Zika IgG/IgM Rapid Test Cassette
(Whole Blood/Serum/Plasma)
Package Insert

A rapid test for the qualitative detection of IgG and IgM antibodies to Zika Virus in human whole blood, serum or plasma specimens. For professional in vitro diagnostic use only.

INTENDED USE

The Zika IgG/IgM Rapid Test Cassette is a lateral flow chromatographic immunoassay for the qualitative detection of IgG and IgM antibodies to Zika Virus in human whole blood, serum or plasma specimen.

SUMMARY

Zika fever, also known as Zika virus disease or simply Zika, is an infectious disease caused by the Zika virus.[1] Most cases have no symptoms, but when present they are usually mild and can resemble dengue fever.[1,4] Symptoms may include fever, red eyes, joint pain, headache, and a maculopapular rash.[1,2,3] Symptoms generally last less than seven days.[2] It has not caused any reported deaths during the initial infection.[4] Mother-to-child transmission during pregnancy can cause microcephaly and other brain malformations in some babies.[5,6] Infections in adults have been linked to Guillain–Barré syndrome (GBS).[4]

Serology for the detection of specific IgM and IgG antibodies to Zika virus can be used. IgM antibodies can be detectable within 3 days of the onset of illness.[7] Serological cross-reactions with closely related flaviviruses such as dengue and West Nile virus as well as vaccines to flaviviruses are possible.[4,8,9]

PRINCIPLE

The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) is a qualitative membrane-based immunoassay for the detection of IgG and IgM antibodies to Zika Virus in whole blood, serum or plasma specimen. This test consists of two components, an IgG component and an IgM component. In the IgG component, anti-human IgG is coated in IgG test line region. During testing, the specimen reacts with Zika NS1 antigen-coated particles in the test cassette. The mixture then migrates upward on the membrane chromatographically by capillary action and reacts with the anti-human IgG in IgG test line region, if the specimen contains IgG antibodies to Zika virus. A colored line will appear in IgG test line region as a result of this. Similarly, anti-human IgM is coated in IgM test line region and if specimen contains IgM antibodies to Zika virus, the conjugate-specimen complex reacts with anti-human IgM. A colored line appears in IgM test line region as a result.

Therefore, if the specimen contains Zika IgG antibodies, a colored line will appear in IgG test line region. If the specimen contains Zika IgM antibodies, a colored line will appear in IgM test line region. If the specimen does not contain anti-Zika antibodies, no colored line will appear in either of the test line regions, indicating a negative result. To serve as a procedural control, a colored line will always appear in the control line region, indicating that the proper volume of specimen has been added and membrane wicking has occurred.

REAGENTS

The test contains anti-human IgM and anti-human IgG as the capture reagent, Zika NS1 as the detection reagent. A goat anti-human IgG is employed in the control line system.

PRECAUTIONS

1. For professional *in vitro* diagnostic use only. Do not use after expiration date.
2. Do not eat, drink or smoke in the area where the specimens or kits are handled.
3. Do not use test if pouch is damaged.
4. Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout all procedures and follow the standard procedures for proper disposal of specimens.
5. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
6. The used test should be discarded according to local regulations.
7. Humidity and temperature can adversely affect results.

STORAGE AND STABILITY

Store as packaged in the sealed pouch at room temperature or refrigerated (2-30°C). The test is stable through the expiration date printed on the sealed pouch. The test must remain in the sealed pouch until use. **DO NOT FREEZE.** Do not use beyond the expiration date.

