Session 1: Educational Session on HCV and HIV

HCV related cancerogenesis

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Hepatitis C is one of the most pressing health emergencies worldwide

The global prevalence of HCV infection has been estimated at 2-3%, which equates to 130-170 million people

>350,000 mortality cases each year for HCV chronic disease related
HCV discovery: one of the most significant biomedical breakthroughs in the last 25 years

This discovery has facilitated the development of effective diagnostics, blood screening tests and the elucidation of promising drug and vaccine targets to control this global pathogen and save the lives of millions of people around the world....
HCV  Flavivirus (genus epacivirus)  Identified in 1989 (nonA-nonB) cloned and sequenced.

The origin of the primate Flaviviridae could be as ancient as the differentiation of primate species some 35 million years ago. HCV could have been coevolving with human populations during their migration out of Africa within the past 100,000 to 150,000 years, but the current HCV genotypes appeared much more recently. A study suggested that types 6 and 4 could have originated 700 years and 350 years ago, respectively, whereas subtypes 1a and 1b could have arisen less than 100 years ago.

Pybus et al Science 2001
Chronic hepatitis associated with HBV and HCV infection is the major risk factor for the development of HCC, being involved in more than 80% of cases of HCC worldwide.
Epidemiology of HCC worldwide

• Fifth most common cancer worldwide

• Third leading cause of cancer-related death worldwide

• Each year, hepatocellular carcinoma (HCC) is diagnosed in more than half a million people worldwide
Incidence of liver cancer worldwide: regional variation

Incidence of liver cancer in different regions of the world (2002)

More frequent in men than in women

Region
- Eastern Asia
- Middle Africa
- Eastern Africa
- South-Eastern Asia
- World
- Western Africa
- Southern Europe
- Caribbean
- Southern Africa
- Western Europe
- Eastern Europe
- Northern America
- Central America
- Western Asia
- Northern Africa
- Australia/New Zealand
- South America
- Northern Europe
- South Central Asia

Incidence rate per 100,000 population*

0 5 10 15 20 25 30 35 40

*Age standardised

GLOBOCAN 2002 database. Available at http://www-dep.iarc.fr/
HCC: a Complex Pathogenesis

Hepatocarcinogenesis is a slow heterogeneous process involving genomic changes progressively altering the hepatocellular phenotype.

During the long preneoplastic stage hepatocyte cycling is accelerated by upregulation of mitogenic pathways, partly through epigenetic changes, producing monoclonal populations of aberrant and dysplastic hepatocytes (often with telomere erosion, microsatellite instability and structural aberrations in genes and chromosomes).

Thorgheirsson SS, Grisham JW. Nature Gen 2002; 31: 339-346; Park YN, Roncalli M. J Hepatol 2006; 45: 734-743
HCC: a Complex Pathogenesis

Dysplastic nodules and HCCs are associated with the accumulation of irreversible structural alterations in genes and chromosomes.

The malignant hepatocyte phenotype may result from the disruption of a number of genes that function in different regulatory pathways, leading to several molecular variants of HCC.

Thorgheimsson SS, Grisham JW. Nature Gen 2002; 31: 339-346; Park YN, Roncalli M. J Hepatol 2006; 45: 734-743
Cirrhosis is the key Risk Factor for HCC

Lok et al. Gastroenterology. 2009
HBV-related risk for HCC

Most HBV-infected patients with HCC have necro-inflammation leading to cirrhosis

But, up to one third of patients with HBV-related HCC do not have cirrhosis!! (The only liver disease!)

HBV DNA may exerts carcinogenic potential by inserting in or near proto-oncogenes or tumor suppressing genes

The risk of HCC is further increased by:

- Male gender
- High viral load
- Alcohol or tobacco use
- Aflatoxin exposure
- HBV or HCV coinfections
**HCV-related risk for HCC**

Virtually all HCV-infected patients with HCC have necro-inflammation leading to cirrhosis!

Persistent necro-inflammation may be carcinogenic by shifting TGF-beta signals from tumor suppression to fibrogenesis, by oncogenicity of core protein, by unbalacing cell-cycle control, or ..........

The risk of developing HCC varies from 1 to 4% per year

The risk of HCC is further increased by:

- Male gender
- Older age at infection (leading to faster disease progression)
- Heavy and long-term alcohol consumption
- Possibly diabetes and obesity
- HBV or HIV coinfections
- Risk reduced by coffee drinking
KEY POINTS

• Cirrhosis patients with chronic HBV and HCV infection is not a prerequisite step for hepatic tumorigenesis.
• The role of HCV and HBV in promoting hepatocellular carcinoma (HCC) development by either direct or indirect effects is still speculative, yet there is compelling evidence that both mechanisms exist.
• Vaccination plays a central role in the prevention of HBV-related HCC.
• Current antiviral therapies for HBV and HCV, if successful, can reduce but not completely eliminate the risk of HCC.
• The introduction of the new HCV direct-acting antiviral agents has not been in practice long enough to permit an estimate of their likelihood of reducing HCC incidence.
HBV & Carcinogenesis

It is believed that the following phenomena:

• High protein X expression
• Integration of viral DNA in a particular position of the host genome
• Active cell division in relation to repair processes
• Inflammatory alterations

are globally responsible for the transformation tumor associated with HBV infection.
The integration of hepadnavirus DNA is presumably by cellular mechanisms. Usually takes place during chronic infection. In the vast majority of cases of hepatocarcinoma, the viral genome is integrated into the DNA of transformed cells. The provirus has deletions, inversions, duplications and other mutations, promoting genetic instability of the cell.

The gene X is retained in the integration process and its product HBX is associated with the protein p53, inactivating its function.

Fig. 1. Hepatitis B viral proteins involved in hepatocarcinogenesis through interaction with specific cellular proteins.
Pre S mutations and stop codons in the HBsAg can induce an oxidative stress thus favoring the neoplastic transformation of the hepatocytes

Pollicino et al., Hepatology 2014
The long-term persistence of HCV infection is unique among RNA viruses that replicate without a DNA form.

