INTENDED USE

The OnSite HCV Ab Plus Combo Rapid Test is a double antigen lateral flow chromatographic immunoassay for the qualitative detection of anti-Hepatitis C virus antibodies (IgG, IgM, IgA) in human serum, plasma or whole blood. It is intended to be used by professionals as a screening test and an aid in the diagnosis of infection with HCV. Any reactive specimen with the OnSite HCV Ab Plus 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 a parenterally transmitted form of non-A, non-B hepatitis (NANBH)
3, becomes a chronic disease in 50% of the cases
4. HCV can also be transmitted through intravenous drug abuse, sexual, and household contact.
5. Hepatitis C virus is a single stranded RNA virus with some structural relations to the flavivirus family. Nucleic acid sequences of HCV cDNA clones provide the basis for the construction of recombinant peptides representing putative Hepatitis C virus proteins
6,7. HCV detection was hampered until the viral genome was cloned and recombinant HCV antigens were expressed so the serological test became practical. The utilization of multiple recombinant protein and/or synthetic peptides has made HCV antibody detection test more sensitive and specific.

The OnSite HCV Ab Plus Combo Rapid Test is the latest generation of antibody rapid test which uses recombinant structure (nucleocapsid) and non-structure proteins with the double antigen sandwich detection method, ensuring superior sensitivity and specificity.

TEST PRINCIPLE

When an adequate volume of test specimen is dispensed into the sample well of the test cassette, the specimen migrates by capillary action across the cassette. The IgG, IgM, or IgA antibodies 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, and C band is pre-coated with goat anti rabbit IgG. The band (which should exhibit a burgundy colored band of goat anti rabbit IgG/rabbit IgG-gold immunocomplex conjugates) regardless of the presence of any antibodies to HCV.

Specimens Ab positive or reactive test result are visible as a burgundy colored band on the control line (C line) which should exhibit a burgundy colored band of goat anti rabbit IgG/rabbit IgG-gold immunocomplex conjugates regardless of the presence of any antibodies to HCV. Otherwise, the test result is invalid and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

1. Each kit contains 25 or 30 test devices, each sealed in a foil pouch with three items inside:
   a. One cassette device
   b. One pipette dropper.
   c. One desiccant.
2. Sample Diluent (1 vial, 5 mL).
3. One package insert (instruction for use)

MATERIALS REQUIRED BUT NOT PROVIDED

1. Clock or Timer
2. Lancet for collection of whole blood

WARNINGS 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 test materials to room temperature (15-30°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, i.e. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test device 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 through the expiration date printed on the sealed pouch. Do not freeze the kit or expose the kit over 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 venipuncture.
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 venipuncture.
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. 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. Once the specimen is thawed, mix well prior to performing assay.

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: Label device with the specimen`s ID number.

Step 4: Fill the pipette dropper with the specimen.

Holding the dropper vertically, dispense 1 drop (about 30-40 µL) of serum/plasma or 1 drop of whole blood (about 45-55 µL) into the sample well making sure that there are no air bubbles. Immediately add 1 drop (about 35-50 µL) of Sample Diluent to the sample well.

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 Plus 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 fall outside of 2°C-30°C.
5. The temperature of the test area falls outside of 15°C-30°C.
2. **Worldwide Performance Panel**

BBI (Boston Biomedica Inc.)'s worldwide performance panel (WWHV301) were tested with the OnSite HCV Ab Plus Combo Rapid Test. The result is shown in the following table.

<table>
<thead>
<tr>
<th>Member ID</th>
<th>Origin</th>
<th>Genotype</th>
<th>Abbott EIA</th>
<th>OnSite HCV Ab Plus Combo Rapid Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>301-01</td>
<td>Argentina</td>
<td>1b</td>
<td>Positive</td>
<td>Positive</td>
</tr>
<tr>
<td>301-02</td>
<td>Argentina</td>
<td>1b</td>
<td>Positive</td>
<td>Positive</td>
</tr>
<tr>
<td>301-03</td>
<td>Argentina</td>
<td>3a/b</td>
<td>Positive</td>
<td>Positive</td>
</tr>
<tr>
<td>301-04</td>
<td>Argentina</td>
<td>2a/c</td>
<td>Positive</td>
<td>Positive</td>
</tr>
<tr>
<td>301-05</td>
<td>Argentina</td>
<td>Not tested</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>301-06</td>
<td>Uganda</td>
<td>4/c/d</td>
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<td>Positive</td>
</tr>
<tr>
<td>301-07</td>
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<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>301-08</td>
<td>Ghana</td>
<td>Not tested</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>301-09</td>
<td>China</td>
<td>1b, 2a/c</td>
<td>Positive</td>
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<td>Positive</td>
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<td>Positive</td>
<td>Positive</td>
</tr>
<tr>
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<td>1a/b, 2a/c</td>
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<td>Positive</td>
</tr>
<tr>
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<td>3a</td>
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</tr>
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<td>Positive</td>
</tr>
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<td>301-18</td>
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</tr>
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<tr>
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<td>Positive</td>
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</table>

3. **Seroconversion Panel**

BBI (Boston Biomedica Inc.)'s seroconversion panel (PHV910 –(M)) were tested with the OnSite HCV Ab Plus Combo Rapid Test. The result is shown in the following table.

<table>
<thead>
<tr>
<th>Member ID</th>
<th>Days bleeding</th>
<th>Abbott HCV EIA 2.0 s/co*</th>
<th>OnSite HCV Ab Plus Combo Rapid Test</th>
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</thead>
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<tr>
<td>910-01</td>
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<td>0.2</td>
<td>Negative</td>
</tr>
<tr>
<td>910-02</td>
<td>4</td>
<td>0.3</td>
<td>Negative</td>
</tr>
<tr>
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<td>8</td>
<td>1.3</td>
<td>Positive</td>
</tr>
<tr>
<td>910-04</td>
<td>11</td>
<td>2.9</td>
<td>Positive</td>
</tr>
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<td>910-05</td>
<td>15</td>
<td>2.4</td>
<td>Positive</td>
</tr>
</tbody>
</table>

* EIA results expressed as specimen absorbance to cut-off ratio (s/co). Ratios > 1.0 are considered reactive.