SPECTRUM

The Creative Approach to Bioscience

HBsAg/HCV/HIV 1.2 Combo Rapid Test Cassette (Whole Blood/Serum /Plasma)

Package Insert
REF: 1235 001 25 test

INTENDED USE

The HBsAg/HCV/HIV 1.2 Combo Rapid Test Cassette (Whole Blood/Serum/Plasma) is a rapid chromatographic immunoassay for the qualitative detection of Hepatitis B surface antigen (HBsAg), antibodies to Hepatitis C Virus and HIV type 1 plus type 2 antibody in whole blood, serum or plasma specimen.

SUMMARY

The HBsAg Rapid Test (Whole Blood/Serum/Plasma) is a rapid test to qualitatively detect the presence of HBsAg in whole blood, serum or plasma specimen. The test utilizes a combination of monoclonal and polyclonal antibodies to selectively detect elevated levels of HBsAg in whole blood, serum or plasma specimen.

Viral hepatitis is a systemic disease primarily involving the liver. Most cases of acute viral hepatitis are caused by Hepatitis A virus, Hepatitis B virus (HBV) or Hepatitis C virus. The complex antigen found on the surface of HBV is called HBsAg. Previous designations included the Australia or Au antigen.[1]The presence of HBsAg in serum or plasma is an indication of an active Hepatitis B infection, either acute or chronic. In a typical Hepatitis B infection, HBsAg will be detected 2 to 4 weeks before the ALT level becomes abnormal and 3 to 5 weeks before symptoms or jaundice develop. HBsAg has four principal subtypes: adw, ayw, adr and ayr. Because of antigenic heterogeneity of the determinant, there are 10 major serotypes of Hepatitis B virus.

The HCV Rapid Test (Whole Blood/Serum/Plasma) is a rapid test to qualitatively detect the presence of antibody to HCV in whole blood, serum or plasma specimen. The test utilizes colloid gold conjugate and recombinant HCV proteins to selectively detect antibody to HCV in whole blood, serum or plasma specimen. The recombinant HCV proteins used in the test kit are encoded by the genes for both structural (nucleocapsid) and non-structural proteins.

Hepatitis C Virus (HCV) is a small, enveloped, positive-sense, single-stranded RNA Virus. HCV is now known to be the major cause of parenterally transmitted non-A, non-B hepatitis. Antibody to HCV is found in over 80% of patients with well-documented non-A, non-B hepatitis.

Conventional methods fail to isolate the virus in cell culture or visualize it by electron microscope. Cloning the viral genome has made it possible to develop serologic assays that use recombinant antigens.[2,3] Compared to the first generation HCV EIAs using single recombinant antigen, multiple antigens using recombinant protein and/or synthetic peptides have been added in new serologic tests to avoid nonspecific cross-reactivity and to increase the sensitivity of the HCV antibody tests.[4,5]

The HIV 1.2 Rapid Test (Whole Blood/Serum/Plasma) is a rapid test to qualitatively detect the presence of antibody to HIV 1 and/or HIV 2 in whole blood, serum or plasma. The test utilizes latex conjugate and multiple recombinant HIV proteins to selectively detect antibodies to the HIV 1&2 in whole blood, serum or plasma.

HIV is the etiologic agent of Acquired Immune Deficiency Syndrome (AIDS). The virion is surrounded by a lipid envelope that is derived from host cell membrane. Several viral glycoproteins are on the envelope. Each virus contains two copies of positive-sense genomic RNAs. HIV 1 has been isolated from patients with AIDS and AIDS-related complex, and from healthy people with high potential risk for developing AIDS.[6] HIV 2 has been isolated from West African AIDS patients and from seropositive asymptomatic individuals.[7] Both HIV 1 and HIV 2 elicit immune response.[8] Detection of HIV antibodies in serum, plasma is the most efficient and common way to determine whether an individual has been exposed to HIV and to screen blood and blood products for HIV.[9] Despite the differences in their biological characteristics, serological activities and genome sequences, HIV 1 and HIV 2 show strong antigenic cross-reactivity.[10,11] Most HIV 2 positive sera can be identified by using HIV 1 based serological tests.

SYMBOLS IN PRODUCT LABELLING

EC REP	Authorised Representative	°C	Temperature Limitation
IVD	For in-vitro diagnostic use		Use by/Expiration Date
LOT	Batch Code/Lot number		CAUTION. Consult instructions for use
REF	Catalogue Number		Manufactured by
	Consult instructions for use		

TEST PRINCIPLE

The HBsAg Rapid Test (Whole Blood/Serum/Plasma) is a qualitative, solid phase, two-site sandwich immunoassay for the detection of HBsAg in whole blood, serum or plasma specimen. The membrane is pre-coated with anti-HBsAg antibodies on the test line region of the cassette. During testing, the whole blood, serum or plasma specimen reacts with the particle coated with anti-HBsAg antibodies. The mixture migrates upward on the membrane chromatographically by capillary action to react with anti-HBsAg antibodies on the membrane and generate a colored line. The presence of this colored line in the test region 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.