- Unlike DNA viruses or retroviruses that are classically associated with latency no episomial or integrated form of HCV has been demonstrated.
- HCV replication occurs only in cytoplasm.

Pereira A A and Jacobson I M
Nat Rev Gastroenterol Hepatol, 2009
Natural history

Female sex, young age at the time of infection >30 years

Acute
- 70-80%

Chronic
- 20-30%

Cirrhosis
- 15%

Hepatic incompetence
- 15%

HCC 1-5%/year

YEAR
- 10
- 20
- 30

FAST alcohol, coinfections (HBV, HIV)

<20 years
HCV & Carcinogenesis

It is believed that the following phenomena:

• Active cell division in relation to repair processes
• Inflammatory alterations
• Direct role of HCV proteins
• miRNA

are globally responsible for the transformation tumor associated with HCV infection.
A major role in developing HCC with HCV infection is played by the core protein.
Figure 1 | A non-exhaustive list of extrahepatic manifestations that have been associated with HCV infection.
The replication of HCV in the extrahepatic organs and, especially, lymphoid cells, might affect the pathogenesis of extrahepatic diseases with HCV infection. HCV persistent infection can cause malignant lymphoma.
HIV, HBV and HCV share several biological similarities, but …

<table>
<thead>
<tr>
<th></th>
<th>HIV</th>
<th>HBV</th>
<th>HCV</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Virus</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>daily production of virions per day</td>
<td>$10^{10}$</td>
<td>$10^{12} - 10^{13}$</td>
<td>$10^{12}$</td>
</tr>
<tr>
<td>half-life of free virions (h)</td>
<td>1</td>
<td>3–24</td>
<td>2–3</td>
</tr>
<tr>
<td>half-life of intracellular virions</td>
<td>days (dependent on infected cells $t_{1/2}$)</td>
<td>months (dependent on infected cells $t_{1/2}$)</td>
<td>hours (not dependent on infected cells $t_{1/2}$)</td>
</tr>
<tr>
<td>mutation rate</td>
<td>very high</td>
<td>high</td>
<td>very high</td>
</tr>
<tr>
<td>constraints due to ORFs in targeted viral enzymes</td>
<td>moderate</td>
<td>high</td>
<td>none</td>
</tr>
<tr>
<td>immune-mediated escape mutants</td>
<td>frequent</td>
<td>infrequent</td>
<td>frequent</td>
</tr>
<tr>
<td><strong>Target cells</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>half-life of infected cells</td>
<td>days</td>
<td>months</td>
<td>weeks</td>
</tr>
<tr>
<td>size of susceptible cells compartment</td>
<td>large</td>
<td>small</td>
<td>probably large</td>
</tr>
<tr>
<td>intracellular viral reservoir</td>
<td>yes (integrated cDNA)</td>
<td>yes (cccDNA)</td>
<td>no</td>
</tr>
</tbody>
</table>

ORFs, open reading frames; cDNA, complementary DNA; cccDNA, covalently closed circular DNA.

_Soriano et al., JAC 2008_
HIV, HBV and HCV share several biological similarities, but …

Differently from HIV and HBV:
• HCV replication occurs only in cytoplasm
• Viral genome is not archived into the genome of infected cells

… This makes HCV curable!!!!

Moradpour D et al., Nature 2007
The increasing burden of mortality from viral hepatitis in USA in the last years

**Figure.** Annual age-adjusted mortality rates from hepatitis B and hepatitis C virus and HIV infections listed as causes of death in the United States between 1999 and 2007.
Estimated number of individuals with HIV, HBV and HCV worldwide

Of the 35 million people living with HIV worldwide, around 20% (~7 million) had chronic hepatitis C and 3 million are chronically infected with HBV.
HIV/HCV coinfection leads to accelerated hepatic fibrosis progression, with higher rates of cirrhosis, liver failure and liver death than does HCV mono-infection.
HIV/HCV coinfection: disease progression and complication

Fig. 1 Pathogenesis of HIV/HCV co-infection: Immune activation and dysregulation, effects on HIV and HCV disease progression, and complications in multiple organ systems.

Operskalski & Kovacs, Curr. HIV/AIDS Rep 2011
Trends in the prevalence of cirrhosis, decompensated cirrhosis, HCC and mortality in 24,040 HIV–infected veterans during period 1996-06 presented according to HCV status

Ioannou V et al HEPATOLOGY 2013
Trends in the prevalence of cirrhosis, decompensated cirrhosis, HCC and mortality in 24,040 HIV–infected veterans during period 1996-06 presented according to HCV status

The incidence of HCC in HIV-infected patients has also increased over time despite improvements to HIV treatment regimens, and in 2009 was estimated at 30 per 100,000 individuals in the AIDS population of the United States
Fig. 1. Mechanisms involved in hepatocarcinogenesis in HIV/hepatitis B virus or hepatitis C virus-coinfected patients. (1) The immunodeficiency associated with HIV infection accelerates the course of HCV and HBV due to a quantitative loss of CD4\(^+\) and CD8\(^+\) T cells and alteration of IFN-\(\gamma\) responses, dendritic and NK cells. (2) Inflammatory mediators upregulate pro-inflammatory and pro-fibrotic cytokines and contribute to accelerating fibrosis by activation of HSC. (3) The HIV gp120 not only induces the apoptosis of hepatocytes through CXCR4 but also fibrosis through binding HSC on its receptors CXCR4 and CCR5. (4) Several other factors due to metabolic syndrome may lead to more severe liver disease. (5) At term, all these factors may accelerate the progression to HCC, and the presence of Tat-induced liver cell dysplasia enhances the genetic alterations that accompany liver regeneration after the necrotic activity.
Transcripts for the chemokine receptors CCR5 and CXCR4, which bind gp120, were detectable in human hepatic stellate cells.

Expression of the HIV co-receptors, CCR5 and CXCR4, in human hepatic stellate cells (HSCs). Total RNA was isolated from two primary HSC lines, the stable human HSC line, LX-2, and from human bone marrow (BM), as indicated. Expression of CCR5 and CXCR4 was assessed by real-time PCR. The scales on the right side refer to data obtained with bone marrow RNA.
CCR5 depletion reduces HCC incidence in the knockout (Mdr2-KO) mice

DKO mice for Mdr2 and CCR5 exhibited a significant decrease in tumor incidence and size
The goal of the current HCV treatment is not the suppression of HCV replication and viremia but the eradication of infection.

