Rapid Response™

Hepatitis C Virus Test Cassette
(Serum/Plasma)

REF HCV-12C40 Product Insert

A rapid test for the qualitative detection of antibodies to Hepatitis C Virus in human serum or plasma.

For professional *in vitro* diagnostic use only.

Intended Use

The Rapid Response™ Hepatitis C Virus Test Cassette is a rapid chromatographic immunoassay for the qualitative detection of antibody to Hepatitis C Virus in serum or plasma.

Summary

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.[1, 2] 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.[3,4]

The Rapid Response™ Hepatitis C Virus Test Cassette is a rapid test to qualitatively detect the presence of antibody to HCV in a serum or plasma specimen. The test utilizes colloid gold conjugate and recombinant HCV proteins to selectively detect antibody to HCV in serum or plasma. The recombinant HCV proteins used in the test kit are encoded by the genes for both structural (nucleocapsid) and non-structural proteins.

Principle

The Rapid Response™ Hepatitis C Virus Test Cassette is a qualitative, membrane based immunoassay for the detection of antibody to HCV in serum or plasma. The membrane is pre-coated with recombinant HCV antigen on the test line region of the Cassette. During testing, the 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.

Reagents

The test Cassette contains recombinant HCV antigen conjugated colloid gold and HCV antigen coated on the membrane.

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.

Materials

Materials provided

- Test cassettes
- Buffer
- Droppers
- Product insert

Materials required but not provided

- Specimen collection containers
- Centrifuge
- Timer

Storage and Stability

The kit can be stored at room temperature or refrigerated (35.6-86°F; 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.

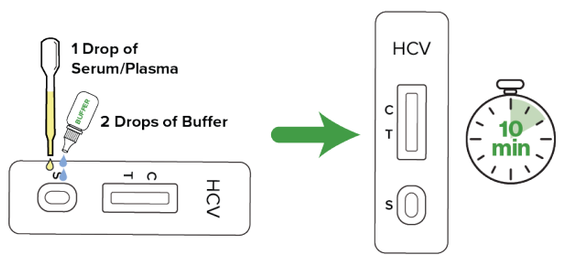
Specimen Collection and Preparation

- The Rapid Response™ Hepatitis C Virus Test Cassette can be performed using serum or plasma.
- Separate the serum or plasma from blood as soon as possible to avoid hemolysis. Only clear, non-hemolyzed specimens can be used.
- Testing should be performed immediately after the specimens have been collected. Do not leave the specimens at room temperature for prolonged periods. Serum and plasma specimens may be stored at 35.6-46.4°F (2-8°C) for up to 3 days. For long term storage, specimens should be kept below -4°F (-20°C).
- 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.
- If specimens are to be shipped, they should be packed in compliance with federal regulations for transportation of etiologic agents.

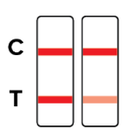
Test Procedure

Allow test Cassette, specimen, and/or controls to equilibrate to room temperature (59-86°F; 15-30°C) prior to testing.

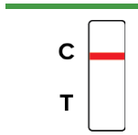
1. Bring the pouch to room temperature before opening it. Remove the test Cassette from the sealed pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one hour.
2. Place the Cassette on a clean and level surface.
3. For **Serum or Plasma** specimen:
   Hold the dropper vertically and transfer **1 drop of serum or plasma (approximately 25µL)** to the specimen well, then **add 2 drops of buffer (approximately 80 µL)**, and start the timer, see illustration below. 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**.



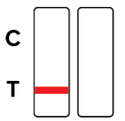
Results Interpretation



POSITIVE: * Two 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 colored in the test line region (T) will vary depending on the concentration of HCV 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 colored 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

1. The Rapid Response™ Hepatitis C Virus Test Cassette is for *in vitro* diagnostic use only. This test should be used for the detection of antibodies to HCV in serum or plasma specimen.
2. The Rapid Response™ Hepatitis C Virus Test Cassette will only indicate the presence of antibodies to HCV in the specimen and should not be used as the sole criteria for the diagnosis of Hepatitis C viral infection.
3. As with all diagnostic tests, all results must be considered with other clinical information available to the physician.
4. 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 Hepatitis C Virus infection.

Performance Characteristics

Sensitivity and Specificity

The recombinant antigen used for the Rapid Response™ Hepatitis C Virus Test Cassette is encoded by genes for both structural (nucleocapsid) and non-structural proteins. The Rapid Response™ Hepatitis C Virus Test Cassette has passed a seroconversion panel and compared with a leading commercial HCV EIA test using clinical specimens.

The results show that the relative sensitivity of the Rapid Response™ Hepatitis C Virus Test Cassette is 98.7%, and the relative specificity is 99.1%.

Method		EIA		Total Result
Rapid Response™	Results	Positive	Negative	
Hepatitis C Virus	Positive	235	6	241
Test Cassette	Negative	3	692	695
Total Result		238	698	936

Relative sensitivity: 98.7% (95%CI:*96.4%-99.7%)
Relative specificity: 99.1% (95%CI:*98.1%-99.7%)
Accuracy: 99.0% (95%CI:*98.2%-99.6%)
*Confidence Intervals

Precision

Intra-Assay

Within-run precision has been determined by using 20 replicates of three specimens: a negative, a HCV low titer positive and a HCV high titer positive. The negative, HCV low titer positive and HCV high titer positive values were correctly identified 100% of the time.

Inter-Assay

Between-run precision has been determined by 20 independent assays on the same three specimens: a negative, a HCV low titer positive and a HCV high titer positive. Three different lots of the Rapid Response™ Hepatitis C Virus Test Cassette have been tested using negative, HCV low titer positive and HCV high titer positive specimens. The specimens were correctly identified 100% of the time.

Cross-reactivity

The Rapid Response™ Hepatitis C Virus Test Cassette 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.

Interfering Substances

The following potentially interfering substances were added to HCV 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.

Bibliography

1. Choo, Q.L., G. Kuo, A.J. Weiner, L.R. Overby, D.W. Bradley,and M. Houghton. Isolation of a cDNA clone derived from a blood-borne non-A, non-B viral hepatitis genome. Science. 1989; 244:359
2. Kuo, G., Q.L. Choo, H.J. Alter, and M. Houghton. An assay for circulating antibodies to a major etiologic Virus of human non-A, non-B hepatitis. Science. 1989; 244:362
3. van der Poel, C. L., H.T.M. Cuypers, H.W. Reesink, and P.N.Lelie.Confirmation of hepatitis C Virus infection by new four-antigen recombinant immunoblot assay. Lancet. 1991; 337:317
4. Wilber, J.C. Development and use of laboratory tests for hepatitis C infection: a review. J. Clin. Immunoassay. 1993; 16:204

Glossary of Symbols

Consult instructions for use	Test per Kit	Do Not Reuse
Store between 35.6°F to 86°F	Use by	Catalogue #
Lot Number	For *in vitro* diagnostic use only	Manufacturer

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