Comparison of Conventional and Revised Isolator Blood Culture Tubes

BARBARA A. BASILLE, KIRK A. EASLEY, GERALDINE S. HALL, AND JOHN A. WASHINGTON II*

Cleveland Clinic Foundation, Cleveland, Ohio 44106

Received 5 June 1987/Accepted 3 August 1987

A paired clinical study compared bacterial and fungal recovery from 4,553 blood cultures processed by the conventional Isolator (Du Pont Co.) and a revised Isolator consisting of a single-stoppered, round-bottom tube containing the same ingredients as the conventional tube except for an inert fluorochemical. Excluding contaminants, there were 425 positive blood cultures with 450 isolates representing 208 patients. There were no statistically significant differences between systems in the number of positive cultures or patients with positive cultures for each organism group studied, nor were there any statistically significant differences between systems in the time required for detection of positive cultures.

Lysis-concentration has proved useful for isolating bacteria, mycobacteria, and fungi (1, 2, 4, 5). In its commercially available form, the lysis-centrifugation tube (Isolator; Du Pont Co., Wilmington, Del.) originally consisted of a doublestoppered tube containing sodium polyanetholsulfonate, disodium ethylenedinitrilotetraacetate, saponin, propylene glycol, and a dense, inert fluorochemical that served as a cushion during centrifugation. After blood was inoculated into the tube, processing involved centrifugation and removal of the spun sediment by syringe and needle for culture. With the advent of the Isostat (Du Pont), it was no longer necessary to remove the sediment by this technique, and the stopper at the bottom of the tube no longer served a useful function. Accordingly, a revised Isolator consisting of a single-stoppered, round-bottom tube without the fluorochemical was developed.

The purpose of this study was to compare the performance characteristics of the revised and conventional Isolator tubes in a clinical study of blood from patients with suspected septicemia.

From 3 November 1986 through 26 January 1987, blood cultures were collected from patients with suspected septicemia in the following manner. The venipuncture site was prepared by using 70% alcohol followed by 3% iodine. A 20-gauge 1-in. (2.54-cm) needle was used on a 20-ml sterile syringe to draw 20 ml of blood for each blood culture requested. Equal volumes of the blood were inoculated into a conventional (10-ml) Isolator tube and a revised (10-ml) Isolator tube. Both tubes were inverted gently three or four times for thorough mixing.

Both tubes were transported to the laboratory in a timely manner and processed simultaneously as previously described (2), except that the centrifugation sediment was neither removed from each tube nor inoculated in a biological safety cabinet. The concentrates from both tubes were inoculated onto each of the following media: two Trypticase soy blood agar plates, one chocolate blood agar plate, and one potato glucose agar plate (Scott Laboratories, Inc., Richmond, Calif.). One Trypticase soy blood agar plate and the chocolate blood agar plate were incubated in 5 to 8% CO₂ at 35°C and examined daily for a full 72 h; one Trypticase soy blood agar plate was incubated in an anaerobic atmosphere at 35°C for 6 days and examined every 48 h; and the potato glucose agar plate was incubated at 30° C and examined every 48 h for 14 days. All plates were incubated with the agar side down for the first 24 h.

The number of CFU per milliliter was calculated for positive blood cultures for each Isolator system. Contamination rates were defined by growth within the streaked area (on streak) of each medium in single positive cultures yielding *Bacillus* spp., *Corynebacterium* spp., and ≤ 1 CFU of coagulase-negative staphylococci per ml.

The number of positive cultures and the time required for detection of growth by each system were analyzed with the McNemar test and the paired Student *t* test, respectively, as described elsewhere (3). The Statistical Analysis System (SAS Institute, Inc., Cary, N.C.) was used for all analyses and management of the data base. Datum analysis was limited to those culture sets in which each Isolator tube in a pair contained ≥ 6 ml of blood and was processed in ≤ 8 h from the time of collection. In the event of polymicrobial septicemia, each isolate was analyzed separately. A single episode of bacteremia or fungemia was defined by the

TABLE 1. Positive cultures with two Isolators

Organism group	No. of positive cultures with the following Isolator(s):		
	Conventional	Revised	Both
Bacteroides spp.	5	0	7
Clostridium spp.	0	2	0
Anaerobic coccus	1	2	1
Staphylococcus aureus	12	9	104
Staphylococcus, coagulase-negative ^a	8	5	18
Streptococcus, group D	6	1	11
Streptococcus, viridans group	15	16	13
Streptococcus, beta-hemolytic	Ò	0	2
Streptococcus pneumoniae	0	0	4
Enterobacteriaceae ^b	17	21	61
Pseudomonas aeruginosa	2	2	13
Candida spp.	7	8	44
Cryptococcus neoformans	0	0	2
Torulopsis spp.	3	5	6
Histoplasma capsulatum	2	2	13

" Data limited to cultures yielding ≥ 1 CFU/ml.

