EnCore Biomedicals

# EnSure Rapid Test For Filariasis IgG/IgM

Rapid Diagnostics Test for Filariasis IgG/IgM  Store at 2°c to 30°c

## INTENDED USE

The Filariasis IgG/IgM Rapid Test Kit is a lateral flow immunoassay for the simultaneous detection and differentiation of IgG and IgM anti-lymphatic filarial parasites (W. Bancrofti and B. Malayi) in human serum, plasma or whole blood. This test is intended to be used as a screening test and as an aid in the diagnosis of infection with lymphatic filarial parasites. Any reactive specimen with the Filariasis IgG/IgM Rapid Test must be confirmed with alternative testing method(s).

## SUMMARY

The lymphatic filariasis known as Elephantiasis, mainly caused by W. bancrofti and B. malayi, affects about 120 million people over 80 countries. The disease is transmitted to humans by the bites of infected mosquitoes within which the microflariae sucked from an infected human subject develops into third-stage larvae. Generally, repeated and prolonged exposure to infected larvae is required for establishment of human infection.

The definitive parasitologic diagnosis is the demonstration of microflariae in blood samples. However, this gold standard test is restricted by the requirement for nocturnal blood collection and lack of adequate sensitivity. Detection of circulating antigens is commercially available. Its usefulness is limited for W. bancrofti. In addition, microfilaremia and antigenemia develop from months to years after exposure. Antibody detection provides an early means to detect filarial parasite infection. Presence of IgM to the parasite antigens suggest current infection, whereas, IgG corresponds to late stage of infection or past infection. Furthermore, identification of conserved antigens allows 'pan-filaria' test to be applicable. Utilization of recombinant proteins eliminates cross-reaction with individuals having other parasitic diseases. The Filariasis IgG/IgM Rapid Test uses conserved recombinant antigens to simultaneously detect IgG and IgM to the W. bancrofti and B. malayi parasites without the restriction on specimen collection.

## PRINCIPLE OF THE ASSAY

The Filariasis IgG/IgM Rapid Test Kit is a lateral flow chromatographic immunoassay. The test cassette consists of: a burgundy colored conjugate pad containing recombinant W. bancrofti and B. malayi common antigens conjugated with colloid gold (Filariasis conjugates) and rabbit IgG-gold conjugates, 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 IgM anti- W. bancrofti and B. malayi, G line is pre-coated with reagents for the detection of IgG anti-W. bancrofti and B. malayi, and the C line is pre-coated with goat anti rabbit IgG.

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. W. bancrofti or B. malayi IgM antibodies if present in the specimen will bind to the Filariasis conjugates. The immunocomplex is then captured on the membrane by the pre- coated anti-human IgM antibody, forming a burgundy colored M line, indicating a W. bancrofti or B. malayi IgM positive test result. W. bancrofti or B. malayi IgG antibodies if present in the specimen will bind to the Filariasis conjugates. The immunocomplex is then captured by the pre-coated reagents on the membrane, forming a burgundy colored G line, indicating a W. bancrofti or B. malayi 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 burgundy colored line of the immunocomplex of goat anti rabbit IgG/rabbit IgG-gold conjugate regardless of the color development on any of the test lines. Otherwise, the test result is invalid and the specimen must be retested with another device.

## PACKAGE CONTAINS

1. Each Pouch contents: Test Cassette, Desiccant.
2. Sample dropper
3. Assay Buffer bottle
4. 1 Instructions for Use

## MATERIALS REQUIRED BUT NOT PROVIDED

1. Timer
2. Calibrated Mioropipette
3. Disposable Gloves

## 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 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 any other test kit as a substitute for components in this kit.
6. Do not use hemolyzed blood 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 results should be read 15-20 minutes after a specimen is applied to the sample well of the device. Any results interpreted outside 15-20 minutes 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.

