Comparative Recovery of Microorganisms from BacT/ALERT Plastic and Glass FA and FN Blood Culture Bottles

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bioMerieux, Inc., has recently introduced plastic bottles to replace glass bottles for use in the BacT/ALERT blood culture system. We compared the performance of the plastic to the glass bottles in a large clinical evaluation. Two blood cultures were collected from each patient, one using glass FA (aerobic) and FN (anaerobic) bottles and one using plastic FA and FN bottles. Of the 4,040 sets of four bottles collected, 3,110 contained the recommended 8 to 12 ml of blood, yielding 524 microorganisms with 359 judged to be clinically significant. Of the 359 significant organisms, 255 were recovered in either one or two bottles from both pairs of bottles in a set while 56 organisms were recovered only from the glass bottles and 48 were recovered only from the plastic bottles (P , not significant [NS]). Of the 286 significant organisms recovered only in the FA bottles (glass and plastic), 180 were recovered in both bottles, 57 in the plastic bottles only, and 49 in the glass bottles only (P , NS). Of the 303 significant organisms recovered in the FN bottles only (glass and plastic), 212 were recovered in both bottles, 46 in the plastic bottles only, and 45 in the glass bottles only (P , NS). For individual organisms, the only significant difference in recovery was obtained for *Escherichia coli*, with more isolates recovered in the FN plastic than in the FN glass bottles ($P = 0.02$). These data suggest that recovery of microorganisms with plastic FA/FN bottles is at least equal to that with glass FA/FN bottles while offering greater safety for users.

bioMerieux, Inc. (Durham, NC), recently introduced plastic blood culture bottles to replace the previously utilized glass blood culture bottles. This change was made to reduce or eliminate the possibility of breakage. The purpose of this study was to compare the performance of the glass and plastic aerobic (FA) and anaerobic (FN) bottles with a full 10-ml blood draw in each bottle.

MATERIALS AND METHODS

The standard of care for adult patients at Geisinger Medical Center is two pairs of blood cultures collected sequentially via separate venipunctures or separate line access. For this study, sets of four BacT/ALERT blood culture bottles were specially prepared in the laboratory. Sets included one each of a plastic FA (PFA), plastic FN (PFN), glass FA (GFA), and glass FN (GFN) blood culture bottle. Each pair of PFA-PFN or GFA-GFN bottles was labeled with the number one or two in order to facilitate collecting first into the PFA-PFN pair for approximately one-half of the patients while the remainder of the patient draws were collected first into the GFA-GFN pair. The standard Geisinger Medical Center protocol was followed for skin preparation prior to collection. Blood volumes were verified (8 to 12 ml) by the use of a visual standard.

For the sake of clarity, the following terminology is consistently utilized in this report: the term “set” refers to all four bottles collected during a patient episode, the term “pair” refers to two bottles collected by a single collection procedure, and the term “bottle” refers to a single bottle. Hence, there are two bottles in a pair and two pairs in a set. All bottles were incubated in BacT/ALERT blood culture cabinets at 35°C, using a 5-day protocol. Culture workup and identification were performed using the standard Geisinger Medical Center microbiology laboratory procedures. All cultures were worked up and reported by the standard protocols used by the microbiology laboratory at Geisinger Medical Center. A 5-day incubation period was used for all negative bottles.

If one or more bottles from a set gave a positive signal by the instrument and there was growth upon subculturing, any remaining bottles from the set that were instrument negative after 5 days of incubation were subcultured.

For the purposes of this study, isolates of coagulase-negative staphylococci, *Bacillus* spp., viridans group streptococci, *Propionibacterium* spp., and aerobic diphtheroids isolated from only one pair of bottles in a set were considered not to be clinically significant. Statistical analysis was performed as described by Ilstrup (1).

