Vers un vaccin bivalent contre les virus des hépatites B et C ?

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but DAA therapies are very expensive and therefore unlikely to be adopted universally.

Low-cost / generic drugs will potentially be available in the future in low-income countries

... but most HCV-infected subjects are not aware of their infection,

then the cost of large-scale HCV screening + DAA treatment will remain very high.

≈ 15% of patients display persistent hepatic inflammation and / or cirrhosis despite virological cure.
→ 180 million people currently infected worldwide

→ 3-4 million new infections (mostly by blood contact: IVDU, unsafe medical practices, health workers) each year
HCV epidemiology in 2015 in the USA

2.7 million HCV chronic carriers
(1.1 diagnosed - 1.6 undiagnosed)

Still 18,000 to 20,000 new infections / year (1 every 30 min)

- Mostly young IVDU
- No symptoms less than 1% reported to health department

1 or 2 cases every month of healthcare transmission
HCV epidemiology in 2015 in China

Estimated HCV prevalence: 1 to 2 %
(15 to 30 millions of HCV chronic carriers)

- Cost for screening + HCV treatment will be tremendous

- Risk factors for new infections:
  - IVDUs (especially in urban areas)
  - Iatrogenic transmission (especially in rural areas)

most are not diagnosed
An HCV prophylactic vaccine is a medical priority ➔ best hope of controlling the world epidemic ➔ opportunity to significantly reduce healthcare cost (especially if the HCV vaccine is associated with the HBV vaccine)

The HBV vaccine has considerably reduced the incidence of HBV-induced HCC

After many years of controversy, a partially-effective HCV vaccine (≈ 60-80% efficacy) appears to be a feasible goal based on:

→ Natural immunity demonstrated in re-exposed humans & chimpanzees

→ Natural immunity linked with viral-specific CD4+ & CD8+ T cell responses & cross-neutralising antibodies

→ Chimpanzee studies demonstrating that vaccinated animals are protected against the development of the carrier state
HCV virology

Adapted from Bartenschlager et al, Nat Rev Microbiol 2013
NS proteins contain numerous mapped T-cell epitopes and are preferentially included in T-cell vaccine candidates.

E1&E2 proteins can induce neutralizing antibodies (nAbs).
Native heterodimer complex comprising both full length envelope glycoproteins E1 (33KDa) & E2 (17KDa)

Produced in CHO or Hela cell lines

E1-E2 retained in the ER via transmembrane domain (TMD)

Primes the induction of viral nAbs a CD4+ T-cell response

Dr. M. Houghton & collaborators

CHIRON

NOVARTIS
Recombinant E1-E2 vaccine

Combined HCV vaccine preclinical data in the chimpanzee model

(combined results from homologous HCV 1a & heterologous HCV 1a challenges)

Chimpanzee studies performed at over the course of 15 years (1994-2009)

with various adjuvants and recombinant E1-E2 of varying purities

<table>
<thead>
<tr>
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<th>Vaccinees</th>
<th>Controls</th>
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<tbody>
<tr>
<td>number</td>
<td>31</td>
<td>24</td>
</tr>
<tr>
<td>acute infection</td>
<td>26 (84%)</td>
<td>24 (100%)</td>
</tr>
<tr>
<td>chronic infection</td>
<td>5 (16%)</td>
<td>15 (62%)</td>
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P < 0.001

Phase I trial with E1-E2 recombinant vaccine (gen 1a) + MF59 as adjuvant

Safety and immunogenicity of HCV E1E2 vaccine adjuvanted with MF59 administered to healthy adults

Sharon E. Frey, Michael Houghton, Stephen Coates, Sergio Abrignani, David Chien, Domenico Rosa, Piero Pileri, Ranjit Ray, Adrian M. Di Bisceglie, Paola Rinella, Heather Hill, Mark C. Wolff, Viola Schultze, Jang H. Han, Bruce Scharschmidt, Robert B. Belshe

Vaccine 28 (2010) 6367-6373

- 60 healthy volunteers (4 groups of 15 individuals with 4x injections of 4, 20 or 100 µg)
- Vaccine safe & well-tolerated
- Elicits anti-E1-E2 titers in the same range as in protected chimpanzees
- Induces a strong lymphoproliferative response to E1-E2
- 20 µg E1-E2 dose administered on month 0, 1 & 6 appears optimal
  (100% of subjects developed a humoral response after the 3rd vaccination)
Phase I trial with E1-E2 recombinant vaccine (gen 1a) + MF59 as adjuvant (neutralization assays with sera collected 2 weeks post-3rd vaccination with 100 μg)

