Geographical clustering of mecC-positive Staphylococcus aureus from bovine mastitis in France

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Keywords: MRSA, veterinary, cattle

Sir,

Staphylococcus aureus is a major bovine pathogen, accounting for ~10% of all pathogenic bacteria isolated each year from diseased cattle through the Resapath surveillance network in France (www.resapath.anses.fr). Most S. aureus strains are responsible for mastitis, a disease causing a major economic burden in the dairy industry. Until recently, mastitis-associated methicillin-resistant S. aureus (MRSA) reported in Europe classically belonged to the livestock-associated clonal complex (CC) 398.1 However, the recent description of a new mecA variant (namely mecC) associated with non-CC398 clones in bovine mastitis and humans in England and Denmark drew attention to the cattle industry.2 Here, following the first description of bovine mecC-positive MRSA in France, we ascertain the prevalence and possible emergence of mecC-positive S. aureus clones in French cattle.

Between 2011 and mid-2013, 1549 coagulase-positive staphylococci (657 in 2011, 704 in 2012 and 188 in early 2013) causing bovine mastitis were gathered by the 65 veterinary laboratories belonging to the Resapath network. All non-duplicate presumptive MRSA, i.e. cefoxitin non-susceptible isolates (diameter <27 mm; Table S1, available as Supplementary data at JAC Online), were sent to the National Reference Laboratory for Antimicrobial Resistance at ANSES Lyon for further analysis. Antimicrobial susceptibility was performed using disc diffusion and interpreted according to the recommendations of the Antibiogram Committee of the French Society of Microbiology (http://www.sfm-microbiologie.fr). Sixteen antibiotics (Table S1) of veterinary and/or human interest were tested, using S. aureus ATCC 25923 as a quality control strain. Methicillin resistance was confirmed using specific mecA/mecC PCRs.2 A total of 10 MRSA isolates (7 in 2011 and 3 in 2013) were identified, further characterized and assigned to specific clones using a microarray-based assay (Alere StaphType DNA microarray; Alere Technologies GmbH, Jena, Germany) (Table 1).

One CC5-SCmec IV isolate belonging to the ‘new paediatric’ clone, classically associated with hospital-acquired infections, was collected in 2011 and most likely exemplifies a human-to-animal transmission. In addition to the mecA gene, this isolate presented the aadD gene (coherent with the antibiotic profile) and several enterotoxin genes whose expression would be a risk for human health in case of raw milk consumption (Table 1). Five isolates collected in 2011 belonged to the CC398 MRSA clone [spa types t011 (n=3) and t899 (n=2)]. All were devoid of enterotoxin genes but displayed the tetracycline resistance gene tet(M) typically associated with CC398 isolates of animal origin. Two isolates additionally harboured the tet(K) gene and one isolate displayed the fexA gene conferring chloramphenicol/ florfenicol resistance (Table 1). Interestingly, the t899 isolates were positive for the immune evasion cluster type (IEC) B (sak, chp and scp but not sea genes), which is considered a human virulence marker. Thus, the codetection of the tet(M) gene and the IEC cluster suggests the humanization of CC398 animal isolates.

The four remaining strains (one collected in 2011 and three in 2013) belonged to the CC130-SCCmec XI MRSA clone, harboured the spa type t1736 (allelic profile 04-82-17-25-16-17) and were positive for the mecC gene. All strains showed identical microarray profiles matching the profiles previously reported for mecC-positive CC130 isolates.4 Of note, all isolates originated from farms located in the countryside within a 30 km perimeter around Nancy (north-east France) and two isolates collected in 2011 and 2013 were from the same farm, although from different animals. Interestingly, the first French case of bovine mecC-positive MRSA, displaying the closely related spa type t843 (04-82-17-25-17-25-16-17), originated from the same tightly focused geographic area.3 PFGE analyses using Smal (data not shown) revealed identical or related (>80% homology according to Tenover’s criteria) profiles for all four isolates and the index case previously published.

