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# Controlled Clinical Comparison of Plastic and Glass Bottles of BacT/ALERT FA Medium for Culturing Organisms from Blood of Adult Patients

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**A new, clear-plastic nonvented aerobic FA bottle, designed to prevent breakage, has been developed for the BacT/ALERT blood culture system. We assessed the new plastic FA bottle by comparing its performance with that of the current glass FA bottle for recovery of microorganisms and time to detection of growth in blood samples obtained for culture from adult patients with suspected bloodstream infections. We conclude that the BacT/ALERT plastic and glass FA bottles are comparable for recovery of microorganisms and that the safety advantage of plastic bottles can be achieved without compromising performance.**

The recovery of microorganisms from blood culture in order to diagnose bacteremia and fungemia remains one of the most important tests performed by a clinical microbiology laboratory. Prompt detection of microorganisms in blood culture bottles provides valuable information for clinicians in guiding therapeutic decisions. Glass bottles have been used routinely in blood culture systems; however, breakage of glass bottles, although a rare event, may compromise the integrity of a blood specimen, delay reporting, and expose health care workers to infectious blood-borne pathogens. To reduce these risks, clear plastic bottles have been developed for use in the BacT/ALERT (bioMérieux, Inc., Durham, N.C.) automated blood culture instrument. The new plastic FA (PFA) bottle contains a casein-soy-based medium with activated charcoal to counter the potential inhibitory activity of antibiotics, as does the glass BacT/ALERT FA bottle. We compared the new PFA bottle to the current FA bottle for the recovery of microorganisms, as well as for the time to detection of the growth of microorganisms in blood samples obtained for culture from adult patients with suspected bloodstream infections.

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From July 2000 to February 2002, blood was collected from adult patients at Duke University Medical Center with suspected bacteremia and fungemia. Institutional review board approval was obtained prior to the study, and all blood cultures were performed as part of standard patient care. Up to 30 ml of blood was collected from each adult patient and divided (10 ml each) among aerobic PFA and FA bottles and anaerobic FN bottles. Although not part of this comparative study, the FN bottle was included to enable recovery of anaerobic microorganisms. Upon receipt in the laboratory, each bottle was measured against known standards to determine the volume of blood contained therein. All bottles were processed regardless of volume received: however, only bottle pairs (one PFA and one FA bottle) containing 8 to 12 ml of blood per bottle were included in the data analysis. All bottles were incubated in the BacT/ALERT instrument for 5 days or until they signaled positive. Based on Gram stain results, aliquots of the blood-broth mixture were subcultured onto appropriate media and incubated per laboratory protocol. Subsequent identification of microorganisms was performed by standard laboratory methods (2). False positives were defined as bottles that were Gram stain and subculture negative after the instrument signaled positive. Gram stain-negative bottles were returned to the instrument for the remainder of the 5-day incubation period or until they were reflagged by the instrument. Specifically for this study, for bottle pairs with a negative bottle and a companion positive bottle, the contents of the negative bottle were subcultured at the end of the 5-day protocol. If microorganisms grew on subculture, these negative bottles were defined as false negative.

An infectious-disease physician reviewed each positive culture and coded it as clinically significant, a contaminant, or an isolate of unknown clinical significance based on previously published criteria (4). Episodes of bloodstream infection were defined by growth of a clinically significant isolate without recovery of a different microorganism during the succeeding 7-day period. If a different clinically significant microorganism was recovered within 72 h, the two isolates were considered evidence of a polymicrobial episode (not included in the analysis). If a different microorganism was recovered after 72 h, the second isolate was considered evidence of a new episode. Patients were considered to be on therapy at the time the blood was collected if they were receiving an agent with activity against the microorganism isolated. Testing for susceptibility of the blood isolate to antimicrobial agents was determined by NCCLS methods (3). In some cases (e.g., *Candida* spp.), resistance was inferred from known patterns of susceptibility. Statistical analysis of results was performed with the modified chi-square test described by McNemar (1).