SPECIMEN COLLECTION AND PREPARATION

- The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) can be performed using whole blood (from venipuncture or fingerstick), serum or plasma.
- To collect **Fingerstick Whole Blood Specimens**:
  - Wash the patient's hand with soap and warm water or clean with an alcohol swab. Allow to dry.
  - Massage the hand without touching the puncture site by rubbing down the hand towards the fingertip of the middle or ring finger.
  - Puncture the skin with a sterile lancet. Wipe away the first sign of blood.
  - Gently rub the hand from wrist to palm to finger to form a rounded drop of blood over the puncture site.
  - Add the Fingerstick Whole Blood specimen to the test by using **a capillary tube**:
    - Touch the end of the capillary tube to the blood until filled to approximately 20µL. Avoid air bubbles.
    - Place the bulb onto the top end of the capillary tube, then squeeze the bulb to dispense the whole blood to the specimen area of the test cassette.
- Separate serum or plasma from blood as soon as possible to avoid hemolysis. Use only clear non-hemolyzed specimens.
- Testing should be performed immediately after the specimens have been collected. Do not leave the specimens at room temperature for prolonged periods. Serum and plasma specimens may be stored at 2-8°C for up to 3 days, for long term storage, specimens should be kept below -20°C. Whole blood collected by venipuncture should be stored at 2-8°C if the test is to be run within 2 days of collection. Do not freeze whole blood specimens. Whole blood collected by fingerstick should be tested immediately.
- Bring specimens to room temperature prior to testing. Frozen specimens must be completely thawed and mixed well prior to testing. Specimens should not be frozen and thawed repeatedly.
- If specimens are to be shipped, they should be packed in compliance with local regulations covering the transportation of etiological agents.
- EDTA K2, Heparin sodium, Citrate sodium and Potassium Oxalate can be used as the anticoagulant for collecting the specimen.

MATERIALS

Materials provided

- Test cassettes
- Droppers
- Package insert
- Buffer

Materials required but not provided

- Specimen collection containers
- Lancets (for fingerstick whole blood only)
- Capillary tubes
- Centrifuge (for plasma only)
- Timer

DIRECTIONS FOR USE

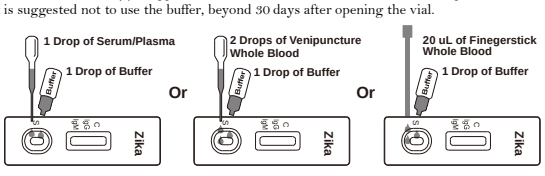
Allow the test, specimen, buffer and/or controls to reach room temperature (15-30°C) prior to testing.

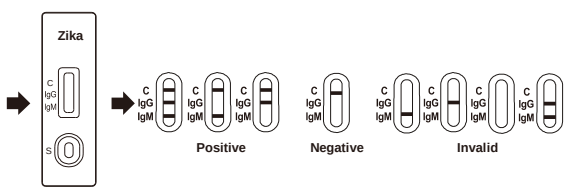
1. Remove the test cassette from the foil pouch and use it within one hour. Best results will be obtained if the test is performed immediately after opening the foil pouch.
2. Place the cassette on a clean and level surface.
   For **Serum or Plasma** specimen:
   - Hold the dropper vertically and transfer **1 full drop of serum or plasma** (approximately 10µL) to the specimen well, then **add 1 drop of buffer** (approximately 40 µL), and start the timer. See illustration below.

   For **Venipuncture Whole Blood** specimen:
   - Hold the dropper vertically and transfer **2 drops of whole blood** (approximately 20µL) to the specimen area, then **add 1 drop of buffer** (approximately 40 µL), and start the timer. See illustration below.

   For **Fingerstick Whole Blood** specimen:
   - To use a capillary tube: Fill the capillary tube **and transfer approximately 20µL of fingerstick whole blood specimen** to the specimen area of test cassette, then **add 1 drop of buffer** (approximately 40 µL) and start the timer. See illustration below.
3. Wait for the colored line(s) to appear. **Read results at 15 minutes.** Do not interpret the result after 20 minutes.

Note: It is suggested not to use the buffer, beyond 30 days after opening the vial.





INTERPRETATION OF RESULTS

IgG POSITIVE:* Two colored lines appear. One colored line should always appear in the control line region (C) and another line should be in the IgG line region.

IgM POSITIVE:* Two colored lines appear. One colored line should always appear in the control line region (C) and another line should be in the IgM line region.

IgG and IgM POSITIVE:* Three colored lines appear. One colored line should always appear in the control line region (C) and two test lines should be in the IgG line region and IgM line region.

***NOTE:** The intensity of the color in the test line regions may vary depending on the concentration of anti-Zika antibodies present in the specimen. Therefore, any shade of color in the test line region should be considered positive.

NEGATIVE: **One colored line appears in the control line region (C)**. No line appears in the IgG region and IgM region.

INVALID: **Control line fails to appear**. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

QUALITY CONTROL

Internal procedural controls are included in the test. A colored line appearing in the control region (C) is an internal procedural control. It confirms sufficient specimen volume and correct procedural technique.