This is so far possible to a larger number of people thanks to the next introduction of new anti-HCV drugs.
EASL: Indications for treatment: who should be treated?

All treatment-naïve and treatment-experienced patients with compensated or decompensated chronic liver disease due to HCV should be considered for therapy (recommendation A1)

- **META VIR F3–F4**: Prioritise treatment
- **META VIR F2**: Treatment is justified
- **META VIR F0–F1**: Individualise treatment
- ** Decompensated cirrhosis**: Urgently treated IFN-free therapy

**Recommendations**
- A1
- A2
- B1
- A1
EASL: Indications for treatment: who should be treated?

“Treatment should be prioritized regardless of the fibrosis stage in patients with HIV or HBV coinfection, patients in the pre- or post-liver transplant setting, patients with clinically significant extra-hepatic manifestations (e.g. symptomatic vasculitis associated with HCV-related mixed cryoglobulinaemia, HCV immune complex-related nephropathy and non-Hodgkin B cell lymphoma), and patients with debilitating fatigue (A1)“

Table 2. Indications for treatment of chronic hepatitis C in 2015: Who should be treated and when?

<table>
<thead>
<tr>
<th>Treatment priority</th>
<th>Patient group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment is indicated</td>
<td>• All treatment-naïve and treatment-experienced patients with compensated and decompensated liver disease</td>
</tr>
</tbody>
</table>
| Treatment should be prioritized | • Patients with significant fibrosis (F3) or cirrhosis (F4), including decompensated cirrhosis  
                                      • Patients with HIV coinfection  
                                      • Patients with HBV coinfection  
                                      • Patients with an indication for liver transplantation  
                                      • Patients with HCV recurrence after liver transplantation  
                                      • Patients with clinically significant extra-hepatic manifestations  
                                      • Patients with debilitating fatigue  
                                      • Individuals at risk of transmitting HCV (active injection drug users, men who have sex with men with high-risk sexual practices, women of child-bearing age who wish to get pregnant, haemodialysis patients, incarcerated individuals) |
| Treatment is justified | • Patients with moderate fibrosis (F2) |
| Treatment can be deferred | • Patients with no or mild disease (F0-F1) and none of the above-mentioned extra-hepatic manifestations |
| Treatment is not recommended | • Patients with limited life expectancy due to non-liver related comorbidities |
The better knowledge of HCV replication cycle allowed the identification of several targeted drugs

Drugs active on viral enzymes
- Drugs active on host cell enzymes

**D Translation**
- miRNA
- ISIS 14803 (antisense)
- AVI-4066 (antisense)
- Heptazyme (ribozyme)
- VGX-410C (small molecules)
- IRES inhibitor TT 033 (siRNA)
- eIF2a phosphorylation inhibitors: Nitazoxanide

**D Post-Translation**
- NS3/4A protease inhibitors

**E RNA Replication**
- NS3H, NS4B, NS5A, NS5B inhibitors
- miR122, CypA
- PI4KIIα inhibitors

**F Assembly**
- Inhibition of core dimerisation, E1/E2 folding, p7, NS2, NS5A inhibitors
- DGAT, MTP inhibitors

**Virion release**
- Lectins, EGCG, blocking Ab, ITX5061, ezetimibe, erlotinib

**Virion release**

**Virus entry**
- Entry factor(s)
- Endocytic compartment
- Receptor-mediated endocytosis
- Fusion
- Uncoating
- Translation and polyprotein processing

**Nucleus**

**Membraneous web**

**(+)**

**(+/-)**

**Ploss A Gut 2012**
### The Standard of Care for HCV Patients Has Greatly Improved

<table>
<thead>
<tr>
<th>Year</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1974</td>
<td>Mystery virus identified</td>
</tr>
<tr>
<td>1989</td>
<td>HCV identified</td>
</tr>
<tr>
<td>1992</td>
<td>HCV Blood Test</td>
</tr>
<tr>
<td>1999</td>
<td>HCV Replicon</td>
</tr>
<tr>
<td>2005</td>
<td>HCV in-vitro Culture</td>
</tr>
<tr>
<td>2011-2013</td>
<td>PegIFN/ribavirin (6-12 mos)</td>
</tr>
<tr>
<td>2012-2014</td>
<td>PegIFN/ribavirin (3-6 mos)</td>
</tr>
</tbody>
</table>

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**SVR (%)**

<table>
<thead>
<tr>
<th>Year</th>
<th>SVR Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1991</td>
<td>8-12%</td>
</tr>
<tr>
<td>1995</td>
<td>15-20%</td>
</tr>
<tr>
<td>1998</td>
<td>38-43%</td>
</tr>
<tr>
<td>2001</td>
<td>25-30%</td>
</tr>
<tr>
<td>2011-2013</td>
<td>50-60%</td>
</tr>
<tr>
<td>2012-2014</td>
<td>70-80%</td>
</tr>
<tr>
<td>2013</td>
<td>83-96%</td>
</tr>
<tr>
<td>2014</td>
<td>89-100%</td>
</tr>
</tbody>
</table>

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**IFN-free DAA +/- RBV (3-6 mos)**

- PegIFN/ribavirin (6-12 mos) [8, 18]
- PegIFN/RBV (3-6-12 mos) [19-24]

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**References**

The elimination of the virus reduces mortality

SVR reduces mortality risk for each genotype:

- Genotype-1 hazard ratio, 0.70; P < .0001  
12,166 patients, SVR 35%
- Genotype-2 hazard ratio, 0.64; P = .006  
2904 patients, SVR 72%
- Genotype-3 hazard ratio, 0.51; P = .0002  
1794 patients, SVR 62%

Backus L, et al CGH 2011

SVR reduces mortality in patients with advanced hepatic fibrosis or cirrhosis (Ishak score 4-6)

- 192 patients (36%) SVR
- 10-year cumulative incidence rate of liver-related mortality or transplantation:
  - 1.9% (95% CI, 0.0%-4.1%) with SVR
  - 27.4% (95% CI, 22.0%-32.8%) without SVR (P < .001)

Achievement of an SVR.....