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TABLE 1. Comparative yields of clinically significant isolates in PFA versus FA aerobic blood culture bottles

| Microorganism(s) | No. of isolates recovered from: | | | $P^a$ |
|---|---|---|---|---|
| | Both bottles | PFA bottles only | FA bottles only | |
| *Staphylococcus aureus* | 50 | 9 | 8 | NS |
| Coagulase-negative staphylococci | 22 | 4 | 2 | NS |
| *Streptococcus* sp.[b] | 1 | 1 | 0 | NS |
| *Enterococcus* sp. | 12 | 6 | 6 | NS |
| Gram-positive bacilli[c] | 6 | 0 | 1 | NS |
| *Enterobacteriaceae*[d] | 52 | 9 | 18 | NS |
| Other gram-negative bacteria[e] | 40 | 7 | 6 | NS |
| Yeasts[f] | 32 | 17 | 13 | NS |
| All microorganisms | 215 | 53 | 54 | NS |

[a] NS, not significant.
[b] Includes viridans group streptococci (one isolate) and *Streptococcus agalactiae* (one isolate).
[c] Includes *Corynebacterium jeikeium* (four isolates), *Listeria monocytogenes* (two isolates), and diphtheroids (one isolate).
[d] Includes *E. coli* (26 isolates), *Enterobacter cloacae* (14 isolates), *Klebsiella pneumoniae* (13 isolates), *Serratia marcescens* (7 isolates), *Enterobacter aerogenes* (7 isolates), *Klebsiella oxytoca* (7 isolates), *Citrobacter freundii* (3 isolates), and *Proteus mirabilis* (2 isolates).
[e] Includes *P. aeruginosa* (16 isolates), *Stenotrophomonas maltophilia* (13 isolates), *Burkholderia cepacia* (9 isolates), *Acinetobacter* sp. (4 isolates), *Chryseobacterium* sp. (4 isolates), *Pandoraea* spp. (3 isolates), *Neisseria* sp. (1 isolate), *Ochrobactrum anthropi* (1 isolate), and unidentified gram-negative rods (2 isolates).
[f] Includes *Candida albicans* (27 isolates), *Candida glabrata* (11 isolates), *Candida krusei* (9 isolates), *Candida parapsilosis* (7 isolates), *Candida lusitaniae* (3 isolates), *Candida tropicalis* (2 isolates), and *Cryptococcus neoformans* (3 isolates).

We received a total of 5,862 blood culture sets that contained both PFA and FA bottles, 4,323 (74%) of which contained an adequate volume of blood. Three hundred twenty-two clinically significant isolates from 162 patients were detected in one or both study bottles (Table 1). Clinically significant isolates were detected with equal frequencies in both study bottles. The results of a subset of blood cultures from patients who were being treated with antimicrobial agents at the time the blood cultures were obtained are summarized in Table 2. There were no significant differences in recovery for any microorganism in this subgroup. In the subgroup of patients not receiving antimicrobial therapy (data not shown), *Escherichia coli* was detected more often in the FA ($n = 8$) than in the PFA ($n = 1$) bottles ($P < 0.05$); however, this difference was not present in the monomicrobial episode analysis (Table 3), which included 130 of 153 total episodes. When both bottles detected clinically significant microorganisms within 72 h the mean times to detection were similar (Table 4). Of the 4,323 paired blood culture bottles, the frequencies of false-positive bottles were similar for PFA ($n = 10$ [0.2%]) and FA ($n = 11$ [0.3%]) bottles. There was one false-negative result (*Stenotrophomonas maltophilia*) in a PFA bottle and one (*Pseudomonas aeruginosa*) in an FA bottle. Of all 185 contaminant isolates detected during this study (detailed data not shown), 53 were detected in both bottles, 52 were detected in PFA bottles only; and 80 were detected in FA bottles only ($P < 0.05$). Of the 131 coagulase-negative staphylococcal contaminants specifically, 38 were detected in both bottles, 36 were detected in PFA bottles only, and 57 were detected in FA bottles only ($P < 0.05$).

