Hepatitis C Virus Therapy Molecular Testing

Indications for Ordering

Predict response to peginterferon (PEG-IFNα)/ribavirin (RBV) therapy for chronic hepatitis C virus genotype 1 (HCV-1) infection

Test Description

- Polymerase chain reaction (PCR) followed by single nucleotide extension (SNE) and capillary electrophoresis
- Variants tested – single nucleotide polymorphisms (SNPs) rs12979860 C/T and rs8099917 T/G near IL28B

Tests to Consider

Typical testing strategy

- Chemiluminescent immunoassay (CIA) or enzyme-linked immunosorbent assay (ELISA) for initial diagnosis of HCV
- Quantitative RNA-PCR to determine whether individual is currently infected
- Genotyping for chronic HCV to determine if HCV-1 genotype present (likely to respond well to treatment)
- IL28B gene testing for treatment decisions

Primary tests

Interleukin 28 B (IL28B)-Associated Variants, 2 SNPs 2004680
- Detect DNA variants associated with response to therapy

Related tests

Hepatitis C Virus (HCV) by Quantitative PCR with Reflex to HCV Genotype by Sequencing 2002685
- Preferred reflex test to confirm active HCV infection following positive HCV screen
- Reflex to genotype aids in prognosis and treatment selection

Hepatitis C Virus (HCV) by Quantitative PCR with Reflex to HCV High-Resolution Genotype by Sequencing 2010793
- Confirm active HCV infection following positive HCV screen when a higher level of subtype resolution is required

Hepatitis C Virus by Quantitative PCR 0098268
- Preferred single test to confirm active HCV infection following positive HCV antibody screen
- Order only after positive HCV screen
- Use to monitor therapy

Hepatitis C Virus Genotype by Sequencing 0055593
- Preferred genotyping test for prognosis and treatment selection
- Do not order prior to molecular confirmation of positive HCV screen
- Assay does not differentiate between type 1a and type 1b

Hepatitis C Virus High-Resolution Genotype by Sequencing 2006898
- Order before initiating HCV therapy to aid in prognosis and therapy selection when a higher level of subtype resolution is required (ie, non 6a/b vs. type 1 and type 1a vs. 1b)
- Do not order prior to molecular confirmation of positive HCV screen

Hepatitis C Virus (HCV) Genotype with Reflex to HCV High-Resolution Genotype by Sequencing 2009255
- Reflex genotyping panel for prognosis and treatment selection when a higher level of subtype resolution is required
- Differentiates between type 1a and type 1b

Disease Overview

Prevalence

Persistent HCV infection
- ~180 million cases worldwide (~3% of population)
- ~4.1 million cases in U.S. (~1.6% of population)
- Estimated favorable allele frequencies

<table>
<thead>
<tr>
<th>Ethnicity</th>
<th>rs12979860 C/T</th>
<th>rs8099917 T/G</th>
</tr>
</thead>
<tbody>
<tr>
<td>African American</td>
<td>0.50</td>
<td>Unknown</td>
</tr>
<tr>
<td>Asian</td>
<td>0.90</td>
<td>0.88</td>
</tr>
<tr>
<td>Caucasian</td>
<td>0.75</td>
<td>0.75</td>
</tr>
<tr>
<td>Hispanic</td>
<td>0.70</td>
<td>Unknown</td>
</tr>
</tbody>
</table>

Physiology

- IL28B gene encodes for lambda or type III interferons (IFN-λ)
- IFN-λ postulated to interact with cellular transmembrane receptor to upregulate the JAK-STAT pathway
- Results in antiviral activity
- SNPs rs12979860 and rs8099917 are located upstream of IL28B gene
- Associated with spontaneous clearance and response to PEG-IFNα/RBV therapy in Caucasians with chronic HCV-1 infection
**Treatment issues**

- Acute HCV infection often leads to chronic disease
  - Therapy to treat disease depends on numerous clinical factors and HCV genotype
- At least 6 major HCV genotypes
  - HCV-1 accounts for 75% of U.S. cases
  - Genotyping helps to predict therapeutic response
- Current recommended therapy for chronic HCV infection
  - PEG-IFNα and RBV combination therapy
    - Eliminates HCV RNA in 40-50% of individuals with HCV-1 and 70-90% of those with HCV-2 or -3
  - Triple therapy (PEG-IFNα/RBV with protease inhibitors)
    - Anticipated to eliminate HCV RNA in ~75% of individuals with HCV-1
- Pretherapeutic identification of factors predicting response is helpful due to
  - High cost of therapy
  - 10-15% discontinuation rate due to adverse side effects of therapy

**Genetics**

**Gene** – *IL28B*
- SNPs rs12979860 C/T and rs8099917 T/G

**Structure/function** – see table below

**Test Interpretation**

**Sensitivity/specificity**
- Clinical sensitivity/specificity – unknown
- Analytical sensitivity/specificity – 99% for SNPs detected

**Results**

See table below

**Limitations**

- SNPs other than those targeted will not be detected
- Usefulness of *IL28B*-associated SNPs for predicting therapy response for HCV genotypes other than HCV-1 is unknown
- Lack of favorable genetic factors should not be used to deny therapy
- Other gene variants and nongenetic factors that may affect response to HCV therapy are not detected
- Diagnostic errors can occur due to rare sequence variations

<table>
<thead>
<tr>
<th><strong>IL28B Genotype Interpretation</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Genotype</strong></td>
</tr>
<tr>
<td>rs12979860 C/C</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>rs8099917 T/T</td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>rs12979860 C/T</td>
</tr>
<tr>
<td>rs12979860 T/T</td>
</tr>
<tr>
<td>rs8099917 T/G</td>
</tr>
<tr>
<td>rs8099917 G/G</td>
</tr>
<tr>
<td>One favorable SNP and one not favorable SNP identified</td>
</tr>
<tr>
<td></td>
</tr>
</tbody>
</table>