Table 4. The achievement of an SVR has an impact on extrahepatic manifestations of HCV.

<table>
<thead>
<tr>
<th>Clinical event</th>
<th>SVR (+)</th>
<th>SVR (-)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diabetes</td>
<td>26/1167 (2.2%)</td>
<td>117/1175 (9.9%)</td>
<td>Arase et al., 2009 [64]</td>
</tr>
<tr>
<td>Malignant lymphoma</td>
<td>0/2161 (0%)</td>
<td>25/1048 (12.6%)</td>
<td>Kawamura et al., 2007 [65]</td>
</tr>
<tr>
<td>Improved neurocognitive functions</td>
<td>8/8 (100%)</td>
<td>0/6 (0%)</td>
<td>Byrnes et al., 2012 [66]</td>
</tr>
</tbody>
</table>

Table 5. Cumulative incidence of clinical events in HCV patients with cirrhosis or advanced fibrosis stratified by treatment response.

<table>
<thead>
<tr>
<th>Endpoint</th>
<th>Reference</th>
<th>Patients</th>
<th>SVR (+)</th>
<th>SVR (-)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hepatocellular carcinoma</td>
<td>Yoshida et al., 1999 [72]</td>
<td>2890</td>
<td>1.9%</td>
<td>17.9%</td>
</tr>
<tr>
<td></td>
<td>Shiratori et al., 2005 [73]</td>
<td>271</td>
<td>17%</td>
<td>35%</td>
</tr>
<tr>
<td></td>
<td>Bruno et al., 2007 [74]</td>
<td>920</td>
<td>5.6%</td>
<td>16%</td>
</tr>
<tr>
<td></td>
<td>Cardoso et al., 2010 [76]</td>
<td>307</td>
<td>5.8%</td>
<td>19.6%</td>
</tr>
<tr>
<td>Decompensation</td>
<td>Veldt et al., 2007 [75]</td>
<td>479</td>
<td>0%</td>
<td>12%</td>
</tr>
<tr>
<td></td>
<td>Cardoso et al., 2010 [76]</td>
<td>307</td>
<td>2.9%</td>
<td>13.7%</td>
</tr>
<tr>
<td>Development of esophageal varices</td>
<td>Bruno et al., 2010 [77]</td>
<td>218</td>
<td>0%</td>
<td>39%</td>
</tr>
<tr>
<td></td>
<td>D’Ambrosio et al., 2011 [78]</td>
<td>127</td>
<td>3.5%</td>
<td>15.1%</td>
</tr>
<tr>
<td>Liver-related death</td>
<td>Yoshida et al., 2002 [69]</td>
<td>2879</td>
<td>0.2%</td>
<td>2%</td>
</tr>
<tr>
<td></td>
<td>Shiratori et al., 2005 [73]</td>
<td>271</td>
<td>0%</td>
<td>15%</td>
</tr>
<tr>
<td></td>
<td>Bruno et al., 2007 [74]</td>
<td>920</td>
<td>1.7%</td>
<td>11.4%</td>
</tr>
<tr>
<td></td>
<td>Cardoso et al., 2010 [76]</td>
<td>307</td>
<td>2.9%</td>
<td>8.8%</td>
</tr>
</tbody>
</table>
167 HIV/HCV-coinfected patients were diagnosed with HCC in the participant Spain hospitals. 65 (39%) of them had been previously treated against HCV. In 13 cases, HCC was diagnosed after achieving consecution of SVR, accounting for 7.8% of the overall cases. The median elapsed time from SVR to diagnosis of HCC was 28 (IQR 20–39) months.

**Table 2. Main features of hepatocellular carcinoma diagnosed in patients with previous sustained virological response (n=13).**

<table>
<thead>
<tr>
<th>Case</th>
<th>Alcohol (g/day)</th>
<th>Months from end of HCV therapy</th>
<th>Months from last normal US</th>
<th>Unilocular</th>
<th>Milan criteria</th>
<th>PT</th>
<th>Extrahepatic disease</th>
<th>BCLC stage</th>
<th>Therapy against HCC</th>
<th>Vital status</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>&gt;100</td>
<td>66</td>
<td>6</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>D</td>
<td>RFA</td>
<td>Alive</td>
</tr>
<tr>
<td>2</td>
<td>0</td>
<td>9</td>
<td>4</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>C</td>
<td>None</td>
<td>Dead</td>
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<tr>
<td>3</td>
<td>0–50</td>
<td>18</td>
<td>30</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>A</td>
<td>Surgical resection</td>
<td>Alive</td>
</tr>
<tr>
<td>4</td>
<td>0</td>
<td>21</td>
<td>3</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>A</td>
<td>LT</td>
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<tr>
<td>5</td>
<td>0–50</td>
<td>22</td>
<td>1</td>
<td>No</td>
<td>No</td>
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<td>No</td>
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<tr>
<td>6</td>
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<td>23</td>
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<td>C</td>
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<td>25</td>
<td>25</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>B</td>
<td>TACE</td>
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<tr>
<td>8</td>
<td>0</td>
<td>31</td>
<td>NA</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>C</td>
<td>None</td>
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<td>0</td>
<td>31</td>
<td>15</td>
<td>No</td>
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<td>Yes</td>
<td>No</td>
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<td>10</td>
<td>0</td>
<td>36</td>
<td>2</td>
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<td>No</td>
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<td>No</td>
<td>C</td>
<td>Sorafenib</td>
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<td>1</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>C</td>
<td>None</td>
<td>Dead</td>
</tr>
<tr>
<td>13</td>
<td>0–50</td>
<td>71</td>
<td>72</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
<td>No</td>
<td>C</td>
<td>None</td>
<td>Dead</td>
</tr>
</tbody>
</table>

BCLC, Barcelona Clinic Liver Cancer; LT, liver transplantation; NA, not available; PT, portal thrombosis; RFA, radiofrequency ablation; TACE, transarterial chemoembolization; US, ultrasound.
Many lessons learnt from HIV can be helpful for designing adequate treatment strategies against viral hepatitis such as HCV….and particularly in the HIV/HCV population….
The personalized medicine

All international guidelines focus on the importance of tailoring antiretroviral therapy to the individual patient, on the basis of HIV-1 genetic data, integrated with clinical, laboratory and therapeutic information.
The HCV genotype and genotype 1 subtype (1a/1b) must be assessed prior to treatment initiation and will determine the choice of therapy (A1).

