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

Leptospira IgG/IgM Lateral Flow Assay Kit

Catalog No: E-HD-C068

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 Leptospira IgG/IgM Test Kit is a lateral flow chromatographic immunoassay. The test strip in the cassette consists of: 1) a burgundy colored conjugate pad containing recombinant L. interrogans antigens conjugated with colloidal gold (Leptospira conjugates) and rabbit IgG-gold conjugates, 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 antibodies for the detection of IgG anti-L. interrogans virus, the M line is pre-coated with antibodies for the detection of IgM anti-L. interrogans virus, and the C line is precoated with a control line antibody.

When an adequate volume of test specimen is dispensed into the sample well of the test cassette, the specimen migrates by capillary action along the cassette. IgG anti-L. interrogans virus, if present in the specimen, will bind to the Leptospira Ag conjugates. The immunocomplex is then captured by the pre-coated anti-human IgG, forming a burgundy colored G line, indicating a L. interrogans IgG positive test result.

Kit components

Item	Specification
Detection card	20/40 T
Sample Diluent	1 vial
Dropper	20/40
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 Do not use product out of date or in a broken aluminum foil, it is disposable and cannot be used repeatedly.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. Please do not use but not limited to the following liquids for negative control: Water, PBS.
4. The tested sample should be fresh and clear. Avoid of using samples of turbidity, polluted, high hemolysis or abnormal viscous.
5. Avoid of touching the chromatography membrane of the sample well and test well.
6. The waste of experiment should be considered as contaminant, and must be properly handled according to the local regulations.
7. The test environment should be kept at a fixed temperature to avoid testing at high temperatures.
8. Each reagent is optimized for use in the **E-HD-C068**. Do not substitute reagents from any other manufacturer into the test kit. Do not combine reagents from other **E-HD-C068** 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

Consider any materials of human origin as infectious and handle them with standard biosafety procedures.

For plasma samples

Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively) by venipuncture.

Separate the plasma by centrifugation.

Carefully withdraw the plasma into a new pre-labeled tube.

For serum samples

Collect blood specimen into a red top collection tube (containing no anticoagulants) by venipuncture.

Allow the blood to clot.

Separate the serum by centrifugation.

Carefully withdraw the serum into a new pre-labeled tube.

Test specimens as soon as possible after collecting. If not tested immediately, store specimens at 2-8 ℃ for up to 5 days. For longer storage, specimens should be kept frozen at -20 ℃. 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 samples demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid interference on result interpretation.

For whole blood samples

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

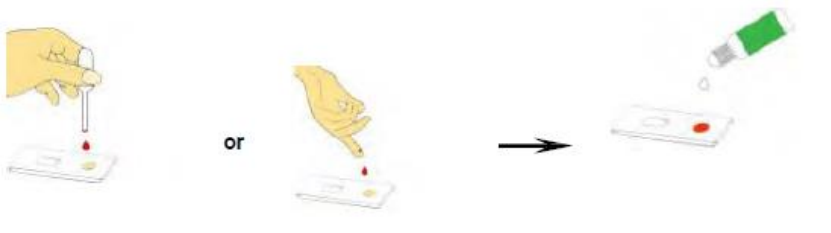
Whole blood specimens should be stored at 2-8 ℃ if not tested immediately. The specimens must be tested within 24 hours of collection.

Assay procedure

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

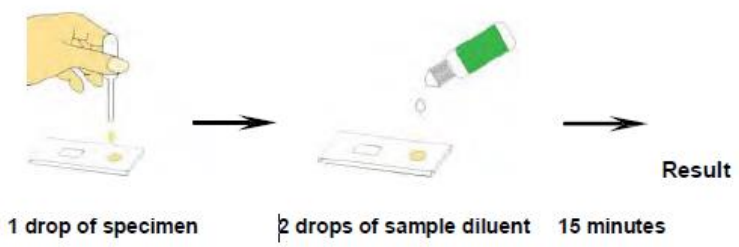
1. Bring the specimen and test components to room temperature if refrigerated or frozen. Once thawed, mix the specimen well prior to performing the assay.
2. When ready to test, open the pouch at the notch and remove the device. Place the test device on a clean, flat surface.
3. Be sure to label the device with specimen's ID number.
4. **For whole blood test**

Apply 1 drop of whole blood (about 40-50 μL) into the sample well. Then add 2 drops (about 70-100 μL) of Sample Diluent immediately.



For serum and plasma test

Fill the dropper with the specimen. Hold the dropper vertically and dispense the 1drop (about 30-45 μL) of specimen into the center of the sample well, making sure that there are no air bubbles. Immediately add 2 drops (about 70-100 μL) of Sample Diluent.



5. Set up the timer.
6. Read the result in 15 minutes. Positive results may be visible in as short as 1 minute. **Don't read results after 15 minute. To avoid confusion, discard the test device after interpreting the result.**

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.

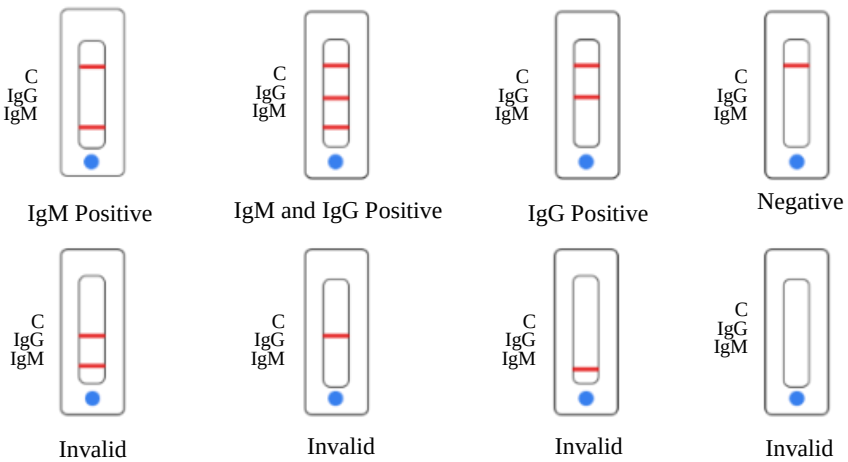
Colored lines should be in the control line region(C) and IgG test line region. No line appears in IgM test line region.

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.

Insufficient buffer volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the procedure with a new cassette. If the problem persists, discontinue using the test kit and contact your local distributor.

NOTE: Don't read result after 15 minutes. To avoid confusion, discard the test device after interpreting the result. Samples with positive results should be confirmed with alternative testing method(s) and clinical findings before a positive determination is made.



NOTE: This figure is only used as a reference for judging results.

Limitations of this test method

1. The Test Procedure and the Interpretation of Results must be followed closely when testing the presence of antibodies to dengue virus in serum, plasma and whole blood from individual subjects. Failure to follow the procedure may lead to inaccurate results.
2. The Leptospira IgG/IgM Test Kit is limited to the qualitative detection of antibodies to L. interrogans in human serum, plasma and whole blood. The intensity of the test band does not have linear correlation with the antibody titer in the specimen.
3. A negative for an individual subject indicates absence of detectable L. interrogans antibodies. However, a negative result does not preclude the possibility of exposure to or infection with L. interrogans.
4. A negative result can occur if the quantity of L. interrogans antibodies present in the specimen is below the limits of detection, or if 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 be interpreted in conjunction with other diagnostic procedures and clinical findings.