# *OnSite*™ Toxo IgG/IgM Combo Rapid Test

**REF R0234C**

## INTENDED USE

The *OnSite* Toxo IgG/IgM Combo Rapid Test is a lateral flow immunoassay for the simultaneous detection and differentiation of IgM and IgG antibodies to *Toxoplasma gondii* (*T. gondii)* in human serum, plasma or whole blood. It is intended to be used by professionals as a screening test and provides a preliminary test result to aid in the diagnosis of infection with *T. gondii*.

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

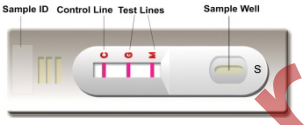
*T. gondii* is an obligate intracellular protozoan parasite with a worldwide distribution[1,2]. Serological data indicates that approximately 6-47% of the population is chronically infected with the organism[3]. A *T. gondii* infection occurs essentially without knowledge of the patient and may be unrelated to direct exposure to a cat (e.g., by ingestion of vegetables or water contaminated with oocysts or ingestion of undercooked meat contaminated with cysts)[4]. An initial *T. gondii* infection and the subsequent chronic infection are clinically undetected in 80% to 89% of healthy individual[5]. In immunosuppressed patients, such as patients infected with HIV, both acute and recurrent toxoplasmosis can have severe clinical manifestations[5]. In pregnant women, an acute *T. gondii* infection may lead to serious fetal congenital mental retardation, blindness and hydrocephaly[6].

Methods for diagnosis of toxoplasmosis include PCR, histological diagnosis, parasite isolation and serology[4]. Serological detection of *T. gondii* specific IgM and IgG antibodies is the primary method for diagnosis of toxoplasmosis. Diagnosis of toxoplasmosis is helpful in determining the risk for congenital toxoplasmosis during pregnancy[4].

IgM anti-*T. gondii* develops during acute primary infection with *T. gondii* and declines generally within a few months[4]. IgG anti-*T. gondii* develops generally within 1–2 weeks post infection, peaks within 1–2 months, and usually persists for life[4]. The absence of IgG antibodies before or early in pregnancy allows the identification of women at risk of acquiring infection[7]. Additionally, the presence of IgG allows the identification of immunocompromised patients at risk for the reactivation of a latent infection[7].

The *OnSite* Toxo IgG/IgM Combo Rapid Test detects IgM and IgG anti-*T. gondii* in human serum, plasma or whole blood by utilizing *T. gondii*-specific antigens. The test can be performed within 10 minutes by minimally skilled personnel without the use of laboratory equipment.

## TEST PRINCIPLE




The *OnSite* Toxo IgG/IgM Combo Rapid Test is a lateral flow chromatographic immunoassay. The test strip in cassette device consists of: 1) a burgundy colored conjugate pad containing a recombinant *T. gondii* antigen conjugated with colloidal gold (Toxo conjugates) and a control antibody conjugated with colloidal gold and 2) a nitrocellulose membrane strip containing two test lines (G and M lines) and a control line (C line). The G line is pre-coated with anti-human IgG for detection of IgG anti-*T. gondii*. The M line is pre-coated with mouse anti-human IgM for detection of IgM anti-*T. gondii*. The C line is pre-coated with a control antibody.

When an adequate volume of test specimen and sample diluent is dispensed into the sample well and buffer well, respectively, the specimen migrates by capillary action across the test strip. IgM anti-*T. gondii,* if present in the specimen, will bind to the Toxo conjugates. The immunocomplex is then captured on the membrane by the pre-coated anti-human IgM forming a burgundy colored M line, indicating an IgM anti-*T. gondii* positive test result. IgG anti-*T. gondii,* if present in the specimen, will bind to the Toxo conjugates. The immunocomplex is then captured on the membrane by the pre-coated anti-human IgG forming a burgundy colored G line, indicating an IgG anti-*T. gondii* positive test result.

Absence of any test lines (G or M) suggests a negative test result. The test contains an internal control (C line) which should exhibit a burgundy colored line of the immunocomplex of the control antibodies, regardless of color development on the test lines (G and M). If no control line (C line) develops, 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. 10 µL capillary tubes
3. Sample diluent (REF SB-R0234, 5 mL/bottle)
4. One package insert (instructions for use)

## MATERIALS MAY BE REQUIRED BUT 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. This package insert must be read completely before performing the test. Failure to follow the insert may lead to inaccurate test results.
2. Do not open the sealed pouch until ready to conduct the assay.
3. Do not use expired devices or components.
4. Bring all reagents to room temperature (15-30°C) before use.
5. Do not use components from another test kit to substitute for components of this kit.
6. Do not use hemolyzed blood specimens 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 negative and positive controls in the same manner as patient specimens.
12. The test result should be read 10-15 minutes after a specimen is applied to the sample well or sample pad of the device. Any results interpreted outside of the 10-15 minute window should be considered invalid and must be repeated.
13. Do not perform the test in a room with strong air flow, e.g. an electric fan or strong air conditioning.

## REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test devices 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**

Step 1: Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively, in Vacutainer®) by venipuncture.
Step 2: Separate the plasma by centrifugation.
Step 3: Carefully withdraw the plasma into new pre-labeled tube.

**Serum**

Step 1: Collect blood specimen into a red top collection tube (containing no anticoagulants in Vacutainer®) by venipuncture.
Step 2: Allow the blood to clot.
Step 3: Separate the serum by centrifugation.
Step 4: Carefully withdraw the serum into a new pre-labeled tube.

Test specimens as soon as possible after collecting. Store specimens at 2-8°C, if not tested immediately for up to 5 days. The specimens should be frozen 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. Do not use specimens demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid interference with result interpretation.

**Whole Blood**

Drops of whole blood can be obtained by either finger tip puncture or venipuncture. Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively in Vacutainer®). Do not use hemolyzed blood for testing.

Whole blood specimens should be stored at 2-8°C if not tested immediately. The specimens must be tested within 24 hours of collection.

## ASSAY PROCEDURE

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

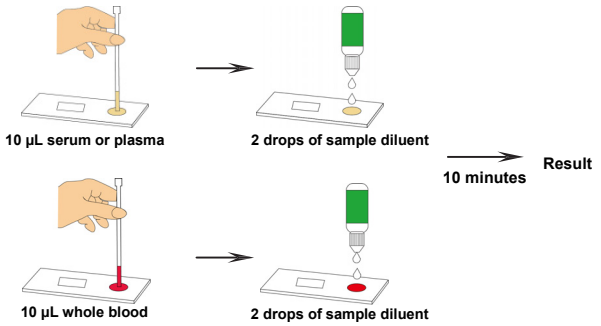
Step 2: When ready to test, open the pouch at the notch and remove the device. Place the test device on a clean, flat surface.

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

Step 4: Fill the capillary tube with specimen not exceeding the specimen line as shown in the images below. The volume of specimen is approximately 10 µL. ***For better precision, transfer specimen using a pipette capable of delivering a 10 µL volume.***

Holding the capillary tube vertically, dispense the entire specimen into the center of the sample well making sure that there are no air bubbles.

Immediately add 2 drops (about 60-80 µL) of sample diluent to the sample well with bottle positioned vertically.




Step 5: Set up the timer.

Step 6: Result should be read at 10 minutes. Positive results may be visible in as soon as 1 minute. Negative results must be confirmed at the end of 15 minutes only. ***Any results interpreted outside of the 10-15 minute window should be considered invalid and must be repeated. Discard used devices after interpreting the result following local requirements governing the disposal of devices.***

## 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 entire 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 the specimens.
   b. A new lot of test kits is used.

c. A new shipment of test kits is used.
d. The temperature during storage of the kits 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 develops, the test indicates that anti-*T. gondii* antibodies are not detected in the specimen. 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 is developed, the test indicates the presence of IgM anti-*T. gondii*. The result is IgM anti-*T. gondii* positive or reactive and IgG anti-*T. gondii* negative or non-reactive.

2.2 In addition to the presence of the C line, if only the G line is developed, the test indicates the presence of IgG anti-*T. gondii*. The result is IgG anti-*T. gondii* positive or reactive and IgM anti-*T. gondii* negative or non-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 IgM anti-*T. gondii* and IgG anti-*T. gondii*. The result is IgM anti-*T. gondii* and IgG anti-*T. gondii* positive or reactive.

*Samples with positive results should be confirmed with alternative testing method(s) and clinical findings before a diagnosis is made.*

**3. INVALID:** If no C line develops, the assay is invalid regardless of any color development on the test lines (M and G) as indicated below. Repeat the assay with a new device.