Rating: Class I, Level A

Testing for HCV genotype is recommended to guide selection of the most appropriate antiviral regimen.
Genotype 1 is by far the most frequent genotype in chronically infected patients worldwide.
Consequences of HCV variability at population level: HCV genotypes

31%–33% nucleotide difference among the 7 known HCV genotypes and 20%–25% among the nearly 67 HCV subtypes (Smith et al., 2014).
Treatment recommendations for HCV-monoinfected or HCV/HIV coinfected patients with chronic hepatitis C with compensated (Child-Pugh A) cirrhosis, including treatment-naïve patients and patients who failed on a treatment based on PegIFN-a and ribavirin (RBV).

<table>
<thead>
<tr>
<th>Patients</th>
<th>PegIFN-α, RBV and sofosbuvir</th>
<th>PegIFN-α, RBV and simeprevir</th>
<th>Sofosbuvir and RBV</th>
<th>Sofosbuvir and ledipasvir</th>
<th>Ritonavir-boosted paritaprevir, ombitasvir and dasabuvir</th>
<th>Ritonavir-boosted paritaprevir, ombitasvir and simeprevir</th>
<th>Sofosbuvir and simeprevir</th>
<th>Sofosbuvir and daclatasvir</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype 1a</td>
<td>12 wk</td>
<td>12 wk (treatment-naïve or relapers) or 24 wk (partial or null responders)</td>
<td>No</td>
<td>12 wk with RBV, or 24 wk without RBV, or 24 wk with RBV if negative predictors of response</td>
<td>24 wk with RBV</td>
<td>12 wk with RBV, or 24 wk without RBV</td>
<td>12 wk with RBV, or 24 wk without RBV</td>
<td>12 wk with RBV, or 24 wk without RBV</td>
</tr>
<tr>
<td>Genotype 1b</td>
<td>12 wk</td>
<td>No</td>
<td>16-20 wk</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>12 wk without RBV</td>
</tr>
<tr>
<td>Genotype 2</td>
<td>12 wk</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Genotype 3</td>
<td>12 wk</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>24 wk with RBV</td>
</tr>
<tr>
<td>Genotype 4</td>
<td>12 wk</td>
<td>12 wk (treatment-naïve or relapers) or 24 wk (partial or null responders)</td>
<td>No</td>
<td>12 wk with RBV, or 24 wk without RBV, or 24 wk with RBV if negative predictors of response</td>
<td>No</td>
<td>24 wk with RBV</td>
<td>12 wk with RBV, or 24 wk without RBV</td>
<td>12 wk with RBV, or 24 wk without RBV</td>
</tr>
<tr>
<td>Genotype 5 or 6</td>
<td>12 wk</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

EASL HCV clinical guidelines 2015
Genotype 1 is by far the most frequent genotype in chronically infected patients worldwide as well as in Europe. 

*Esteban JI et al J Hepatol 2008;48:148-162*
HCV genotype was the most important baseline predictor for response to Peg-IFN + Ribavirin Combination Therapy.

HCV genotypes 2 and 3:
- SVR = 78-86%
- HCV-2 = 80-95%
- HCV-3 Low viremia = 75-80%
- HCV-3 High viremia = 60-70%

HCV genotype 1:
- SVR = 35-65%
- HCV-1 Low viremia = 50%
- HCV-1 High viremia = 30-35%

HCV genotypes 4 and 6:
- SVR = 42-52%
Genotype 3 HCV-infected patients had poor SVR rates following treatment for 12 or 16 weeks with Sofosbuvir+RBV.
HCV-3 is associated with increased risk of HCC in patients with cirrhosis

Probability of HCC-Free Survival According to HCV Genotype


Log-rank test and Cox model were used to compare the actuarial incidence of HCC between genotype subgroups.

Higher incidence of HCC seems to be independent of well-known risk factors, including grade of steatosis. Steatosis was more frequent in GT-3 patients, but its grade was not associated with HCC development.

GT=genotype; HCC=hepatocellular carcinoma

Nkontchou et al. J Viral Hepat. 2011
Is Genotype 3 of the Hepatitis C Virus the New Villain?

