Clonal spread of MRSA CC398 sublineages within and between Danish pig farms

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Publication date:
2010

Document Version
Early version, also known as pre-print

Citation for published version (APA):
Clonal spread of MRSA CC398 sublineages within and between Danish pig farms

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Introduction
In 2008 participants at a national conference for Danish pig producers were screened for MRSA and 19 out of 759 (2.5%) individuals resulted to be positive for MRSA CC3981, the MRSA lineage associated with livestock. One year after the conference, we visited the farms of six of the 19 positive farm workers and investigated the occurrence of MRSA in different age groups of pigs and environmental sites.

Objectives
1. To evaluate the frequency and diversity of MRSA CC398 within and between Danish pig farms.
2. To assess whether porcine and environmental isolates were epidemiologically related to the human isolates collected in the previous study.

Sampling
A total of 311 porcine nasal swabs and 80 dust samples were collected from the six farms including:
- 41 pregnant sows shortly before farrowing
- 69 sows and 160 piglets after farrowing
- 90 growers 3 weeks after weaning
- 60 finishers shortly before slaughtering.
- 80 dust samples collected from three production units (i.e. farrowing, growing and finishing)

MRSA isolation & Identification

<table>
<thead>
<tr>
<th>Enrichment</th>
<th>Brilliance</th>
<th>Oxoida</th>
<th>5% Blood agar</th>
<th>PFGE using CRIP</th>
<th>spa-typing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mueller Hinton broth 6.5% NaCl</td>
<td>MRSA agar</td>
<td>(Presumptive MRSA)</td>
<td>MecA &amp; CC398-specific PCR</td>
<td>spa-typing</td>
<td></td>
</tr>
</tbody>
</table>

Acknowledgments
This study was supported by the EU-HEALTH Project PILGRIM (223050) of the Seventh Framework Programme (FP7)

Results
1. MRSA CC398 was isolated from all farms, pig age groups and production units
2. MRSA CC398 frequency within farms varied between 50 and 82% of the samples tested
3. The frequency of MRSA CC398 carriage was lower in pregnant sows (34.1%) compared with farrowed sows (76%), piglets (68%), growers (98%) and finishers (66%) (Figure 1)
4. MRSA-positive dust samples were less frequent in the units where pregnant sows were kept compared with other production units (Table 1).
5. Isolates from different farms displayed differences in their PFGE profiles (up to 20% diversity)
6. Within each farm, the PFGE and spa-types displayed by porcine isolates were indistinguishable or closely related to those obtained from human isolates (Figure 2)
7. Forty-two of the 53 isolates (79%) typed by spa-typing belonged to t034. The remaining isolates were spa-type 011 and originated almost exclusively (10 out of 11 isolates) from one farm located on the north-west region of Jutland (Figure 3). Human isolates had the same spa-type as that found in the farm.

Table 1. Frequency of MRSA CC398 among pigs belonging to different age groups, and dust samples

<table>
<thead>
<tr>
<th>Farm</th>
<th>Dust</th>
<th>Pregnant</th>
<th>Farrowed</th>
<th>Piglets</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3/5</td>
<td>7/10</td>
<td>10/10</td>
<td>10/10</td>
</tr>
<tr>
<td>2</td>
<td>3/5</td>
<td>3/10</td>
<td>9/10</td>
<td>7/10</td>
</tr>
<tr>
<td>3</td>
<td>4/5</td>
<td>2/6</td>
<td>8/10</td>
<td>8/10</td>
</tr>
<tr>
<td>4</td>
<td>0/5</td>
<td>1/10</td>
<td>4/10</td>
<td>5/10</td>
</tr>
<tr>
<td>5</td>
<td>2/5</td>
<td>0/2</td>
<td>6/10</td>
<td>2/10</td>
</tr>
<tr>
<td>6</td>
<td>4/5</td>
<td>1/3</td>
<td>9/10</td>
<td>9/10</td>
</tr>
<tr>
<td>Total</td>
<td>16/30</td>
<td>14/41</td>
<td>46/60</td>
<td>41/60</td>
</tr>
</tbody>
</table>

Figure 1. Frequency of MRSA CC398 found in dust from three production units, and animals from 5 age groups.

Figure 2. PFGE dendrogram of MRSA CC398 isolated from human and animals in farms 1-5.

Figure 3. Distribution of farms (1-6) and spa-types from human, pig and environmental samples.

Conclusions
1. The study provided evidence of the association between human and pig isolates at the farm
2. Frequency of MRSA CC398 within farms seems to increase along the production flow, i.e. it is more frequent in the growing and finishing units than in the farrowing units
3. t034 is the predominant spa-type in Danish pig farming
4. Each farm is associated with a specific CC398 sublineage that differs from those occurring at other farms
5. Such CC398 sublineage may persist for long periods of time (at least one year) within the farm