initial clinical suspicion and radiographic findings with subsequent laboratory confirmation by sputum examination and culture are the traditional method(s) in the diagnosis of active TB[5,6]. However, these methods either lack sensitivity or are time consuming, and are particularly not suitable for patients who are unable to produce adequate sputum, smear-negative or are suspected to have extra-pulmonary TB.

The *OnSite* TB IgG/IgM Combo Rapid Test is developed to alleviate these obstacles. The test detects anti-*M. TB* IgM and IgG in serum, plasma or whole blood in 15 minutes. An IgM positive result indicates a fresh *M. TB* infection, while an IgG positive result suggests a previous or chronic infection. Since the test uses *M. TB* specific antigens[7-9], it can still detect IgM antibodies in individuals who were recently exposed to *M. TB* but were vaccinated with BCG. In addition, the test can be performed by minimally skilled personnel without cumbersome laboratory equipment.

TEST PRINCIPLE



The *OnSite* TB IgG/IgM Combo Rapid Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a colored conjugate pad containing *M. TB* antigens conjugated with colloidal gold (M. TB conjugates) and a control antibody conjugated with colloidal gold, 2) a nitrocellulose membrane strip containing two test lines (M and G lines) and a control line (C line). The M line is pre-coated with monoclonal anti-human IgM for the detection of anti-*M. TB* IgM, the G line is pre-coated with reagents for the detection of anti-*M. TB* IgG, and the C line is pre-coated with a control line antibody.

When an adequate volume of test specimen is dispensed into the sample well of the cassette, the specimen migrates by capillary action across the cassette. Anti-*M. TB* IgM if present in the specimen will bind to the *M. TB* conjugates. The immunocomplex is then captured on the membrane by the pre-coated anti-human IgM antibody forming a colored M line, indicating an IgM positive test result.

Anti-*M. TB* IgG if present in the specimen, will bind to the *M. TB* conjugates. The immunocomplex is then captured by the pre-coated reagents on the membrane forming a colored G line, indicating an IgG positive test result.

Absence of any test lines (M and G) suggests a negative result. The test contains an internal control (C line) which should exhibit a colored line of the immunocomplex of the control antibodies regardless of color development on any of the T lines. Otherwise, the test result is invalid and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

1. Individually sealed foil pouches containing:
   a. One cassette device
   b. One desiccant
2. Plastic droppers
3. Sample diluent (REF SB-R0053, 5 mL/bottle)
4. Instructions for Use

MATERIALS MAY BE REQUIRED AND NOT PROVIDED

1. Positive Control
2. Negative Control

MATERIALS REQUIRED BUT NOT PROVIDED

1. Clock or timer
2. Lancing device for whole blood test

WARNINGS AND PRECAUTIONS

For *In Vitro* Diagnostic Use

1. Read these instructions for use completely before performing the test. Failure to follow the instructions could lead to inaccurate test results.
2. Do not open the sealed pouch unless ready to conduct the assay.
3. Do not use expired devices.
4. Bring all reagents to room temperature (15-30°C) before use.
5. Do not use the components in any other type of test kit as a substitute for the components in this kit.
6. Do not use hemolyzed blood specimen for testing.
7. Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after performing the test.
8. Users of this test should follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other blood-borne pathogens.
9. Do not smoke, drink, or eat in areas where specimens or kit reagents are being handled.
10. Dispose of all specimens and materials used to perform the test as bio-hazardous waste.
11. Handle the negative and positive controls in the same manner as patient specimens.
12. The test result should be read 15 minutes after a specimen is applied to the sample well or sample pad of the device. Any results interpreted outside of the 15 minute window should be considered invalid and must be repeated.
13. Do not perform the test in a room with strong air flow, i.e. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test device unopened at 2-30°C. If stored at 2-8°C, ensure that the test device is brought to room temperature before opening. The test device is stable through the expiration date printed on the sealed pouch. Do not freeze the kit or expose the kit to temperatures above 30°C.

SPECIMEN COLLECTION AND HANDLING

Consider any materials of human origin as infectious and handle them using standard bio-safety procedures.

Plasma/Serum

Step 1: Collect venous blood by venipuncture into collection tubes containing EDTA, citrate or heparin anticoagulants for plasma, or collection tubes containing no anticoagulants for serum.

Step 2: A) To prepare plasma specimens, centrifuge the blood and carefully withdraw the plasma into a new pre-labeled tube.

B) To prepare serum specimens, allow blood to clot, centrifuge and carefully withdraw the serum into a new pre-labeled tube.

Test serum/plasma specimens as soon as possible after collection, or store refrigerated at 2-8°C for up to 5 days or freeze at -20°C for longer storage.