Nicolas Goossens\textsuperscript{1} and Francesco Negro\textsuperscript{1,2}

Association between HCV genotype and prevalence of liver steatosis
<table>
<thead>
<tr>
<th>Clinical Outcome</th>
<th>Type of Study</th>
<th>Characteristics of G3 Patients</th>
<th>Characteristics of Comparator Group</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increased steatosis</td>
<td>Individual data meta-analysis of 3068 HCV patients with a liver biopsy</td>
<td>669 patients with G3</td>
<td>2399 patients with non-G3 (G1, 2 and 4)</td>
<td>(18)</td>
</tr>
<tr>
<td></td>
<td>Retrospective cohort study of 141 HCV patients without cirrhosis with a liver biopsy</td>
<td>28 patients with G3</td>
<td>113 patients with non-G3 (G1, 2, 4-9)</td>
<td>(19)</td>
</tr>
<tr>
<td></td>
<td>Retrospective cohort study of 180 HCV patients with a liver biopsy</td>
<td>28 patients with G3</td>
<td>154 patients with non-G3 (G1, 2 and &quot;mixed&quot;)</td>
<td>(20)</td>
</tr>
<tr>
<td></td>
<td>Retrospective cohort study of 101 HCV patients without other risk factors for steatosis (70 immunotolerant patients and 31 post-liver transplant patients)</td>
<td>24 patients with G3</td>
<td>46 patients with non-G3 (G1, 2, 5 and &quot;mixed&quot;)</td>
<td>(21)</td>
</tr>
<tr>
<td></td>
<td>Retrospective case-control of 62 HCV patients having undergone antiviral therapy with pre- and posttransplant liver biopsy</td>
<td>34 patients with G3</td>
<td>28 patients with G1</td>
<td>(22)</td>
</tr>
<tr>
<td>Increased liver fibrosis progression</td>
<td>Retrospective cohort study of 1189 HCV patients with a liver biopsy and an assessable date of infection</td>
<td>312 patients with G3</td>
<td>877 patients with non-G3 (G1, 2 and 4)</td>
<td>(44)</td>
</tr>
<tr>
<td></td>
<td>Meta-analysis of 15 studies (8 studies with a single liver biopsy and 8 studies with paired liver biopsies)</td>
<td>Single biopsy studies: 730 patients with G3, Paired biopsy studies: 115 patients with G3</td>
<td>Singles biopsy studies: 1,619 patients with non-G3, Paired biopsy studies: 551 patients with non-G3</td>
<td>(45)</td>
</tr>
<tr>
<td></td>
<td>Retrospective/prospective cohort of 724 Native Alaskan patients with detectable viremia for HCV</td>
<td>109 patients with G3</td>
<td>615 patients with non-G3 (G1 and 2)</td>
<td>(47)</td>
</tr>
<tr>
<td>Increased risk of HCC</td>
<td>Retrospective cohort study of 4065 drug users with treatment-naive HCV</td>
<td>1,077 patients with G3</td>
<td>1,986 patients with non-G3 (G1, 2, 4 and 5)</td>
<td>(48)</td>
</tr>
<tr>
<td></td>
<td>Retrospective cohort study of 353 patients with biopsy-proven HCV cirrhosis</td>
<td>25 patients with G3</td>
<td>328 patients with non-G3 (G1, 2 and 4)</td>
<td>(51)</td>
</tr>
<tr>
<td></td>
<td>Long-term follow-up study of 530 HCV patients treated with an interferon-based regimen</td>
<td>13 patients with G3</td>
<td>407 patients with non-G3 (G1, 2, 4 and &quot;other&quot;)</td>
<td>(52)</td>
</tr>
<tr>
<td>Reduced SVR after interferon-α/ribavirin dual therapy</td>
<td>RCT of 224 G2 or G3 patients treated with pegylated interferon-α and ribavirin for a duration of 24 weeks</td>
<td>182 patients with G3</td>
<td>42 patients with G2</td>
<td>(55)</td>
</tr>
<tr>
<td></td>
<td>RCT of 283 G2 or G3 patients treated with pegylated interferon-α and ribavirin for a duration of 12-24 weeks</td>
<td>70 patients with G3</td>
<td>213 patients with G2</td>
<td>(56)</td>
</tr>
<tr>
<td></td>
<td>RCT of 1469 G2 or G3 patients treated with pegylated interferon-α and ribavirin for a duration of 18-24 weeks</td>
<td>727 patients with G3</td>
<td>728 patients with G2</td>
<td>(57)</td>
</tr>
<tr>
<td></td>
<td>Meta-analysis of 6 RCTs, 1 retrospective study and 1 prospective study studying the differential SVR rate between G2 and G3.</td>
<td>1,225 patients with G3</td>
<td>1,050 patients with G2</td>
<td>(58)</td>
</tr>
<tr>
<td></td>
<td>Meta-analysis of 17 RCTs studying the effect of different therapy duration on SVR</td>
<td>843 rapid virologic responders patients with G3</td>
<td>739 rapid virologic responders patients with G2</td>
<td>(59)</td>
</tr>
</tbody>
</table>
ALLY 3: SVR$_{12}$ in Patients With Cirrhosis

- Among patients with cirrhosis, 34% (11/32) had baseline platelet counts < 100,000/mm$^3$

SVR$_{12}$=sustained virologic response at posttreatment week 12.

$^a$ HCV RNA <LLOQ (25 IU/mL), detectable or undetectable; error bars reflect 95% Confidence Intervals (CI).

$^b$ Cirrhosis status determined in 141 patients by liver biopsy (METAVIR F4), FibroScan (> 14.6 kPa), or FibroTest score ≥ 0.75 and APRI (aspartate aminotransferase to platelet ratio index) > 2.

$^c$ Cirrhosis status for 11 patients was missing or inconclusive (FibroTest score > 0.48 to < 0.75 or APRI > 1 to ≤ 2).

ID_490  HCV genotype: 3a  Age: 48  Sex: M  Relapse to SOC  Cirrhotic  HCC  Waiting list for OLT

HCV- RNA (log IU/ml)

GRT Day 0
NS5B Resistance Mutations: None

GRT at failure
NS5B Resistance Mutations: S282T

Sofosbuvir

708,007 IU/ml

763 IU/ml

364 IU/ml

277 IU/ml

382 IU/ml

591 IU/ml

321 IU/ml

793 IU/ml

LLOQ (12 IU/ml)

<12 IU/ml

Day 0 1 w 2 w 3 w 4 w 5 w 7 w 10 w 12 w 15 w
A correct determination of HCV-genotype is relevant prior to treatment initiation

- Several commercial assays are available for determining genotype/subtype.
- All assays target the 5’NCR gene for genotypes 1-6, in addition, the 2 assays more used in diagnostics, Abbott and INNO-LiPA-HCV-2.0, target also the NS5B and the core gene, respectively, providing additional information also in subtyping: for genotype 1 (1a/1b, both), and for all genotypes (only Innolipa).