## SPECIMEN COLLECTION

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

**Plasma**

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

**Serum**

1. Collect blood specimen into a red top collection tube (containing no anticoagulants in Vacutainer) by veinpuncture.
2. Allow the blood to clot.
3. Separate the serum by centrifugation.
4. Carefully withdraw the serum into a new pre-labeled tube.
5. Test specimens as soon as possible after collecting. Store specimens at 2°c to 8°c if not tested immediately.
6. Store specimens at 2°c to 8°c up to 5 days. The specimens should be frozen at -20°c for longer storage.

**Blood**

Drops of whole blood can be obtained by either finger tip puncture or veinpuncture. Do not use any hemolized blood for testing. Whole blood specimens should be stored in refrigeration (2°c-8°c) if not tested immediately. The specimens must be tested within 24 hours of collection. 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.

## TEST PROCEDURE

1. Bring the kit components to room temperature before testing.
2. Open the pouch and remove the test, dropper and silica gel pouch. Check the color of the silica gel. It should be blue, if it has turned colorless or pink, discard the test and use another test. Once opened, the test must be used immediately.

**For whole blood test**

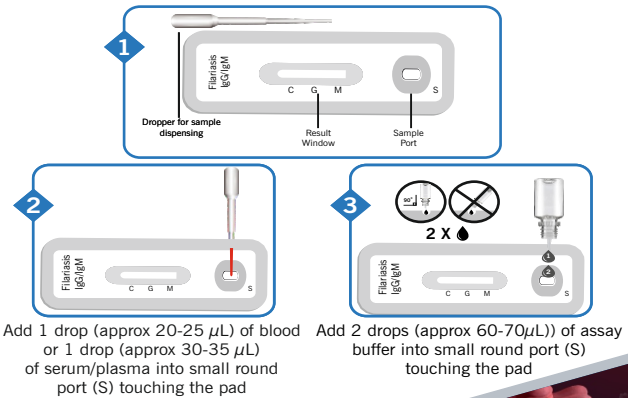
- Add 1 drop of whole blood (approx 20-25 $\mu$L) into the sample well.
- Then add 2 drops (approx 60-70 $\mu$L) of Assay buffer immediately.

**For serum or plasma**

- Add 1 drop (approx 30 $\mu$L-35 $\mu$L) of specimen into the sample well making sure that there are no air bubbles.
- Then add 2 drops (approx 60-70 $\mu$L) of Assay Buffer immediately.

5. Set up timer.
6. Results can be read in 20 minutes.

- Don't read results after 30 minutes. To avoid confusion, discard the test device after interpreting the result.




Add 1 drop (approx 20-25 $\mu$L) of blood or 1 drop (approx 30-35 $\mu$L) of serum/plasma into small round port (S) touching the pad

Add 2 drops (approx 60-70$\mu$L)) of assay buffer into small round port (S) touching the pad

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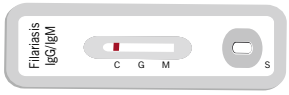
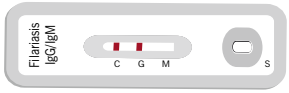
## INTERPRETATION OF ASSAY RESULT

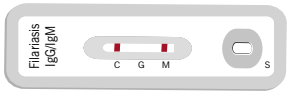
**NEGATIVE :** Only the C line shows color development, the two test line (M and G) show no color development.



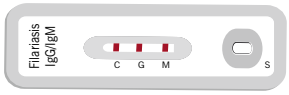

**IgG Positive:** In addition to the presence of C line, if only G line is developed, the test indicates for the presence of anti-W. bancrofti or B. malayi IgG antibody. The result is positive.




**IgM Positive:** IgM Positive: In addition to the presence of C line, if only M line is developed, the test indicates for the presence of anti-W. bancrofti or B. malayi IgM antibody. The result is positive.



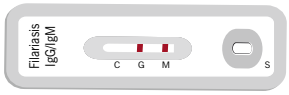

**IgG/IgM Positive:** In addition to the presence of C line, both M and G line are developed, the test indicates for the presence of both IgG and IgM anti-W. bancrofti or B. malayi. The result is also positive.



