（**FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS !**）

Leishmania IgG/IgM Lateral Flow Assay Kit

Catalog No: E-HD-C069

20T/40T

This manual must be read attentively and completely before using this product.

If you have any problems, please contact our Technical Service Center for help.

Toll-free: 1-888-852-8623 Tel: 1-832-243-6086 Fax: 1-832-243-6017

Email: techsupport@elabscience.com

Website: www.vetassay-elab.com

Please kindly provide us the lot number (on the outside of the box) of the kit for more efficient service.

Test principle

The Leishmania IgG/IgM Test is a lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing recombinant rK39 antigen conjugated with colloidal gold (Leishmania conjugates) and rabbit IgG-gold conjugates, 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-L. donovani IgM, the G line is pre-coated withreagents for the detection of anti-L. donovani IgG, 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. L. donovani IgM, if present in the specimen, will bind to the Leishmania 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 L. donovani IgM positive test result.

L. donovani IgG if present in the specimen will bind to the Leishmania conjugates. The immunocomplex is then captured by the pre-coated reagents on the membrane forming a burgundy colored G line, indicating a L. donovani 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 conjugates regardless of color development on any of the test lines. If the C line does not develop, the test result is invalid and the specimen must be retested with another device.

Kit components

Item	Specification
Detection card	20/40 T
Sample Diluent	1 vial
Manual	1 copy

Note: All reagent bottle caps must be tightened to prevent evaporation and microbial pollution.

Notes

1. Please read the manual carefully before use, changes of operation may result in unreliable results.
2. Do not use product out of date or in a broken aluminum foil, it is disposable and cannot be used repeatedly.
3. The detection card should be brought to room temperature before opening after take it out from the refrigerator. The opening detection card should be used as soon as possible.
4. Please do not use but not limited to the following liquids for negative control: Water, PBS.
5. The tested sample should be fresh and clear. Avoid of using samples of turbidity, polluted, high hemolysis or abnormal viscous.
6. Avoid of touching the chromatography membrane of the sample well and test well.
7. The waste of experiment should be considered as contaminant, and must be properly handled according to the local regulations.
8. The test environment should be kept at a fixed temperature to avoid testing at high temperatures.

9. Each reagent is optimized for use in the **E-HD-C069**. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other **E-HD-C069** with different lot numbers.

Storage and expiry date

Storage: Store at 4-30℃. With cool and dry environment, avoid freeze.

Expiry date: expiration date is on the packing box.

Sample preparation

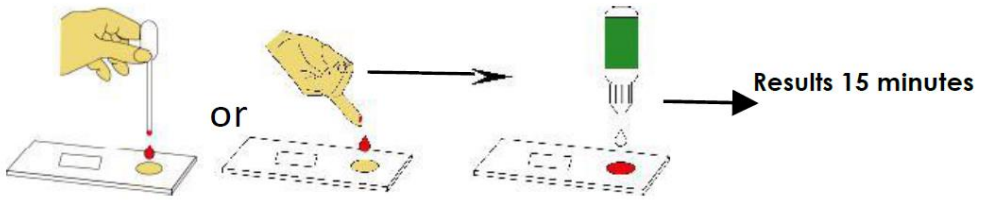
1. Leishmania IgG/IgM Test Kit can be performed with serum, plasma and whole blood.
2. Serum and plasma samples could be collected by conventional methods. After blood collection, serum and plasma should be immediately separated for analysis. EDTA or heparin sodium anticoagulant plasma could be used.
3. The whole blood samples could be collected by conventional methods and the test should be performed immediately after sample collection.
4. It is recommended to use serum or plasma as the priority sample types for testing, and whole blood samples can be used in urgent or special cases.
5. For serum or plasma, please determine within 4 hours after separation; If the sample cannot be tested within 4 hours, please store it at 2℃ - 8℃ (valid for up to 5 days) or at -20℃ (valid for at least 3 months).
6. For whole blood, please determine within 4 hours after collection; If the sample cannot be tested within 4 hours, please store it at 2℃ -8℃ (valid for 3 days). Do not freeze whole blood samples.
7. Bring samples to room temperature prior to testing. Frozen serum or plasma samples must be completely thawed and mixed well prior to testing. Do not freeze and thaw samples repeatedly.

Assay procedure

Allow all kit components and sample to reach room temperature (25℃) prior to testing.