Avoid multiple freeze-thaw cycles. Prior to testing, bring frozen specimens to room temperature slowly and mix gently. Specimens containing visible particulate matter should be clarified by centrifugation before testing.

Whole Blood

Collect whole blood by either fingertip puncture, or by venipuncture. Collect venous whole blood into a collection tube containing EDTA, citrate or heparin anticoagulants. Do not use hemolyzed blood for testing.

Test whole blood specimens as soon as possible after collection. Do not freeze whole blood specimens

Whole blood specimens should be stored in refrigeration (2-8°C), if not tested immediately. The specimens must be tested within 24 hours of collection.

Note: Do not test venous specimens demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid possible interference with the assay result.

ASSAY PROCEDURE

Step 1: Bring the specimen and test components to room temperature if refrigerated or frozen. Once thawed, mix the specimen well prior to performing the assay.

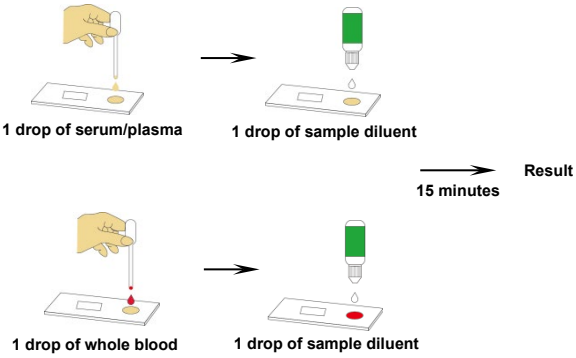
Step 2: When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.

Step 3: Be sure to label the device with the specimen's ID number.

Step 4: Fill the plastic dropper with the specimen.

Holding the dropper vertically, dispense 1 drop (about 30-45 µL) of serum/plasma or 1 drop of whole blood (about 40-50 µL) into the sample well making sure that there are no air bubbles.

Immediately add 1 drop (about 35-50 µL) of sample diluent to the sample well with bottle positioned vertically.



Step 5: Set up timer.

Step 6: Read results at 15 minutes. Positive results may be visible as soon as 2 minutes. All results must be confirmed at the end of 15 minutes only. ***However, any results interpreted outside 15 minutes should be considered invalid and must be repeated. Discard used device after interpreting the results following local laws governing the disposal of device.***

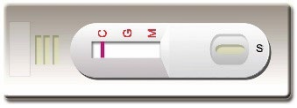
QUALITY CONTROL

1. **Internal Control:** This test contains a built-in control feature, the C line. The C line develops after adding the specimen and the sample diluent. If the C line does not develop, review the whole procedure and repeat the test with a new device.

2. **External Control:** Good Laboratory Practice recommends using external controls, positive and negative, to assure the proper performance of the assay, particularly under the following circumstances:
   a. A new operator uses the kit, prior to performing the testing of specimens.
   b. A new lot of test kits is used.
   c. A new shipment of kits is used.
   d. The temperature during storage of the kit falls outside of 2-30°C.
   e. The temperature of the test area falls outside of 15-30°C.
   f. To verify a higher than expected frequency of positive or negative results.
   g. To investigate the cause of repeated invalid results.

INTERPRETATION OF ASSAY RESULT

1. **NEGATIVE RESULT:** If only the C line is present, the absence of any color in both the test lines (M and G) indicates that there is no detectable anti-*M. TB* IgG and IgM antibodies. The result is negative or non-reactive.



2. **POSITIVE RESULT:**

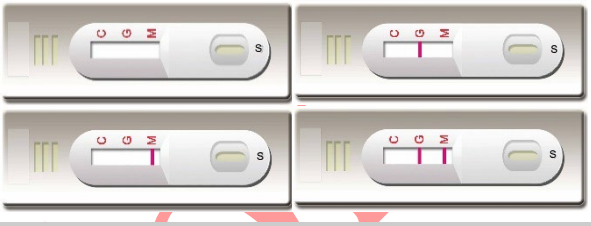
2.1 In addition to the presence of the C line, if only the M line develops, the test indicates the presence of anti-*M. TB* IgM. The result is IgM positive or reactive.

2.2 In addition to the presence of the C line, if only the G line develops, the test indicates the presence of anti-*M. TB* IgG. The result is IgG positive or reactive.

2.3 In addition to the presence of the C line, if both the M and the G lines develop, the test indicates the presence of anti-*M. TB* IgG and IgM. The result is IgG and IgM positive or reactive.

Specimens with positive results should be confirmed with alternative testing method(s) and clinical findings before a diagnostic decision is made.

