Evaluation of the GENSPEED® MRSA Test Kit and its ability to detect the novel mecA homologue mecC

Andreas Petersen*, Alexandra Medina & Anders Rhod Larsen

Microbiology and Infection Control, Statens Serum Institut, Copenhagen, Denmark * email: aap@ssi.dk, phone: +45 3268 8168

P 0360

Introduction
Early detection of methicillin-resistant Staphylococcus aureus (MRSA) is important in infection control. Traditionally phenotypic criteria and resistance genes mecC and mecA, as well as mecC homologue mecA(LGA251) are used. Since mecA(LGA251) is non-susceptible to norfloxacin, it has been described as part of the resistance genotype mecC. In addition, mecC homologue mecA(LGA251) is detected in most systems. The objective of this study was to evaluate the GENSPEED® MRSA Test Kit using a well characterized MRSA collection including isolates harboring the mecC gene. The GENSPEED® MRSA Test Kit is designed to detect both homologues directly from nasal swabs.

Methods
In Denmark all new cases of MRSA are sent to the National Reference Laboratory for Staphylococci (NRS) for identification. In this retrospective study all submitted MRSA isolates from Nasal swabs were tested from May 1, 2010 to April 30, 2011 were included. The NRS serves an area in Denmark where the MRSA prevalence is high (Figure 3).

Figure 1. Design of the GENSPEED® microfluidic test cartridge
The GENSPEED® MRSA test kit combines four immobilized target hybridization probes for S. aureus, methicillin resistant S. aureus (MRSA) and Staphylococcus epidermidis. The immobilized target hybridization probes are detected with a compact Reader (Figure 2). The GENSPEED® MRSA Test Kit is designed to detect both mecA homologues directly from nasal swabs.

Results and discussion
The set represented 27 spa types assigned to CC1, CC2, CC30, CC45, CC5, CC59, CC8, CC80, CC93, ST123237, CC130, and CC398. Results from the 95 submitted human MRSA cases are shown in Table 1.

Table 1. Number of positive results for the PCR assay and the GENSPEED® MRSA Test Kit

<table>
<thead>
<tr>
<th>PCR results</th>
<th>mecA</th>
<th>mecC</th>
</tr>
</thead>
<tbody>
<tr>
<td>GENSPEED results</td>
<td>89</td>
<td>0</td>
</tr>
<tr>
<td>mecA</td>
<td>0</td>
<td>6</td>
</tr>
</tbody>
</table>

The GENSPEED development software correctly identified all mecA-MRSA from various genetic lineages and all six mecC-MRSA. The latter is of importance since PCEs and a single commercial system have so far been able to provide fast and reliable detection of the mecC type of MRSA. In addition, all four mecC positive animal isolates and five MSSA were correctly identified by the GENSPEED Test Kit.

The evaluation was performed satisfactorily with pure cultures and is expected it to perform equally well directly on nasal swabs.

Reference
Steager M, Andersen PS, Knorva P, Pichon B, Holmes MA, Edwards G, Laurent F, Teale C, Skov R, Larsen AR. Rapid detection, differentiation and typing of methicillin-resistant Staphylococcus aureus (MRSA) based on mecA and mecC. The Reference MRSA Test Kit was used instead of the standard software. The raw signals were used to determine an appropriate threshold for culture-based detection.

Figure 2. The GENSPEED® Reader
The GENSPEED®-platform combines a disposable microfluidic test cartridge (test chip) with a compact Reader. Different capture oligo-nucleotides are deposited onto the microfluidic test cartridge (Figure 1).

Figure 3. Location of DCM Slagelse and the catchment area
The MRSA status was confirmed by multiplex PCR detecting both the mec and mecC genes in addition to spa (Steager et al, 2012). The target DNA was sequenced to give the spa type, which was annotated to S. aureus clonal complex (CC). In addition, four mecC-positive isolates from sheep and cattle and five mecC-negative S. aureus (MSSA) were included. The isolates were tested according to the manufacturer’s instruction and training. With the exception that pure cultures were prepared before use. The results from the microbial identification showed that the software was used instead of the standard software. The raw signals were used to determine an appropriate threshold for culture-based detection.

Acknowledgment
The study was funded by Greiner Bio-One Diagnostics GmbH, Rainbach, Austria.