Overview

Useful For
Determining hepatitis C virus (HCV) genotype (1 to 5) to guide antiviral therapy in patients with chronic hepatitis C

Differentiating between HCV subtypes 1a and 1b

This assay should **not be used** as a screening test for HCV infection. It should be performed only on specimens obtained from patients confirmed to have HCV RNA levels in serum of 500 IU/mL or higher.

Reflex Tests

<table>
<thead>
<tr>
<th>Test ID</th>
<th>Reporting Name</th>
<th>Available Separately</th>
<th>Always Performed</th>
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</thead>
<tbody>
<tr>
<td>HCVGR</td>
<td>HCV Genotype Resolution, S</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

Testing Algorithm

The following algorithms are available in Special Instructions:

- Chronic Hepatitis C Treatment and Monitoring Algorithm: Direct Antiviral Antigen (DAA) Combination
- Hepatitis C: Testing Algorithm for Screening and Diagnosis

Special Instructions

- Hepatitis C: Testing Algorithm for Screening and Diagnosis
- Chronic Hepatitis C Treatment and Monitoring Algorithm: Direct Antiviral Agent (DAA) Combination

Method Name
Reverse Transcriptase-Polymerase Chain Reaction (RT-PCR) followed by Hybridization with Sequence-Specific, Fluorescent-Labeled Oligonucleotide Probes

NY State Available
Yes

Specimen

Specimen Type
Serum SST

Shipping Instructions
Ship specimen frozen on dry ice only. If shipment will be delayed for more than 3 days, freeze serum at -20 degrees C or colder (up to 42 days) until shipment on dry ice.

Specimen Required
Supplies: Aliquot Tube, 5 mL (T465)

Collection Container/Tube: Serum gel
**Submission Container/Tube:** Plastic vial

**Specimen Volume:** 5 mL

**Collection Instructions:**

1. Centrifuge blood collection tube per collection tube manufacturer's instructions (eg, centrifuge and aliquot within 2 hours of collection for BD Vacutainer tubes).

2. Transfer serum into aliquot tube.

**Additional Information:**

1. Specimens should contain a recommended minimum HCV viral load of 500 IU/mL.

2. Serum specimens previously submitted to other laboratories for non-microbiology tests are NOT acceptable for add-on test requests, due to possible sample-to-sample carryover from automation used for those tests.

**Forms**

If not ordering electronically, complete, print, and send a Gastroenterology and Hepatology Client Test Request (T728) with the specimen.

**Specimen Minimum Volume**

1.5 mL

**Reject Due To**

<table>
<thead>
<tr>
<th>Gross hemolysis</th>
<th>OK</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gross lipemia</td>
<td>OK</td>
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</table>

**Specimen Stability Information**

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Temperature</th>
<th>Time</th>
<th>Special Container</th>
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<tbody>
<tr>
<td>Serum SST</td>
<td>Frozen (preferred)</td>
<td>42 days</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Refrigerated</td>
<td>72 hours</td>
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**Clinical and Interpretive**

**Clinical Information**

Unique nucleotide sequences of certain regions (eg, 5’-noncoding, core, NS5b) of the hepatitis C virus (HCV) genome allow classification of HCV into 6 major genotypes or clades (1-6), based on the most recently proposed HCV genotype nomenclature. In the United States, the most commonly encountered HCV genotypes are 1a and 1b, followed by genotypes 2 and 3. Worldwide geographic distribution, disease outcome, and response to antiviral therapy differ among the genotypes. Therefore, reliable methods for genotype determination are important for proper selection of antiviral therapy and optimal patient management. Infections with HCV genotypes 2 and 3 have better therapeutic response rates (80%-90%) than genotypes 1 and 4 (40%-50%) to previous standard combination therapy (ribavirin plus pegylated interferon alpha-2a or alpha-2b). Duration of such combination therapy is 24 weeks for chronic HCV genotype 2 and 3 infections in patients who show early virologic response (>2 log or 100-fold decrease...
in HCV RNA or no detectable HCV RNA at week 12 of therapy), while patients with chronic HCV genotype 1 and 4 infections receive a minimum of 48 weeks of such combination therapy if early virologic response is achieved (undetectable HCV RNA at week 4 of therapy).

Therapeutic response rates for HCV genotype 1 infection are improved significantly (80%-90%) when oral direct acting antiviral agents (eg, daclatasvir, sofosbuvir, ledipasvir + sofosbuvir, velpatasvir + sofosbuvir, glecaprevir + pibrentasvir, elbasvir + grazoprevir, velpatasvir + voxilaprevir + sofosbuvir) are added or used in lieu of interferon-based combination therapy.

The American Association for the Study of Liver Diseases (AASLD) and Infectious Disease Society of America (IDSA) recommendations for testing, managing, and treating hepatitis C are available at www.hcvguidelines.org/full-report-view

The following algorithms are available in Special Instructions:

- Chronic Hepatitis C Treatment and Monitoring Algorithm: Direct Antiviral Antigen (DAA) Combination

- Hepatitis C: Testing Algorithm for Screening and Diagnosis

**Reference Values**

**Undetected**

**Interpretation**

An "Undetected" result indicates the absence of detectable hepatitis C virus (HCV) RNA in the specimen.

