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
- Do not order prior to molecular confirmation of positive HCV screen
- 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

Ethnicity	rs12979860 C/T	rs8099917 T/G
African American	0.50	Unknown
Asian	0.90	0.88
Caucasian	0.75	0.75
Hispanic	0.70	Unknown

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-INFα/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

***IL28B* Genotype Interpretation**	
Genotype	**Interpretation**
• rs12979860 C/C	**Favorable** • 2- to 3-fold greater rate of sustained virological response (SVR) following PEG-IFNα/RBV therapy • 3-fold increase in natural clearance of HCV
• rs8099917 T/T	**Favorable** • Higher rate of SVR following PEG-IFNα/RBV therapy • Increased natural clearance of HCV
• rs12979860 C/T • rs12979860 T/T • rs8099917 T/G • rs8099917 G/G	**Not favorable** • Less likely to respond to treatment and achieve SVR
One favorable SNP and one not favorable SNP identified	**Indeterminate** • Likelihood of SVR following PEG-IFNα/RBV therapy is not well defined