^{*} Corresponding author.

^b Includes 44 Escherichia coli cultures, 12 Klebsiella pneumoniae cultures, 12 Enterobacter cloacae cultures, 11 Serratia marcescens cultures, 6 Proteus mirabilis cultures, 5 Citrobacter freundii cultures, 5 Klebsiella oxytoca cultures, 2 Morganella morganii cultures, and 2 Enterobacter agglomerans cultures.

isolation of the same microorganism from one or multiple cultures from the same patient within 2 weeks from the time of the initial positive culture with that microorganism.

A total of 4,553 blood cultures was obtained during the study. There were 425 positive cultures (9%) yielding 450 clinically significant isolates, representing 208 episodes of sepsis: Accordingly, there were 4,128 negative cultures representing 158 patients. The numbers of positive cultures from the conventional and revised Isolator tubes, separately and together, are listed in Table 1. No statistically significant differences were detected between methods for positive cultures of each organism group (Table 1) or for patients with positive cultures of each organism group (data not shown). Whether the trends toward more frequent recovery of Bacteroides spp. and group D streptococci from the conventional Isolator would achieve statistical significance (P <0.05) with a larger sample is unknown. There was no statistically significant difference between systems for each organism group or for all organism groups combined (P <0.64) in the interval required for the detection of positive cultures. Isolates of Histoplasma capsulatum required, on the average, 8 days (standard deviation, ± 3.2 days) and 8.7 days (standard deviation, ± 3.2 days) for isolation from the conventional and revised Isolators, respectively, and were recovered only from potato glucose agar plates incubated at 30°C. The overall contamination rate was 7% and did not differ significantly between systems.

In conclusion, the revised Isolator tube displayed performance characteristics that were virtually identical to those of the conventional Isolator tube. The major advantage of the revised system is greater convenience in the centrifugation process, since placement of the tube in a fixed-head centrifuge is no longer required.

We are grateful to personnel in the Central Processing, Bacteriology, and Mycology laboratories for processing blood cultures concurrently with the two lysis-centrifugation systems and to Frank Bican and Barbara Kirkley for assistance in datum collection.

We are grateful to the Du Pont Co., Wilmington, Del., for financial support.

LITERATURE CITED

- Bille, J., L. Stockman, G. D. Roberts, C. D. Horstmeier, and D. M. Ilstrup. 1983. Evaluation of a lysis-centrifugation system for the recovery of yeasts and filamentous fungi from blood. J. Clin. Microbiol. 18:469–471.
- Henry, N. K., C. A. McLimans, A. J. Wright, R. L. Thompson, W. R. Wilson, and J. A. Washington II. 1983. Microbiological and clinical evaluation of the Isolator lysis-centrifugation blood culture tube. J. Clin. Microbiol. 17:864–869.
- 3. Ilstrup, D. M. 1978. Statistical methods employed in the study of blood culture media, p. 31–39. *In* J. A. Washington (ed.), The detection of septicemia. CRC Press, Inc., Boca Raton, Fla.
- Kelly, M. T., M. F. Fojtasek, T. M. Abbott, D. C. Hale, J. R. Dizikes, R. Boshard, G. E. Buck, W. J. Martin, and J. M. Matsen. 1983. Clinical evaluation of a lysis-centrifugation technique for detection of septicemia. J. Am. Med. Assoc. 250:2185–2188.
- Kiehn, T. E., J. W. M. Gold, P. Brannon, R. J. Timberger, and D. Armstrong. 1985. Mycobacterium tuberculosis bacteremia detected by the Isolator lysis-centrifugation blood culture system. J. Clin. Microbiol. 21:647–648.