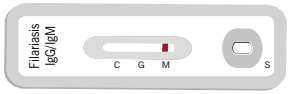

**INVALID: The test should be considered invalid if,**

**A) No line appears at 'C', 'IgG' and 'IgM' region**


**B) No line appears at 'C' region and line appear at 'IgG' and 'IgM' region.**




**C) No line appears at 'C' and 'IgG' region and line appear at 'IgM' region**




**D) No line appears at 'C' and 'IgM' region and line appear at 'IgG' region.**


## PERFORMANCE CHARACTERISTICS

Internal Evaluation:

In an in-house study, total 249 samples were evaluated for sensitivity and specificity. We found the relative sensitivity was 99 % (i. e. 99/100) and the relative specificity was 99.33% (i. e. 150/151). The results are summarized in the following table:

| Sample | Total Number of samples tested | Rapid Filariasis IgG/IgM Test | | Sensitivity (%) | Specificity (%) |
|---|---|---|---|---|---|
| | | Positive | Negative | | |
| Filariasis IgG Positive Serum Samples | 12 | 12 | - | 99% | |
| Filariasis IgG Positive Plasma Samples | 10 | 10 | - | | |
| Filariasis IgG Positive Whole Blood Samples | 10 | 9 | 1 | | |
| Filariasis IgM Positive Serum Samples | 15 | 15 | - | | |
| Filariasis IgM Positive Plasma Samples | 10 | 10 | - | | |
| Filariasis IgM Positive Whole Blood Samples | 10 | 10 | - | | |
| Filariasis IgG/IgM Positive Serum Samples | 12 | 12 | - | | |
| Filariasis IgG/IgM Positive Plasma Samples | 10 | 10 | - | | |
| Filariasis IgG/IgM Positive Whole Blood Samples | 10 | 10 | - | | |
| Negative Human Serum | 50 | 1 | 49 | | 99.33% |
| Negative Human Plasma | 50 | - | 50 | | |
| Negative Human Whole Blood | 50 | - | 50 | | |

## LIMITATIONS

1. The Assay Procedure and the Test Result Interpretation must be followed closely when testing the presence of antibodies to filarial parasites in serum, plasma or whole blood from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The Filariasis IgG/IgM Rapid Test Kit is limited to the qualitative detection of antibodies to W. bancrofti and B. malayi in human serum, plasma or whole blood. The intensity of the test line does not have linear correlation with the antibody titer in the specimen.
3. A negative result for an individual subject indicates absence of detectable W. bancrofti and B.malayi antibodies. However, a negative test result does not preclude the possibility of exposure to W. bancrofti and B. malayi.
4. A negative result can occur if the quantity of W. bancrofti and B. malayi antibodies 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.
5. Some specimens containing unusually high titer of heterophile antibodies or rheumatoid factor may affect expected results.
6. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

## DISCLAIMER

All precaution shall be taken to ensure the diagnostic ability and accuracy of this product. This product is utilized outside the control of manufacturer and distributors. The various factors including storage temperature, environmental conditions and procedure error may affect the results. This test provides presumptive diagnosis of Filariasis IgG and IgM infection. A confirmed Filariasis infection diagnosis should only be made by a physician after all clinical and laboratory Findings have been evaluated

## REFERENCES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5. Lammie PJ, Weil G, et al: Recombinant antigen-based antibody assays for the diagnosis surveillance of lymphatic filariasis-a multiplecenter trial. Flaria Jornal 2004: 3: 9-18.
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**SYMBOL KEY**

| | | | | | |
|---|---|---|---|---|---|
| IVD | In Vitro Diagnostic Use | 2°C – 30°C | Temperature limitation | | Expiry Date |
| | Manufacturer | | Single Use | LOT | Lot Number |
| | Manufacturing Date | | Number of tests in the pack | | Do not use if pouch or kit damaged |
| | Read package insert before use | | | | |


Manufacturer By

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