



A COMPARATIVE STUDY OF CONVENTIONAL METHOD VERSUS BACTEC METHOD IN BACTERIOLOGICAL PROFILE OF SEPTICEMIA

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Abstract: Sepsis is common and also more dangerous in elderly, immune-compromised, and critically ill patients. It occurs in 1–2% of all hospitalizations and accounts for as much as 25% of intensive-care unit (ICU) bed utilization. It is a major cause of death in intensive-care units worldwide, with mortality rates that range from 20% for sepsis to 40% for severe sepsis to >60% for septic shock. A total of 160 blood samples were collected from patients with suspected septicemia from several wards of Medwin Hospital. Alternate samples were Cultured by Bactec 9050 (Becton Dickinson Microbiology Systems, Sparks, Md) and conventional methods. And 12 random samples were cultured by both Bactec 9050 and conventional methods. It was observed that out of 160 cases 43 (26.9%) of septicemia positive cases were detected by one or the other method. out of 80 samples cultured by Bactec method 29 (36.2%) were Bactec positive cases. And out of 80 samples cultured by Conventional method 14 (17.5%) were Conventional positive cases. In a statistical comparison of Bactec and Conventional method, Sensitivity for Bactec method was seen as 90% whereas for conventional method it was 60%. Bactec method has the highest specificity i.e. 100% whereas Conventional method has 75%. The Bactec performed better than the conventional in overall detection, time to detection, number of false-negatives, and missed episodes of simulated septicemia.

Keywords: Septicemia; Bactec method; Conventional method

INTRODUCTION

Septicemia is a serious medical condition characterized by inflammation of the whole body leading to various organs functional damage. It can arise from infections throughout the body, including infections in the lungs, abdomen, and urinary tract. It may come before or at the same time as infections of the bone (osteomyelitis), central nervous system (meningitis), heart (endocarditis), or other tissues.

Severe sepsis occurs, when sepsis leads to organ dysfunction, low blood pressure (hypotension), or insufficient blood flow (hypoperfusion) to one or more organs (causing, for example, lactic acidosis, decreased urine production, or altered mental status). Sepsis can lead to septic shock, Multiple Organ Dysfunction Syndrome (MODS) (formerly known as multiple organ failure), and death. Organ dysfunction results from sepsis-induced hypotension (< 90 mmHg or a reduction of ≥ 40 mmHg from baseline) and diffuse intravascular coagulation, among other things.

Common bacterial causes of sepsis are gram-negative bacilli (for example, *E. coli*, *P. aeruginosa*, *E. corrodens*), *S. aureus*, *Streptococcus* species and *Enterococcus* species; however, there are a large number of bacterial genera that have been known to cause sepsis. *Candida* species are some of the most frequent fungi that cause sepsis. In general, a person

with sepsis can be contagious, so precautions such as hand washing, sterile gloves, masks, and clothing coverage should be considered depending on the patient's infection source.¹

According to the American College of Chest Physicians and the Society of Critical Care Medicine, there are different levels of sepsis²:

* Systemic inflammatory response syndrome (SIRS):

Defined by the presence of two or more of the following findings:

- Body temperature < 36 °C (97 °F) or > 38 °C (100 °F) (hypothermia or fever).
- Heart rate > 90 beats per minute (tachycardia).
- Respiratory rate > 20 breaths per minute or, on blood gas, a PaCO₂ less than 32 mm Hg (4.3 kPa) (tachypnea or hypocapnia due to hyperventilation).
- White blood cell count < 4,000 cells/mm³ or > 12,000 cells/mm³ (< 4 × 10⁹[citation needed] or > 12 × 10⁹ cells/L), or greater than 10% band forms (immature white blood cells). (Leukopenia, leukocytosis, or bandemia).

***Sepsis:** Defined as SIRS in response to a confirmed infectious process. Infection can be suspected or proven (by culture, stain, or polymerase

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chain reaction (PCR)), or a clinical syndrome pathognomonic for infection.

***Severe sepsis.** Defined as, sepsis with organ dysfunction, hypo perfusion, or hypotension.

***Septic shock.** Defined as, sepsis with refractory arterial hypotension or hypo perfusion abnormalities in spite of adequate fluid resuscitation.

Sepsis is common and also more dangerous in elderly, immunocompromised, and critically ill patients. It occurs in 1–2% of all hospitalizations and accounts for as much as 25% of intensive-care unit (ICU) bed utilization. It is a major cause of death in intensive-care units worldwide, with mortality rates that range from 20% for sepsis to 40% for severe sepsis to >60% for septic shock.²

With this background this study is an attempt to know the bacteriological profile of septicemia cases by comparative diagnosis by conventional method and bactec method.

