VERSATREK® for Use in Determination of Fungemia

ABSTRACT

With the increasing number of patients with complex clinical conditions and undergoing complex medical therapy, more unusual and infectious disease processes have emerged. Fungal pathogens are agents of a variety of infections, including fungemia, which are part of this trend. Fungi are now being recognized as an important cause of infection in patients with cancer, hematopoietic stem cell transplantation, solid organ transplant, and severe trauma. Major predisposing factors leading to these infections include immunosuppression, use of broad-spectrum antibiotics, use of central vascular catheters (especially in association with the administration of hyperalimentation solutions), and more aggressive attempts to prolong the survival of patients with complicated, serious diseases. Recent data suggest that fungi are more prevalent than anticipated bacteremias. There are also frequent reports of illnesses associated with a long list of opportunistic, and environmental molds and yeasts including moulds and yeasts which are not typically considered bloodstream pathogens. The decision-making process for the correct diagnosis and isolate identification can be challenging. The VersaTREK® (VT) Microbial Detection System is FDA-cleared for use in the recovery of bacteria and yeast from blood and normally sterile body fluids. Additionally, the system has the flexibility to detect Mycobacteria and perform first-line drug testing for Mtb isolates. Although it is extremely difficult to evaluate a blood culture instrument using patient specimens as the seed/seeded specimens is necessary.

RESULTS

Table 1 lists the organisms tested in this study and the range of time-to-detection of the organism. During the initial phase of the testing, there was some trial and error in obtaining the correct dilution (McFarland Standard) in order to achieve the appropriate inoculum size as the current literature only defines dilutions for bacteria. Several fungi spores are much larger than bacteria which affects the initial turbidity needed to make the correct inoculum. yeast was inoculated to wells where the inoculum was less than 10 cfu/ml but growth was detected. If the inoculum size was greater than 100 cfu/ml, the test was repeated with the correct supension. (For some molds the inoculum size was greater than 200 cfu/ml yet included in the study). These isolates were difficult to achieve the correct inoculum size due to reduced/maintain the sensitivity of the test.

Four of the organisms tested failed the detection by the VersaTREK system: Cryptococcus albidos, Cryptococcus laurentii, B. dermatidis, and C. tropicalis. The growth of these organisms was not confirmed via subculture plates. This could be due to the fact that the current media formulation does not support the growth and/or the temperature of the instrument was not optimal for growth of these organisms. For this reason, we recommend that these isolates be plated directly and ultimately isolate identification.

BACKGROUND

The VersaTREK® (VT) Microbial Detection System is FDA cleared for use in the recovery of bacteria and fungi from blood and normally sterile body fluids. Additionally, the system has the flexibility to detect Mycobacteria and perform first-line drug testing for Mtb isolates. Although the system is not specifically cleared for fungal testing, the VersaTREK system has been added to the list of systems that can be used for fungal testing. The VersaTREK system has been added to the list of systems that can be used for fungal testing. The VersaTREK system has been added to the list of systems that can be used for fungal testing. The VersaTREK system has been added to the list of systems that can be used for fungal testing. The VersaTREK system has been added to the list of systems that can be used for fungal testing.

METHODS

It is extremely difficult to evaluate a blood culture instrument using patient specimens as the test vastly yield poor results. Therefore, for the evaluation of the ability of the blood culture media to support growth AND the analyzer to detect the growth of pathogens using seeded specimens is necessary.

More than one hundred isolates, both pathogenic and ATCC reference strains, of yeasts and molds were used, along with aseptic patient specimens that were positive for molds for the study. Solutions were made in sterile water to achieve an inoculum of 100 cfu/ml (as outlined in the quality control section of the VersaTREK Technical bulletin). In addition to the organism suspensions, 5.0% sterile human blood was inoculated into clinical conditions to assure a unique blood culture environment. The organism suspensions were inoculated into the VersaTREK blood culture bottle. This allowed for the detection of fungi directly from the specimen (blood collected in a 10 ml SPS vacutainer tube).

REFERENCES


DISCUSSIONS AND CONCLUSIONS

Due to the high mortality rate from fungemia, the rapid detection and identification of fungi from a patient’s blood can have great diagnostic and prognostic importance. Risk factors for fungemia and confounding factors include: isolation, but are not limited to, fever of unknown origin, cancer and transplant, severe trauma, neutropenia, severe immunosuppression, and those individuals with burns or invasive intravascular devices.

Though most fungi are caused by yeast and most isolates were detected in the REDC1 (aerobic blood culture bottle) within the usual 3 day incubation period, there is the potential for the indeterminate result to be a slower growing mold, of which some have better recovery and faster detection in the MYCO bottles. For this reason, we have divided the following protocol for use when a fungal blood culture is entered or delayed.

• Collect one 10 ml Vacutainer tube for SPS anticoagulant
• Transfer 5 ml of blood to a REDC 1 aerobic blood culture bottle (length of incubation set at 3-5 days normal length of culture for a fungal culture)
• Transfer 0.5 ml of blood to a MYCO blood culture bottle (which is 1 ml of Growth Supplement has been added - incubation preset at 42 days)

• Centrifuge the remaining blood at 3600 rpm for 15 minutes and place the sediment to an enriched, non-inhibitory fungal media (i.e. BHI 10% sheep blood) and incubate at 35°C for 48 hours (total incubation time of the MYCO bottle)

The combination of these features should give the fastest and least labor intensive results. Adding a culture of the sediment should enable the detection of molds and yeasts that do not grow at the standard blood culture temperature of 35°C and/or those that do not produce or consume enough gas to be detected by the analyzer.

Unlike the BD BACTEC FX® and bioMerieux BacT/Alert, the VersaTREK Microbial Detection System detects a change in the pressure of this headspace of the blood culture and/or MYCO bottle. This allows for the detection of any gas production and gas consumption. The BACTEC FX and BacT/Alert use sensors to detect CO2 production only. VersaTREK uses otherwise real-time detection to detect the production of many species of bacteria, yeast and molds compared to competitive systems due to the detection of gas consumption.

We will continue to collect data from patient specimens as the protocol is put in place. During the 177 (or so) years that we have had the VersaTREK analyzers (originally known as ESP by Diag), we have never recovered a fungal isolate from the currently used Septicheck system (discontinued product of BD) that was not recovered by the TREK system.