

## Bacteriological Profile and Antibigram of Bloodstream Infection in a Tertiary Care Hospital, India

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### ABSTRACT

**Purpose:** Blood stream infections (BSIs) are an important cause of morbidity and mortality worldwide. Continuous or intermittent presence of microorganisms in the circulating blood is a danger to every organ in the body. BSIs range from self-limiting infections to life threatening sepsis requiring rapid and aggressive antimicrobial treatment.[1] Culture of blood is a vital tool to diagnose such infections. Antibiotic susceptibility patterns help in rationalizing therapy.

**Objective:** The objective of this study was to determine the bacteriological profile and their antibiotic sensitivity patterns of isolates from blood stream infections.

**Materials and Methods:** This is a retrospective study conducted from February 2018 to January 2019 at a tertiary care hospital, RIMS, India. Blood samples were aseptically collected and incubated in BD Bactec system, a fully automated blood culture system for detection of aerobic growth and incubated for 7 days at 37°C. Identification of microbial growth was done by standard methods (biochemical tests) and antibiotic sensitivity test was carried out by Kirby-Baur disc diffusion method as per Clinical Laboratory Standards Institute guidelines (CLSI guidelines).

**Results:** A total of 82 (28%) pathogens were isolated from 289 bacteremia suspect patient blood specimens. Gram - positive cocci (65.85%) were predominant organisms recovered followed by Gram-negative bacilli (34.15%). Staphylococcus

aureus, Klebsiella spp and CoNS, were the primary pathogens isolated. Staphylococcus aureus (48%) was the predominant among all. Glycopeptides, aminoglycosides, and carbapenems, were the most effective drugs for treating bacteremia.

**Conclusions:** Early diagnosis and appropriate antimicrobial treatment is the basis for the successful treatment of sepsis. The understanding of local bacteriological profile and antimicrobial susceptibility patterns may help the clinician in rationalizing the empirical treatment strategies.

**Keywords:** Bloodstream Infection, Antimicrobial Sensitivity, Blood Culture, Bacterial Profile, Empirical Therapy.

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### INTRODUCTION

Continuous or intermittent presence of microorganisms in the circulating blood is a menace to every organ in the body. Roughly 200,000 cases of bacteraemia and fungemia occur yearly with mortality rates ranging from 20-50%.<sup>1</sup> In current era, one of the major causes of morbidity and mortality, ranging from self-limiting to life threatening sepsis are Bloodstream infection (BSI) that requires rapid antimicrobial treatment.<sup>2</sup> But the great concern is emergence of resistance among the bacterial pathogens causing these infections. Antimicrobial resistance among bacterias are emerging grave public health concern in both developed and developing countries.<sup>3,4</sup> Although due to lack of surveillance systems, inadequate resources, poor devotion to infection control strategies, use of antibiotics without doctor prescription and limited antimicrobial formularies, monitoring and controlling antimicrobial

resistance is difficult particularly in developing countries like India.<sup>5</sup> The multidrug resistant bacterial infection leads to outcome in terms of prolonged hospital stay, increased risk of death and necessitate treatment with more costly antibiotics. Both Gram negative and Gram positive bacteria may be accountable for these infections.<sup>6,7</sup> In hospital setting, empirical start of antimicrobial therapy is a norm before the results of blood culture are available. But due to high mortality and morbidity associated with septicaemia, precise choice of empiric therapy is of importance. The diagnosis of these infections can easily be made with blood culture. Since blood is a sterile fluid, the positive predictive value of a blood culture is high. Early identification of the causative pathogen and start of appropriate treatment can significantly reduce the morbidity, hospital stay and mortality among patients

with BSIs. A wide spectrum of bacteria has been described according to geographical alteration that leads to blood stream infections.<sup>6,8</sup>

This study was carried out in our medical college and hospital in North India to know the profile of bacterial pathogens causing BSIs in the patients admitted to the critical care units and also to know the trends of antibiotic sensitivity pattern among these agents.

So that early identification of the causative pathogen/diagnosis and initiation of appropriate treatment of these infections can make the difference between life and death.

## MATERIALS AND METHODS

This retrospective study was conducted in the Department of Microbiology, RIMS, Ranchi. A total of 289 patients of all age group (up to 70 years) and both genders who were admitted to medical general wards and ICUs were included during the period of February 2018 to January 2019.

### SPECIMEN COLLECTION

**A. Timing:** Blood samples were collected from clinically suspected cases of BSIs admitted in various inpatient departments of the hospital for routine blood culture and sensitivity. Blood was collected prior to starting empirical antimicrobial therapy. If antimicrobial agent was already started, then the best time of collection is just before the next dose of antimicrobial agent.

**B. Volume:** The volume of the draw is critical as the concentration of organisms in most of the bacteremias is low, especially if the patient is on antimicrobial therapy. However, in infants and children, the number of bacteria in the blood is higher so, less blood is required. Higher the volume of blood, greater is the yield of isolation.

