OnSite HCV Ab Combo Rapid Test- (Serum / Plasma / Whole Blood)

INTENDED USE

The OnSite HCV Ab Combo Rapid Test is an indirect lateral flow chromatographic immunoassay for the qualitative detection of IgG and IgM anti-Hepatitis C virus (HCV) in human serum, plasma or whole blood. It is intended to be used as a screening test and as an aid in the diagnosis of infection with HCV. Any reactive specimen with the OnSite HCV Ab Combo Rapid Test must be confirmed with alternative testing method(s) and clinical findings.

SUMMARY AND EXPLANATION OF THE TEST

Hepatitis C virus (HCV), which was formerly described as the parentally transmitted form of non-A, non-B hepatitis (NANBH), becomes a chronic disease in 50% of the cases. HCV can also be transmitted through intravenous drug abuse, sexual, and household contact. HCV is a single stranded RNA virus with some structural relations to the flavivirus family. Nucleic acid sequences of HCV cDNA clones provided the basis for the construction of recombinant peptides representing putative Hepatitis C virus proteins. Anti-hepatitis C virus antibody screening of blood using synthetic or recombinant proteins, helped to identify apparently healthy blood donors with anti-HCV antibodies who otherwise might have transmitted the virus.

TEST PRINCIPLE

The OnSite HCV Ab Combo Rapid Test is an indirect lateral flow chromatographic immunoassay. The test cassette consists of: 1) a burgundy colored conjugate pad containing proprietary recombinant protein A conjugated with colloidal gold (Protein A conjugates), 2) a nitrocellulose membrane strip containing a test band (T band) and a control band (C band). The T band is pre-coated with recombinant HCV antigens, and the C band is pre-coated with anti-protein A antibodies.

When an adequate volume of test specimen is dispersed into the sample well of the test cassette, the specimen migrates by capillary action across the cassette. The IgM or IgG antibody to HCV if present in the specimen will bind to the HCV conjugates. The immunocomplex is then captured on the membrane by the pre-coated non-conjugated HCV antigens, forming a burgundy colored T band, indicating a HCV Ab positive test result.

Absence of the T band suggests a negative result. The test contains an internal control (C band) which should exhibit a burgundy colored band of the immunocomplex of anti-protein A antibodies.

ASSAY PROCEDURE

1. Each kit contains 30 test devices, each sealed in a foil pouch with three items inside:
   a. One cassette device.
   b. One plastic drizzler.
   c. One desiccant.
2. Sample diluent (1 bottle, 5 mL)
3. One package insert (instruction for use).

MATERIALS REQUIRED AND AVAILABLE FOR PURCHASE

1. Positive Control (1 vial, red cap, 1 mL, Cat # R0021-P)
2. Negative Control (1 vial, green cap, 1 mL, Cat # R0021-N)

MATERIALS REQUIRED BUT NOT PROVIDED

1. Clock or Timer
2. Lancing device for whole blood test
3. Pipette and tips capable of delivering 20 µL volumes with a precision better than 1.5%.

WARRANTS AND PRECAUTIONS

For In Vitro Diagnostic Use

1. This package insert must be read completely before performing the test. Failure to follow the insert gives inaccurate test results.
2. Do not open the sealed pouch, unless ready to conduct the assay.
3. Do not use expired devices.
4. Bring all reagents to room temperature (15°C-30°C) before use.
5. Do not use the components in any other type of test kit as a substitute for the components in this kit.
6. Do not use hemolized blood specimen for testing.
7. Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after performing the test.
8. Users of this test should follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other blood-borne pathogens.
9. Do not smoke, drink, or eat in areas where specimens or kit reagents are being handled.
10. Dispose of all specimens and materials used to perform the test as biohazardous waste.
11. Handle the Negative and Positive Control in the same manner as patient specimens.
12. The testing results should be read within 15 minutes after a specimen is applied to the sample well or sample pad of the device. Read result after 15 minutes may give erroneous results.
13. Do not perform the test in a room with strong air flow, ie. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test devices unopened at 2°C-30°C. The positive and negative controls should be kept at 2°C-8°C. If stored at 2°C-8°C, ensure that the test device is brought to room temperature before opening. The test device is stable for up to 30°C.

