CE IVD

FRENOVO

Leishmania IgG/IgM Rapid Test Kit

Instructions For Use

PRODUCT NAME

Leishmania IgG/IgM Rapid Test Kit

PACKAGE SPECIFICATION

20 tests/kit

INTENDED USE

Leishmania IgG/IgM Rapid Test is an in vitro qualitative immunochromatographic assay for the rapid detection of IgG and IgM antibodies to Leishmania in human whole blood, serum and plasma. The test results are intended to aid in thediagnosis of Leishmania infection.

SUMMARY AND PRINCIPLES OF THE PROCEDURE

Visceral leishmaniasis, or Kala-azar, is a disseminated infection caused by several subspecies of the L. donovani. The disease is estimated by the World Health Organization (WHO) to affect approximately 12 million people in 88 countries. It is transmitted to humans by bites of the Phlebotomus sandflies, which acquire infection from feeding on infected animals. Though it is a disease found in poor countries, in Southern Europe, it has become the leading opportunistic infection in AIDS patients. Identification of L. donovani organism from the blood, bone marrow, liver, lymph nodes or the spleen provides a definite mean of diagnosis. Serological detection of anti-L. donovani IgM is found to be an excellent marker for the acute Visceral leishmaniasis. Tests used in clinic are included ELISA, fluorescent antibody or direct agglutination tests. Recently, utilization of L. donovani specific protein in the test has improved the sensitivity and specificity dramatically. The Leishmania IgG/IgM Rapid Test is a recombinant protein based serological test, which detects IgG and IgM antibodies to the L. Donovani simultaneously. The test provides a reliable result within 15 minutes without any instruments.

The Leishmania IgG/IgM Rapid Test is a qualitative, membrane based immunoassay for the detection of IgG and IgM antibodies to Leishmania in whole blood, serum or plasma. The membrane is pre-coated with mouse anti-human IgG and mouse anti-human IgM on the test line region of the cassette. During testing, the whole blood, serum or plasma specimen reacts with recombinant Leishmania antigen conjugated colloid gold. The mixture migrates upward on the membrane chromatographically by capillary action to react with mouse anti-human IgG or/and mouse anti-human IgM on the membrane and generate a colored line. Presence of this colored line indicates a positive result, while its absence indicates a negative result. To serve as a procedural control, a colored line will always appear in the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

MATERIALS PROVIDED

Each kit contains:

1. test cassettes: 20 pieces test cassettes individually pouched.
2. Wash Buffer Solution: 3.0 ml in dropper bottle.
3. Droppers: 20 pieces droppers of 10 ul.
4. Package insert: 1 piece attached.

MATERIALS REQUIRED BUT NOT PROVIDED

- Timer or stopwatch.
- Specimen collection containers
- Disposable gloves and/or protective clothing
- Centrifuge(for plasma only)
- Micro-pipette

WARNINGS

1. Read the package insert completely before using the product. The instructions must be followed carefully as not doing so may result in inaccurate results.
2. The kits for diagnostic use only.
3. Perform test at room temperature.

PRECAUTIONS

1. Leishmania IgG/IgM Rapid Test Kit is for professional use only.
2. The package insert instructions must be followed to ensure optimum test performance.
3. The kit is intended for in vitro diagnostic use.
4. As with all screening assays, any results should be considered presumptive until confirmatory assays have been performed according to local practice or WHO guidelines.

Safety Precautions

1. Standard precautions for handling infectious agents should be observed when using this kit.
2. Wear protective clothing such as lab coat, safety glasses and disposable gloves when handling specimens and assay reagents.
3. Wash hands thoroughly after use.
4. In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.

Bio safety Precautions

Appropriate bio safety practices should be used when handling specimens and reagents. These precautions include, but are not limited to the following:

1. Do not smoke,eat,drink,apply cosmetics or handle contact lenses in areas in which specimens are handled.
2. Dispose of all specimens, used devices and tubes as though they are capable of transmitting infection. The preferred methods of disposal are by autoclave at 121°C for a minimum of 60 minutes or by incineration. Disposable materials may be incinerated. Liquid waste may be mixed with appropriate chemical disinfectants. A solution of 10% bleach is recommended. Allow 60 minutes for effective decontamination. NOTE: Do not autoclave solutions containing bleach.
3. When disposing of solution, avoid contact with acid to prevent liberation of a toxic gas.
4. All spills should be wiped thoroughly using a suitable disinfectant such as a sodium hypochlorite solution.
5. Use a separate dropper and device for each specimen tested.

