Hepatitis C Virus (HCV) Genotyping Reflex to NS3 Drug Resistance
Test Code: 10001

Clinical and Procedure

Clinical Utility
The HCV Genotyping with NS3 Drug Resistance assay detects NS3 (nonstructural protein 3) mutations and polymorphisms in HCV genotypes 1a and 1b that are associated with resistance to direct-acting protease inhibitors (antivirals) glecaprevir (in Mavyret®), grazoprevir (in Zepatier®), paritaprevir (in Viekira Pak/XR® and Technivie®), simeprevir (Olysio®), and voxilaprevir (in Vosevi®).
The assay is intended to be used for patients with HCV viral loads who are being screened prior to treatment with the direct-acting HCV protease antivirals, or during treatment with these antivirals when drug resistance is suspected.

About HCV
HCV infection is the most common chronic bloodborne infection in the U.S., with approximately 3.2 million people chronically infected and an additional 17,000 (approx.) new infections acquired annually. The HCV NS3 serine protease plays a critical role in viral pathogenesis and serves to cleave the viral polyprotein at several points, releasing the component viral proteins. Multiple mutations causing resistance to FDA-approved HCV protease inhibitors have been defined, and the levels of increased resistance for each mutation have been calculated using HCV replicons or reporter constructs. Some of the mutations cause resistance to multiple protease inhibitors. Knowledge of these mutations is important in patients since an incomplete suppression of viral replication by an ineffective drug combination could prevent a sustained viral response (“cure”), and could readily support the development of antiviral drug resistance. In addition to detection of drug resistance during treatment, a screen for the protease Q80K variant in subtypes 1a is recommended prior to the use of simeprevir. The manufacturer’s package insert for the protease inhibitor simeprevir (Olysio®) states as follows: “Screening patients with HCV genotype 1a infection for the presence of virus with the NS3 Q80K polymorphism at baseline is strongly recommended. Alternative therapy should be considered for patients infected with HCV genotype 1a containing the Q80K polymorphism.”

Procedure
HCV Genotyping Reflex to NS3 Drug Resistance assay utilizes RT-PCR amplification with primers in highly conserved viral genomic regions to amplify HCV genotypes 1a and 1b. The fragments are purified and sequenced using sequencing primers from conserved regions of the fragments. The sequence is compared to a database of mutations associated with antiviral resistance. This test has not been cleared or approved for diagnostic use by the U.S. Food and Drug Administration.

Turnaround Time
4-11 business days from receipt of specimen.

Specimen Information

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Order Code</th>
<th>CPT Code</th>
<th>NY Approved</th>
<th>Volume</th>
<th>Assay Range</th>
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</thead>
<tbody>
<tr>
<td>plasma</td>
<td>10001</td>
<td>87902</td>
<td>Yes</td>
<td>2 mL (min. 1mL, min. viral load on 1000 IU/mL)</td>
<td>Resistant/None Detected</td>
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Special Instructions
- Collect 4-5 mL whole blood in EDTA, ACD or PPT, centrifuge and transfer 2 mL plasma to a sterile, screw top tube.
- If the specimen was collected in PPT, the entire tube can be shipped following centrifugation.
- Ship frozen in dry ice Monday through Friday.
Mutations in the NS3 gene will be reported as Resistant/None Detected. Interpretation of gene mutations and association with antiviral resistance, including glecaprevir (in Mavyret®), grazoprevir (in Zepatier®), paritaprevir (in Viekira Pak/XR® and Technivie®), simeprevir (Olysio®), and voxilaprevir (in Vosevi®) will be provided with the report. See HCV NS3 Reportable Mutations in the test information navigation. Additionally the presence or absence of mutation Q80K is reported individually.

Shipping
Ship Monday through Friday on dry ice. Friday shipments must be labeled for Saturday delivery. All specimens must be labeled with patient's name and collection date. A Viracor Eurofins test requisition form must accompany each specimen. Multiple tests can be run on one specimen. Ship specimens FedEx Priority Overnight® to: Viracor Eurofins, 1001 NW Technology Dr., Lee's Summit, MO 64086.

Causes for Rejection
HCV RNA concentrations too low to allow antiviral resistance testing (see above for minimum volume and viral load), subtypes other than HCV 1a or 1b, whole blood frozen, specimens beyond their acceptable length of time from collection as listed in the specimen handling, or specimen types other than those listed.

Disclaimer
Specimens are approved for testing in New York only when indicated in the Specimen Information field above. The CPT codes provided are based on Viracor Eurofins’ interpretation of the American Medical Association's Current Procedural Terminology (CPT) codes and are provided for informational purposes only. CPT coding is the sole responsibility of the billing party. Questions regarding coding should be addressed to your local Medicare carrier. Viracor Eurofins assumes no responsibility for billing errors due to reliance on the CPT codes illustrated in this material.

References

2 OLYSIO™ (simeprevir) capsules, for oral use, FULL PRESCRIBING INFORMATION Viekira