FRENOVO One Step HBV-5 Combo Rapid Test Kit

Instructions For Use

PRODUCT NAME

FRENOVO One Step HBV-5 Combo Rapid Test Kit

PACKAGE SPECIFICATION

25 tests/kit

INTENDED USE

FRENOVO One Step HBV-5 Combo Rapid Test Kit is a rapid chromatographic immunoassay for the qualitative detection of hepatitis B virus markers, Hepatitis B Surface Antigen (HBsAg), Hepatitis B Surface Antibody (HBsAb), Hepatitis B Envelope Antigen (HBeAg), Hepatitis B Envelope Antibody (HBeAb), and Hepatitis Core Antibody (HBcAb) in human whole blood, serum, or plasma specimens to aid in the diagnosis of infection with hepatitis B virus (HBV). The test only provides preliminary analysis results but not critical diagnosis criteria.

For professional in vitro diagnostic use only.

SUMMARY AND PRINCIPLES OF THE PROCEDURE

Hepatitis B virus (HBV) is the most common cause of persistent viremia and the most important cause of chronic liver disease and hepatocellular carcinoma. Clinically apparent HBV infections may have been in existence for several millennia. It is estimated that there are 300 million chronic carriers of HBV in the world. The carrier rates vary from as little as 0.3% (Western countries) to 20% (Asia, Africa). HBV is a hepatotropic DNA virus. The core of the virus contains a DNA polymerase, the core antigen (HBcAg) and the e antigen (HBeAg). The core of HBV is enclosed in a coat that contains lipid, carbohydrate and protein including an antigen termed hepatitis B surface antigen (HBsAg). Most people fight off the infection themselves, but approximately 5-10 percent of those infected with the virus become carriers, and an additional 5-10 percent of those infected each year will progress to chronic liver disease, cirrhosis and possibly liver cancer.

The FRENOVO One Step HBV-5 Combo Rapid Test is a qualitative membrane-based immunoassay for the detection of HBsAg, HBsAb, HBeAg, HBeAb and HBcAb in human whole blood, serum, or plasma specimens.

HBsAg and HBeAg

The HBsAg and HBeAg tests are qualitative, two-site sandwich immunoassays for the detection of HBsAg or HBeAg in whole blood, serum or plasma specimens. The membrane is pre-coated with anti-HBsAg or anti-HBeAg antibodies on the test line region (T) of the strip. During testing, the specimen reacts with the particle coated with anti-HBsAg or anti-HBeAg antibodies. The mixture migrates upward on the membrane chromatographically by capillary action to react with anti-HBsAg or anti-HBeAg antibodies on the membrane and generate a colored line. The presence of this colored line in the test line region (T) indicates a positive result, while its absence indicates a negative result.

HBsAb

Hepatitis B surface Antibody (HBsAb) is also known as antiHepatitis B surface Antigen (anti-HBs). This test is a qualitative, lateral flow immunoassay for the detection of HBsAb inwhole blood, serum or plasma specimens. The membrane is pre-coated with HBsAg on the test line region (T) of the strip. During testing, the specimen reacts with the particle coated with HBsAg. The mixture migrates upward on the membrane chromatographically by capillary action to react with HBsAg on the membrane and generate a colored line. The presence of this colored line in the test line region (T) indicates a positive result, while its absence indicates a negative result.

HBeAb and HBcAb

Hepatitis B envelope Antibody (HBeAb) is also known as antiHepatitis B envelope Antigen (anti-HBe). Hepatitis B core Antibody (HBcAb) is also known as anti-Hepatitis B core Antigen (anti-HBc). These tests are immunoassays based on the principle of competitive binding. During testing, the mixture migrates upward on the membrane chromatographically by capillary action. The membrane is precoated with HBeAg or HBcAg on the test line region (T) of the strip. During testing, anti-HBe antibody or anti-HBc antibody, if present in the specimen, will compete with particle coated anti-HBe antibody or anti-HBc antibody for limited amount of HBeAg or all HBcAg on the membrane, and no line will form in the test line region (T), indicating a positive result. A visible colored line will form in the control line (C) and test line region (T) if there is no anti-HBe antibody or anti-HBc antibody in the specimen because all the antibody coated particles will be captured by the antigen coated in the test line region (T).

An internal quality control is included in the test, in the form of a colored line appearing in the control line region (C), indicating that the test is functional, and proper and sufficient volume of specimen has been applied to enable migration through the test and control lines, regardless of whether there is a test line or not. If the control line (C) does not appear within the testing time, test result is invalid and the test should be repeated with a new test device.

MATERIALS PROVIDED

Each kit contains:

1. Test Devices: 25 pieces test devices individually pouched.
2. Wash Buffer Solution: 3 dropper bottles of 3.0 ml in each.
3. Droppers: 25 pieces droppers of 25 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)
- Micropipette
- Lancets(for fingertip whole blood only)

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 kitis for diagnostic use only.
3. Perform test at room temperature.

