**Introduction:**

SCCmec (staphylococcal cassette chromosome mec) elements are large, mobile genetic elements in staphylococci that carry methicillin resistance genes mecA or mecC. Additionally, they harbor recombinase and accessory genes. Twelve main SCCmec types and several variants have been described based on the identity of the mec complex and of the ccr recombinase genes [http://www.sccmec.org/Pages/SCCmecTypesEn.html](http://www.sccmec.org/Pages/SCCmecTypesEn.html). Methicillin-resistant Staphylococcus aureus (MRSA) has been for many years a serious and global problem to infection control and healthcare. While there is a multitude of data available on the population structure of MRSA and on its changes from several regions, such as Western Europe, Australia, Japan, and the US, there are only limited data from other parts of the world including Pakistan. The prevalence of MRSA in Pakistan is known to be high, but very few studies have described the molecular epidemiology of different MRSA clones circulating in the country applying molecular typing tools.

**Objectives:**

To characterize, monitor and to type *S. aureus* isolates, nosocomial MRSA infections from dialysis/kidney transplant units of two tertiary care hospitals of Rawalpindi in Pakistan were investigated.

**Method:**

<table>
<thead>
<tr>
<th>CC</th>
<th>Strain</th>
<th>n</th>
<th>%</th>
<th>SCCmec type</th>
<th>Other resistance genes</th>
</tr>
</thead>
<tbody>
<tr>
<td>CC1</td>
<td>5 &lt;/br&gt; 5</td>
<td>5</td>
<td>5</td>
<td>SCCmecIV</td>
<td>blaZ, cadX, aphA3, aacA2, tetK, dfrA</td>
</tr>
<tr>
<td>CC2</td>
<td>5 &lt;/br&gt; 5</td>
<td>5</td>
<td>5</td>
<td>SCCmecIV</td>
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</table>

**Results:**

Six CCs and nineteen strains (as defined by CC affiliation, toxin gene carriage and SCCmec subtype) were identified. All isolates were mecA positive and resistant to all tested beta-lactams. MRSA with mecC were not identified. Furthermore, a high PVL rate (34%) was observed, with PVL-positives mainly belonging to the CC722 lineage (Table 1). The most common (Figure 1) strains were i) PVL-positive CC722-MRSA-V, "Bengal Bay Clone" (ten isolates, 23%), ii) ST239-MRSA-III [ccrC] (five isolates; 11%) and iii) CCE-MRSA-IV strain similar to UK EMRSA-15 as well as CC6-MRSA-IV "WA MRSA-51" (both with four isolates; 9% each). A detailed strain description and an overview of the typed strains and relevant target genes are listed in Table 1. The population structure of MRSA observed herein indicated some connections to the Middle Eastern/Arabian Gulf region. Some isolates were assigned to strains known from there such as here CC5-MRSA-IV [MW2-like] "Maltese Clone", CC6-MRSA-IV, and tetracycline-positive CC22-MRSA-II "Gaza Clone". This may be due to the fact that approximately 3,600,000 Pakistani citizens are currently working in the Middle Eastern/Arabian Gulf region. Furthermore, two variants of the ancient pandemic strain ST239-II were identified, which differed, for instance, in the presence of mercury resistance genes merA/B and SCC-associated cadmium resistance gene cadB (as well as of the gene encoding surface-anchored protein X [sasX] and in some SCCmec genes. One isolate (merA/B and sasX positive) showed a SCCmec pattern as predicted for BMB393, GenBank CP005288.1 and CUMU_HX2007, GenBank ACM2. Therefore, this clone might be introduced into the clade within ST239 together with South American, Portuguese, South-East Asian and South-East Asian by "USA300" strains. The other five isolates (merA/B and sasX negative) yielded dfrA alleles as predicted for CNA9, GenBank ANC1 16K, GenBank BAZ2. These places them into a lineage known to occur in Turkey, Eastern Europe, Russia and Northern China. These observations might indicate several, and independent, importations of ST239-II strains to Pakistan.

**Conclusion:**

Methicillin-resistant *S. aureus* is a major issue in Pakistan that contain a variety of clonal complexes. Dominating strains include variants of pandemic strain ST239-MRSA-III, Panton-Valentine leukocidin (PVL)-positive CC722-MRSA (23 %, Figure 1) and several CC8-MRSA-IV as well as CC6-MRSA-IV. Many strains indicated epidemiological links to Middle Eastern/Arabian Gulf region. In addition, the observation of different variants of ST239-III hint that several, and independent, importations to Pakistan might have occurred. The monitoring of MRSA from countries with less known epidemiology (e.g., Pakistan) is necessary to observe the distribution and spread of multi-resistant bacteria, as well as possible links to global travel, migration and commerce.

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### Table 1: Clonal complex CCs affiliation, numbers, geographic prevalence, and description of the MRSA strain detected.

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