LIMITATION

1. The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) is for in vitro diagnostic use only. This test should be used for detection of IgG and IgM antibody to Zika virus in whole blood, serum or plasma specimens. Neither the quantitative value nor the rate of increase in the concentration of IgG or IgM antibodies to Zika can be determined by this qualitative test.
2. The Zika IgG/IgM Rapid Test Cassette (Whole blood/Serum/Plasma) will only indicate the presence of IgG and IgM antibodies to Zika in the specimen and should not be used as the sole criteria for the diagnosis of Zika infections.
3. As with all diagnostic tests, all results must be considered with other clinical information available to the physician.
4. If the test result is negative and clinical symptoms persist, additional follow-up testing using other clinical methods is suggested. A negative result at any time does not preclude the possibility of Zika infection.
5. The hematocrit level of the whole blood can affect the test results. Hematocrit level needs to be between 25% and 65% for accurate results.

EXPECTED VALUES

The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) has been compared with a leading commercial EIA Zika IgG tests and Zika IgM tests. The correlation between these two systems is over 99%.

PERFORMANCE CHARACTERISTICS

Sensitivity and Specificity

The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) was compared with a leading commercial EIA Zika IgG tests and Zika IgM tests; the results show that Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) has a high sensitivity and specificity.

IgG Result

Method		EIA		Total Results
Zika IgG/IgM Rapid Test for IgG	Results	Positive	Negative	
	Positive	22	1	23
	Negative	0	89	89
Total Result		22	90	112

Relative Sensitivity: > 99.9% (95%CI*: 87.3%-100%) *Confidence Interval

Relative Specificity: 98.9% (95%CI*: 97.1%-99.8%)

Accuracy: 99.1% (95%CI*: 95.1%-99.9%)

IgM Result

Method		EIA		Total Results
Zika IgG/IgM Rapid Test for IgM	Results	Positive	Negative	
	Positive	17	0	17
	Negative	1	90	91
Total Result		18	90	108

Relative Sensitivity: 94.4% (95%CI*: 72.7%-99.9%) *Confidence Interval

Relative Specificity: > 99.9% (95%CI*: 96.7%-100%)

Accuracy: 99.1% (95%CI*: 94.9%-100%)

Precision

Intra-Assay

Within-run precision has been determined by using 10 replicates of three specimens: negative, low positive, and high positive. The negative, low positive, and high positive values were correctly identified >99% of the time.

Inter-Assay

Between-run precision has been determined by 10 independent assays on the same specimens: negative, low positive, and high positive. Three different lots of the Zika IgG/IgM Rapid Test cassette (Whole Blood/Serum/Plasma) have been tested over a 3-days period using negative, low positive, and high positive specimens. The specimens were correctly identified >99% of the time.

Cross-reactivity

The Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) has been tested for anti-HAV IgM, HBsAg, anti-HCV IgG, anti-HIV IgG, anti-RF IgG, anti-Syphilis IgG, anti-H. Pylori IgG, anti-Rubella IgG, anti-Rubella IgM, anti-Toxo IgG, anti-Toxo IgM, anti-HSV 1 IgG, anti-HSV 1 IgM, anti-HSV 2 IgG, anti-HSV 2 IgM, anti-Dengue IgG+IgM and anti-Chikungunya IgG+IgM positive specimens. The results showed no cross-reactivity.

Interfering Substances

The following compounds have been tested using the Zika IgG/IgM Rapid Test Cassette (Whole Blood/Serum/Plasma) and no interference was observed.

Acetaminophen: 20 mg/dL	Caffeine: 20 mg/dL
Acetylsalicylic Acid: 20 mg/dL	Gentisic Acid: 20 mg/dL
Ascorbic Acid: 2g/dL	Albumin: 2 g/dL
Creatin: 200 mg/dL	Hemoglobin 1000mg/dL
Bilirubin: 1g/dL	Oxalic Acid: 60mg/dL

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Index of Symbols

	Attention, see instructions for use	Σ	Tests per kit	EC REP	Authorized Representative
IVD	For in vitro diagnostic use only		Use by		Do not reuse
2°C 30°C	Store between 2-30°C	LOT	Lot Number	REF	Catalog #
	Do not use if package is damaged				

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