### Target Regions: HCV 5’ UTR, CORE & NS5B region

- **5’ UTR**: C, E1, E2, p7, NS2, NS3, NS4A, NS4B, NS5A, NS5B
- **3’ UTR**: aa. 1 192 384 742 810 1027 1658 1712 1973 2421 3011

**Assays**

- **Trugene HCV Genotyping assay**: Direct sequencing
- **INNO-LiPA HCV 1.0**: Reverse hybridization
- **INNO-LiPA HCV 2.0**: Reverse hybridization
- **Abbott RealTime HCV Genotype II assay**: Real time PCR
A correct determination of HCV-genotype is relevant prior to treatment initiation

- Several commercial assays are available for determining genotype/subtype

However, not all genotypes can be resolved, with results being reported as: ‘indeterminate’, ‘mixed’, ‘genotype X reactivity with Y’, or just the major genotype 1 alone.

Concordance of the TRUGENE assay with NS3/4A or NS5B sequence-based genotype subtyping assays is 79.6%. Sarrazin et al., 2015

Out of 1052 samples, tested with Abbott m2000 HCV genotype II, 89 (8.5%) underwent further sequencing to determine the HCV genotype. Benedet et al., 2014
HCV-sequencing and commercial-assays were concordant in 91.84% of cases analysed

<table>
<thead>
<tr>
<th>Genotype/subtype confirmed</th>
<th>Patients (N)</th>
<th>Patients (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype 1 with no subtype</td>
<td>6</td>
<td>1.75%</td>
</tr>
<tr>
<td>Discordant genotypes</td>
<td>4</td>
<td>1.17%</td>
</tr>
<tr>
<td>Genotype 1 with discordant subtype</td>
<td>10</td>
<td>2.91%</td>
</tr>
<tr>
<td>Mixed genotypes</td>
<td>8</td>
<td>2.33%</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>343</strong></td>
<td><strong>100%</strong></td>
</tr>
</tbody>
</table>

We reanalyzed genotype data by phylogenetic analysis of 343 HCV-infected patients candidate to DAA-treatment, who performed a genotypic-resistance-test between 2011 and 2014. To confirm the appropriate genotype allocation, HCV-sequencing was performed by home-made protocols, specific for each genotype, on NS3-protease (95% samples), together with/in alternative to NS5A (9%) and/or NS5B (14%). Notably, all patients with a previous result of ‘mixed’ or ‘indeterminate’ HCV-genotype or subtype by commercial-assays were instead precisely resolved by HCV-sequencing.

Ceccherini-Silberstein F. et al, Hepatology 2015
Different prevalence of RAVs among plasma and liver tissues (not tumoral and HCC)

15 HCC/LT pts infected by HCV GT1a [N=4], 1b [N=6], 3a [N=4], and 4d [N=1] underwent LT due to HCC, in 2014-2015. Plasma sequences of NS3 (N=14), NS5a (N=15) and NS5b (N=11) were successfully obtained. NT and TT sequences of NS3 (N=8 and 7), NS5a (N=5 and 6) and NS5b (N=2 and 1) were also successfully obtained.

RAVs were found in at least one compartment in 5/15 patients (NS3 RAVs: 2 pts, 1 HCV-1b, 1 HCV-3; NS5a RAVs: 3 HCV-1b pts, NS5b RAVs: 2 HCV-1b pts).

<table>
<thead>
<tr>
<th>Patient</th>
<th>Genotype</th>
<th>Compartments</th>
<th>NS3-Protease</th>
<th>NS5A</th>
<th>NS5B-Polymerase</th>
<th>HCV-RNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT.2</td>
<td>1b</td>
<td>TT</td>
<td>none</td>
<td>Q54H</td>
<td>n.a.</td>
<td>N.A.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>none</td>
<td>Q54H, Y93H/P/Y/S</td>
<td>n.a.</td>
<td>N.A.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>none</td>
<td>Q54H</td>
<td>L159F, C316N, V499T</td>
<td>410 IU/ml</td>
</tr>
<tr>
<td>PT.22*</td>
<td>1b</td>
<td>TT</td>
<td>none</td>
<td>T54T/S</td>
<td>Y93 (H/Y)</td>
<td>none</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>none</td>
<td>Y93 (H/Y)</td>
<td>none</td>
<td>1284809 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>none</td>
<td>none</td>
<td>none</td>
<td>54503 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 26 w post-LT</td>
<td>none</td>
<td>none</td>
<td>none</td>
<td>3433 IU/ml</td>
</tr>
<tr>
<td>PT.1</td>
<td>3a</td>
<td>TT</td>
<td>none</td>
<td>S122A</td>
<td>none</td>
<td>38 IU/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>11652 IU/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>6643 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 49 w post-LT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>2153271 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 64 w post-LT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>5238593 IU/ml</td>
</tr>
<tr>
<td>PT.18</td>
<td>1b</td>
<td>PL</td>
<td>none</td>
<td>Q54H</td>
<td>none</td>
<td>171419 IU/ml</td>
</tr>
<tr>
<td>PT.27**</td>
<td>1b</td>
<td>TT</td>
<td>none</td>
<td>none</td>
<td>none</td>
<td>642 IU/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 4 w pre-LT</td>
<td>none</td>
<td>none</td>
<td>S556G</td>
<td>212875 IU/ml</td>
</tr>
</tbody>
</table>

Prevalence of NS3, NS5Aa and NS5b RAVs found in different compartments of HCC/transplanted pts: tumorous tissue (TT), not-tumoral tissue (NT), plasma (PL), 26, 49 and 64 weeks post-LT plasma (PL26w, PL 49w and PL64 w). N.A. Not available.

*PT.22 began Sofosbuvir/Ribavirin treatment 26 weeks post-LT. **PT.27 was TND at the time of OLT.

Sorbo MC et al., ICAR 2015
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15 HCC pts infected by HCV GT1a [N=4], 1b [N=6], 3a [N=4], and 4d [N=1] underwent LT due to HCC, in 2014-2015. Plasma sequences of NS3 (N=14), NS5a (N=15) and NS5b (N=11) were successfully obtained. NT and TT sequences of NS3 (N=8 and 7), NS5a (N=5 and 6) and NS5b (N=2 and 1) were also successfully obtained.

RAVs were found in at least one compartment in 5/15 patients (NS3 RAVs: 2 pts, 1HCV-1b, 1 HCV-3; NS5a RAVs: 3 HCV-1b pts, NS5b RAVS: 2 HCV-1b pts).

