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Performance Evaluation of the VersaTREK Blood Culture System for Quality Control Testing of Platelet Units

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The performance of the VersaTREK blood culture system for the detection of a bacterial contamination of platelet concentrates was assessed using samples spiked with serial dilutions of nine bacterial species associated with platelet contamination. The system detected growth for all organisms in <20 h, and the detection sensitivity was <20 CFU/ml.

Four million platelet concentrates (PLT) are transfused in the United States annually. To preserve their function, PLT are stored at 20 to 24°C—a range that also permits bacterial growth (4). Bacterial contamination is reported to occur in 1 in 1,000 to 2,000 PTL units (4). A study conducted by the Red Cross over 26 months found that the PLT contamination rate was 1:5,399 units and that PLT contamination resulted in 20 septic reactions, including three fatalities (6).

To limit adverse events, the American Association of Blood Banks (AABB) mandated that member institutions adopt measures to detect bacterial contamination in PLT (1). Of the commercially available automated microbial detection systems, only the BacT/Alert (bioMérieux, Durham, NC) and the Pall eBDS (Pall Corporation, East Hills, NY) systems received FDA clearance for the quality control (QC) testing of PLT, necessitating that laboratories using other systems perform in-house verification studies (7). To this end, we evaluated the Bactec 9240 system (BD Microbiology, Cockeysville, MD) prior to using it for the detection of PLT contamination in our facility by using single-donor PLT spiked with dilutions of nine species of bacteria commonly associated with PLT contamination (2, 3, 5, 9). The Bactec system was found to have a detection limit of <10 CFU/ml for all but one test organism (Streptococcus mitis) and detected growth in <18 h.

When the VersaTREK system (TREK Diagnostic Systems, Cleveland, OH) replaced the Bactec 9240 system in our laboratory as our primary blood culture system, the PLT protocol used previously was repeated and the results are presented here. VersaTREK monitors bacterial growth by detecting pressure changes in the headspace of the blood culture bottle secondary to gas consumption/production.

Nine bacterial species, including Escherichia coli (ATCC 35922), Pseudomonas aeruginosa (ATCC 27853), Staphylococcus aureus (ATCC 14990), Staphylococcus epidermidis (ATCC 14990), and Klebsiella pneumoniae (ATCC 13883) and clinical isolates of Bacillus cereus, Serratia marcescens, Enterobacter cloacae, and Streptococcus mitis, were used for the study. The bacteria were cultured overnight in 10 ml of Trypticase soy broth with shaking and harvested by centrifugation at 3,000 × g for 5 min. The cells were washed in phosphate-buffered saline, centrifuged, and suspended in phosphate-buffered saline to approximate a 0.5 McFarland standard (approximately 1 × 10⁸ to 5 × 10⁶ CFU/ml). Tenfold serial dilutions (10⁻¹ to 10⁻¹⁰) were made from each suspension in single-donor PLT. Blood culture bottles (VersaTREK Redox 1 [80 ml]; TREK) were inoculated with 4 ml of each dilution, ranging from 10⁻⁴ to 10⁻¹⁰. A blood culture bottle inoculated with 4 ml of the PLT product served as a negative control. Dilutions ranging from 10⁻¹ to 10⁻⁴ showed consistent positive growth and were not included in this study. Inoculated bottles were incubated in the VersaTREK system for a maximum of 5 days. Signal-positive bottles were subcultured to blood agar to confirm the identity of the isolates.

To determine the inoculum size, 0.1 ml of each dilution was plated in duplicate onto blood agar plates. Colony counts were obtained after 48 h of incubation, and the number of organisms injected into each blood culture bottle was determined (Table 1).

The detection sensitivity of the VersaTREK system was <10 CFU/ml for eight of the nine species tested (Table 1). The VersaTREK system was the most sensitive for the detection of S. epidermidis (calculated at 0.025 CFU/ml) and the least sensitive for S. mitis (20 CFU/ml). The highest dilution (lowest inoculum) of each organism to generate a positive signal did so in ≤18.1 h. The range of signal positivity varied from 6.0 h for S. marcescens (10⁻⁴ dilution; 5.08 × 10⁴ CFU per ml) to 18.1 h for S. epidermidis (10⁻¹⁰ dilution; 0.025 CFU per ml) (Table 1).