Handling Precautions

1. Do not use if the kit safety seal is absent, damaged or broken.
2. Do not use any device if the pouches have been perforated.
3. Each device is for single use only.
4. Do not mix wash buffer solution/test cassettes from different kit lots.
5. Do not use the kit past the expiration date (this date is printed on the kit box).
6. Adequate lighting is required to read the test results.
7. The result should be read immediately after the end of the 10 minutes incubation time following the addition of specimen and wash buffer solution. Do not read results beyond 15 minutes.

STORAGE INSTRUCTIONS

1. The kit should be stored between 2-30°C and the shelf life is 24 months.
2. The kit components are stable until the expiration date printed on the outer label, when stored as directed. The kit expiry date is determined by whichever of the components has the shortest expiry date. The kit expiry date is not impacted once the wash buffer solution has been opened. Do not use kit components beyond overall kit expiry date.
3. If stored refrigerated, ensure that the pouched device is brought to room temperature before opening.
4. Do not freeze the kit.

SAMPLE COLLECTION AND PREPARATION

1. Applicable samples： Whole Blood/Serum/Plasma.
2. Separate serum or plasma from whole blood as soon as possible to avoid hemolysis. Use only clear nonhemolysis specimens.
3. Testing should be performed immediately after the specimens have been collected as soon as possible. Do not leave the specimens at room temperature for prolonged periods.
4. Serum and plasma specimens may be stored at 2~8 ℃ for up to 7 days, for long term storage, serum/plasma specimens should be kept below -20 ℃. Whole blood collected by venipuncture should be stored at 2~8 ℃ if the test is to be run within 2 days of collection.
5. Do not freeze whole blood specimens.
6. 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.
7. If specimens are to be shipped, they should be packed in compliance with local regulations covering the transportation of etiological agents.
8. EDTA K2, Heparin sodium, Citrate sodium and Potassium Oxalate can be used as the anticoagulant for collecting the specimen.

QUALITY CONTROL

An internal procedural control is included in the test. a colored line appearing in the control line region (C) is an internal valid procedural control, it confirming adequate membrane wicking. Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

TEST PROCEDURE

Allow the test cassette, specimen, buffer solution to equilibrate to room temperature (15-30°C) prior to testing.

1. 1.Remove the test cassette from the sealed pouch and use it within one hour. Place the test cassette on a clean and level surface.
2. For Serum or Plasma Specimens
   To use a dropper: Hold the dropper vertically, draw the specimen and transfer the specimen to the sample well of the test cassette (one drop/approximately 10ul), then squeeze the wash buffer solution bottle, add three drops wash buffer solution inside (approximately 90ul) to the sample well and start the timer. Avoid trapping air bubbles in the sample well.

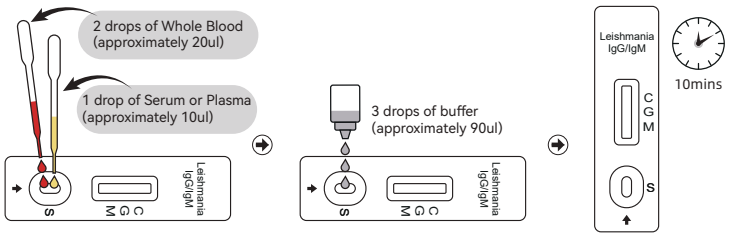
To use a micro-pipette: Pipette and dispense 10ul of specimen to the sample well of the test cassette, then squeeze the wash buffer solution bottle, add three drops wash buffer solution inside (approximately 90ul) to the sample well and start the timer.

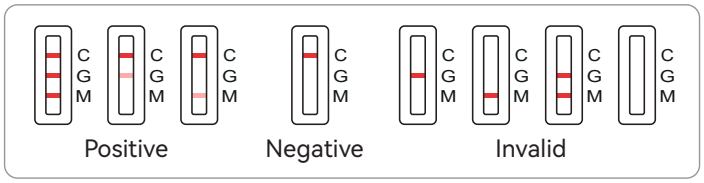
For Whole Blood Specimens

To use a dropper: Hold the dropper vertically, draw the specimen and transfer the specimen to the sample well of the test cassette (two drops/approximately 20ul), then squeeze the wash buffer solution bottle, add three drops wash buffer solution inside (approximately 90ul) to the sample well and start the timer. Avoid trapping air bubbles in the sample well.

To use a micro-pipette: Pipette and dispense 20ul of specimen to the sample well of the test cassette, then squeeze the wash buffer solution bottle, add three drops wash buffer solution inside (approximately 90ul) to the sample well and start the timer.