An "Indeterminate" result may be due to 1 or more of the following causes: 1) low HCV RNA level (ie,<500 IU/mL), 2) HCV genotype 6, 3) probe reactivity with multiple HCV genotypes, or 4) variation in patient's HCV target sequences with mismatches to PCR primers and/or probes. Specimens generating indeterminate results with this assay will be automatically evaluated with the subsequent test HCVGR / Hepatitis C Virus Genotype Resolution, Serum.

An HCV genotype result of "1" without a subtype result may be due to 1 or more of the following causes: 1) low HCV RNA level (ie, <500 IU/mL), 2) probe reactivity with multiple genotype 1 subtypes, 3) variation in HCV genotype 1 target sequence, or 4) misclassification of some true genotype 6 strains.

This assay is able to differentiate between HCV subtypes 1a and 1b. However, subtypes are not reported for HCV genotypes 2 to 5 due to limitations of the current genotyping assay in accurately differentiating the various subtypes of these genotypes.

Results with multiple or mixed HCV genotypes (eg, 1, 5; 1a, 2; or 3, 5) may be due to mixed genotype infection or assay probe cross-reactivity. Only those specimens with multiple or mixed genotype results containing genotype 1 but no subtype will be automatically evaluated with the subsequent test HCVGR / Hepatitis C Virus Genotype Resolution, Serum.

**Cautions**

An "Undetected" or "Indeterminate" hepatitis C virus (HCV) genotype result does not rule-out active HCV infection. Test results should be correlated with routine serologic and molecular-based testing, as well as clinical presentation. Specimens with indeterminate results will be automatically evaluated with the subsequent test HCVGR / Hepatitis C Virus Genotype Resolution, Serum.

Known cross-reactivity between the assay probes and various HCV genotypes limits the ability of this assay to identify multiple HCV genotypes present in a given specimen. Such cross-reactivity or the actual presence of multiple HCV genotypes in the same specimen may result in an "Indeterminate" or multiple/mixed genotype result.
Clinical Reference


Performance

Method Description

Sample Preparation:

The Abbott mSample Preparation System kit is used with the Abbott m2000sp, an automated sample preparation system using the magnetic microparticle processes to extract and purify viral nucleic acids from human serum specimens. An internal control is incorporated in the nucleic acid extraction and purification procedure for processing the assay controls and clinical specimens. After capture of nucleic acids onto magnetic microparticles, the microparticles are washed to remove unbound sample components. Then, the bound nucleic acids are eluted from the microparticles and the eluates are transferred to a 96-well microtiter plate containing PCR mastermix reagents for amplification and detection.

Amplification, Detection, and Genotyping:

The Abbott RealTime HCV Genotype II assay is used to amplify the 5’ noncoding (5’ NC), nonstructural 5b (NS5b), and core regions of the hepatitis C virus (HCV) genome, with several PCR primer sets that are optimized to amplify all HCV isolates. An internal control primer set is included to amplify a portion of the hydroxypyruvate reductase gene of the pumpkin plant, Cucurbita pepo. The assays positive control is an armored RNA particle diluted in negative human plasma.

During the amplification reaction, cDNA sequences are generated from the target RNA sequences by the reverse transcriptase activity of the thermostable rTth DNA polymerase. First, the HCV and internal control reverse primers anneal to their respective target sequences and are extended during a prolonged incubation period. After a denaturation step, in which the temperature of the reaction is raised above the melting point of the double-stranded cDNA:RNA product, a second primer anneals to the cDNA strand and is extended by the DNA polymerase activity of the rTth enzyme to create a double-stranded DNA product. During each round of thermal cycling, amplification products dissociate to single strands at a high temperature, allowing primer annealing and extension as the temperature is lowered. Exponential amplification of the product is achieved through repeated cycling between high and low temperatures, resulting in a billion-fold or greater amplification of target sequences. Fluorescent probes specific for HCV genotypes 1 to 5 and subtypes 1a and 1b anneal to the amplified sequence products in 4 separate reaction wells for each specimen. (Package inserts: Abbott RealTime HCV Genotype II; Abbott Molecular Inc., Des Plaines, IL, 6/2013)

PDF Report

No

Day(s) and Time(s) Test Performed

Monday through Friday; 7 a.m.-4 p.m.

Analytic Time

Monday through Thursday, 1 day; Friday, 3 days
Maximum Laboratory Time
6 days

Specimen Retention Time
2 months

Performing Laboratory Location
Rochester

Fees and Codes

Fees
- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact Customer Service.

Test Classification
This test has been cleared, approved or is exempt by the U.S. Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

CPT Code Information
87902

LOINC® Information

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<td>HCV Genotype, S</td>
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<table>
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