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<th>HCV-RNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>PT.2</td>
<td>1b</td>
<td>TT</td>
<td>none</td>
<td>Q54H</td>
<td>n.a.</td>
<td>N.A.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>none</td>
<td>Q54H, Y93H/P/Y/S</td>
<td>n.a.</td>
<td>N.A.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>none</td>
<td>Q54H</td>
<td>L159F, C316N, V499T</td>
<td>410 IU/ml</td>
</tr>
<tr>
<td>PT.22*</td>
<td>1b</td>
<td>TT</td>
<td>T54T/S</td>
<td>Y93 (H/Y)</td>
<td>none</td>
<td>1284809 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>none</td>
<td>Y93 (H/Y)</td>
<td>none</td>
<td>1810598 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>none</td>
<td>none</td>
<td>none</td>
<td>54503 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 26 w post-LT</td>
<td>none</td>
<td>none</td>
<td>3433 IU/ml</td>
<td></td>
</tr>
<tr>
<td>PT.1</td>
<td>3a</td>
<td>TT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>38 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>NT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>11652 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL</td>
<td>S122A</td>
<td>n.a.</td>
<td>n.a.</td>
<td>6643 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 49 w post-LT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>2153271 IU/ml</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 64 w post-LT</td>
<td>S122A</td>
<td>none</td>
<td>n.a.</td>
<td>5238593 IU/ml</td>
</tr>
<tr>
<td>PT.18</td>
<td>1b</td>
<td>PL</td>
<td>none</td>
<td>Q54H</td>
<td>none</td>
<td>171419 IU/ml</td>
</tr>
<tr>
<td>PT.27**</td>
<td>1b</td>
<td>TT</td>
<td>none</td>
<td>none</td>
<td>n.a.</td>
<td>642 UI/µg</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PL 4 w pre-LT</td>
<td>none</td>
<td>none</td>
<td>S556G</td>
<td>212875 IU/ml</td>
</tr>
</tbody>
</table>

Prevalence of NS3,NS5Aa and NS5b RAVs found in different compartments of HCC/transplanted pts: tumorous tissue (TT), not-tumoral tissue (NT), plasma (PL), 26, 49 and 64 weeks post-LT plasma (PL26w, PL 49w and PL64 w). N.A. Not available.
*PT.22 began Sofosbuvir/Ribavirin treatment 26 weeks post-LT. **PT.27 was TND at the time of OLT.

Sorbo MC et al., ICAR 2015
Natural RAVs were detected in 55/286 (19.2%) NS3 sequences, in 30/96 (31.3%) NS5A sequences and 4/90 (4.4%) NS5B sequences from DAA-naïve patients with an available baseline resistance test.

DAA naïve pts without HCC/LT (control group) were analysed for NS3 (1a=143, 1b=176, 3=7, 4=7), NS5a (1a=42, 1b=42, 3=9, 4=7) and NS5B (1a=34, 1b=44, 3=7, 4=5) as control.

<table>
<thead>
<tr>
<th>NS3 RAVs</th>
<th>Genotype</th>
<th>1a (N=143)</th>
<th>1b (N=176)</th>
<th>3a (N=6)</th>
<th>4d (N=2)</th>
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<th>1b (N=42)</th>
<th>3a (N=9)</th>
<th>4d (N=7)</th>
<th>NS5b RAVs</th>
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<th>1b (N=44)</th>
<th>3a (N=7)</th>
<th>4d (N=5)</th>
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</table>

NS3, NS5Aa and NS5b RAVs found in control group of DAA naïve pts without HCC and LT.

- Interestingly, the NS3 RAV S122A found in 1 HCC/LT GT3, was instead detected in only 1 control pt, infected with GT1b (prevalence in HCC pts 6.6% vs 0.3% in controls p= 0.08 Fisher test).

Sorbo MC et al., ICAR 2015
# HCV-RNA comparison among SERUM, NO-HCC and HCC samples

<table>
<thead>
<tr>
<th>ID Patient</th>
<th>Age (years)</th>
<th>Sex</th>
<th>Cirrhosis</th>
<th>HCC</th>
<th>LT</th>
<th>HCV Genotype</th>
<th>VIREMIA (IU/ml)</th>
<th>HCV-RNA NO-HCC (IU/μg RNA)</th>
<th>HCV-RNA HCC (IU/μg RNA)</th>
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</table>

**UNTREATED**

**TREATED SOF+RBV**

| PT2        | 63          | M   | 1         | 1   | 1  | 1b           | TND            | 59                       | 642                        |

SOF, sofosbuvir; RBV, ribavirin; HCC, Hepatocellular Carcinoma; LT, Liver Transplantation; IU, International Units; TND, Target Not Detected.

---

**PT.1**  
**HCV genotype: 1a**  
**Sex: M**  
**Age: 48**  
**HCC**  
**LIVER TRANSPLANTED**

GRT Day 0  
NS3 Mutations PI: T40T/A, Q89H, S91A, L153I  
NS3 Mutations TT: T40T/A, Q89H, S91A, L153I  
NS3 Mutations NT: N.D.  
NO NS3 RESISTANCE MUTATIONS

Antonucci et al., ICAR 2015
HCV - a curable disease

We can cure HCV; **SVR** a validated surrogate of clinical efficacy because it predicts long-term clinical benefit

To cure everyone with HCV we need to find it

When we have found it we need to treat it properly

Accurate diagnostics and treatment will be key to reduce HCV infections **and therefore to reduce the HCC HCV-related**

However, only a small proportion of infected persons are likely to have access to **new therapies** in most countries!!!!!
Conclusions

Today in the era of new DAAs, we can achieve in the majority of patients the SVR, and therefore the cure of HCV-associated disease and hopefully the reduction of mortality.

However we are still in a Phase of learning process…..

Not all patients are identical…

We phase complexity: virus, host, previous treatment outcome, DAA, and clinical aspects

HCV-RNA decay at early time points (i.e. 48h? week-1, week-2) may help in predicting treatment outcome?

The performance of a baseline sequencing of HCV can provide important virological information for clinical management of patients with chronic HCV infection?