METHODS FOR THE IDENTIFICATION OF Staphylococcus spp

Author(s) Maria de Lourdes Ribeiro de Souza da Cunha, Adriano Martison Ferreira, Mariana Fávero Bonesso, Alessandro Lia Mondelli

Institution(s) 1. UNESP, Universidade Estadual Paulista, Rubião Júnior, Botucatu-SP

Abstract:

The emergence of Staphylococcus spp. not only as human pathogens but also as reservoirs of antibiotic resistance determinants requires the development of methods for their rapid and reliable identification in Culture Collection of samples of medical importance. In recent years, several commercial systems for the rapid identification of staphylococci have been developed as an alternative to the classical identification protocols, which are too laborious and time-consuming to be used in most laboratories. The commercial systems, based on miniaturized biochemical or immunologic reactions, are widely used today for both clinical and research purposes. However, these diagnostic systems present problems, such as cost and response time, but more importantly, they often provide unreliable results. Several of the problems associated with these systems result from the variable expression of phenotypic characters that are used as diagnostic parameters. Additionally, many of these kits are based on colorimetric results, and subjectivity in their interpretation may lead to ambiguity. For these reasons, significant efforts have been made in order to develop alternative identification methods combining speed, reliability, and low cost. These criteria are met by methods based on molecular rather than phenotypic characters. One of these methods is ribosome spacer PCR or internal transcribed spacer-PCR (ITS-PCR). Based on the above considerations and in view of the need for rapid, simple, and reliable methods, the objective of the present study was to compare three techniques for the identification of Staphylococcus spp. isolated from patients with Urinary Tract Infection (UTI), i.e., a simplified method developed in our laboratory, the commercial Vitek and the ITS-PCR method. The amplicons were resolved in high-resolution agarose gels and visually compared with the patterns obtained for the control strains of ATCC staphylococcal species. Of the 100 staphylococcus samples studied, the following were identified: 57 S. saprophyticus, 16 S. epidermidis, 8 S. haemolyticus, 3 S. warneri and 16 S. aureus. Compared to the ITS-PCR, the simplified method identified 97% of the CNS species, respectively, while this rate was 80% for the Vitek. Inaccurate identification by the Vitek method was observed for the three Staphylococcus warneri isolates, S. saprophyticus (84.2%), S. epidermidis (62.5%), S. haemolyticus (75%) . All species analyzed had unique ITS-PCR patterns, ITS-PCR and simplified method proved to be valuables alternatives for the identification of staphylococci, offering, within the same response time and at lower cost, higher reliability than the currently available commercial systems.

Key words: Staphylococcus, Identification, Methods