RESULTS

A total of 4,040 sets of four bottles were collected. Of these 4,040 sets, 3,110 contained the recommended 8 to 12 ml of blood in each of the four bottles. These 3,110 sets yielded 524 microorganisms, including 359 judged to be clinically significant and 165 judged to be probable contaminants. Of the 359 significant organisms, 255 were recovered in either one or two bottles from both pairs of bottles in a set while 56 were recovered only from the glass bottles and 48 were recovered only from the plastic bottles ($P > 0.05$; not significant [NS]) (Table 1). Separate analyses were performed to compare the performances of the two aerobic (PFA and GFA) and the two anaerobic (PFN and GFN) bottles. Of the 286 significant organisms recovered in the FA bottles (both glass and plastic), 180 were recovered in both bottles, 57 in the PFA bottles only, and 49 in the GFA bottles only (P , NS) (Table 2). Of the 303 significant organisms recovered in the FN bottles (both glass and plastic), 212 were recovered in both bottles, 46 in the plastic bottles only, and 45 in the glass bottles only (P , NS) (Table 3). For individual organisms, the only significant difference in recovery was obtained for *Escherichia coli*, with more isolates recovered in the plastic FN bottles ($P = 0.02$). There were no significant differences in the number of probable contaminants isolated between the glass and the plastic bottle sets overall or between the aerobic bottles. However, more contaminants were recovered from the plastic anaerobic bottles than were recovered from the glass anaerobic bottles ($P = 0.03$).

Subculture of bottles that were reported as negative by the

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TABLE 1. Comparative yield of 524 isolates from paired plastic FA-FN and glass FA-FN blood culture bottles

Isolate	No. of isolates from:			<i>P</i> value ^a
	Both bottle pairs	Plastic bottles only	Glass bottles only	
Significant isolates				
Gram-positive cocci	183	18	30	NS
<i>S. aureus</i>	102	14	21	NS
Coagulase-negative <i>Staphylococcus</i>	43	0	0	NC
Enterococci ^b	18	3	6	NS
<i>Streptococcus pneumoniae</i>	8	0	1	NC
Other streptococci ^c	12	1	2	NS
<i>Listeria monocytogenes</i>	1	0	0	NC
Gram-negative bacilli	55	17	16	NS
<i>E. coli</i>	30	10	7	NS
Other <i>Enterobacteriaceae</i> ^d	21	2	5	NS
Other gram-negative bacilli ^e	4	5	4	NS
Anaerobes ^f	10	7	8	NS
Yeasts ^g	6	6	2	NS
Total	255	48	56	NS
Probable contaminants				
Coagulase-negative <i>Staphylococcus</i>	0	72	57	NS
<i>Micrococcus</i> sp.	0	1	3	NC
Viridans group streptococci	0	8	2	NS
Aerobic gram-positive bacilli	0	11	8	NC
Anaerobic gram-positive bacilli	0	2	1	NC
Total	0	94	71	NS

^a NC, not calculated if total number of isolate is ≤10.^b Includes 20 *Enterococcus faecalis*, 5 *Enterococcus faecium*, and 2 *Enterococcus* sp. isolates.^c Includes seven group B streptococci, one group G isolate, one *Streptococcus bovis* variant, and six viridans group streptococcal isolates.^d Includes two *Citrobacter freundii*, one *Citrobacter koseri*, one *Citrobacter* sp., three *Enterobacter aerogenes*, four *Enterobacter cloacae*, one *Enterobacter gergoviae*, one *Klebsiella oxytoca*, nine *Klebsiella pneumoniae*, one *Morganella morganii*, one *Proteus mirabilis*, one *Salmonella* sp., and three *Serratia marcescens* isolates.^e Includes three *Achromobacter xylosoxidans*, one *Flavimonas oryzae*, one *Shewanella putrefaciens*, six *Pseudomonas aeruginosa*, and two unidentified oxidase-positive, gram-negative bacillus isolates.^f Includes one *Bacteroides caccae* isolate, four *Bacteroides fragilis* isolates, one *Bacteroides fragilis* group isolate, one *Bacteroides thetaiotaomicron* isolate, two *Bacteroides vulgatus* isolates, one *Campylobacter gracilis* isolate, two *Clostridium* sp. isolates, one *Clostridium perfringens* isolate, one *Clostridium septicum* isolate, two *Eubacterium lentum* isolates, one *Fusobacterium* sp. isolate, two anaerobic gram-negative bacilli, four anaerobic gram-positive cocci, and two anaerobic gram-positive bacillus isolates.^g Includes nine *Candida albicans*, three *Candida glabrata*, one *Candida tropicalis*, and one *Cryptococcus neoformans* isolate.