SPECIMEN COLLECTION AND HANDLING

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

Plasma
1. Collect blood specimen into a lavender, blue or green top collection tube (containing EDTA, citrate or heparin, respectively in Vacutainer®) by veinpuncture.
2. Separate the plasma by centrifugation.
3. Carefully withdraw the plasma into new pre-labeled tube.

Serum
1. Collect blood specimen into a red top collection tube (containing no anticoagulants in Vacutainer®) by veinpuncture.
2. Allow the blood to clot.
3. Separate the serum by centrifugation.
4. Carefully withdraw the serum into a new pre-labeled tube.

Whole blood specimens should be stored in refrigeration (2°C-8°C) if not tested immediately. Store specimens at 2°C-8°C up to 5 days. The specimens should be frozen at -20°C for longer storage.

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.

Blood

Drops of whole blood can be obtained by either finger tip puncture or vein puncture. Do not use any hemolized blood for testing.

Whole blood specimens should be stored in refrigeration (2°C-8°C) if not tested immediately. The specimens must be tested within 24 hours of collection.

ASSAY PROCEDURE

Step 1: Bring the specimen and test components to room temperature if refrigerated or frozen. Mix the specimen well prior to assay once thawed.

Step 2: When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.

Step 3: Be sure to label the device with specimen’s ID number.

Step 4: For whole blood test
- Dispense 1 drop (about 40-50 µL) of the whole blood specimen into the sample well
- Then add 1 drop (about 35-50 µL) of Sample Diluent immediately

For serum or plasma test
- Dispense 20 µL of the specimen into the sample well
- Then add 2 drops (about 70-100 µL) of Sample Diluent immediately

20 µL of serum/plasma 2 drops of sample diluent 15 minutes

RESULT
Step 5: Set up timer.

Step 6: Results can be read in 15 minutes. Positive results can be visible in as short as 1 minute. Don’t read result after 15 minutes. To avoid confusion, discard the test device after interpreting the result.

**QUALITY CONTROL**

Using individual OnSite HCV Ab Combo Rapid Test cassettes as described in the Assay Procedure above, run 1 Positive Control and 1 Negative Control (provided upon request) under the following circumstances to monitor test performance:

1. A new operator uses the kit, prior to performing testing of specimens.
2. A new test kit is used.
3. A new shipment of kits is used.
4. The temperature used during storage of the kit falls outside of 2°C -30°C.
5. The temperature of the test area falls outside of 15° C -30°C.

Expected results are as follows:

**Negative Control**

Only the C band shows color development. The T band shows no color development.

**Positive Control**

Both C and T bands show color development.

The appearance of any burgundy color in the T band, regardless of intensity, must be considered as presence of the band.

**INTERPRETATION OF ASSAY RESULT**

1. **NEGATIVE RESULT**: If only the C band is developed, the test indicates that no detectable antibodies to HCV are present in the specimen. The result is negative.

2. **POSITIVE RESULT**: If both C and T bands are developed, the test indicates for the presence of antibodies to HCV in the specimen. The result is positive.

3. **INVALID**: If no C band is developed, the assay is invalid regardless of color development on the T band as indicated below. Repeat the assay with a new device.

Samples with positive results should be confirmed with alternative testing method(s) and clinical findings before a positive determination is made.

**PERFORMANCE CHARACTERISTICS**

**Clinical Performance**

A total of 700 samples from susceptible subjects were tested with the OnSite HCV Ab Combo Rapid Test and by a commercial HCV IgG ELISA kit. Comparison for all subjects is showed in the following table.

<table>
<thead>
<tr>
<th>OnSite HCV Ab Combo Rapid Test</th>
<th>HCV IgG ELISA Positive</th>
<th>HCV IgG ELISA Negative</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>40</td>
<td>2</td>
<td>42</td>
</tr>
<tr>
<td>Negative</td>
<td>13</td>
<td>645</td>
<td>658</td>
</tr>
<tr>
<td>Total</td>
<td>53</td>
<td>647</td>
<td>700</td>
</tr>
</tbody>
</table>

Relative Sensitivity: 95.2%, Relative Specificity:98.3%, Overall Agreement: 98.1%

**REFERENCES**