TABLE 2. Comparative yields of clinically significant isolates in PFA versus FA aerobic blood culture bottles from patients receiving antimicrobial therapy

| Microorganism(s) | No. of isolates recovered from: | | | $P^a$ |
|---|---|---|---|---|
| | Both bottles | PFA bottles only | FA bottles only | |
| *Staphylococcus aureus* | 14 | 3 | 3 | NS |
| Coagulase-negative staphylococci | 9 | 2 | 1 | NS |
| *Enterococcus* sp. | 5 | 0 | 1 | NS |
| *Corynebacterium jekeium* | 1 | 0 | 1 | NS |
| *Enterobacteriaceae*[b] | 11 | 3 | 4 | NS |
| Other gram-negative bacteria[c] | 9 | 1 | 3 | NS |
| Yeasts[d] | 16 | 7 | 5 | NS |
| All microorganisms | 65 | 16 | 18 | NS |

[a] NS, not significant.
[b] Includes *E. coli* (eight isolates), *K. pneumoniae* (four isolates), *Enterobacter aerogenes* (three isolates), *Enterobacter cloacae* (two isolates), and *Proteus mirabilis* (one isolate).
[c] Includes *P. aeruginosa* (four isolates), *Stenotrophomonas maltophilia* (three isolates), *B. cepacia* (three isolates), *Acinetobacter* sp. (one isolate), *O. anthropi* (one isolate), and an unidentified gram-negative rod (one isolate).
[d] Includes *Candida albicans* (10 isolates), *Candida krusei* (9 isolates), *Candida parapsilosis* (6 isolates), *Cryptococcus neoformans* (2 isolates), and *Candida lusitaniae* (1 isolate).

The overall performance of the aerobic plastic (PFA) bottle was similar to that of the glass (FA) bottle for detection of clinically significant microorganisms from patients with suspected bacteremia or fungemia. Fewer contami-

TABLE 3. Comparative levels of detection of episodes of monomicrobial bacteremia or fungemia by PFA and FA aerobic culture bottles paired with an anaerobic FN bottle

| Cause of episode | No. of episodes detected by: | | | $P^a$ |
|---|---|---|---|---|
| | Both bottles[h] | PFA bottles only | FA bottles only | |
| *Staphylococcus aureus* | 37 | 3 | 1 | NS |
| Coagulase-negative staphylococci | 14 | 0 | 0 | NS |
| *Streptococcus* sp.[b] | 1 | 1 | 0 | NS |
| *Enterococcus* sp. | 6 | 1 | 2 | NS |
| Gram-positive bacilli[c] | 4 | 0 | 0 | NS |
| *Enterobacteriaceae*[d] | 36 | 3 | 5 | NS |
| Other gram-negative bacteria[e] | 20 | 0 | 2 | NS |
| Yeasts[f] | 10 | 5 | 1 | NS |
| Anaerobes[g] | 2 | 0 | 0 | NS |
| All microorganisms | 130 | 13 | 11 | NS |

[a] NS, not significant.
[b] Includes viridans group streptococci (one isolate) and *Streptococcus agalactiae* (one isolate).
[c] Includes *Listeria monocytogenes* (two isolates), *Cornyebacterium jeikeium* (one isolate), and diphtheroids (one isolate).
[d] Includes *E. coli* (20 isolates), *K. pneumoniae* (9 isolates), *Enterobacter cloacae* (5 isolates), *Enterobacter aerogenes* (3 isolates), *Serratia marcescens* (3 isolates), *K. oxytoca* (2 isolates), *Proteus mirabilis* (1 isolate), and *Pantoea agglomerans* (1 isolate).
[e] Includes *P. aeruginosa* (six isolates), *B. cepacia* (five isolates), *Stenotrophomonas maltophilia* (four isolates), *Acinetobacter* sp. (one isolate), *Chryseobacterium* sp. (two isolates), *Pandoraea* sp. (one isolate), *Neisseria* sp. (one isolate), *O. anthropi* (one isolate), and an unidentified gram-negative rod (one isolate).
[f] Includes *Candida albicans* (eight isolates), *Candida glabrata* (four isolates), *Candida krusei* (one isolate), *Candida parapsilosis* (one isolate), *Candida tropicalis* (one isolate), and *Cryptococcus neoformans* (one isolate).
[g] Includes *Clostridium perfringens* (one isolate) and *Bacteroides fragilis* (one isolate).
[h] When the anaerobic FN bottle was positive, the episode was considered to be positive in both systems.

