R319
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Molecular biology, including diagnostics: Molecular typing

Evaluation of semi-automated *rep*-polymerase chain reaction (DiversiLab®) for typing of outbreak-related methicillin-resistant *Staphylococcus aureus* in a Belgian hospital

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Typing of bacterial strains is essential to confirm an outbreak, to describe its spread and to guide control measures.

Objectives: To assess the performance of the Diversilab® (DL) rep-PCR system for typing of methicillin-resistant *Staphylococcus aureus* (MRSA) causing nosocomial outbreaks in a 800-bed secondary care hospital (2009-2010).

Methods: A total of 23 MRSA strains isolated from 8 nosocomial outbreaks, each including 2-4 colonized or infected patients were investigated. An outbreak was defined as two or more patients with new, nosocomial acquired MRSA at the same hospital ward at the same time. DL analysis was performed according to the manufacturer's instructions. The peak-patterns were analyzed with the web-based DL software using Pearson correlation coefficients to determine the degree of similarity between the strains. Patterns showing one peak difference were designated as closely-related and profiles with more than one peak difference were categorized as different. Spa-typing was performed as previously described (Hallin et al. J Clin Microbiol. 2007) and spa types were determined with Ridom StaphType software (Ridom GmbH) by the National Reference Center for *Staphylococcus aureus*. Concordance between methods was calculated (Robinson et al. J Mol. Evol. 1998).

Results: Seven different DL patterns were observed, 3 DL types were found in only one strain and 4 other types were found in 3-7 strains. Eight spa-types were found clustering in 4 different spa clonal complexes (CC). More than 85% of the strains belonged to the two most prevalent clonal complexes circulating in Belgium, respectively spa CC0038 (73%) and spa CC008 (13%). Nine (39%) discordant results were observed between DL- and spa typing. DL type 5 (n = 7 strains) included 4 spa types (t038, t739, t12334 and t131) belonging to the same spa CC038 underlining the lower discriminatory power of the DL system. DL type 9 (n = 6 strains) included 3 distinct spa CC (CC038, CC008 and CC6898) showing misclassification of the DL system. Concordance of DL with spa typing was 70%. For outbreak investigation, DL led to misinterpretation concerning ruling out or ruling in of 2 of the 8 outbreaks examined.

Conclusion: A limited total number of isolates reflecting a restricted strain selection per outbreak was studied. 4 distinct spa clonal complexes were identified, reflecting the hospital's low clonal MRSA diversity. DL showed a lower discriminatory power for MRSA typing compared to spa-typing. With only a marginal clonal diversity in MRSA strains, a highly discriminative typing technique is needed to confirm nosocomial transmission of MRSA strains. We conclude that DL typing is not discriminatory enough for MRSA outbreak investigation in our hospital.