Following this study, the VersaTREK system was introduced for the routine QC of apheresis PLT using a 4-ml inoculum volume (Table 2). The products with negative growth after 24 h of monitoring were released for transfusion but were incubated for a total of 5 days. Over a 6-month period, 1,970 apheresis PLT were tested, and 5 were positive for bacterial growth. Positive cultures included three viridans-group Streptococcus species, one coagulase-negative Staphylococcus species, and one Bacillus species. Three of the five positive PLT were detected within 24 h of culture and discarded. The other two were transfused without evidence of unfavorable outcomes based on chart review.
In 2004, the AABB introduced a standard requiring all member institutions to implement QC testing of PLT to decrease the transfusion of contaminated units (1). Alternatives for testing included the measurement of pH or glucose level, microscopy, or culture, and the target level of detection was 10 CFU per ml (2). Although measurements of pH or glucose level give rapid results, the sensitivity of these methods is poor (10⁶ to 10⁷ CFU/ml [4]). Two commercial bacterial detection systems, the BacT/Alert (bioMérieux) and the eBDS (Pall), were shown to detect 10 CFU/ml (4) and received FDA clearance for this application. Perhaps it would be reasonable to reassess the FDA clearance process.

After implementation, the VersaTREK system showed an overall positivity rate of approximately 1 in 394 PLT compared to 1 in 554 for the Bactec 9240 system (5). The higher frequency of detection of the viridans group Streptococcus by VersaTREK (three of five positive cultures) compared to that of the Bactec 9240 system (one of seven positive cultures) (5) suggests either an increased sensitivity of the former or a greater propensity for contamination. Mirrett et al. showed a significant increase in the detection of streptococci and enterococci from blood using VersaTREK compared with that of the BacT/Alert system (8).

We conclude that VersaTREK is comparable to Bactec 9240 for detecting the low-level contamination of PLT prior to transfusion.

REFERENCES

TABLE 1. Results of the VersaTREK detection of mock-infected PLT with nine bacterial test strains

<table>
<thead>
<tr>
<th>Organism (primary inoculum size [CFU/ml])</th>
<th>Time (h) to signal bottle detection for indicated dilution</th>
<th>Sensitivity of detection (CFU/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10⁻¹</td>
<td>10⁻²</td>
</tr>
<tr>
<td>P. aeruginosa (9.21 × 10⁷)</td>
<td>9.3</td>
<td>11.1</td>
</tr>
<tr>
<td>S. aureus (5.4 × 10⁸)</td>
<td>8.2</td>
<td>9.2</td>
</tr>
<tr>
<td>E. coli (1.21 × 10⁸)</td>
<td>6.7</td>
<td>7.5</td>
</tr>
<tr>
<td>S. marcescens (1.27 × 10⁹)</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>S. epidermidis (6.4 × 10⁷)</td>
<td>9.9</td>
<td>11.6</td>
</tr>
<tr>
<td>K. pneumoniae (1 × 10⁷)</td>
<td>7.3</td>
<td>8.1</td>
</tr>
<tr>
<td>B. cereus (3.2 × 10⁸)</td>
<td>6.5</td>
<td>7.3</td>
</tr>
<tr>
<td>E. cloacae (9 × 10⁷)</td>
<td>7.3</td>
<td>8.3</td>
</tr>
<tr>
<td>S. mitis (2 × 10⁷)</td>
<td>11</td>
<td>13.2</td>
</tr>
</tbody>
</table>

Table 1. Results of the VersaTREK detection of mock-infected PLT with nine bacterial test strains

* NG, no growth after 5 days of incubation.


**TABLE 2. Positive bacterial culture in apheresis PLT detected by VersaTREK during the first 6 months (1,970 donors)**

<table>
<thead>
<tr>
<th>Collection date</th>
<th>Initial culture date</th>
<th>Initial culture result</th>
</tr>
</thead>
<tbody>
<tr>
<td>January 22, 2008</td>
<td>January 22, 2008</td>
<td>Viridans group</td>
</tr>
<tr>
<td>February 7, 2008</td>
<td>February 8, 2008</td>
<td>Viridans group</td>
</tr>
<tr>
<td>April 23, 2008</td>
<td>April 24, 2008</td>
<td>Coagulase-negative</td>
</tr>
<tr>
<td>May 14, 2008</td>
<td>May 15, 2008</td>
<td>Bacteroides species, not B. anthracis</td>
</tr>
<tr>
<td>June 7, 2008</td>
<td>June 8, 2008</td>
<td>Viridans group</td>
</tr>
</tbody>
</table>

* Apheresis PLT transfused before turning positive.