PRECAUTIONS

1. The kit rapid test 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 extraction 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 box 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 devices 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

Consider any materials of human origin as infectious and handle them using standard biosafety procedures. The test can be performed using whole blood (from venipuncture or fingerstick), serum or plasma specimens. Follow standard laboratory procedures to collect specimens.

Plasma/Serum

1. Collect blood specimen into collection tube containing EDTA, citrate or heparin for plasma or collection tube containing no anticoagulants for serum by venipuncture.
2. To make plasma specimen, centrifuge collected specimens and carefully withdraw the plasma into a new pre-labeled tube.
3. To make serum specimen, allow blood to clot, then centrifuge collected specimens and carefully withdraw the serum into a new pre-labeled tube.

Test specimens as soon as possible after collecting. Store specimens at 2-8° C if not tested immediately. Specimens can be stored at 2-8 ° C for up to 3 days, and should be frozen at -20 ° C for longer storage.
Avoid multiple freeze-thaw cycles (no more than 3 times). Prior to testing, equilibrate 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 so as to avoid interference on result interpretation.

Whole Blood

Collect whole blood by either fingertip puncture or by venipuncture into collection tube containing EDTA, citrate or heparin for plasma. Do not use any hemolyzed blood for testing.

Do not freeze a whole blood specimen, otherwise the red blood cell will break, which may cause hemolysis. Whole blood specimens should be stored in refrigeration (2-8 ° C) if not tested immediately. The specimens must be tested within 24 hours after collection.

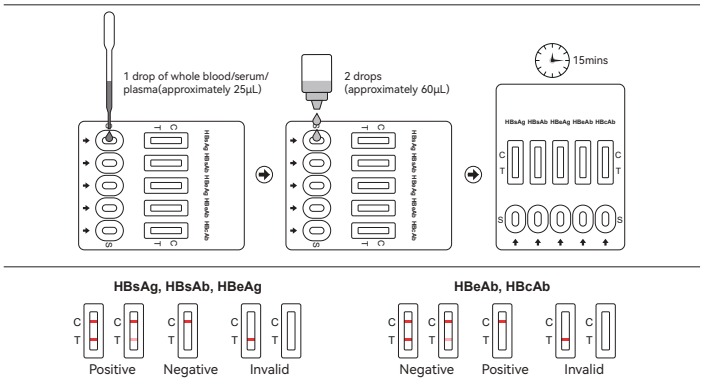
QUALITY CONTROL

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 device, specimen, wash Buffer solution to equilibrate to room temperature (15-30°C) prior to testing.

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. Fill the pipette dropper with the specimen. Hold the dropper vertically and transfer one drop of whole blood/serum/plasma specimen (approximately 25μL) into the sample well making sure that there are no air bubbles. Then add two drops of sample diluent (approximately 60μL) to the sample well immediately. See illustration below.
3. Start the timer.
4. Wait for the colored line(s) to appear. Read test results at 15 minutes. Do not interpret the result after 20 minutes.



INTERPRETATION OF RESULTS

Warning: Do not interpret all 5 test strips at the same criterion. Please follow the directions below carefully.

For HBsAg, HBsAb, HBeAg

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

***NOTE:** The intensity of the color in the test line region (T) will vary depending on the concentration of HBsAg, HBsAb, HBeAg present in the specimen. Therefore, any shade of color in the test region (T) should be considered positive.

NEGATIVE: One colored line appears in the control region (C).No apparent colored line appears in the test region (T).

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

For HBeAb, HBcAb

NEGATIVE:* Two distinct colored lines appear. One colored line should be in the control region (C) and another colored line should be in the test region (T).

***NOTE:** The intensity of the color in the test line region (T) may vary. But it should be considered negative whenever there is even a faint pink line.

POSITIVE: One colored line appears in the control region (C).No apparent colored line appears in the test region (T).

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

LIMITATIONS

1. The test is only used for the qualitative detection of HBsAg, HBsAb, HBeAg, HBeAb and HBcAb in human whole blood, serum, or plasma specimens by healthcare professionals. The intensity of the test line does not have a linear correlation with the HBsAg, HBsAb, HBeAg, HBeAb and HBcAb level in the specimen.
2. The test does not indicate the level of HBsAg, HBsAb, HBeAg, HBeAb and HBcAb in the specimens or the rate of increase in HBsAg, and should not be used as the sole criteria for the diagnosis of infection with HBV.
3. A negative result indicates that HBsAg, HBsAb, HBeAg, HBeAb and HBcAb is not present in the specimen. However, a negative test result at any time does not preclude the possibility of exposure to or infection with HBV.
4. A negative result may occur if the level of HBsAg, HBsAb, HBeAg, HBeAb and HBcAb present in the specimen is below the detection limits of the assay or the antigens/antibodies that are detected are not present during the stage of disease when a sample is collected.
5. A positive result using the FRENOVO One Step HBV-5 Combo Rapid Test suggests the presence of HBsAg, HBsAb, HBeAg, HBeAb and HBcAb in the sample and the positive test result should be interpreted as preliminary positive for HBsAg, HBsAb, HBeAg, HBeAb and HBcAb. Positive test results must be confirmed by additional testing.
6. If the test result is negative and clinical symptoms persist, re-sample the patient and additional testing using alternative clinical methods is recommended.
7. Test results obtained with this test should only be interpreted in conjunction with other diagnostic procedures and clinical findings.