The HCV Rapid Test (Whole Blood/Serum/Plasma) is a qualitative, membrane based immunoassay for the detection of antibody to HCV in whole blood, serum or plasma. The membrane is pre-coated with recombinant HCV antigen on the test line region of the cassette. During testing, the whole blood, serum or plasma specimen reacts with recombinant HCV antigen conjugated colloid gold.

The mixture migrates upward on the membrane chromatographically by capillary action to react with recombinant HCV antigen 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 at the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

The HIV 1.2 Rapid Test (Whole Blood/Serum/Plasma) is a qualitative, membrane based immunoassay for the detection of antibodies to HIV 1.2 in whole blood, serum or plasma.

The membrane is pre-coated with recombinant HIV antigens. During testing, the whole blood, serum or plasma specimen reacts with HIV antigen coated particles in the test device. The mixture then migrates upward on the membrane chromatographically by capillary action and reacts with recombinant HIV antigen on the membrane in the test line region. If the specimen contains antibodies to HIV 1 and/or HIV 2, a colored line will appear in the test line region, indicating a positive result. If the specimen does not contain HIV 1 and/or HIV 2 antibodies, a colored line will not appear in the test line region, indicating 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.

REAGENTS AND MATERIALS PROVIDED

The test cassette contains anti-HBsAg conjugated particles, anti-HBsAg coated on the membrane, recombinant HCV antigen conjugated particles, HCV antigen coated on the membrane and HIV 1 antigen plus HIV 2 antigen conjugated particles and HIV 1 antigen plus HIV 2 antigen coated on the membrane.

Materials provided

- Test cassettes
- Buffer
- Droppers
- Package insert

Materials required but not provided

- Specimen collection containers
- Centrifuge (for plasma only)
- Timer

WARNINGS AND PRECAUTIONS

- For professional in vitro diagnostic use only. Do not use after expiration date.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout the procedure and follow the standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
- Humidity and temperature can adversely affect results.

REAGENT PREPARATION AND STORAGE INSTRUCTION

The kit can be stored at room temperature or refrigerated (2-30°C). The test cassette is stable through the expiration date printed on the sealed pouch. The test cassette must remain in the sealed pouch until use. DO NOT FREEZE. Do not use beyond the expiration date.

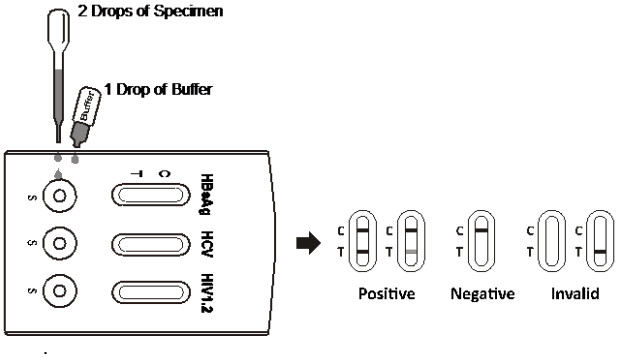
SPECIMEN COLLECTION AND PREPARATION

- The HBsAg/HCV/HIV 1.2 Combo Rapid Test Cassette (Whole Blood/Serum /Plasma) can be performed using whole blood (from venipuncture or fingerstick) serum or plasma specimen.
- To collect **Fingerstick Whole Blood specimens**:
  - Wash the patient's hand with soap and warm water or clean with an alcohol swab. Allow to dry.
  - Massage the hand without touching the puncture site by rubbing down the hand towards the fingertip of the middle or ring finger.
  - Puncture the skin with a sterile lancet. Wipe away the first sign of blood.
  - Gently rub the hand from wrist to palm to finger to form a rounded drop of blood over the puncture site.
  - Add the Fingerstick Whole Blood specimen to the test by using **a capillary tube**:
    - Touch the end of the capillary tube to the blood until filled to approximately 50 μL. Avoid air bubbles.
    - Place the bulb onto the top end of the capillary tube, then squeeze the bulb to dispense the whole blood to the specimen area of the test cassette.
  - Add the Fingerstick Whole Blood specimen to the test by using **hanging drops**:
    - Position the patient's finger so that the drop of blood is just above the specimen area of the test cassette.
    - Allow 2 hanging drops of fingerstick whole blood to fall into the center of the specimen area respectively, or move the patient's finger so that the hanging drop touches the center of the specimen area. Avoid touching the finger directly to the specimen area.
- Separate serum or plasma from blood as soon as possible to avoid hemolysis. Use only clear non-hemolyzed specimens.

TEST PROCEDURE

Allow test cassette, specimen, and/or controls to equilibrate to room temperature (15-30°C) prior to testing.