1. Before the test, read the operation manual completely and equilibrate the test reagent to room temperature (15℃ -30℃). The test should be conducted at room temperature.
2. Take out the test strip from the aluminum foil packaging bag, and use it within 1 hour after unsealing to prevent moisture of test cassette.
3. For whole blood test: Apply 1 drop of whole blood (about 40-50 μL) into the sample well. Then add 1 drop (about 40-50 μL) of Sample Diluent immediately.
   For serum or plasma test: Dispense 45 μL of the specimen into the sample well. Then add 1 drops (about 35-50 μL) of Sample Diluent immediately.
4. Set up timer.
5. Results can be read in 15 minutes. Positive results can be visible in as short as 1 minute.

NOTE: Don't read result after 15 minutes. To avoid confusion, discard the test device after interpreting the result.



For serum or plasma test

Dispense 45 μL of the specimen into the sample well.
Then add 1 drops (about 35-50 μL) of Sample Diluent immediately.

Judgment of result

1. IgM POSITIVE: Two lines appear.

Colored lines should be in the control line region (C) and IgM test line region. No line appears in IgG test line region.

2. IgG and IgM POSITIVE: Three lines appear.

Colored lines should be in the control line region(C), IgG line test region and IgM test line region. The color intensities of the lines do not have to match.

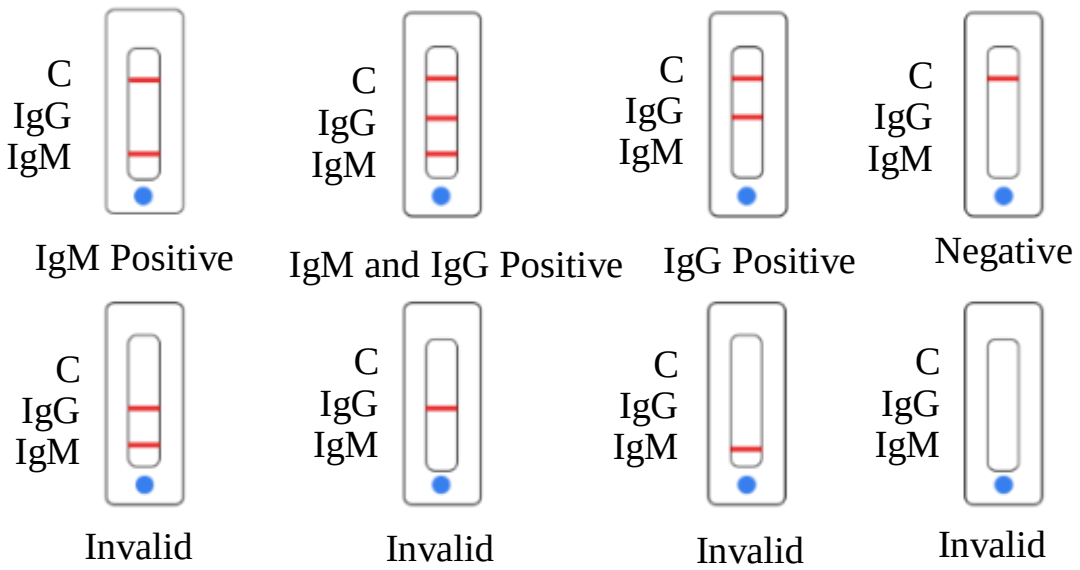
3. IgG POSITIVE: Two lines appear.

Colored lines should be in the control line region(C) and IgG test line region. No line appears in IgM test line region.

NOTE: The intensity of the color in the IgG and/or IgM test line region(s) will vary depending on the concentration of COVID-19 antibodies in the sample. Therefore, any shade of color in the IgG and/or IgM test line region(s) should be considered positive.

4. NEGATIVE: One colored line should be in the control line region (C). No line appears in IgG and IgM test line region(s).

5. INVALID: Control line fails to appear.



Limitations of this test method

1. The Assay Procedure and the Interpretation of Assay Result sections must be followed closely when testing for 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 Leishmania IgG/IgM Test is limited to the qualitative detection of antibodies to L. donovani in human serum, plasma or whole blood. The intensity of the test line does not have a linear correlation with the antibody titer in the specimen.
3. A non-reactive result for an individual subject indicates absence of detectable anti-L. donovani antibodies. However, a non-reactive test result does not preclude the possibility of exposure tothe visceral leishmaniasis causative species of L. donovani.
4. A non-reactive result can occur if the quantity of the L. donovani 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 titers 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.