instrument but had a companion positive bottle from the set detected by the instrument yielded 17 organisms. There were 10 *Pseudomonas aeruginosa* isolates (5 from PFN and 5 from GFN bottles) and 1 *Alcaligenes* sp. isolate (PFN) detected by subculture from an anaerobic bottle of a pair that had a positive aerobic bottle. One isolate of *Eubacterium lentum* was detected in a PFN bottle but not in the GFN bottle, while one isolate of *Pseudomonas aeruginosa* was instrument positive in a

TABLE 2. Comparative yield of 388 isolates from plastic aerobic (FA) and glass aerobic (FA) blood culture bottles by bottle type

Isolate	No. of isolates from:			P value ^a
	Both bottles	Plastic aerobic bottles	Glass aerobic bottles	
Significant isolates				
Gram-positive cocci	121	33	31	NS
<i>S. aureus</i>	64	19	17	NS
Coagulase-negative <i>Staphylococcus</i>	26	6	5	NS
Enterococci ^b	14	4	7	NS
<i>Streptococcus pneumoniae</i>	7	1	1	NC
Other streptococci ^c	10	3	1	NS
<i>Listeria monocytogenes</i>	1	0	0	NC
Gram-negative bacilli	51	19	16	NS
<i>E. coli</i>	24	9	8	NS
Other <i>Enterobacteriaceae</i> ^d	24	5	4	NS
Other gram-negative bacilli ^e	3	5	4	NS
<i>Eubacterium lentum</i>	1	0	0	NC
Yeasts ^f	6	5	2	NS
Total	180	57	49	NS
Probable contaminants				
Coagulase-negative <i>Staphylococcus</i>	0	46	38	NS
<i>Micrococcus</i> sp.	0	1	3	NC
Viridans group streptococci	0	5	1	NS
Aerobic gram-positive bacilli	0	5	3	NC
Total	0	57	45	NS

^a NC, not calculated when total number of isolates is ≤10.^b Includes 17 *Enterococcus faecalis*, 6 *Enterococcus faecium*, and 2 *Enterococcus* sp. isolates.^c Includes six group B, one group G, one *Streptococcus bovis* variant, and six viridans group streptococcal isolates.^d Includes two *Citrobacter freundii*, one *Citrobacter koseri*, one *Citrobacter* sp., three *Enterobacter aerogenes*, three *Enterobacter cloacae*, six *Enterobacter gergoviae*, one *Klebsiella oxytoca*, eight *Klebsiella pneumoniae*, one *Morganella morganii*, four *Proteus mirabilis*, one *Salmonella* sp., and two *Serratia marcescens* isolates.^e Includes two *Achromobacter xylosoxidans*, one *Flavimonas oryzae*, six *Pseudomonas aeruginosa*, one *Shewanella putrefaciens*, and two unidentified oxidase-positive, gram-negative bacillus isolates.^f Includes eight *Candida albicans*, three *Candida glabrata*, one *Candida tropicalis*, and one *Cryptococcus neoformans* isolate.

PFA bottle but detected only by subculture from the GFA bottle. One *Alcaligenes* sp. isolate was detected by subculture of the PFN bottle, while the GFA bottle from the set was instrument positive for an isolate of *Klebsiella pneumoniae* and the GFN bottle from the same set was instrument positive for an isolate of *Serratia marcescens*. Two *Propionibacterium* sp. isolates and one *Corynebacterium* sp. isolate were detected by subculture of bottles while each of the paired bottles was instrument positive for isolates of a *Staphylococcus* sp. other than *Staphylococcus aureus*.

DISCUSSION

Since the introduction of the original BacT/ALERT blood culture system, several “generations” of new bottles have been

TABLE 3. Comparative yield of 391 isolates from plastic anaerobic (FN) and glass anaerobic (FN) blood culture bottles by bottle type

