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Comparative study of a non-radiometric BACTEC system and a conventional blood culture system in a clinical microbiology laboratory

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Abstract

The BACTEC NR730 blood culture system is a commercially available system which utilizes infrared spectrophotometry to detect bacterial growth in broth media inoculated with the blood specimen. The objective of this study is to compare this system with the conventional system used in our laboratory. A total of 479 blood samples were collected from adult patients in the Klang TAR Hospital and inoculated simultaneously into the two systems. Clinically significant isolates were recovered from 59 (12.3%) specimens by one or both systems yielding a total of 65 isolates of which 41 isolates were recovered in both systems, 18 in BACTEC only and 6 in the conventional system ($P < 0.01$). Of the 40 (8.3%) blood cultures that were judged as contaminated, 2 were found in both systems, 12 in the BACTEC only and 26 in the conventional system. After 48 hours (day 2) the BACTEC system had recovered 93.2% of the total significant isolates while the conventional system recovered 61.7% only. Our results showed that the BACTEC system yielded more significant isolates in a shorter time and with a lower incidence of contamination as compared to the conventional system.

Key works: Blood culture system, infrared spectrophotometry.

INTRODUCTION

The rapid detection of bacteremia is one of the important roles of a clinical microbiology laboratory. Various commercial and automated systems for blood culture have been designed to enable rapid recovery of microorganisms. These systems vary in techniques used for detecting microbial growth, the types of broth media and media supplements available for use with each system, bottle atmospheres, the blood-to-broth ratio, the volume of blood to be inoculated and the use of shakers or agitation for aerobic bottles. [1-4] Many studies have been done to evaluate a variety of commercially available blood culture systems [2-5] but as yet no agreement has been reached as to which of these systems is optimal for the isolation of a wide variety of microorganisms.

A study by Courcol *et al*[2] in evaluating a non-radiometric blood culture system BACTEC NR660 (Becton Dickinson) has shown that it provided a quicker detection of positive blood cultures and produced a highly acceptable level of reliability.

Our present study is to compare another non-radiometric blood culture system BACTEC NR730 (Becton Dickinson) which utilizes infrared spectrophotometry to detect carbon dioxide generated by microorganisms in specially formulated culture media with the conventional system used in our laboratory.

MATERIALS AND METHODS

Collection of samples

A total of 479 paired blood culture sets were colected from adult patients with suspected bacteremia from several wards of Klang Tengku ampuan Rahimah Hospital (TAR) Hospital. Each set consisted of a pair of BACTEC NR730 bottles, one containing aerobic medium NR16A and the other containing anaerobic medium NR17A and a pair of conventional bottles (IMR media), that consisted of trytic soya broth with 0.03% of sodium polyanetholesulfonate (SPS) as the aerobic medium and thiolglycollate broth with 0.05% of SPS and 0.01% agar as the anaerobic medium. Approximately 12ml of blood was collected aseptically by venipuncture from each patient and an equal volume (3 ml) was inoculated into each bottle and dispatched to the laboratory on the same day it was collected.

Processing of samples

Upon receipt at the Klang TAR Hospital pathological laboratory, the conventional bottles were incubated at 37°C overnight. A blind subculture was done at the end of 24 hrs incubation for both the aerobic bottles and anaerobic bottles and a gram stained smear was made for microscopic examination. Visual inspection was done once daily from day 1 to 6 for evidence of

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microbial growth. After the first blind subculture, unless the visual inspection showed macroscopic evidence of microbial growth, a final blind subculture was done at the end of 6 days incubation before discarding.

The BACTEC bottles were incubated at 37°C overnight (the aerobic bottle in an orbital shaker) and dispatched to IMR microbiology laboratory for further processing. These bottles were then tested once daily on BASTEC NR730 instrument. Aerobic bottles with a growth value of ≥ 30 and anaerobic bottle of ≥ 15 were subcultured on appropriate media and a gram stained smear was made for microscopic examination. When only one bottle in a set was positive, processing of the negative bottle was continued. Terminal subcultures were done on day 7 on all bottles before discarding.

Recording and analysis of data

The following information was recorded on a report sheet for each set of positive bottles for both the conventional and BACTEC systems: bottle identification, time of sample collection, nature of antimicrobial therapy and time of antibiotic administration, results for conventional and BACTEC bottles, subculture results and organism identifications. A paired comparison was performed on positive culture bottles and the identified microorganisms were judged as significant isolates or contaminants according to clinical evaluations. All contaminants were not included in the statistical calculations.

