Hepatitis C – Monitoring and Complications (and Treatment!)

Dr Mark Douglas
Hepatitis C Virus

- Positive single strand RNA virus
- *Flaviviridae* family, *Hepacivirus* genus
- 9.6 kbp genome
- ~3000 amino acid polyprotein
- 60 nm enveloped virions
- Lipo-viral particles
- Until recently, unable to grow in cell culture

Shimizu et al., 1996
Features of the HCV Genome

- **Structural proteins**
  - Core (capsid)
  - E1 and E2 (envelope glycoproteins)

- **Non-structural proteins**
  - P7
  - NS4A
  - NS2
  - NS3
  - NS4B
  - NS5A
  - NS5B

**Cellular proteases**

**Viral proteases**
HCV Proteins - ER Membrane Associations

Moradpour et al, 2007
<table>
<thead>
<tr>
<th>Protein</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Core</td>
<td>Highly basic core protein</td>
</tr>
<tr>
<td>E1*gp31</td>
<td>Envelope glycoprotein, heterodimer in virion</td>
</tr>
<tr>
<td>E2* gp70</td>
<td>Envelope glycoprotein, heterodimer in virion</td>
</tr>
<tr>
<td>p7</td>
<td>Cleaved from E2, behaves as ion channel</td>
</tr>
<tr>
<td>NS2</td>
<td>Serine protease (NS2/3)</td>
</tr>
<tr>
<td>NS3</td>
<td>Serine protease, helicase and NTPase activities</td>
</tr>
<tr>
<td>NS4A</td>
<td>Cofactor essential for NS3 serine protease activity</td>
</tr>
<tr>
<td>NS4B</td>
<td>Induces a membranous replication complex at the ER</td>
</tr>
<tr>
<td>NS5A</td>
<td>Serine phosphoprotein, interacts with cell pathways</td>
</tr>
<tr>
<td>NS5B</td>
<td>RNA Polymerase – (RNA dependent)</td>
</tr>
</tbody>
</table>
HCV – Life Cycle

- Cytoplasmic replication
- Replication complexes at ER
- Membranous web induced by NS4B
- Interactions with lipid metabolism
- Some export via VLDL pathway

Moradpour et al, 2007
Hepatitis C - Epidemiology

- 3% prevalence worldwide (1% Australia, USA, UK)
- Causes more deaths than HIV in the USA, Australia
- Main cause of cirrhosis requiring liver transplant: Australia, USA, UK
- 3 to 4 million new cases per year (most IDU here)

Estimated Total Chronic HCV Infections Worldwide:

Global Chronic Hepatitis C Infection (Millions)

- North America 4.0
- Europe 7.0
- Asia Pacific 80
- Latin America 7.0
- Africa and Middle East 72

Global Distribution of HCV Genotypes

Genotypes 1-3 Worldwide
Genotypes 4 & 5 Africa
Genotype 6 Asia

Webster et al., 2000
Hepatitis C - Transmission

● **Source**
  - human blood
  - blood contaminated body fluids
  - ?infected lymphocytes

● **Route**
  - inoculation
  - Sexual transmission rare, but reported (well described in HIV +ve MSM)
  - vertical transmission 5% (at birth)
Typical Course of HCV Infection

- **Minor (anicteric) acute illness**
  - 20-40% virus clearance
  - 60-80% chronic viral replication

- **Chronic HCV**
  - 20-30% no evidence of disease
  - 60-80% chronically elevated LFTs
  - 3-9% develop cirrhosis over 20-30y
  - HCC 1-3% per year once cirrhotic
Metabolic Complications of HCV

- Steatosis – HCV Genotype 3
- Insulin resistance – HCV Genotype 1
  - Predicts progression, poor response to interferon-based treatment
- Mechanisms are poorly understood
  - effects of HCV core on metabolic pathways?
  - chronic inflammation?
Monitoring HCV

● Infection
  – Anti-HCV antibodies, antigen (ELISA)
  – Detect HCV RNA by RT-PCR
  – Quantitative PCR, genotype

● Disease
  – LFTs (especially ALT)
  – Liver biopsy (Fibroscan)
  – HCC screen (ultrasound, AFP)
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Serum Markers of HCV

HEPATITIS C

- viraemia
- anti-HCV

ALT

CHRONIC COURSE (60%)

SELF LIMITING COURSE (40%)

EXPOSURE ONSET months
Ranges of Linear Quantification of HCV RNA Assays (IU/ml)

10  10^2  10^3  10^4  10^5  10^6  10^7  10^8

Untreated hepatitis C
Ranges of Linear Quantification of HCV RNA Assays (IU/ml)

- Cobas Amplicor HCV Monitor v2.0
- Versant HCV RNA 3.0 (bDNA)

Untreated hepatitis C
Ranges of Linear Quantification of HCV RNA Assays (IU/ml)

- Cobas Amplicor HCV Monitor v2.0
- Versant HCV RNA 3.0 (bDNA)
- Cobas TaqMan HCV Test (Roche)
- Abbott Real-Time HCV Assay (Abbott)

Untreated hepatitis C

- 50 IU/ml
Roche Real-Time PCR

Cobas Ampliprep®

Cobas Taqman®

Cobas Ampliprep®/Cobas Taqman® (CAP/CTM) platform
Real-Time PCR Quantification

- Automation
  - sample preparation
  - PCR cycling, nucleic acid quantification
- Improved sensitivity
- No carryover contamination
- Extended dynamic range of quantification
- Precision and reproducibility
That sounds great!

