First community-acquired meticillin-resistant *Staphylococcus aureus* in Malaysia

Recent reports of increasing community-acquired meticillin-resistant *Staphylococcus aureus* (CA-MRSA) infection in our neighbouring country Singapore (Hsu et al., 2006) indicate the possibility of such a serious infection in Malaysia as well. This prompted us to investigate the presence of CA-MRSA strains in the Malaysian population. For the purposes of this study, CA-MRSA was defined as an isolate from a subject who had no health-care exposure or antibiotic therapy in the previous 6 months which contained staphylococcal chromosomal cassette *mec* (SCC*mec*) type IV or V. The presence of PVL was an added value.

A nasal culture survey among 100 university students from the faculty of medicine and health sciences (Universiti Putra Malaysia, Serdang) was conducted with ethical approval to screen for the presence of CA-MRSA. All participating subjects were students who did not attend clinical wards. Moreover, the faculty where the screening took place was not directly affiliated to the hospital, thereby minimizing the risk of acquisition from the hospital.

Only subjects without obvious risk factors (exposure to the health-care system and usage of any antibiotic during the previous 6 months) for CA-MRSA carriage or infection were included in the study. *S. aureus* was isolated from the nasal swabs of participating subjects by standard procedures. Among the 100 subjects, 26 (26 %) were colonized with *S. aureus*, which is in agreement with previously reported data (Choi et al., 2006).

The susceptibilities of the *S. aureus* carriage isolates to oxacillin, erythromycin, gentamicin, rifampicin, fusidic acid, mupirocin and vancomycin were determined by Kirby–Bauer disc diffusion and interpreted according to the CLSI (2005) standards. Antibiotic susceptibility testing revealed one (3.8 %) isolate to be resistant to rifampicin and erythromycin while six (23 %) were intermediately resistant to erythromycin and fusidic acid (Table 1). All isolates were susceptible to gentamicin, mupirocin and vancomycin. Antibiotic susceptibility profiles indicated that 85 % of the carriage isolates were sensitive to all antibiotics tested. Not a single strain was resistant to more than three classes of antibiotics such as β-lactams, macrolides and fusidic acid.

MRSA screening by oxacillin susceptibility testing and *mec*A PCR revealed three (11.5 %) positives. This MRSA carriage rate [3/100 subjects (3 %) or 3/26 *S. aureus* isolates (11.5 %)] is higher than that suggested by previous data from a recent study from Malaysia (1.2 % of 81 *S. aureus* isolates from healthy individuals) or the United States (0.8 % of 2964 *S. aureus* isolates) (Choi et al., 2006; Kuehnert et al., 2006). Surprisingly, the carriage rate defined in this study is comparable to that of New York state prisoners (Lowey et al., 2007).

Further characterization of the three MRSA isolates by SCC*mec* typing (Oliveira & de Lencastre, 2002) and multilocus sequence typing (MLST) identified all three isolates as putative CA-MRSA (Table 1). One isolate, ST80-MRSA-IVa, gave a positive signal in Panton–Valentine leukocidin (PVL) toxin gene PCR (Lina et al., 1999). Isolates 5 and 36 harboured *PVL* gene and *mec*A and could be considered as putative *PVL*+ CA-MRSA strains with CA-MRSA-IVa type.

### Table 1. Antimicrobial susceptibility profiles and molecular patterns of *S. aureus* and CA-MRSA

<table>
<thead>
<tr>
<th>No.</th>
<th>Strain</th>
<th>Antimicrobial susceptibility*</th>
<th>Molecular pattern</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Ox</td>
<td>Ery</td>
</tr>
<tr>
<td>1</td>
<td>Isolate 5</td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td>2</td>
<td>Isolate 15</td>
<td>R</td>
<td>I</td>
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<tr>
<td>3</td>
<td>Isolate 19</td>
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<td>5</td>
<td>Isolate 24</td>
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<td>S</td>
</tr>
<tr>
<td>6</td>
<td>Isolate 36</td>
<td>R</td>
<td>I</td>
</tr>
<tr>
<td>7</td>
<td>Isolate 38</td>
<td>I</td>
<td>S</td>
</tr>
<tr>
<td>8</td>
<td>Isolate 52</td>
<td>I</td>
<td>S</td>
</tr>
</tbody>
</table>

NT, Not tested.

*Ox, Oxacillin; Ery, erythromycin; Gen, gentamicin; Van, vancomycin; Mup, mupirocin; Fus, fusidic acid; Rif, rifampicin; S, sensitive; R, resistant; I, intermediate.

†SCC*mec*, staphylococcal chromosomal cassette *mec*.

‡PVL, Panton–Valentine leukocidin.

§MLST, Multilocus sequence typing allelic profile.

‖ST, Sequence type based on multilocus sequence typing.
SCC\textit{mec}-V but were negative in the PVL PCR. Although the isolates studied were not from people with infections, the presence of PVL would suggest a certain degree of virulence in this isolate. If the strain becomes pathogenic, it may contribute to dissemination in the community. MLST identified the MRSA-IVa strain as ST80 (European clone), while the MRSA-V isolates did not show a perfect match with the sequence types deposited in the MLST database. ST1004 (8-163-129-19-6-125-117) was assigned to the CA-MRSA-V strain from Malaysia. The two ST1004 isolates were isolated from students taking the same course, hence there could have been cross-transmission. The different antibiogram pattern for both strains, particularly with erythromycin and fusidic acid, indicates that although they share the same sequence type, antibiotic susceptibility could vary. The unique sequence type of these MRSA isolates confirms that new strains of CA-MRSA are continuously emerging and disseminating.

In conclusion, we report a relatively high incidence of CA-MRSA with SCC\textit{mec} type IVa or V among healthy Malaysian carriers. The PVL-negative ST1004-MRSA-V may be an important new clone in Malaysia. The presence of PVL-positive ST80-MRSA-IVa in Malaysia indicates the spread of the ecologically successful European clone to the Asian continent.

Isolation of CA-MRSA from healthy university students demonstrates the importance of continued surveillance and decolonization of colonized students in our institution. High-level surveillance of clinical samples for MRSA is required to define the instance and spread of CA-MRSA in hospital settings and among patients and health-care workers.

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