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Comparative evaluation of the Vitek-2 Compact and Phoenix systems for rapid identification and antibiotic susceptibility testing directly from blood cultures of Gram-negative and Gram-positive isolates

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Abstract

We performed a comparative evaluation of the Vitek-2 Compact and Phoenix systems for direct identification and antimicrobial susceptibility testing (AST) from positive blood culture bottles in comparison to the standard methods. Overall, 139 monomicrobial blood cultures, comprising 91 Gram-negative and 48 Gram-positive isolates, were studied. Altogether, 100% and 92.3% of the Gram-negative isolates and 75% and 43.75% of the Gram-positive isolates showed concordant identification between the direct and the standard methods with Vitek and Phoenix, respectively. AST categorical agreements of 98.7% and 99% in Gram-negative and of 96.2% and 99.5% in Gram-positive isolates with Vitek and Phoenix, respectively, were observed. In conclusion, direct inoculation procedures for Gram-negative isolates showed an excellent performance with both automated systems, while for identification of Gram-positive isolates they proved to be less reliable, although Vitek provided acceptable results. This approach contributes to reducing the turnaround time to result of blood cultures, with a positive impact on patient care.

Keywords: Direct inoculation method; Blood cultures; Vitek; Phoenix; Gram positive; Gram negative

1. Introduction

Bloodstream infections (BSIs) are associated with high morbidity and mortality. The importance of early administration of adequate empirical antimicrobial therapy in the survival of patients with sepsis is demonstrated by several studies (Leibovici et al., 1998; Lodise et al., 2003). The administration of inappropriate initial antimicrobial therapy has been observed to be associated with an adverse outcome in specific populations of patients, such as those with severe neutropenia (Lin et al., 2008), those in the intensive care unit (Garrouste-Orgeas et al., 2006; Ibrahim et al., 2000), those with septic shock (Kumar et al., 2006), those infected with specific microorganisms (Anderson et al., 2006; Lodise et al., 2003; Micek et al., 2005; Morrell et al., 2005), those with bacteraemias associated with antibiotic-resistant bacteria (Kang et al., 2005; Kim et al., 2004), and those with a high-risk source of bacteraemia (Rodríguez-Baño et al., 2009; Zaragoza et al., 2003). For the administration of an early appropriate empirical therapy, it is primarily important to ensure a correct identification of the microorganisms responsible for BSIs for accurate diagnosis (Beekmann et al., 2003; Peters et al., 2004). Appropriate antimicrobial therapy has been shown also to lead to cost savings (Beekmann et al., 2003; Coleman et al., 1991; Hirschman et al., 1988; Tumbarello et al., 2010) and to help prevent the spread of antimicrobial resistance (Fraser et al., 2006; Goldmann et al., 1996; McGowan, 1994). Such consequences have led physicians to use more broad-spectrum antibiotics, sometimes using combination therapy, to decrease the probability

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of inadequate empirical regimens. However, a potential consequence of the use of unnecessarily broad-spectrum antimicrobials is the emergence of further antimicrobial resistance, greater cost, and more side effects.

In the light of this, the key role of microbiological findings in the diagnosis of BSIs seems evident (Barenfanger et al., 2008; Seifert, 2009; Towns et al., 2010). The introduction of reliable microbiological tests in the clinical microbiology laboratory that provide rapid identification (ID) and antimicrobial susceptibility testing (AST) of the causative agent of BSI is among the most important task in the clinical microbiology laboratory. At present, Gram stains of positive blood cultures are the most important factor influencing appropriate therapy in the majority of clinical microbiology laboratories (Barenfanger et al., 2008). Currently, the standard procedure for ID and AST of bacteria that have grown in blood cultures in liquid medium first requires an overnight subculture on solid agar medium, thus delaying the exact identification of the bacteria by 24 to 48 h. In order to decrease the time required for identification and AST of bacteria, positive blood culture bottles may be directly inoculated using an automated system. Several studies have compared the direct inoculation method with the standard procedure using different types of blood culture bottles and several automated systems (Bruins et al., 2004; Chen et al., 2008; de Cueto et al., 2004; Funke and Funke-Kissling, 2004; Ling et al., 2003; Lupetti et al., 2010; Quesada et al., 2010; Waites et al., 1998). These articles revealed acceptable results obtained mostly for Gram-negative rods, but less reliable ones for Gram-positive isolates (Bruins et al., 2004; de Cueto et al., 2004; Funke and Funke-Kissling, 2004; Ling et al., 2003; Quesada et al., 2010), although recently a good performance by the direct method compared to the currently used method was also obtained among Gram-positive cocci (Lupetti et al., 2010).

