Frequency of occurrence of HIV-1 superinfection in MSM

Laura Hebberecht, Leen Vancoillie, Marlies Schauvliege, Kenny Dauwe, Virginie Mortier, Chris Verhofstede
Introduction

• HIV-1 superinfection
  • Re-infection after initial infection

• Challenges
  • Methodology
  • Temporary observation
  • Intrasubtype >< Intersubtype
- Incidence rate of HIV-1 superinfection: 0-7%
  - ~ Population studied & detection method

*Redd AD, Quinn TC, Tobian AAR. Frequency and implications of HIV superinfection. The Lancet Infectious Diseases. 2013;13(7):622-8*
Study of HIV-1 superinfection

- Importance
  - Frequency of occurrence
  - In vivo model to study the host immune system
    - Viral immune escape
  - Recombination
    - CRF (88)

Los Alamos HIV sequence database
Patient selection

- MSM (2008-2013)
- Subtype B
- 2 plasma samples (> 6 m interval)
- 74 patients
Methods

- Roche 454 deep sequencing
  - *env* V3 region (228 nucleotides/76 amino acids)
  - 2 samples from each patient
  - 34 samples pooled per run

- Manual data cleaning, editing and aligning
  - BioEdit + in-house software

- Maximum Likelihood phylogenetic trees construction (PhyML)
Identification of Superinfection

• Tree visualisation in iTol

• Superinfected patient
  • At least 1 of the 2 samples
    → 2 or more monophyletic clusters
    → Bootstrap value ≥ 90%
    → Separated by sequences of ≥ 1 patient
Results

- Pt 1
- Pt 2
- Pt 3
- Pt 4

1st sample
2nd sample
Bootstrap ≥ 90%
Difference in mean pairwise genetic distance between both samples

Time interval between both samples (months)

-0.05

0

0.05

0.1

0.15

0.2

4 patients superinfected
## Confirmation of superinfection with longitudinal samples

<table>
<thead>
<tr>
<th>Patient ID</th>
<th>Sample dates</th>
<th>Date interval (days)</th>
<th>Virus population</th>
<th>% strain 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>08</td>
<td>6/06/2008</td>
<td>0</td>
<td>strain 1</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>20/02/2009</td>
<td>259</td>
<td>strain 1</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>22/02/2010</td>
<td>626</td>
<td>strain 1 + 2</td>
<td>42%</td>
</tr>
<tr>
<td></td>
<td>24/01/2011</td>
<td>962</td>
<td>strain 1 + 2</td>
<td>11%</td>
</tr>
<tr>
<td></td>
<td>19/12/2011</td>
<td>1291</td>
<td>strain 1 + 2</td>
<td>79%</td>
</tr>
<tr>
<td>16</td>
<td>26/01/2009</td>
<td>0</td>
<td>strain 1</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>9/06/2009</td>
<td>134</td>
<td>strain 1 + 2</td>
<td>35%</td>
</tr>
<tr>
<td></td>
<td>30/10/2009</td>
<td>277</td>
<td>strain 1 + 2</td>
<td>62%</td>
</tr>
<tr>
<td></td>
<td>19/01/2010</td>
<td>358</td>
<td>strain 1 + 2</td>
<td>74%</td>
</tr>
<tr>
<td>35</td>
<td>24/02/2010</td>
<td>0</td>
<td>strain 1</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>10/03/2010</td>
<td>14</td>
<td>strain 1</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>16/06/2010</td>
<td>112</td>
<td>strain 1 + 2</td>
<td>4%</td>
</tr>
<tr>
<td></td>
<td>9/01/2013</td>
<td>1050</td>
<td>strain 1 + 2</td>
<td>23%</td>
</tr>
<tr>
<td>66</td>
<td>19/03/2012</td>
<td>0</td>
<td>strain 1</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>20/02/2013</td>
<td>338</td>
<td>strain 1 + 2</td>
<td>23%</td>
</tr>
<tr>
<td></td>
<td>3/07/2013</td>
<td>471</td>
<td>strain 1 + 2</td>
<td>6%</td>
</tr>
<tr>
<td></td>
<td>27/11/2013</td>
<td>618</td>
<td>strain 1 + 2</td>
<td>19%</td>
</tr>
<tr>
<td></td>
<td>26/03/2014</td>
<td>737</td>
<td>strain 1 + 2</td>
<td>39%</td>
</tr>
</tbody>
</table>
Viral load evolution in superinfected patients

- Strain 1
- Strain 1 + 2

+ 2.19
+ 0.94
Conclusions

• Frequency of occurrence of HIV-1 superinfection (intrasubtype)
  • 4/74 patients = 5.4%
  • Likely an underestimation

• Clinical consequences
  • Viral load increase

• Baseline drug resistance analysis
  • Sanger sequencing
    → Missing one of the variants
    → Deep sequencing (in high risk populations)
Acknowledgements

• AIDS Reference Laboratory
  Kenny Dauwe, Els Demecheeleer, Sylvie Dinakis, Laura Hebberecht, Virginie Mortier, Marlies Schauvliege, Delfien Staelens, Leen Vancoillie, Chris Verhofstede

• AIDS Reference Centre