Isolate	No. of isolates from:			<i>P</i> value ^a
	Both bottles	Plastic anaerobic bottles	Glass anaerobic bottles	
Significant isolates				
Gram-positive cocci	161	21	26	NS
<i>S. aureus</i>	95	15	15	NS
Coagulase-negative <i>Staphylococcus</i>	31	3	4	NS
Enterococci ^b	18	1	5	NS
<i>Streptococcus pneumoniae</i>	7	2	0	NC
Other streptococci ^c	10	0	2	NS
<i>Listeria monocytogenes</i>	1	0	0	NS
Gram-negative bacilli	44	13	7	NS
<i>E. coli</i>	24	10	2	0.02
Other <i>Enterobacteriaceae</i> ^d	20	3	5	NS
Anaerobes ^e	6	11	12	NS
<i>C. albicans</i>	0	1	0	NC
Total	212	46	45	NS
Probable contaminants				
Coagulase-negative <i>Staphylococcus</i>	0	40	27	NS
Viridans group streptococci	0	6	1	NS
Aerobic gram-positive bacilli	0	6	5	NC
Anaerobic gram-positive bacilli	0	2	1	NC
Total	0	54	34	0.03

^a NC, not calculated when total number of isolates is ≤10.

^b Includes 19 *Enterococcus faecalis*, 3 *Enterococcus faecium*, and 2 *Enterococcus* sp. isolates.

^c Includes seven group B, one group G, and four viridans group streptococcal isolates.

^d Includes one *Citrobacter freundii*; one *Citrobacter koseri*, one *Citrobacter* sp., three *Enterobacter aerogenes*, four *Enterobacter cloacae*, one *Enterobacter gergoviae*, one *Klebsiella oxytoca*, seven *Klebsiella pneumoniae*, one *Morganella morganii*, four *Proteus mirabilis*, one *Salmonella* sp., and three *Serratia marcescens* isolates.

^e Includes two *Bacteroides caccae*, four *Bacteroides fragilis*, one *Bacteroides fragilis* group, one *Bacteroides thetaiotaomicron*, three *Bacteroides vulgatus*, one *Campylobacter gracilis*, one *Clostridium perfringens*, one *Clostridium septicum*, two *Clostridium* sp., one *Fusobacterium* sp., and two *Eubacterium lentum* isolates, two anaerobic gram-negative bacilli, five anaerobic gram-negative cocci, and three anaerobic gram-positive bacilli.

introduced. Initially, only bottles with the designations “standard aerobic” and “standard anaerobic” were available; at a later time, additional bottles with the designations “FAN aerobic” and “FAN anaerobic” were introduced. The FAN bottles were designed to enhance the recovery of fastidious bacteria, bacteria from patients receiving antimicrobial therapy, and yeasts in comparison to recovery with the standard BacT/ALERT blood culture bottles (4, 5). Subsequently, nonvented BacT/ALERT bottles, designated FA and FN, replaced the

FAN aerobic and FAN anaerobic blood culture bottles. The FA and FN bottles have a different medium formulation and a different type of sensor than the FAN bottles (2). The FA bottles contain a different concentration of charcoal, a smaller volume of medium, and more headspace gas than the FAN aerobic bottles. Lastly, the FA bottles are unvented, while the FAN aerobic bottles required transient venting.

In addition to the obvious change from glass to plastic in the bottles, the plastic bottle necessitated a change in the liquid sensor used in the bottles (Steven Rothenberg, personal communication). No changes were made in media or atmosphere in the bottles. Consequently, it is not unexpected that there would be little if any difference in recovery between the glass and plastic bottles. Petti et al. reported no difference in recovery between plastic and glass PF bottles (3).

The plastic bottles weigh approximately 4 pounds less per 100 bottles than glass bottles. In our laboratory, which performs around 25,000 blood cultures per year, use of plastic bottles results in approximately 2,000 pounds less of biohazard waste per year than use of glass bottles. The potential cost savings for biohazard waste disposal would be determined by disposal costs for each laboratory.

When we routinely used glass bottles in our laboratory, we periodically received a bottle that was cracked during transit to the laboratory. Because the glass bottles had a plastic wrapper to curtail shattering if dropped, cracks that developed as a result of dropping were not always visually apparent. On several occasions, we did not detect cracked bottles until after they were loaded into the instruments. Subsequent leaks required decontamination of individual cells and, on one occasion, decontamination of an entire block. This potential problem should be eliminated with plastic bottles.

In conclusion, in our evaluation, which included 3,110 blood culture sets, we found no significant difference in recovery of clinically significant microorganisms between the glass and plastic FA and FN bottles, although the plastic bottles did recover more organisms. Moreover, plastic bottles offer greater safety and, potentially, lower disposal costs than glass bottles.

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