Statistical analysis

The chi-square test was used to test for significant difference in the recovery of organisms between the two systems and the fisher's exact test was used for small numbers.

RESULTS

During the study period, a total of 479 sets of four culture bottles were received and codmpared. There were 52, 40, 33 and 34 positive culture bottles with BACTEC NR16A, NR17A, conventional aerobic and anaerobic bottles respectively giving a total of 59 sets positive by one or both systems, 16 from BACTEC system alone and 6 from conventional system.

A total of 65 isolates were recovered from these positive cultures of which 40 isolates were recovered from both systems, 17 were recovered in BACTEC system and 6 were recovered from the conventional system (P<0.01) (Table 1).

TABLE 1: Comparative yield of clinically signiicant isolates from BACTEC and conventional system

Organism	No. of isolates recovered from BACTEC and Conventional	BACTEC only	Conventional only	P value
Facultative bacteria (gram positive)				<0.01
Staphylococcus aureus	4	1		
Enterococcus		1		
Viridans Streptococcus		1		
Streptococcus Group A	1			
Aerobic and facultative bacteria (gram negative)				<0.01
Enterobacter spp.	1	2	4	
Escherichia coli	11	4		
Klebsiella spp.	13	2	2	
Pseudomonas spp.	3			
Pseudomons aeruginosa	1			
Haemophilus influenzae	1			
Proteus spp.	1	1		
Salmonella enteritidis		1		
Acinetobacter spp.	4	4		

All the isolates recovered were aerobic and facultativeanaerobic organisms. Recovery of gram positive organisms ($P<0.01$) was significantly better with the BACTEC system. When analysing the results, an evaluation of the clinical history of the patients was taken into consideration and probable contaminants were excluded from the analysis of positive cultures to define more precisely the performance of the BACTEC and conventional systems.

The total number of blood cultures that were judged as contaminated were 40 (8.3%), 2 of them were found in both systems, 12 in BACTEC only and 26 in the conventional system. The predominant contaminant encountered by the BACTEC system was the Bacillus sp. Whereas in the conventional system probable contaminants were usually mixed growths of a few types of organisms (Table 2).

The speed for the detection of positivity by the two systems was compared in terms of the cumulative number of significant isolates detected per day and the cumulative percentage per day was. calculated from days 1 to 6. The time to positivity was calculated from the day the specimen was collected (Day 0) to the end of 6 days incubation period (Day 6).

The BACTEC system recovered 72.9% of the total significant isolates on Day 1 and 93.6% by Day 2 whilst the corresponding percentages for the conventional system were 55.3% and 61.7% respectively. The BACTEC system recovered all the isolates by Day 4 but the conventional system by Day 6 (Fig. 1).

DISCUSSION

The BACTEC NR660 system was the first non-radiometric blood culture system introduced which had its accuracy and sensitivity documented in various evaluations. This system was designed to meet the needs of laboratories with larga test volumes, but for the smaller volume laboratory the BACTEC NR730 system would be a more suitable choice.

One of the main advantages of the BACTEC system found from the results of our study was that this system yielded more significant isolates in a shorter time as compared to the conventional system. The BACTEC system recovered 93.6% of the total significant isolates by 48 hours (Day 2) yielding a total of 55 isolates as compared to the conventional system which recovered 61.7% on the same day yielding a total of 29 isolates (Fig. 1).

The reason for the difference in the yield and the speed of recovery of microorganisms between the two systems was not clear but the BACTEC bottles differed in the broth base used which was soybean-casein digest with nutritional supplements and antibiotic binding resins.

TABLE 2: Comparative of the types of contaminants found in the BACTEC and conventional system

Organism	No. of sets contaminated		
	BACTEC and Conventional	BACTEC only	Conventional only
Bacillus spp.	2	5	5
Coagulase negative Staphlycoccus		3	3
Non-fermentative gram negative rods			2
Diphteroids		1	2
Micrococcus spp.		1	
Asporegenous fungi		2	
Mixed growth of more than 3 types			14