3. Wait for the colored line(s)to appear. The test result should be read at 10 minutes. Do not interpret the result after 15 minutes.





INTERPRETATION OF RESULTS

IgG and IgM POSITIVE: Three lines appear. One colored line should be in the control line region(C), and two colored lines should appear in IgG test line region and IgM test line region. The color intensities of the lines do not have to match.

IgG POSITIVE: Two lines appear. One colored line should be in the control line region(C), and a colored line appears in IgG test line region.

IgM POSITIVE: Two lines appear. One colored line should be in the control line region (C), and a colored 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 Leishmania antibodies in the specimen. Therefore, any shade of color in the IgG and/or IgM test line region(s)should be considered positive.

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

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 test cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

LIMITATIONS

1. The Assay Procedure and the Test Result Interpretation must be followed closely when testing the presence of antibodies to the L. donovani in serum, plasma or whole blood from individual subjects. Failure to follow the procedure may give inaccurate results.
2. The Leishmania IgG/IgM Rapid Testis limited to the qualitative detection of antibodies to L. donovani in human serum, plasma or whole blood. The intensity of the test band does not have linear correlation with the antibody titer in the specimen.
3. A negative result for an individual subject indicates absence of detectable anti-L. donovani antibodies. However, a negative test result does not preclude the possibility of exposure to Visceral leishmaniasis causative species of the L. donovani .
4. A negative 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. The results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical finding.
6. The hematocrit level of the whole blood can affect the test results. Hematocrit level needs to be between 25% and 65% for accurate results.

PERFORMANCE CHARACTERISTICS

Sensitivity and Specificity

A total of 269 samples from susceptible subjects were tested by the Leishmania IgG/IgM Rapid Testand by a commercial L. donovani IgM ELISA. Comparison for all subjects is shown in the following table.

Leishmania IgM Study		ELISA		
Leishmania IgG/IgM Rapid test	Results	IgM Positive	IgM Negative	Total Results
	IgM Positive	31	4	35
	IgM Negative	3	231	234
Total Results		34	235	269

Leishmania IgM Study Summary Results:
Clinical sensitivity =91.18% (76.32% ~ 98.14%)
Clinical specificity = 98.30% (95.70% ~ 99.53%)
Accuracy=97.40% (94.71% ~ 98.95%)

A total of 314 samples from susceptible subjects were tested by the Leishmania IgG/IgM Rapid Testand by a commercial L. donovani IgG ELISA. Comparison for all subjects is shown in the following table.

Leishmania IgG Study		ELISA		
Leishmania IgG/IgM Rapid test	Results	IgG Positive	IgG Negative	Total Results
	IgG Positive	13	6	19
	IgG Negative	1	294	295
Total Results		14	300	314

Leishmania IgG Study Summary Results:
Clinical sensitivity =92.86% (66.13% ~ 99.82%)
Clinical specificity =98.00% (95.70% ~ 99.26%)
Accuracy=97.77% (95.46% ~ 99.10%)

Interference Substances

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

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

Cross Reaction

The Leishmania IgG/IgM Rapid Test has been tested by HAMA, RF, HBsAg, HBsAb, HBeAg, HBeAb, HBcAb,anti-Syphilis, anti-HIV, anti-H.Pylori, anti-MONO, anti-CMV IgG, anti-CMV IgM, anti-Rubella IgG, anti-Rubella IgM, anti-Toxo IgG and anti-Toxo IgM positive specimens. The results showed no cross-reactivity.

INDEX OF SYBOML

IVD	In vitro diagnostic medical device		single-use, Please don't reuse it
	Use-by date		Consult instructions for use
	Cautions		Manufacturer
2°C 30°C	Temperature limit	LOT	Batch code
	Date of manufacture		Keep Dry
	Avoid overexposure to the sun		Don't use the product when the package is damaged
	Biological risks	CE	CE mark

Hangzhou Frenovo Biotech Co., Ltd.
Address: Room 401,Building 26, Room 401&301,Building 27,No.488-1,Donghu North Road, Donghu Street,LinPing District,Hangzhou City,Zhejiang Province.China.
Tel: 86-571-89170657
Email: business@frenovo.com

EC REP
CMC Medical Devices & Drugs S.L
Address: C/Horacio Lengo Nº 18, CP 29006, Málaga, Spain
Emai: info@cmcmedicaldevices.com

INSTRUCTION APPROVAL AND REVISION DATE

Revision Date: 2023.01.23
Approval Date: 2023.01.23
Date of Issue: 2023.01.23