Although MRSA are rare among clinical staphylococci causing bovine mastitis in France (10/1549, 0.6%) as previously suggested,5 the proportion of CC130-mecC isolates was unexpectedly high (4/10). Moreover, the occurrence of all mecC-positive MRSA in a very small geographic area is intriguing and gives credit to the recent hypothesis of a possible clustering of mecC-positive cases.6 This might even be the hidden part of the iceberg since, in France, susceptibility tests are only performed on a limited number of mastitis-causing S. aureus isolates. Also, the detection of two mecC-positive isolates on the same farm in 2011 and 2013...
Table 1. Characteristics of the 10 MRSA isolates

<table>
<thead>
<tr>
<th>Isolate</th>
<th>Year</th>
<th>Geographical origin</th>
<th>mec gene</th>
<th>Assigned clonea</th>
<th>SCCmec cassette</th>
<th>spa type</th>
<th>Resistance genes</th>
<th>Enterotoxins</th>
</tr>
</thead>
<tbody>
<tr>
<td>26988</td>
<td>2011</td>
<td>Aveyron</td>
<td>mecA</td>
<td>CC398</td>
<td>V</td>
<td>t011</td>
<td>tet(M), tet(K)</td>
<td>none</td>
</tr>
<tr>
<td>26998</td>
<td>2011</td>
<td>Côtes d’Armor</td>
<td>mecA</td>
<td>CC398</td>
<td>V</td>
<td>t011</td>
<td>tet(M), tet(K)</td>
<td>none</td>
</tr>
<tr>
<td>27243</td>
<td>2011</td>
<td>Meurthe et Moselle</td>
<td>mecC</td>
<td>CC130</td>
<td>XI</td>
<td>t1736</td>
<td>none</td>
<td>none</td>
</tr>
<tr>
<td>29676</td>
<td>2011</td>
<td>Côtes d’Armor</td>
<td>mecA</td>
<td>CC5 paediatric clone</td>
<td>IV</td>
<td>t002</td>
<td>aadD</td>
<td>sea, seg, sei, sem, seo, seu</td>
</tr>
<tr>
<td>31745b</td>
<td>2011</td>
<td>Orne</td>
<td>mecA</td>
<td>CC398</td>
<td>IV</td>
<td>t011</td>
<td>tet(M)</td>
<td>none</td>
</tr>
<tr>
<td>31747b</td>
<td>2011</td>
<td>Orne</td>
<td>mecA</td>
<td>CC398</td>
<td>IV</td>
<td>t899</td>
<td>tet(M), fexA</td>
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<tr>
<td>31933</td>
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<td>Midi-Pyrénées</td>
<td>mecA</td>
<td>CC398</td>
<td>IV</td>
<td>t899</td>
<td>tet(M), vga(A)</td>
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</tr>
<tr>
<td>33619</td>
<td>2013</td>
<td>Meurthe et Moselle</td>
<td>mecC</td>
<td>CC130</td>
<td>XI</td>
<td>t1736</td>
<td>none</td>
<td>none</td>
</tr>
<tr>
<td>33620</td>
<td>2013</td>
<td>Meurthe et Moselle</td>
<td>mecC</td>
<td>CC130</td>
<td>XI</td>
<td>t1736</td>
<td>none</td>
<td>none</td>
</tr>
<tr>
<td>33621</td>
<td>2013</td>
<td>Meurthe et Moselle</td>
<td>mecC</td>
<td>CC130</td>
<td>XI</td>
<td>t1736</td>
<td>none</td>
<td>none</td>
</tr>
</tbody>
</table>

aOn the basis of Alere StaphyType DNA microarray.

bThese two isolates originated from different distant farms.

suggests the persistence capacity of the mecC-positive CC130 clone over years, even though repeated introductions of mecC-positive isolates cannot be excluded. Finally, the description of divergent but closely related spa types and PFGE patterns among the mecC-positive isolates potentially reflects local microevolution or the circulation of different subclones. No epidemiological link was evidenced between the three colonized farms, neither at the animal level (exchanges and contiguous fields) nor at the farmer level (shared employees and relatives). Also, no human mecC-positive MRSA was reported in the nearby hospitals (F. Laurent, personal observation). Since the dynamics of expansion of this clone is not known, further studies are required to explore the capacity of mecC-positive isolates to persist and propagate on and between cattle farms and eventually disseminate from animals to humans.

Acknowledgements

We gratefully thank all peripheral laboratories of the Resapath network involved in the study.

Funding

This work was supported by the French Agency for Food, Environmental and Occupational Health Safety (ANSES), the French Ministry of Health and the French Institute for Public Health Surveillance (InVS).

Transparency declarations

None to declare.

Supplementary data

Table S1 is available as Supplementary data at JAC Online (http://jac.oxfordjournals.org/).

References


Emergence of Escherichia coli ST131 sub-clone H30 producing VIM-1 and KPC-3 carbapenemases, Italy

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