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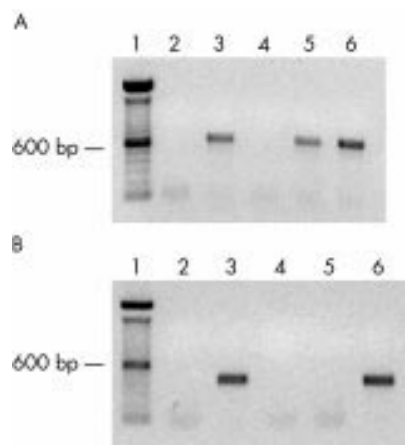
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### Molecular determination of carriage of the *mecA* locus in coagulase negative staphylococci in screening swabs from patients in an intensive care unit

Staphylococci are important nosocomial pathogens, which are easily transmitted from patient to patient, and there is concern over the recent emergence of antibiotic resistance



**Figure 1** Gel electrophoresis of amplicons resulting from polymerase chain reaction amplification of DNA extracted from salt meat broth containing multiple sets of screening swabs from patients in the intensive care unit. Lane 1, 100 bp DNA molecular weight ladder (Gibco, Paisley, UK); lane 2, negative control (molecular grade water); lane 3, positive control for respective loci; lane 4, coagulase negative *Staphylococcus* sp.; lane 5, methicillin sensitive *Staphylococcus aureus* (MSSA); lane 6, methicillin resistant *S aureus* (MRSA). (A) Amplification of *mecA* locus, (B) amplification of the *femB* locus.

to the  $\beta$  lactam class of antimicrobial drugs, particularly within methicillin resistant *Staphylococcus aureus* (MRSA). Coagulase negative staphylococci, although relatively less pathogenic than *S aureus*, are important pathogens, particularly in immunocompromised or immunosuppressed patients, where they can cause line associated bacteraemia and infections of prosthetic devices, such as heart valves. Recently, several studies have suggested that *S aureus* may acquire the *mecA* locus through the horizontal gene transfer of this locus from methicillin resistant (*mecA* +ve) coagulase negative staphylococci to methicillin sensitive *S aureus*, thus resulting in the emergence of MRSA phenotypes.<sup>1,2</sup> Therefore, the aim of our study was to determine the incidence of carriage of the *mecA* locus in coagulase negative staphylococci isolated from screening swabs from an intensive care unit (ICU) in a large acute teaching hospital.

Seventy five patients within the ICU at Belfast City Hospital were examined during the period February to June 1998 and screening swabs (nose, throat, perineum, skin lesions, and other sites) were obtained. Swabs were taken from patients on first admission to the ICU. Swabs from multiple sites from each patient were pooled and cultured within four hours of collection in salt meat broth (20 ml) (Oxoid CM94; Oxoid Ltd, Basingstoke, UK) at 37°C for 17 hours. An aliquot (1000  $\mu$ l) was removed and, after centrifugation at 14 000  $\times$ g for five minutes, the pellet was resuspended in polymerase chain reaction (PCR) grade water (100  $\mu$ l) (LAL grade water; Biowhittaker Inc, Walkersville, Maryland, USA) for one hour. After this, the pellet was recentrifuged at 10 000  $\times$ g for five minutes and 2  $\mu$ l DNA template was removed for PCR amplification. *MecA* and *femB* PCR amplifications were performed on all extracted DNA, as described previously,<sup>3</sup> in addition to an extra control PCR based on the amplification of a highly conserved region of eubacterial 16S rDNA.<sup>3</sup> Of the 75 patients examined, two were positive for MRSA by PCR (*mecA* and *femB* positive; fig 1), and the organisms were also detected simultaneously by conventional techniques. However, 38 of 75 patients were positive for methicillin resistant coagulase negative staphylococci (*mecA* +ve; *femB* -ve; fig 1). This is in agreement with a larger study examining the incidence of *mecA* positive coagulase negative staphylococci in five hospitals in Greece, where 59.5% of coagulase negative staphylococci isolates were *mecA* positive.<sup>4</sup>

To date, there have been several reports<sup>5</sup> on the molecular detection of MRSA in ICU patients using combinations of the *mecA* and *femB* gene loci; however, there have been relatively few reports on the prevalence of methicillin resistant coagulase negative staphylococci from patients in the ICU.<sup>4</sup> Although methicillin resistant coagulase negative staphylococci are not as important an infection control problem in the ICU as MRSA, they may act as an eventual source of methicillin resistance in MRSA, given their ability to share resistance determinants with their close phylogenetic neighbour, MRSA. Therefore, it is important that such resistance should be monitored continuously in coagulase negative staphylococci to determine the

following: (1) whether there is an association between methicillin resistance in coagulase negative staphylococci and downstream resistance in MRSA and (2) the time required for the emergence of MRSA via this mechanism. In addition, it is important to estimate whether or not this time period is of clinical consequence to the patient, in terms of infection control procedures and antibiotic intervention. Such an approach as this may be useful in developing specific infection control procedures and antimicrobial treatment protocols in the local setting.

Overall, this is the first report on the prevalence of methicillin resistance in coagulase negative staphylococci in patients from an ICU in Northern Ireland and it demonstrated that approximately half of all patients admitted to the ICU were in possession of the *mecA* locus. Further studies are required to help elucidate the mechanism and epidemiology of such potential interspecies horizontal gene transfer to maintain effective infection control and antibiotic treatment regimens.

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