TABLE 4. Comparative times to positivity of clinically important bacteria and fungi when both the aerobic PFA and FA culture bottles were positive within 72 h

| Microorganism(s) | No. of isolates | Mean time to positivity (h) | |
|---|---|---|---|
| | | PFA bottles | FA bottles |
| *Staphylococcus aureus* | 50 | 17.3 | 17.7 |
| Coagulase-negative staphylococci | 21 | 24.4 | 23.0 |
| Viridans group streptococcus | 1 | 46.2 | 50.4 |
| *Enterococcus* sp. | 12 | 14.4 | 14.3 |
| Gram-positive bacilli[a] | 6 | 26.1 | 27.5 |
| *Enterobacteriaceae*[b] | 52 | 11.9 | 12.3 |
| Other gram-negative bacteria[c] | 35 | 20.2 | 19.9 |
| Yeasts[d] | 27 | 38.3 | 41.2 |
| All microorganisms | 204 | 20.2 | 20.6 |

[a] Includes *Comyebacterium jeikeium* (three isolates), *L. monocytogenes* (two isolates), and diphtheroids (one isolate).

[b] Includes *E. coli* (13 isolates), *Enterobacter cloacae* (13 isolates), *K. pneumoniae* (8 isolates), *K. oxytoca* (7 isolates), *Serratia marcescens* (6 isolates), *Citrobacter freundii* (2 isolates), *Enterobacter aerogenes* (1 isolate), *Proteus mirabilis* (1 isolate), and *Pantoea agglomerans* (1 isolate).

[c] Includes *P. aeruginosa* (12 isolates), *Stenotrophomonas maltophilia* (8 isolates), *B. cepacia* (7 isolates), *Acinetobacter* sp. (3 isolates), *Chryseobacterium* sp. (3 isolates), *Neisseria* sp. (1 isolate), and *O. anthropi* (1 isolate).

[d] Includes *Candida albicans* (14 isolates), *Candida krusei* (6 isolates), *Candida parapsilosis* (4 isolates), *Candida lusitaniae* (2 isolates), and *Candida tropicalis* (1 isolate).

nants were found in the PFA bottles. When both bottles were positive within 72 h, the times to detection were also similar. Because plastic bottles provide a performance comparable to that of glass bottles and have the potential for added safety, we recommend their use in the BacT/ALERT blood culture system.

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## REFERENCES

1. **McNemar, Q.** 1962. Psychological statistics, 3rd ed., p. 209–239. John Wiley & Sons, Inc., New York, N.Y.
2. **Murray, P. R., E. J. Baron, J. H. Jorgensen, M. A. Pfaller, and R. H. Yolken (ed.).** 2003. Manual of clinical microbiology, 8th ed. American Society for Microbiology, Washington D.C.
3. **NCCLS.** 2004. Performance standards for antimicrobial susceptibility testing, 14th informational supplement. Document M100-S14. NCCLS, Wayne, Pa.
4. **Weinstein, M. P., M. L. Towns, S. M. Quartey, S. Mirrett, L. G. Reimer, G. Parmigiani, and L. B. Reller.** 1997. The clinical significance of positive blood cultures in the 1990s: a prospective comprehensive evaluation of the microbiology, epidemiology, and outcome of bacteremia and fungemia in adults. Clin. Infect. Dis. **24:**584–602.