Assessing prion infectivity in the skin of sporadic CJD patients

Wenquan Zou, MD/PhD

Departments of Pathology and Neurology
National Prion Disease Pathology Surveillance Center
Case Western Reserve University
Cleveland, Ohio, USA
Email: wxz6@case.edu
Background

Cunningham et al., 2004

Table 2. Bioassay results for greater kudu tissues injected into C57Bl-J6 mice

<table>
<thead>
<tr>
<th>Kudu identification no.</th>
<th>Tissue injected</th>
<th>Positive mice/total</th>
<th>Mean survival period post injection (days) ± SD</th>
<th>Survival period range (d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1212</td>
<td>Rostral cerebrum</td>
<td>13/20</td>
<td>595±84</td>
<td>426-745</td>
</tr>
<tr>
<td></td>
<td>Cranial thoracic spinal cord</td>
<td>19/20*</td>
<td>557±121</td>
<td>413-621</td>
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<tr>
<td></td>
<td>Lumbar spinal cord</td>
<td>15/19</td>
<td>521±69</td>
<td>432-634</td>
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<td></td>
<td>Spleen</td>
<td>3/11*</td>
<td>619±41</td>
<td>773-651</td>
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<tr>
<td></td>
<td>Retropharyngeal lymph node (biceps brachii + vastus lateralis)</td>
<td>6/11*</td>
<td>784±77</td>
<td>691-921</td>
</tr>
<tr>
<td></td>
<td>Skin (flank)</td>
<td>2/18*</td>
<td>713±22</td>
<td>697-728</td>
</tr>
</tbody>
</table>

Thomzig et al., 2007

Notari et al., 2010
**PrPres in patient-specific skin-derived fibroblasts**

<table>
<thead>
<tr>
<th></th>
<th>E200K</th>
<th>D178N</th>
<th>F198S</th>
<th>E200K</th>
<th>sCJD</th>
<th>5-Insert</th>
<th>CTL</th>
<th>CTL</th>
<th>AD</th>
<th>Brain</th>
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</table>

- Untreated
- +PK
- +PNGase F

**kDa**

<table>
<thead>
<tr>
<th>27-</th>
<th>34-</th>
<th>19-</th>
<th>17</th>
</tr>
</thead>
</table>

**Brain**

- E200G
- MD
- 2-Insert
- sCJD
- APN
- PLY
- sCJD
Background/Aim

- Histories of non-CNS-surgeries could be a risk factor of sporadic CJD

- There are dermal prions in sCJD patients, which are infectious and can be a biomarker for diagnosis of human prion diseases
Subjects and Methods

Skin

Western blotting

Bioassay

RT-QuIC
Western blotting of PrPSc in skin of vCJD patients

**US vCJD**

<table>
<thead>
<tr>
<th>PK</th>
<th>US vCJD</th>
<th>PK + PNGase F</th>
</tr>
</thead>
<tbody>
<tr>
<td>brain</td>
<td>skin</td>
<td>skin</td>
</tr>
<tr>
<td>27-</td>
<td>17-</td>
<td>27-</td>
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</tbody>
</table>

Expos. time

- 5 min
- 50 min

**US vCJD**

+PK

<table>
<thead>
<tr>
<th>PNGase F</th>
<th>UK vCJD</th>
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</thead>
<tbody>
<tr>
<td>-</td>
<td>Brain</td>
</tr>
<tr>
<td>+</td>
<td>Skin</td>
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</table>

-27
-19
Detection of PrPSc in the skin of sCJD patients

+PK

PK + PNGase F
RT-QuIC assay of PrP seeding activity in the skin and brain of CJD patients

- **Patient 1**: Brain $10^{-5}$, S1 Skin $10^{-3}$, P4 Skin $10^{-3}$
- **Patient 2**: Brain normal
- **Patient 3**: P4 Skin, AD $10^{-4}$
- **Patient 4**: Brain normal
- **Patient 5**: S1 & P4 Skin: normal $10^{-2}$ Patients 8-17
- **Patient 6**: (vCJD)
- **Patient 7**: (vCJD)
RT-QuIC analysis of PrP seeding activity in the skin and brain of CJD patients

![Graph showing ThT fluorescence over hours for different groups: CJD LB (n=12), CJD Apex (n=16), CJD Ear (n=16), Non-CJD LB (n=4), Non-CJD Apex (n=5), and Non-CJD Ear (n=5). The graph includes error bars for each data point.](image-url)
RT-QuIC analysis of PrP seeding activity in the skin and brain of CJD patients

![Graph showing maximum ThT fluorescence (Mean % ± SD) for sCJD and Non-CJD patients compared to overall outcome.](image)
Bioassay of sCJD skin samples with humanized transgenic mice

- **TgNN6H/sCJDMM2 (5/5)**
- **TgWV/sCJDVV1 (7/7)**
- **TgWV/non-CJDVV (0/7)**

Days post-inoculation vs. Percentage survival
Western blotting and histology of TgNN6h mouse brains inoculated with skin samples
Western blotting and histology of TgWV mouse brains inoculated with skin samples

<table>
<thead>
<tr>
<th>Skin-inoculated</th>
<th>Brain Inoc</th>
<th>NonCJD Inoc</th>
<th>sCJD Brain</th>
</tr>
</thead>
<tbody>
<tr>
<td>#6017-1 &amp; #6017-2</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>#6017-2 &amp; #6017-3 &amp; #6017-4</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

- PK: - means negative, + means positive
- Lanes: 1-16
- Images a, b, c, d, e, f, g, h are histological sections showing different regions or stages of the study.
Conclusions

- Prion seeding activity is detectable in the skin of sCJD patients, which may set up a basis for developing skin-based antemortem and postmortem RT-QuIC assay for CJD
- Dermal prions in sCJD are infectious, which may raise concerns about the potential for iatrogenic sCJD transmission through skin during non-CNS surgeries
- In no way does our study imply that prion transmission can occur via casual contact
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Participants