1. Bring the pouch to room temperature before opening it. Remove the test device from the sealed pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one hour.
2. Place the test device on a clean and level surface. Hold the dropper vertically and transfer **2 drops of specimen (approximately 50μL)** to the specimen area each, then add **1 drop of buffer (approximately 40μL)** respectively. Start the timer. See the illustration below.
3. Wait for the colored line(s) to appear. The test result should be read at 10 minutes. Do not interpret the result after 20 minutes



INTERPRETATION OF RESULTS

(Please refer to the illustration above)

POSITIVE: * Two distinct colored lines appear. One color line should be in the control region (C) and another color 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 antigen and/or HCV antibodies and/or HIV 1.2 antibodies present in the specimen. Therefore, any shade of red in the test region should be considered positive.

NEGATIVE: One color line appears in the control region (C). No apparent red or pink 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.

QUALITY CONTROL

Internal procedural controls are included in the test. A color line appearing in the control region (C) is an internal positive procedural control. It confirms sufficient specimen volume and correct procedural technique.

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.

LIMITATIONS OF TEST

1. This test is for in vitro diagnostic use only.
2. This test has been developed for testing whole blood, serum or plasma specimen. The performance of the test using other specimens has not been substantiated.
3. This test is a qualitative screening assay. It is not designed to determine the quantitative concentration of HBsAg antigen, HCV antibody and HIV antibody.
4. The HBsAg Rapid Test cannot detect less than 1 PEI ng/ml of HBsAg in specimens.
5. As with all diagnostic tests, all results must be considered with other clinical information available to the physician.
6. If the test result is negative and clinical symptoms persist, additional follow-up testing using other clinical methods is recommended. A negative result at any time does not preclude the possibility of HBsAg and/or Hepatitis C Virus and/or HIV infectious.

Expected Values

The HBsAg/HCV/HIV 1.2 Combo Rapid Test Cassette (Whole Blood/Serum/Plasma) has been compared with a leading commercial EIA test , respectively. The correlation between these two systems is 99%.

PERFORMANCE CHARACTERISTICS

Sensitivity and Specificity

1.HBsAg

Method		EIA		Total Results
HBsAg Rapid Test (Whole Blood/Serum /Plasma)	Results	Positive	Negative	
	Positive	99	2	101
	Negative	1	298	299
Total Results		100	300	400

Relative Sensitivity: 99.0% (95%CI:*94.6%-100%)

Relative Specificity: 99.3% (95%CI:*97.6%-99.9%)

Overall accuracy: 99.3% (95%CI:*97.8%-99.8%)
*Confidence Intervals

2.HCV

Method		EIA		Total Results
HCV Rapid Test (Whole Blood/Serum/Plasma)	Results	Positive	Negative	
	Positive	98	2	100
	Negative	2	298	300
Total Result		100	300	400

Relative Sensitivity: 98.0% (95%CI:*93.0%-99.8%)

Relative Specificity: 99.3% (95%CI:*97.6%-99.9%)

Overall accuracy: 99.3% (95%CI:*97.8%-99.8%)
*Confidence Intervals

3.HIV 1.2

Method		EIA		Total Results
HIV 1.2 Rapid Test (Whole Blood/Serum/Plasma)	Results	Positive	Negative	
	Positive	99	3	102
	Negative	1	297	298
Total Result		100	300	400

Relative Sensitivity: 99.0% (95%CI:*94.6%-100%)

Relative Specificity: 99.0% (95%CI:*97.1%-99.8%)

Overall accuracy: 99.0% (95%CI:*97.5%-99.7%)
*Confidence Intervals

Precision

Intra-Assay

Within-run precision has been determined by using 10 replicates of four different specimens containing different concentrations of HBsAg antigen, HCV antibody and HIV antibody. The negative, positive values were correctly identified 100% of the time.

Inter-Assay

Between-run precision has been determined by 10 independent assays on the same four different specimens containing different concentrations of HBsAg, and HCV antibody. Three different lots of the HBsAg/HCV/HIV 1.2 Combo Rapid Test (Whole Blood/Serum/Plasma) have been tested over a 10 days period using above negative and positive specimens. The specimens were correctly identified 100% of the time.

Cross-reactivity

The HBsAg Rapid Test (Whole Blood/Serum/Plasma) has been tested by HAMA, Rheumatoid factor (RF), HAV, Syphilis, HIV, H. Pylori, MONO, CMV, Rubella, HCV, HEV and TOXO positive specimens. The results showed no cross-reactivity

The HCV Rapid Test (Whole Blood/Serum/Plasma) has been tested by HAMA, RF, HBsAg, HBsAb, HBeAg, HBeAb, HBcAb, Syphilis,HIV, H. Pylori, MONO, CMV, Rubella and TOXO positive specimens. The results showed no cross-reactivity.

The HIV 1.2 Rapid Test(Whole Blood/Serum/Plasma) has been tested by HAMA, RF, HBsAg, HBsAb, HBeAg, HBeAb, HBcAb, HCV, Syphilis, H. Pylori, MONO, CMV, Rubella and TOXO positive specimens. The results showed no cross-reactivity.

Interfering Substances

The following potentially interfering substances were added to HBsAg, HCV antibody negative and positive specimens.

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

None of the substances at the concentration tested interfered in the assay.

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