... but is there a catch?
Cost!! (Medicare restrictions)

- **HCV PCR (qualitative)**
  - $92.80
  - 4 × per year on treatment (or 1 × per year)

- **HCV PCR (quantitative – viral load)**
  - $181.45
  - 2 × per year

- **HCV Genotype**
  - $206.20
  - 1 × per year
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Liver Biopsy
Biopsy - Metavir Score

Activity
- A0 = no activity
- A1 = mild activity
- A2 = moderate activity
- A3 = severe activity

Fibrosis
- F0 = no fibrosis
- F1 = portal fibrosis without septa
- F2 = few septa
- F3 = numerous septa without cirrhosis
- F4 = cirrhosis
Fibroscan - Transient Elastography

INNOVATION in liver disease management
Fibroscan - correlation

Gut 2006;55:403–408
Fibroscan – predictive value

Table 2  Cut off values of liver stiffness according to fibrosis stage for a positive predictive value of at least 90%

<table>
<thead>
<tr>
<th></th>
<th>Moderate (F≥2)</th>
<th>Severe (F≥3)</th>
<th>Cirrhosis (F=4)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cut off</td>
<td>7.2</td>
<td>12.5</td>
<td>17.6</td>
</tr>
<tr>
<td>PPV</td>
<td>90</td>
<td>90</td>
<td>91</td>
</tr>
<tr>
<td>NPV</td>
<td>52</td>
<td>80</td>
<td>92</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>64</td>
<td>65</td>
<td>77</td>
</tr>
<tr>
<td>Specificity</td>
<td>85</td>
<td>95</td>
<td>97</td>
</tr>
<tr>
<td>PLR</td>
<td>4.2</td>
<td>13.7</td>
<td>28.4</td>
</tr>
</tbody>
</table>

PPV, positive predictive value; NPV, negative predictive value; PLR, positive likelihood ratio.
HCV - Treatment

- Interferon (pegylated), ribavirin
- “Cure” - Sustained Virological Response
- Treatment & outcome depend on genotype
- Genotypes 1 (4, 5, 6)
  - 48 weeks treatment
  - 50% SVR
- Genotypes 2, 3
  - 24 weeks treatment
  - 80% SVR
Hepatitis C Treatment Outcomes

Peginterferon and Ribavirin

Non-Response
Relapse

Undetectable
SVR

HCV RNA (Log IU/mL)

-8 -4 -2 0 4 8 12 16 20 24 32 40 48 52 60 72

Weeks After Start of Therapy
Factors associated with SVR

- Genotype 2 or 3 (not 1, 4)
- Lower HCV viral load
- Milder degree of fibrosis (F0-F1 vs. F3-4)
- Lower body weight / Insulin resistance
- Younger age
- Adherence to treatment
- Female
- Asian > Caucasian > African
Evolution of SVR rates

Patients achieving SVR (%)

<table>
<thead>
<tr>
<th>Weeks</th>
<th>Peg-IFN</th>
<th>IFN + ribavirin</th>
<th>Peg-IFN + ribavirin</th>
</tr>
</thead>
<tbody>
<tr>
<td>24</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>48</td>
<td>6-19</td>
<td>35-43</td>
<td>76-82</td>
</tr>
<tr>
<td>78</td>
<td>10-22</td>
<td>33-36</td>
<td>42-46</td>
</tr>
<tr>
<td>11-19</td>
<td>18-39</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10-22</td>
<td></td>
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<td></td>
</tr>
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*Range of values reported; lower bar represents lower value

Manns, Foster et al., Nature Reviews Drug Discovery 2007
Genome-wide association studies

Treatment response

- Genetic variation in IL28B is associated with response to PEG-IFN and ribavirin treatment
  - SNP rs12979860
  - SNP rs8099917
    ▸ Suppiah V, et al. Nat Genet 2009; 41:1100-04 (Australia)
    ▸ Rauch A, et al. Gastroenterology 2010: January 7 (Switzerland)
Genetic variation in IL28B and HCV response

IL28B encodes IFN-λ (cytokine with antiviral activity)
Association of rs12979860 with SVR

Favourable IL28B polymorphism is common in Asia

Evolution of SVR rates

Patients achieving SVR (%)

<table>
<thead>
<tr>
<th>Peg-IFN</th>
<th>Peg-IFN + ribavirin</th>
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<tbody>
<tr>
<td>61-79</td>
<td>76-82</td>
</tr>
<tr>
<td>42-46</td>
<td>G1</td>
</tr>
</tbody>
</table>

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Manns, Foster et al., Nature Reviews Drug Discovery 2007
Potential New Drug Targets

Direct Acting Antivirals (DAAs)

- **NS3/4A protease inhibitors**
  - telaprevir, boceprevir

- **NS5B polymerase inhibitors**
  - Nucleoside analogues
  - Non-nucleoside analogues

- **NS5A inhibitors**

Phase 3 Studies (NEJM, 2011)

- Boceprevir untreated (SPRINT-2)
- Telaprevir untreated (ADVANCE)
- Boceprevir prior relapse (RESPOND-2)
- Telaprevir prior relapse (REALIZE)
- Boceprevir prior non-response (RESPOND-2)
- Telaprevir prior non-response (REALIZE)

PEG-IFN + RBV vs. DAA added
Conclusions – Hepatitis C

- Hepatitis C is now a curable disease
  - Screen high risk patients and refer for Rx
- HCV PCR is key marker of active infection
- 70-80% SVR gt 2/3 with PEG-IFN/RBV
- 45% SVR gt 1 with PEG-IFN/RBV (30-70% IL28B)
- 70-80% SVR gt 1 when adding DAA
- IFN-free treatment likely to become a reality in the next 5 years
- 90% SVR with 12 weeks of IFN-free oral treatment may soon be possible