## PERFORMANCE CHARACTERISTICS

**1. Analytical Sensitivity of IgG Detection**

Twenty groups of matrix were spiked with the WHO International Standard Anti-Toxoplasma Serum Ig (TOXM) concentrations of 0.625, 1.25, 2.5, 5, and 10 IU/mL. The specimens were run on the *OnSite* Toxo IgG/IgM Combo Rapid Test. Defined as the 95% detection level, the limit of detection or sensitivity for the *OnSite* Toxo IgG/IgM Combo Rapid Test IgG test line is 2.5 IU/mL.

| IgG (IU/mL) | 0.625 | 1.25 | 2.5 | 5 | 10 |
|---|---|---|---|---|---|
| Number Positive | 0 | 11 | 19 | 20 | 20 |
| Number Negative | 20 | 9 | 1 | 0 | 0 |

**2. Accuracy of IgG Detection**

A total of 237 clinical specimens were collected and tested on the *OnSite* Toxo IgG/IgM Combo Rapid Test and by commercial ELISA. Comparison for all subjects showed 94.9% overall agreement for the IgG test line.

**3. Accuracy of IgM Detection**

A total of 231 clinical specimens were collected and tested on the *OnSite* Toxo IgG/IgM Combo Rapid Test and by commercial ELISA. Comparison for all subjects showed 97.8% overall agreement for the IgM test line.

**4. Performance on BBI Anti-*T. gondii* Mixed Titer Performance Panel**

The performance of the *OnSite* Toxo IgG/IgM Combo Rapid Test was evaluated using the BBI (Boston Biomedical Inc) Anti-*T. gondii* Mixed Titer Performance Panel (PTT202) and compared with three commercial immunoassays. The comparison is shown in the following table:

| BBI Reference Panel (PTT202) | Abbott ARCHITECT | bioMerieux VIDAS | Diasorin LIAISON | *OnSite* Toxo IgG/IgM Combo Rapid Test |
|---|---|---|---|---|
| IgG Positive | 20 | 20 | 20 | 20 |
| IgG Negative | 1 | 1 | 1 | 1 |
| IgM Positive | 8 | 7 | 6 | 6 |
| IgM Negative | 13 | 14 | 15 | 15 |

**5. Cross-Reactivity**

No false positive anti-*T. gondii* IgG and IgM results were observed on 3-14 specimens from the following disease states or special conditions, respectively:

| | | | | |
|---|---|---|---|---|
| CMV | Dengue | HAV | HBV | HCV |
| HIV | HSV-1 | HSV-2 | hCG | *H. pylori* |
| Malaria | TB | *T. pallidum* | Rubella | ANA |
| HAMA | RF (up to 8400 IU/mL) | | | |

**6. Interference**

Common substances (such as pain and fever medication and blood components) may affect the performance of the *OnSite* Toxo IgG/IgM Combo Rapid Test. This was studied by spiking these substances into IgM positive, medium-level IgG positive, weak-level IgG positive, and IgM and IgG negative specimens, respectively. The results demonstrate that at the concentrations tested, the substances studied do not affect the performance of the *OnSite* Toxo IgG/IgM Combo Rapid Test.

List of potentially interfering substances and concentrations tested:

| | | | |
|---|---|---|---|
| 1. Albumin | 60 g/L | 6. Hemoglobin | 2 g/L |
| 2. Bilirubin | 20 mg/dL | 7. Heparin | 3,000 U/L |
| 3. Creatinine | 442 µmol/L | 8. Salicylic acid | 4.24 mmol/L |
| 4. EDTA | 3.4 µmol/L | 9. Sodium citrate | 3.8% |
| 5. Glucose | 55 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 *T. gondii* in serum, plasma or whole blood from individual subjects. Failure to follow the procedure may lead to inaccurate test results.
2. The *OnSite* Toxo IgG/IgM Combo Rapid Test is limited to the qualitative detection of antibodies to *T. gondii* in serum, plasma or whole blood. The intensities of the test lines do not have linear correlation with the antibody titers in the specimen.
3. A negative or non-reactive result for an individual subject indicates absence of detectable *T. gondii* antibodies. However, a negative test result does not preclude the possibility of exposure to or infection with *T. gondii*.
4. A negative or non-reactive result can occur if the quantity of the anti-*T. gondii* IgG or IgM 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 the disease in which a sample is collected.
5. The *OnSite* Toxo IgG/IgM Combo Rapid Test has not been validated on specimens from neonates.
6. Infection may progress rapidly. If the symptom persists, while the result from *OnSite* Toxo IgG/IgM Combo Rapid Test is negative or non-reactive, it is recommended to test with an alternative test method.
7. Some specimens containing unusually high titers of heterophile antibodies or rheumatoid factor may affect expected results.
8. 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 | | Do not reuse | | |
| | Manufacturer | | Date of manufacture | | |


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