

Hepatitis C Resistance Testing at Quest Diagnostics

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November 17, 2015



HCV Genotypic Drug Resistance Tests I

Technology: reverse transcription, PCR amplification and Sanger sequencing

- HCV Genotype 1 NS3 Protease (1a: ~84%)
 - Resistance predictions: BOC, TVR → +SMV (2013) → BOC, PTV, SMV (2015, current) → GZR, PTV, SMV (2016, TBD)
 - Q80K: ~45% of genotype 1a
 - 2015 SMV resistance mutations (R155K, D168E/V, S122K/R): ~1.5%

More NS3 data to be presented at HepDart, Dec 6-10, 2015

HCV Genotypic Drug Resistance Tests II

Technology: reverse transcription, PCR amplification and Sanger sequencing

- Genotype 1 NS5a (1a: ~79%)
 - Resistance predicted: DCV, LDV, OBV → +elbasvir in 2016
 - Major resistance-associated codons (28, 30, 31, 58, 93)
 - Y93C/H/N: 14% 1a, 39% 1b

More NS5a data to be presented at HepDart, Dec 6-10, 2015

- Genotype 3a NS5a (DCV resistance)
 - Validated, coming soon!
 - Y93H detected in 4/30 (13%) randomly selected genotype 3a patient samples
- Genotype 1 NS5b Polymerase (1a: ~78%)
 - Resistance predicted: SOF (S282T)
 - S282T detected in 7 patient samples (~1.4%; 6/7 1a)
 - S282C found in 1 sample (1b)

Case Study: Consult for HCV Resistance Testing

Patient: 63 yr old man, genotype 1a, cirrhotic, failed therapy on SOF+LDV

- Tests ordered: NS3 and NS5a resistance tests
- Test Results:
 - NS5a: Y93N
 - Likely resistant to all three approved NS5a inhibitors
 - NS3: genotype 1a and Q80K polymorphism
 - SMV is not recommended for cirrhotic patients infected with genotype 1a and Q80K
- Outcome:
 - Clinician stated that he will defer retreatment until he can get the patient into a clinical trial setting
 - Hcvguidelines.org: “For patients who have both NS3 and NS5A inhibitor RAVs detected, retreatment should be conducted in a clinical trial setting, as an appropriate treatment regimen cannot be recommended at this time” (August 20, 2015).

Perspective on Hepatitis C Resistance Testing – When and What to Test

Treatment-naïve patients*

- NS3 resistance test (Q80K) if genotype 1a, cirrhotic and considering simeprevir-based regimen
- NS5a genotype 3 resistance test (Y93H) if genotype 3 and considering a daclatasvir-based regimen
- New DAAs perform resistance testing if warranted (e.g: grazoprevir + elbasvir for genotype 1a NS5a)

Treatment-failure patients*

- NS3 and NS5a resistance testing to guide retreatment decisions

* Consult <http://hcvguidelines.org/> for the latest recommendations

HCV Resistance Testing Challenges and Open Questions

- Clinical interpretation of resistance mutations
 - Interpretation (“resistance predicted”/“not predicted”) differs according to the clinical context of the patient
 - Baseline NS5a mutations in treatment-naïve genotype 1 patients vs treatment-failure emergent mutations
 - NS3 Q80K in genotype 1a cirrhotic vs non-cirrhotic patients
 - Potency of DAA in regimen
- Mixed genotype infections
 - Approx 2-4% of patients are dually infected (e.g: 1a+2b, 1a+3a, 1a+1b)
 - Bulk sequencing may only give a partial answer
- Minority variants
 - NGS enables detection of minority resistant variants but unclear what are the cutoffs (20%? 5%? 1%?).