Herpes virus reactivation in brain-injured patients: role and mechanisms of CD4 T cell defects. The IBIS virus study

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Déclaration d’intérêts de 2014 à 2017

- Intérêts financiers : néant
- Liens durables ou permanents : néant
- Interventions ponctuelles : néant
- Intérêts indirects : MSD
Critical-illness related immunosuppression / Virome

Hotchkiss et al. Nature Immunol 2013
De Vlaminck et al. Cell 2013
The IBIS Virus project

Herpes simplex Virus

249 patients
- 159 Traumatic brain injuries
- 51 SubArachnoid Hemorrhage
- 39 Stroke

77 (32%) serology negative
- 2 (3%) in-ICU acquisition
  - 2 blood
  - 2 lungs
- 11 (14%) in-ICU death
- 12 (16%) death (6 months)

172 (71%) serology positive
- 104 (60%) without reactivation
  - NA blood
  - NA lungs
- 18 (17%) in-ICU death
- 20 (19%) death (6 months)
- 68 (40%) with reactivation
  - 13 blood
  - 66 lungs
- 17 (25%) in-ICU death
- 20 (29%) death (6 months)
Risk of HSV and CMV reactivations in 249 brain-injured patients

**Graph Details:**
- **HR 2.4 (95% CI 1.5-4.0), p<0.0001**
- **Probability of viral reactivation**
- **Days**
- **HSV sero-positive**: n=172
  - 0: 124
  - 7: 104
- **CMV sero-positive**: n=91
  - 0: 87
  - 14: 77
Association of HSV reactivation with poor in-ICU outcomes

B. Probability of survival

<table>
<thead>
<tr>
<th>Days</th>
<th>HSV sero-negative (n=77)</th>
<th>Without reactivation (n=104)</th>
<th>With reactivation (n=68)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>100</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>30</td>
<td>80</td>
<td>75</td>
<td>80</td>
</tr>
<tr>
<td>60</td>
<td>65</td>
<td>60</td>
<td>65</td>
</tr>
<tr>
<td>90</td>
<td>50</td>
<td>45</td>
<td>50</td>
</tr>
</tbody>
</table>

HR 1.4 (95% CI 0.7-2.7), p=0.13

C. Probability of ICU hospitalization

<table>
<thead>
<tr>
<th>Days</th>
<th>HSV sero-negative (n=77)</th>
<th>Without reactivation (n=104)</th>
<th>With reactivation (n=68)</th>
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<td>0</td>
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<td>50</td>
<td>45</td>
<td>50</td>
</tr>
</tbody>
</table>

HR 0.6 (95% CI 0.4-0.8), p=0.005
Association of CMV reactivation with outcomes
HSV reactivations are associated with poor neurological outcome

<table>
<thead>
<tr>
<th></th>
<th>Bad neurological recovery (GOS-E 1-5)</th>
<th>Good neurological recovery (GOS-E 6-8)</th>
<th>p-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients</td>
<td>163</td>
<td>86</td>
<td>0.002</td>
</tr>
<tr>
<td>Age, years</td>
<td>53 (34-64)</td>
<td>42 (27-56)</td>
<td></td>
</tr>
<tr>
<td>Male, yes</td>
<td>125 (77)</td>
<td>54 (63)</td>
<td>0.02</td>
</tr>
<tr>
<td>Pathology</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trauma</td>
<td>101 (62)</td>
<td>58 (67)</td>
<td>0.61</td>
</tr>
<tr>
<td>Subarachnoid hemorrhage</td>
<td>34 (21)</td>
<td>17 (20)</td>
<td></td>
</tr>
<tr>
<td>Stroke</td>
<td>28 (17)</td>
<td>11 (13)</td>
<td></td>
</tr>
<tr>
<td>Glasgow Coma Scale</td>
<td>5 (3-8)</td>
<td>8 (5-10)</td>
<td>0.0004</td>
</tr>
</tbody>
</table>

