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COMPARISON OF DIRECT AND INDIRECT SUSCEPTIBILITY TEST METHODS FOR DETECTION OF BACTERAEMIC METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS*

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ABSTRACT

Introduction: The clinical laboratory is required to rapidly identify *Staphylococcus aureus* as a cause of bacteraemia, and in particular, to detect methicillin resistance amongst bacteraemic isolates, to facilitate prompt initiation of appropriate therapy which may directly impact on patient survival, and to allow for implementation of appropriate infection control measures. Hence, the laboratory needs to choose tests to detect methicillin-resistant *S. aureus* (MRSA) bacteraemia which are rapid, accurate, simple, cost-effective and appropriate for the setting.

Primary study objective: To determine the accuracy of four phenotypic susceptibility tests to directly detect MRSA from blood culture specimens (BC) compared with detection of the *mecA* gene by the polymerase chain reaction (PCR) from *S. aureus* cultured from the same BC.

Materials and Methods: BCs were selected from patients with incident, *S. aureus* bacteraemic episodes at two hospitals, during January and February 2006. *S. aureus* was identified by standard phenotypic tests, including the presence of a deoxyribonuclease (DNAse). Direct susceptibility tests (DST) were performed (oxacillin (1µg) and cefoxitin (30µg) disk diffusion (DD), oxacillin Etest® (AB bioMérieux) and CHROMagar®-MRSA (CHROMagar® Microbiology)), and repeated on stored cultures. Detection of *nuc* and *mecA* genes by PCR confirmed *S. aureus* and methicillin resistance respectively. The sensitivity and specificity of the DST were calculated with reference to the *mecA* PCR result, to fulfil the primary study objective.

Results: During the two-month study period, 9,400 BC were submitted to the clinical laboratories at the 2 hospitals; *S. aureus* was isolated from 156 specimens. Of these, 89 BC

from 89 incident cases were included in the study, and 65 were subjected to all tests, including PCR. Of the 65 *nuc*-positive *S. aureus* isolates from 65 BC, all were positive with the direct DNAse test, and 25 (38%) were *mecA* positive. Compared to PCR, sensitivity and specificity for the direct oxacillin DD, cefoxitin DD, oxacillin Etest® and CHROMagar®-MRSA was 100% and 90%, 98% and 100%, 100% and 100%, and 96% and 42% respectively.

Discussion: In this study, we found that, compared to PCR for the *nuc* and *mecA* genes, the combination of a direct DNAse test and oxacillin Etest®, facilitated accurate detection of MRSA bacteraemia. The direct oxacillin Etest® result did not appear to be influenced by a non-standardised inoculum, in contrast to the other direct tests, and provided an oxacillin minimum inhibitory concentration. The direct cefoxitin DD test produced more accurate results than the direct oxacillin DD test, was easier to read and distinguished MRSA from MSSA with zone diameters clustering into more clearly defined susceptibility categories. Although the chromogenic agar performed well when used to identify methicillin resistance amongst cultured *S. aureus* isolates, it was apparent that this test, read at 24 hours, could not be used reliably as a DST. Since the Etest® is more costly than the DD test; its use should be reserved for BC from patients in "high-risk" hospital areas, e.g. intensive care units. The direct cefoxitin DD could be used for all BC positive for GPCC, and could be used without a direct identification test because of its lower cost; it is further recommended that the direct cefoxitin DD test replace the direct oxacillin test.