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Rapid Extraction and Direct Identification of Methicillin-Resistant Staphylococci in Clinical Samples Using PCR

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Methicillin-resistant staphylococci (MRS) are one of the most common causes of nosocomial infections and bacteremia. Standard bacterial identification and susceptibility testing frequently require as long as 72 hours to report results, and there may be difficulty in rapidly and accurately identifying methicillin resistance. The use of polymerase chain reaction (PCR) is a rapid and simple process for the amplification of target DNA sequences, which can be used to identify and test bacteria for antimicrobial resistance. However, many sample preparation methods are unsuitable for PCR utilization in the clinical laboratory, because they either are not cost-effective, take too long to perform, or do not provide a satisfactory DNA template for PCR. Jaffe and coinvestigators from David Grant Medical Center, Travis AFB, California, conducted tests whose goal was to provide same-day results to facilitate rapid diagnosis and therapy. In this report, they describe a rapid method for extraction of bacterial DNA directly from blood-culture bottles that gave quality DNA for PCR in as little as 20 minutes. They compared this extraction method to the standard QIAGEN method for turnaround time (TAT), cost, purity, and use of template in PCR.

Specific identification of MRS was determined using intragenic primer sets for bacterial and *Staphylococcus* 16S rRNA and *mec*A gene sequences. The PCR primer sets were validated with 416 isolates of staphylococci, including methicillin-resistant *Staphylococcus aureus* (n=106), methicillin-sensitive *S aureus* (n=134), and coagulase-negative *Staphylococcus* (n=176). The total supply cost of the extraction method and PCR was $2.15 per sample, with a result TAT of less than 4 hours. The methods represent a rapid and accurate DNA extraction and PCR-based identification system, which makes the system an ideal candidate for use under austere field conditions and one that may have utility in the clinical laboratory.

FROM: Jaffe RI, Lane JD, Albury SV, Niemeyer DM. Rapid extraction from and direct identification in clinical samples of methicillin-resistant staphylococci using the PCR. *J Clin Microbiol* 2000;38:3407-3412.