MATERIALS AND METHODS

This study was conducted at Medwin Hospital, Hyderabad. A total of 160 blood samples were collected from patients with suspected septicemia from several wards of Medwin Hospital. Alternate samples were Cultured by Bactec 9050 (Becton Dickinson Microbiology Systems, Sparks, Md) and conventional methods. Thus a total of 80 samples were cultured by Bactec 9050 and 80 samples were cultured by conventional methods. And 12 random samples were cultured by both Bactec 9050 and conventional methods. Approximately 10ml of blood was collected aseptically by venipuncture from each patient, each 5ml was transferred into Brain heart infusion broth and BACTEC plus aerobic/F (soybean casein digest broth). According to the manufacturer's guide the inoculated culture bottles were loaded into Bactec 9050 instrument and incubated at 37°C identification of etiological pathogens was based on routine bacteriological procedures. Routinely, the bottles were incubated in Bactec 9050 for seven days. During these days, if the system alerted for positive blood culture, 2 to 4 drops of blood culture samples were inoculated on Mac Conkey's agar and chocolates agar and 5% sheep blood agar.

Then, after 24- 48hrs of incubation aerobically, the samples were stained by Gram's Method. The organisms were identified by using preliminary tests and Bio-chemical reactions for different organisms.

RESULTS

In the present study of 6 months from January 2010 to July 2010 at Medwin Hospital Hyderabad, the total no. of cases taken for blood tests was 160.

Table.1: Showing gender distribution of septicemia cases by bactec and conventional methods

Sex	Positive cases		Negative cases		Total	
	No	%	No	%	No	%
Male	26	60	59	50	85	53
Female	17	40	58	50	75	47
Total	43	27	117	73	160	100

Table 1 indicated that out of 160 cases cultured 43 (26.9%) of septicemia positive cases were detected by one or the other method and Out of this 43 positive cases 26 (60%) were males and 17 (40%) were females.

Table.2: Showing distribution of cases as per different diagnostic method

Cases	Conventional		Bactec		Total	
	No	%	No	%	No	%
Positive	14	17.5	29	36.2	43	27
Negative	66	82.5	51	63.8	117	73
Total	80	100	80	100	160	100

Table 2 indicated that out of 80 samples cultured by Bactec method 29 (36.2%) were Bactec positive cases. And out of 80 samples cultured by Conventional method 14 (17.5%) were Conventional positive cases. Out of 29 Bactec positive cases equal number of cases were detected in males 15 (51.7%) and females 14 (49.3%). In males the most effected age group was found to be 51 to 80 whereas in females septicemia was found almost equally in all age groups. Out of 14 conventional positive cases most of them were detected in males 9 (64.3%) in the age group of 41 to 70.

Table.3: Bacteriological profile of septicemia positive cases

Profile	No	%
Methicillin sensitive <i>Staphylococcus aureus</i>	14	32.5
Klebsiella	10	23.5
Escherichia coli	5	11.6
Enterococci	4	9.3
Pseudomonas	4	9.3
Acinetobacter	3	6.9
Others	3	6.9
Total	43	100

In Table 3 all positive cases were taken into account. Gram-positive cocci and Gram-negative bacilli were involved in the infections of 39.5% and 60.5% respectively. Methicillin sensitive *Staphylococcus aureus* 11 (25.5%), Klebsiella 10 (23.5%), Escherichia coli 5 (11.6%) and Enterococci 4 (9.3%) were the most frequently isolated pathogens. Other organisms isolated were Pseudomonas 3(6.9%), Methicillin resistant *Staphylococcus aureus* 2 (4.6%), Acinetobacter 2 (4.6%),

Enterobacter 1 (2.3%), Salmonella 1 (2.3%) and Burkholderia cepacia 1 (2.3%).

Each positive blood culture was critically assessed and categorized as clinically significant or not clinically significant, by correlating with clinical signs and symptoms and various lab parameters like white blood cell count, number of blood samples culture positive out of the total number drawn, results of other site cultures etc. All indeterminate cases were reviewed by an infectious disease physician prior to classification. Only clinically significant positive blood cultures were included in our study.

In a comparative academic study relatively for 12 cases taken, we found 9 cases as Bactec positive and 3 cases as Bactec negative, whereas in Conventional method 4 cases were positive and 8 cases were negative. In addition to this 3 cases were found to be Bactec as well as Conventional negative. 3 cases were both Bactec as well as conventional positive and 1 case was found to be Bactec negative but conventional positive (false positive). 5 cases were Bactec positive and conventional negative.

In a statistical comparison of Bactec and Conventional method, Sensitivity for Bactec method was seen as 90% whereas for conventional method it was 60%. Bactec method has the highest specificity i.e. 100% whereas Conventional method has 75%.

Among 43 septicemia cases, 6 were found to be IV canula (canula tip was sent for culture and sensitivity) positive. Organisms isolated in IV canula culture positive were MSSA 3, MRSA 1, Klebsiella 1, and Enterococci 1.

In our study we also found some of the septicemia cases who had other samples like pus (bedsore) (3), urine (4) and sputum (2) showing the growth of the same bacteria which were found in blood. 9 (22.5%) out of 43 total cases were showing growth in other samples with same organism and same antibiogram.