Moreover, in order to decrease the turnaround time needed to obtain the results for the ID of microorganisms in blood cultures, various molecular methods have been proposed, although all these methods have proved to be quite expensive and labour intensive, and to require highly trained personnel (Hansen et al., 2010; Jordan et al., 2009; Parta et al., 2009; Peters et al., 2006; Qian et al., 2001; Tenover, 2007; Tissari et al., 2010; Wallet et al., 2010; Wellinghausen et al., 2004). More recently, the use of matrix-assisted laser desorption ionization–time of flight mass spectrometry as a valid tool for rapid ID of bacteria and yeasts directly from positive blood culture broths has been proposed also (Ferroni et al., 2010; Prodhom et al., 2010; Stevenson et al., 2010).

The objective of this study was to evaluate the reliability and accuracy of 2 automated systems, the Vitek-2 Compact (bioMérieux, Marcy l'Etoile, France) and Phoenix (Becton Dickinson [BD], Sparks, MD, USA), for direct ID and AST of Gram-negative rods and Gram-positive bacteria directly from positive Bactec blood culture bottles.

2. Materials and methods

2.1. Samples

This study was performed during normal working days between April and October 2010 in a university teaching hospital in Rome, Italy. A total of 815 consecutive sets of blood cultures were collected during the study period. Each set comprised 3 aerobic and anaerobic bottles (Bactec Plus Aerobic/F, Bactec Plus Anaerobic/F, BD) per patient obtained during a 1-h period from cases with suspected bacteraemia. Blood bottles were incubated into the Bactec 9120 instrument (software version V4.91A), which is capable of incubating, agitating, and continuously monitoring aerobic and anaerobic bacterial growth by using the standard growth detection algorithms provided by it. Only the first positive blood culture containing bacteria that appeared to be monomicrobial in the Gram stain was included in the study. If samples in both the aerobic and anaerobic bottles for 1 blood culture were detected as positive and the organisms showed identical Gram staining morphologies, only the aerobic bottle was used for the study. A total of 4 blood cultures with anaerobic bacteria, 11 blood cultures with yeasts, and 17 polymicrobial blood cultures (confirmed after subculturing on agar plates) were observed, and they were excluded from the present study.

A total of 139 monomicrobial blood cultures were used in the present study, comprising 91 Gram-negative and 48 Gram-positive species. Gram-negative species consisted of 48 *Escherichia coli* (52.7%), 15 *Pseudomonas aeruginosa* (16.5%), 10 *Klebsiella pneumoniae* (11%), 6 *Enterobacter cloacae* (6.6%), 3 *Serratia marcescens* (3.3%), 2 *Klebsiella oxytoca* (2.2%), and 1 each (1.1%) of *Citrobacter braakii*, *Citrobacter freundii*, *Enterobacter aerogenes*, *Raoultella planticola*, *Proteus mirabilis*, *Pantoea agglomerans*, and *Stenotrophomonas maltophilia*. Among Gram-positive bacterial species, 22 isolates were represented by *Staphylococcus epidermidis* (45.8%), 12 by *Staphylococcus aureus* (25%), 4 by *Enterococcus faecium* (8.3%), 3 by *Staphylococcus hominis* (6.25%), 2 by *Enterococcus faecalis* (4.2%), and 1 each (2.1%) by *Streptococcus pneumoniae*, *Streptococcus agalactiae*, *Streptococcus anginosus*, *Micrococcus luteus*, and *Corynebacterium striatum*.

2.2. Direct inoculation system

For direct testing, 8 mL of the positive blood sample was processed. Samples were centrifuged first at 600 × *g* for 10 min to pellet red cells. The supernatant was then transferred into a new tube and centrifuged at 3000 × *g* for 10 min to pellet bacteria. The bacterial pellet was resuspended in sterile 0.9% saline, and some drops of this bacterial suspension were added with a sterile Pasteur pipette into the ID broth (BD) for the Phoenix system, and in 0.45% sodium chloride (bioMérieux) for Vitek-2 Compact so that the suspension matched the turbidity of McFarland 0.5–0.6 and 0.53–0.63 standard for Phoenix and Vitek-2 Compact systems,