3. **INVALID:** If no C line develops, the assay is invalid regardless of any color in the test lines as indicated below. Repeat the assay with a new device.



PERFORMANCE CHARACTERISTICS

1. **Clinical Performance**

1.1. Positive Agreement in confirmed TB cases

A total of 289 specimens from TB patients confirmed with bacteriology tests were tested with the *OnSite* TB IgG/IgM Combo Rapid Test. The results for all subjects are shown in the following table:

Result	*OnSite* TB IgG/IgM Combo Rapid Test		
	Positive for IgG	Positive for IgM	Positive for IgG or IgM
Positive	84.1% (243/289)	2.4% (7/289)	84.1% (243/289)

The positive agreement is 84.1% (95% CI: 79.4%-88.1%)

1.2. Diagnostic accuracy in comparison with ELISA

A total of 223 specimens were tested by the *OnSite* TB IgG/IgM Combo Rapid Test and a commercial TB IgG ELISA kit. Comparison of the results for all subjects is shown in the following table:

TB IgG ELISA	*OnSite* TB IgG/IgM Combo Rapid Test		
	Positive	Negative	Total
Positive	34	5	39
Negative	10	174	184
Total	44	179	223

Relative Sensitivity: 87.2% (95% CI: 72.6%-95.7%),
Relative Specificity: 94.6% (95% CI: 90.2%-97.4%),
Relative Agreement: 93.2% (95% CI: 89.2%-96.2%)

2. **Cross-Reactivity**

No false positive results were observed on 5-10 specimens from the following disease states or special conditions, respectively:

CMV	Malaria	HCV	HIV	Syphilis
ANA	HAMA	RF (up to 2,500 IU/mL)		

3. **Interference**

Common substances (such as pain and fever medication and blood components) may affect the performance of the *OnSite* TB IgG/IgM Combo Rapid Test. The results are presented in the following table and demonstrate that at the concentrations tested, the substances studied do not affect the performance of *OnSite* TB IgG/IgM Combo Rapid Test.

List of potentially interfering substances and concentrations tested:

1. Albumin	50 g/L	6. Heparin	3,000 U/L
2. Bilirubin	20 mg/dL	7. EDTA	3.4 µmol/L
3. Creatinine	442 µmol/L	8. Ibuprofen	20 mg/dL
4. Glucose	55 mmol/L	9. Sodium citrate	3.8%
5. Salicylic acid	4.34 mmol/L		

LIMITATIONS OF TEST

1. The Assay Procedure and the Interpretation of Assay Result sections must be followed closely when testing for the presence of antibodies to *M. TB* in serum or plasma or whole blood from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The *OnSite* TB IgG/IgM Combo Rapid Test is limited to the qualitative detection of anti-*M. TB* IgG and IgM in human serum or plasma or whole blood. The intensity of the test line does not have a linear correlation with the antibody titer in the specimen.
3. The test also recognizes antibodies to *M. bovis* and *M. africanum.*
4. An IgG positive or reactive response may be detected in BCG vaccinated personnel.
5. A negative or non-reactive result for an individual subject indicates absence of detectable antibodies to *M. TB*. However, a negative or non-reactive test result does not preclude the possibility of exposure to or infection with *M. TB.*
6. A negative or non-reactive result can occur if the quantity of the antibodies to *M. TB* present in the specimen is below the detection limits of the assay or the antibodies that are detected are not present during the stage of disease in which a sample is collected.
7. Immunocompromised conditions such as HIV infection may reduce the test sensitivity.
8. Severe forms of infection may progress rapidly. If symptoms persist while the result from the *OnSite* TB IgG/IgM Combo Rapid Test is negative or non-reactive, it is recommended to test with an alternative test method, such as PCR.
9. Some specimens containing unusually high titers of heterophile antibodies or rheumatoid factor may affect expected results.
10. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

REFERENCES

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Index of Symbols

	Consult instructions for use	IVD	For *in vitro* diagnostic use only		Use by
REF	Catalog #	LOT	Lot Number	Σ N	Tests per kit
2°C 30°C	Store between 2-30°C	EC REP	Authorized Representative		Do not reuse
	Manufacturer		Date of manufacture		

CTK Biotech, Inc.
13855 Stowe Drive
Poway, CA 92064, USA
Tel: 858-457-8698
Fax: 858-535-1739
E-mail: info@ctkbiotech.com

EC REP **MDSS GmbH**
Schiffgraben 41
30175 Hannover, Germany

PI-R0053C Rev E3.0
Date released: 2022-08-30
English version

For Export Only, Not for Re-sale in the USA.