A cluster-based protein coevolution method uncovers critical features of the original HCV fusion mechanism

Comment l’étude de la co-évolution des protéines virales a permis de révéler le mécanisme de fusion membrane du VHC

HCV E1E2 : Assembly on infectious virions
HCV E1E2: Interaction with receptors

HCV E1E2: Internalization within cells
HCV E1E2: Membrane fusion

Viral membrane
E1 et E2
Post-fusion structure
Endosomal membrane
Cytoplasm

E1 and E2 functions are poorly characterized

Adapted from Krey et al, 2010
Dengue envelope glycoproteins

HCV

E1                 E2

Dengue E

Dengue PrM

Dengue E domains

Class II fusion protein-induced fusion

HCV

E1                 E2

Dengue E

• Cell surface binding
• Membrane fusion

Dengue PrM

• Structural chaperone

Virion genome

Cell

Homodimer dissociation ➔ Domain III Fold Back ➔ Membrane merging ➔ Fusion
E2 is not a Class 2 fusion protein

hepacivirus
HCV
E1     E2

Kong et al. Lat, Science 2013
Khan Marcotrigiano Nature 2014
El Omari Stuart Nat Com 2014

E1 and E2 functions are still poorly characterized

hepacivirus
HCV
E1     E2

Kong et al. Lat, Science 2013
Khan Marcotrigiano Nature 2014
El Omari Stuart Nat Com 2014
HCV fusion mechanism: how do E1 and E2 act?

**hepativirus**
- HCV
- E1
- E2
- Binding: E2
- Fusion: E1?

**flavivirus**
- Dengue E
- Dengue PrM
- Binding: E
- Fusion: E

**pestivirus**
- BVDV
- E1
- E2
- Binding: E2
- Fusion: E1

Hepatitis C Virus E1E2 coevolution networks analysis unveils their functional dialog and structural organization

Identification

Molecular virology

Theoretical modelling

Function, molecular entry mechanism

Entry inhibitors development

Application
Protein coevolution signals are strong mediator of protein functions.

Such signals represent interesting tools to unveil viral protein dynamic rearrangements and uncover potential therapeutic targets.
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The BIS Model
A computational detection of amino acid co-evolution

- Allows identification of networks and dialogs within proteins
- Principle: Small protein fragments can be used as basic building blocks to reconstruct networks of co-evolving amino acids

Can we identify E1E2 functional dialogs faster and in a larger extent?
Can we predict potential E1E2 structural organization and rearrangements?
Application of BIS model to two envelope glycoproteins: HCV E1E2

<table>
<thead>
<tr>
<th>Number of sequenced protein</th>
<th>Number of clusters detected</th>
<th>Clusters location</th>
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</thead>
<tbody>
<tr>
<td>1a</td>
<td>25</td>
<td>intra-E1 (p&lt;0.05)</td>
</tr>
<tr>
<td>1b</td>
<td>15</td>
<td>intra-E1</td>
</tr>
<tr>
<td>1a</td>
<td>25</td>
<td>intra-E2 (p&lt;0.05)</td>
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<td>1b</td>
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<td>intra-E2</td>
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<tr>
<td>2a</td>
<td>15</td>
<td>intra-E1-E2 (p&lt;0.05)</td>
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<tr>
<td>2b</td>
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<td>intra-E1-E2</td>
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<td>2a</td>
<td>30</td>
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<tr>
<td>2b</td>
<td>15</td>
<td>inter-E1-E2</td>
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</tbody>
</table>

Clusters map important residues described in the literature.

Douam et al. in press.
E1E2 clusters map important residues described in the literature

<table>
<thead>
<tr>
<th>Genotype 1a clusters</th>
<th>Structural Clusters (folding, heterodimerisation, binding)</th>
<th>Membrane fusion Clusters</th>
<th>Multifunctional Clusters</th>
<th>Undefined role</th>
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<tbody>
<tr>
<td></td>
<td>2 X</td>
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<td></td>
<td></td>
<td>15 7</td>
</tr>
</tbody>
</table>

HCV fusion cluster support E1E2-interdependent rearrangements

Douam et al. In press.
HCV fusion cluster support E1E2 interdependent rearrangements

Douam et al. In press.

Soluble BL inhibits HCV entry

Douam et al., In Revision