A Hepatitis C Virus (HCV) Vaccine Comprising Envelope Glycoproteins gpE1/gpE2 Derived from a Single Isolate Elicits Broad Cross-Genotype Neutralizing Antibodies in Humans

John Lok Man Law¹, Chao Chen¹, Jason Wong¹, Darren Hockman¹, Deanna M. Santer¹, Sharon E. Frey², Robert B. Belshe², Takaji Wakita³, Jens Bukh⁴, Christopher T. Jones⁵, Charles M. Rice⁶, Sergio Abrignani⁷, D. Lorne Tyrrell¹, Michael Houghton¹

Neutralization normalized using the pre-vaccination sera of the same individual

5/13 human sera neutralized over 50% of heterologous HCVcc 1a
(2 of which neutralized up to 80% of viral infectivity)

(Dr Houghton) is planning to test the efficacy of an improved recombinant E1-E2 vaccine in Canadian IVDUs
Hepatitis B & hepatitis C viruses

<table>
<thead>
<tr>
<th>HBV</th>
<th>HCV</th>
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<tr>
<td><strong>Family / genome</strong></td>
<td><strong>Family / genome</strong></td>
</tr>
<tr>
<td>Hepadnavirus / DNA</td>
<td>Flavivirus / RNA</td>
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</table>

**Structure**

**Envelope**

**Prophylactic vaccine**
Chimeric Hepatitis B Virus/Hepatitis C Virus Envelope Proteins Elicit Broadly Neutralizing Antibodies and Constitute a Potential Bivalent Prophylactic Vaccine

Elodie Beaumont, Romuald Patient, Christophe Houriez, Isabelle Dimier-Poisson, and Philippe Rougeret

Stable production of $\approx 10 \mu g/ml$ HBs Ag in the supernatant of CHO clones

subviral particles are secreted and used for the HBV vaccine

Stable production of anti-S

anti-E1 + anti-E2
Productive folding and heterodimerization of the HCV envelope proteins in the context of the fusion proteins

mAb AR3A
recognize a conformation-dependent discontinuous epitope on E2
neutralize different HCV genotypes in vitro

mAb AR5A
recognize the folded E1-E2 heterodimer

Beaumont et al, Hepatology 2013
The best immunogen for anti-HCV response is the subviral particle containing the E2-S chimeric protein (rabbits & mice data, with different adjuvants).

3 x 15 μg (week 0, 2, 4) of particles serum collected week 6 + controls with Engerix + controls with adjuvant alone (MF 59 Addavax®)

anti-E1 or E2

anti-HBs

The anti-HBs response is equivalent to the response induced by a commercial HBV vaccine, suggesting that the chimeric particles could replace existing HBV vaccines whilst providing the additional benefit of protection against HCV.
Evaluation of the neutralizing response in the HCVcc chimeric viruses Huh7.5 model

Genotypes 1a, 1b, 2a & 3

or adjuvant alone

Beaumont et al, Hepatology 2013
Evaluation of the neutralizing response in the HCVcc chimeric viruses Huh7.5 model

Serum dilutions

or adjuvant alone

Beaumont et al, Hepatology 2013
Chimeric HBV-HCV particles could be used in **two different strategies**:

- **In primary vaccination**, to induce protective immunity to both HBV & HCV
- **As booster doses** in individuals previously vaccinated against HBV, to ensure full protection against HBV and induce protective immunity to HCV
Conclusions

The entire HCV E1 and/or E2 env proteins, are incorporated in secreted subviral particles resembling the HBV vaccine.

Sera containing anti-E1 and anti-E2 elicited by the chimeric HBV-HCV (genotype 1a) particles neutralize different HCV heterologous strains of various genotypes (1a = 1b > 2 & 3).

This vaccine candidate could be produced by the same procedures established for HBV vaccines, reducing the time and cost of its industrial development.

The anti-HBs response induced by the chimeric particles is equivalent to the response induced by a commercial HBV vaccine, suggesting that this vaccine could replace existing HBV vaccines whilst providing the additional benefit of protection against HCV.

This bivalent HBV-HCV prophylactic vaccine could be used in primary vaccination for both viruses or as booster doses in individuals previously vaccinated against HCV.
Perspective 1: Production of subviral particles containing HCV envelope from different genotypes to immunize with a mix of particles and increase the cross-neutralizing properties of this vaccine candidate.

Massima et al, Hepatology 2014
Perspective 2: Preclinical assay in a primate model

- Compare HBV vaccine with the HBV-HCV chimeric particles
- Tolerability
- T-cell response (ELISPOT)
- Humoral immune response (anti-HBs & anti-E1-E2)
- Various HCV genotypes neutralization
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