PERFORMANCE CHARACTERISTICS

1. Clinical Performance

The FRENOVO One Step HBV-5 Combo Rapid Test Kit been has correctly identified specimens of a performance panel and has been evaluated with a reference commercial assay using clinical specimens, respectively. Test results are presented in the table below.

Clinical performance compared to ELISA: HBsAg

One Step HBV-5 Combo Rapid Test	ELISA		
	Positive	Negative	Total
Positive	710	5	715
Negative	10	2120	2130
Total	720	2125	2845

Sensitivity (Positive Percent Agreement): 98.6% (95% CI: 97.46%~99.24%)
Specificity (Negative Percent Agreement): 99.7% (95% CI: 99.45%~99.90%)
Accuracy (Overall Percent Agreement): 99.4% (95% CI: 99.13%~99.68%)

Clinical performance compared to ELISA: HBsAb

One Step HBV-5 Combo Rapid Test	ELISA		
	Positive	Negative	Total
Positive	678	2	680
Negative	2	708	710
Total	680	710	1390

Sensitivity (Positive Percent Agreement): 99.7% (95% CI: 98.93%~99.92%)
Specificity (Negative Percent Agreement): 99.7% (95% CI: 98.98%~99.92%)
Accuracy (Overall Percent Agreement): 99.7% (95% CI: 99.26%~99.89%)

Clinical performance compared to ELISA: HBeAg

One Step HBV-5 Combo Rapid Test	ELISA		
	Positive	Negative	Total
Positive	610	8	618
Negative	12	900	1002
Total	622	908	1530

Sensitivity (Positive Percent Agreement): 98.0% (95% CI: 96.66%~98.99%)
Specificity (Negative Percent Agreement): 99.1% (95% CI: 98.27%~99.55%)
Accuracy (Overall Percent Agreement): 98.6% (95% CI: 97.99%~99.15%)

Clinical performance compared to ELISA: HBeAb

One Step HBV-5 Combo Rapid Test	ELISA		
	Positive	Negative	Total
Positive	489	7	496
Negative	11	793	804
Total	500	800	1300

Sensitivity (Positive Percent Agreement): 97.8% (95% CI: 96.66%~98.99%)
Specificity (Negative Percent Agreement): 99.1% (95% CI: 98.27%~99.55%)
Accuracy (Overall Percent Agreement):98.6% (95% CI: 97.99%~99.15%)

Clinical performance compared to ELISA: HBcAb

One Step HBV-5 Combo Rapid Test	ELISA		
	Positive	Negative	Total
Positive	547	8	555
Negative	13	812	825
Total	560	820	1380

Sensitivity (Positive Percent Agreement): 97.6% (95% CI: 96.07%~98.64%)
Specificity (Negative Percent Agreement): 99.0% (95% CI: 98.09%~99.50%)
Accuracy (Overall Percent Agreement):98.4% (95% CI: 97.68%~99.00%)

2. Analytical Sensitivity

The analytical sensitivity of the FRENOVO One Step HBV-5 Combo Rapid Test was evaluated by testing reference panel. The results demonstrated that the assay could detect a concentration of 0.5ng/mL (1.0 IU/mL) HBsAg, 10 mIU/mL HBsAb, 2.0 NCU/mL HBeAg, 2.0 NCU/mL HBeAb, 2.0 NCU/mL HBcAb.

3. Precision

Intra-Assay

Within-run precision has been determined by using 15 replicates of three specimens containing negative, low positive, high positive of HBsAg, HBsAb, HBeAg, HBeAb, HBcAb. The negative and positive values were correctly identified 99% of the time.

Inter-Assay

Between-run precision has been determined by using the same three specimens of negative, low positive, high positive of HBsAg, HBsAb, HBeAg, HBeAb, HBcAb in 15 independent assays. Three different lots of theFRENOVO One Step HBV-5 Combo Rapid Test has been tested over a 10 days period using negative, low positive and high positive specimens. The specimens were correctly identified 99% of the time.

4. Cross-reactivity

The FRENOVO One Step HBV-5 Combo Rapid Test has been tested by HAMA, Rheumatoid factor (RF), HAV, Syphilis, HIV, H. Pylori, HCV, CMV, Rubella and TOXO positive specimens. The results showed no cross-reactivity.

5. Interfering Substances

The FRENOVO One Step HBV-5 Combo Rapid Test has been tested for possible interference from visibly hemolyzed and lipemic specimens. No interference was observed. In addition, no interference was observed in specimens containing up to 2,000 mg/dL Hemoglobin, 1,000 mg/dL Bilirubin, and 2,000 mg/dL human serum Albumin.

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		

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INSTRUCTION APPROVAL AND REVISION DATE

Revision Date: 2025.10.22

Approval Date: 2025.10.22

Date of Issue: 2025.10.22