Table.4: Comparative study between Bactec & Conventional methods (Total cases studied – 12)

Cases	Bactec +ve	Bactec -ve	Total
Conventional +ve	4	1	5
Conventional -Ve	5	2	7
Total	9	3	12

Statistical comparative study and significance of Bactec and conventional method

Statistical Significance	Bactec method	Conventional method
Sensitivity	90% (44.4)	60% (80%)
Specificity	100% (33.3%)	75% (71.43%)

DISCUSSION

Advances in the field of blood culture media and detection technologies have considerably improved the sensitivity of bloodstream infections. Standard bacteriological media for the growth of bacteria and several studies have compared the efficacy of various bacteriological media and different detection systems for the isolation of bacteria in the blood. Parameters such as medium composition and volume, incubation conditions, and method of detection of a positive culture must be evaluated to determine the technology best suited to the needs of a particular institution.

In this study, 26.9% cases were having positive blood culture, which is near to the findings of Gul Durmaz (32%)³ but more than James J. Plorde (15.2%)⁴ and Larry G. Carlson (6.5%)⁵

In current study Gram negative bacilli (60.5%) were the most common organisms found in blood culture positive septicemia cases. Similarly Mitra Barati⁶ and Karen K. Krisher⁷ also reported Gram negative bacilli as the most common organisms found in blood culture positive septicemia cases. But in contrast to our findings, Gul Durmaz et al.,³ and James J. Plorde et al.,⁴ reported Gram positive cocci as the most common organisms found in blood culture positive septicemia cases. Diekma DJ et al.,⁸ Uslan DZ⁹ and Valles J¹⁰ also reported Gram positive cocci as commonest pathogen.

The expedient laboratory diagnosis of bacteraemia is fundamental to quality patient care. However, the selection of a blood culture system that is both cost-effective and reliable for the rapid detection of important blood pathogens often poses a dilemma for many laboratories. In current study total of 43 cases were detected and screened by routine conventional blood culture and Bactec method. Bactec positive cases were 29 (67.4%) whereas conventional positive cases were 14 (32.6%). Among total 43 cases, 12 cases were taken for the comparative study of Bactec and Conventional methods. 9 cases were Bactec positive whereas only 3 cases were found to be conventional negative. This comparison reveals that Bactec is more sensitive than conventional method. Karen K. Krisher⁷ also reported Bactec as more sensitive than conventional method. Whereas in conventional method 4 cases were Conventional positive and 8 cases were conventional negative. In addition to these 2 cases were found to be Bactec as well as conventional negative. 3 cases were both Bactec as well as conventional positive, 1 case was found to be Bactec negative but conventional positive (false positive), 5 cases were Bactec positive and conventional negative. The Bactec performed better and fast in missed episodes of simulated septicemia. The fact that the blind sub culturing procedure of conventional methods has not been applied in the automated systems

minimizes the risks of contamination also the most important advantage of this system is the time it gains for treatment, due to the rapid isolation, especially for slow-growing microorganisms.¹¹

Drawing of blood through a central venous catheter (CVC) for the diagnosis of bacteremia is highly debated^{12, 13}. Due to the possibility of culturing blood contaminated by organisms adhering to CVC lumen, Out of total 43 septicemia positive cases, 6 IV device related infection cases were found. The common isolates found were Gram positive cocci (62.5%) followed by Gram negative bacilli (37.5%). Since the contamination of a blood sample is usually associated with a low inoculum, quantitative blood cultures could be useful in differentiating contamination from bacteremia. Therefore, the utilization of clinical findings with stricter laboratory criteria may contribute to a more accurate interpretation of blood culture results.

Usually in septicemia cases blood is the only sample for growth of the pathogens. But In our study pus, urine and sputum from 9 (20.9%) cases showed the same growth as that of blood of the septicemia cases. 4 (44.4 %) cases were catheter induced septicemia cases in which Methicillin sensitive *Staphylococcus aureus* was commonest isolate.

Amongst the Gram positive organisms maximum resistance was seen with ampicillin (70%) and Erythromycin (64.3%). An increased Ampicillin resistance of 64%, 87%, was also reported by Guha et al., and Karki et al., respectively in their studies.¹⁴

Most of the Staphylococci cases were Methicillin sensitive. Vancomycin, Imipenem, Gatifloxacin, Cephalexin were the drugs to which Gram positive cocci were most sensitive. Gram negative bacilli were most sensitive to Meropenem, Imipenem and cefepazone.

This study has implications for clinical laboratories and in situations where a significant proportion of blood cultures from patients are carried out while the patient is receiving therapy. As antimicrobial agents change, it is important to reevaluate the efficacy of these systems.

CONCLUSION

In conclusion, the results of this study support the use of Bactec method in conjunction with conventional for culture of blood. We were able to recover significantly more microorganisms from the Bactec than from the conventional method, indicating that the benefit of the Bactec goes beyond what might have been expected merely from the culture of blood by conventional method. The development and quick

access of laboratories have provided clinical microbiologists convenient and sensitive methods for blood cultures and have renewed interest in the quantitation of microorganisms in patient blood. Recent attention has been focused on the diagnostic and prognostic significance of determining the magnitude of bacteremia in various clinical circumstances and patient populations. The Bactec performed better than the conventional in overall detection, time to detection, number of false-negatives, and missed episodes of simulated septicemia.

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