**Herpes simplex virus reactivation, yes**

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<tr>
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<th>Bad neurological recovery (GOS-E 1-5)</th>
<th>Good neurological recovery (GOS-E 6-8)</th>
<th>p-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>HSV sero-positive</td>
<td>55 (34)</td>
<td>13 (15)</td>
<td>0.0009</td>
</tr>
<tr>
<td>Lung reactivation, yes</td>
<td>55 (34)</td>
<td>13 (15)</td>
<td>0.002</td>
</tr>
<tr>
<td>Blood reactivation, yes</td>
<td>12 (7)</td>
<td>2 (2)</td>
<td>0.10</td>
</tr>
</tbody>
</table>

**Cytomegalovirus reactivation, yes**

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<th>Good neurological recovery (GOS-E 6-8)</th>
<th>p-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>HSV sero-positive</td>
<td>55 (34)</td>
<td>13 (15)</td>
<td>0.002</td>
</tr>
<tr>
<td>Lung reactivation, yes</td>
<td>55 (34)</td>
<td>13 (15)</td>
<td>0.002</td>
</tr>
<tr>
<td>Blood reactivation, yes</td>
<td>12 (7)</td>
<td>2 (2)</td>
<td>0.10</td>
</tr>
<tr>
<td>CMV sero-positive</td>
<td>10</td>
<td>2</td>
<td>0.18</td>
</tr>
<tr>
<td>Lung reactivation, yes</td>
<td>9</td>
<td>2</td>
<td>0.24</td>
</tr>
<tr>
<td>Blood reactivation, yes</td>
<td>6</td>
<td>1</td>
<td>0.25</td>
</tr>
</tbody>
</table>

**Virus reactivation (Herpes simplex virus OR Cytomegalovirus), yes**

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<thead>
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<th>Good neurological recovery (GOS-E 6-8)</th>
<th>p-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>HSV reactivation</td>
<td>61 (37)</td>
<td>15 (17)</td>
<td>0.01</td>
</tr>
</tbody>
</table>
Working hypothesis:

BI-induced immunosuppression is responsible for Herpes Simplex Virus reactivation

WP 1: Increased viral replication or loss of immune control?
Decreased circulating viral load of Torque Teno Virus in BI patients without HSV reactivation
Working hypothesis:

BI-induced T cell defects are responsible for Herpes Simplex Virus reactivation

WP 2: Does the number of T cells matter?
Lymphopenia after brain injury
No proof of apoptosis / necrosis of lymphocytes in Spleen Tissue after brain-injury

Activated Caspase 3

Sham
Brain injury

Healthy human
Brain-dead patient

Classical histology
Activated Caspase 3
Lymphopenia is not associated with HSV reactivation, just a marker of severity?
Working hypothesis:

BI-induced T cell defects are responsible for the severity of HSV reactivation

WP 3 : Do functions of T cells matter ?
Defective production of IFN-γ by memory CD4 T cells
Altered T cells are not exhausted

- CD4 CFSE en %
  - Healthy
  - Patient

- CD8 CFSE en %
  - Healthy
  - Patient

- PD1 MFI
  - CD4
  - CD8
  - CD3
Brain injury alters the transcriptional factor program of CD4 T cells.
CD4 T cells re-programming is associated with immunosuppression and HSV reactivation
Working hypothesis:

BI-induced T cell defects are responsible for the severity of HSV reactivation

WP 4: Several mechanisms the CD4 T cells reprogramming?
CD4 T cells reprogramming and soluble circulating mediators?
CD4 T cells reprogramming is continuously maintained by CD14+ cells, and other mediators are involved.
CD4 T cells reprogramming is partially IL-4 dependent, but not TGF-β dependent.
Conclusions

• HSV reactivations are common in brain injured patients, associated with poor outcome

• Long-lasting lymphopenia
  • No apoptosis (caspase ko mice ?)
  • Not associated with HSV reactivation

• Long lasting TF reprogramming of CD4 T cells after BI
  • No exhaustion

• Altered TF reprogramming of CD4 T cells is associated with HSV reactivation
  • Microenvironment : CD14, IL-4.
  • Other mechanisms under the scope : epigenetic modifications ? CD14 cells ?
  • Role of direct activation by brain-released DAMPs ?
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