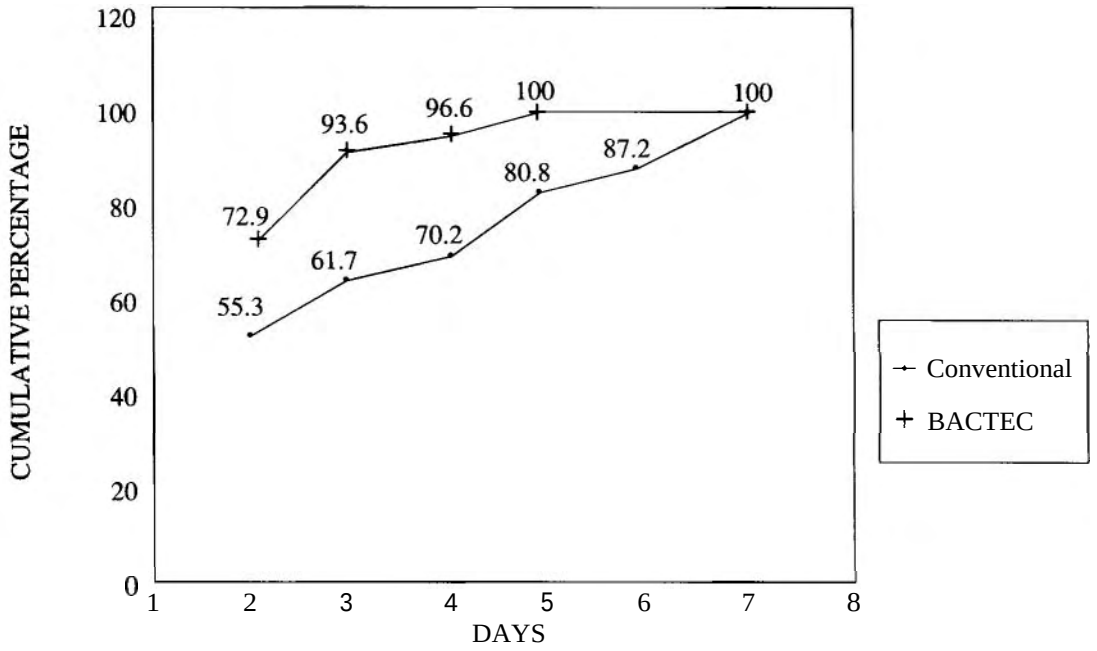

FIG. 1: Comparison of the cumulative percentage of significant isolates from BACTEC and conventional systems during the 6 days incubation period.

In a routine evaluation study of the BACTEC resin media (NR16A and NR17A) versus the BACTEC media without resin (NR6A and NR7A), it was reported that the former recovered significantly more microorganisms in a shorter time from the blood of patients undergoing antimicrobial therapy.[3] despite their inefficiency in removing some antibiotics, the resins in blood culture medium could increase the recovery of microorganisms because of the enhanced lysis of leukocytes.[6]

The shorter detection time for microbial growth obtained by the BACTEC system could be due to the difference in the technique of processing the blood cultures. The BACTEC bottles (both aerobic and anaerobic) were examined by an instrument once daily to detect the presence of C 0 2 generated by microorganisms, giving a specific growth value for the decision to perform subcultures, but for the conventional bottles, visual inspection for microbial growth is an important procedure to decide subsequent subcultures when the first blind subculture is negative. The detection of turbidity or other evidence of microbial growth could be masked, delayed or missed by factors such as lipemic blood, light penetration in the bottle, frosty bottles caused by repeated usage or certain organisms which tend to clump and settle to the bottom of the culture broth.

During our study period, we performed terminal subcultures at the end of 6 days incubation period of the BACTEC aerobic and anaerobic bottles. Our data showed that none grew up any significant isolates other than those recovered earlier. We recovered 4 isolates from the aerobic medium, which by clinical evaluations we had judged as contaminants and one significant isolate from the anaerobic bottle, which also grew in the aerobic bottle. This shows that terminal subcultures are not necessary for the BACTEC system as they do not increase the number of significant positive blood cultures. Our results are consistent with those reported by Wilson *et al*[5] that terminal subcultures are unnecessary with BACTEC blood culture bottles.

The conventional system was significantly more likely to be contaminated than the BACTEC system. Our results showed that out of the 40 positive specimens that were judged as contaminated, 2 were found in both systems, 12 in the BACTEC only and 26 in the conventional system. Contamination in blood cultures could occur in many ways such as skin contamination when the blood specimen was obtained, inadequate sterilization of the broth media during production or exogenous contamination during subculture of the blood specimen in the laboratory. The need for more subcultures by the conventional system with frequent exposures of the incubating broth during the process of subculturing could be one of the probable reason for the higher contamination rate in this study.

In summary, we would like to conclude that the use of the BACTEC NR730 system offers several advantages over the conventional blood culture system. Besides providing a higher yield of significant microorganisms in a shorter time and with a lower incidence of contamination, the BACTEC system requires less technician time.

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