BACTEC blood culture bottle and recommended volume- At least 8–10 ml per bottle for adult and 1-3 ml per bottle for pediatric. In this study only aerobic bottles were used not the anaerobic.

**C. Collection Method:** All blood cultures were collected by venipuncture. Blood cultures must always be collected first, prior to any other blood order. Bottles must remain upright or on a slight slant only to ensure no fluid is in the neck of bottle during the venipuncture process.

**D. Preparation of Site and Antiseptics:** Blood was collected under strict aseptic conditions using sterile disposable syringe.

Skin was treated with 70% isopropyl alcohol and then a second antiseptic solution tincture iodine or chlorhexidine was applied. The rubber cap of blood culture bottle was cleaned with alcohol and was left for drying. Tourniquet was re tied without touching the prepped area, needle was used to withdraw blood from the vein. After bleeding stops, if any iodine remains on the skin, venipuncture site was recleaned with alcohol to remove.

**E. Labelling:** All bottles were properly labelled.

**TRANSPORT:** Blood cultures were transported at room temperature. Blood cultures were not refrigerated if there was a delay in transporting to laboratory.

**PROCESSING:** BSI was confirmed by using microbiological blood culture. The blood specimens were collected aseptically and inoculated into blood culture bottle system and loaded to BD Bactec automated blood culture system and incubated for 7 days at 37°C. This continuous detection system eliminates the need for daily inspection and the requirement for terminal subculture. Based on the patient's clinical condition, blood samples were obtained from different sites by venepuncture (Left brachial, right brachial vein etc.).<sup>7,8</sup>

For those bottles which flagged positive, a Gram stain was performed and an inoculum sub-cultured. If no organisms were seen on Gram stain, after subculture the BACTEC™ bottle was returned to the machine for further monitoring. If there was no growth on subculture, this was recorded. BACTEC™ bottles were monitored until the end of the 7 day incubation. The obtained positive blood culture bottles were gram stained and immediately reported to respective wards (Critical alerts). The finding was further confirmed by growth on Blood agar and MacConkey agar. Both plates were read after 5 days incubation and all bacteria isolated were identified on the basis of growth characteristics, and biochemical profiles. And antibiotic sensitivity test was carried out by Kirby-Baur disc diffusion method as per Clinical Laboratory Standards Institute guidelines (CLSI guidelines) using antibiotic discs (HiMedia Mumbai).<sup>8</sup>

### Statistical Analysis

All the relevant data were analysed by MS office and results were expressed in frequency and percentages. Tables and graphs were used to summarize the results. The chi-square test was applied to know the association between variables. A p-value of < 0.05 was considered as statistically significant.

**Table 1: Overall adult and pediatric blood culture results (n=289)**

Result	Adults (%)	Pediatrics (%)	Total (%)
Growth of microorganisms	7 (17.94)	75 (30)	82 (28.37)
No growth (sterile)	32 (82.05)	175 (70)	207 (71.63)
Total	39	250	289

**Table 2: Studied maximum blood samples Number of Positive, Negative and Percentages**

Sample category/department	Hospitalised patients Positive culture
Pediatrics (medicine + surgery)	75 (91.46%)
Medicine adult	06 (7.31%)
Surgery adult	01 (1.21%)

Table 3: Antibiotic susceptibility pattern of gram negative bacteria

Antibiotics	Pseudomonas spp. n=3	Klebsiella spp. n=16	Acinetobacter spp. n=3	E.coli n=4	Enterobacter cloacae n=2
Ciprofloxacin	1	8	0	2	-
Tobramycin	0	1	2	0	-
Piperacillin-tazobactam	3	4	1	2	-
Cefotaxime	1	1	1	2	-
Imipenem	1	11	1	3	1
Amikacin	3	2	2	2	1
Cotrimoxazole	1	9	0	0	1
Netilmicin	2	1	1	0	--
Ampicillin	1	3	0	0	
Gentamycin	1	1	2	0	-
Levofloxacin	2	10	2	0	1

Table 4: Antibiotic susceptibility pattern of gram positive bacteria.

Antibiotics	S.aureus n=39	CoNS n=12	Enterococcus n=2	Streptococcus n=1
Linezolid	39	12	1	
Vancomycin	39	12	-	
Erythromycin	10	2	-	
Ciprofloxacin	22	4	-	
Piperacillin-tazobactam	11	8	-	
Cefotaxime	7	2	-	
Imipenem		10	-	
Amikacin	-	-	1	
Cotrimoxazole	11	4	-	
Gentamycin	28	8	1	
Levofloxacin	-	8	-	

Table 5: Bacterial species isolated from blood cultures

Bacteria	Total no of isolates (%)
Staphylococcus aureus	39 (47.56)
Klebsiella spp.	16 (19.51)
CoNS (Coagulase Negative Staphylococcus spp.)	12 (14.63)
Escherichia coli	4 (4.87)
Pseudomonas spp.	3 (3.65)
Acinetobacter spp.	3 (3.65)
Enterobacter cloacae	2 (2.44)
Enterococcus spp.	2 (2.44)
Streptococcus spp.	1 (1.22)

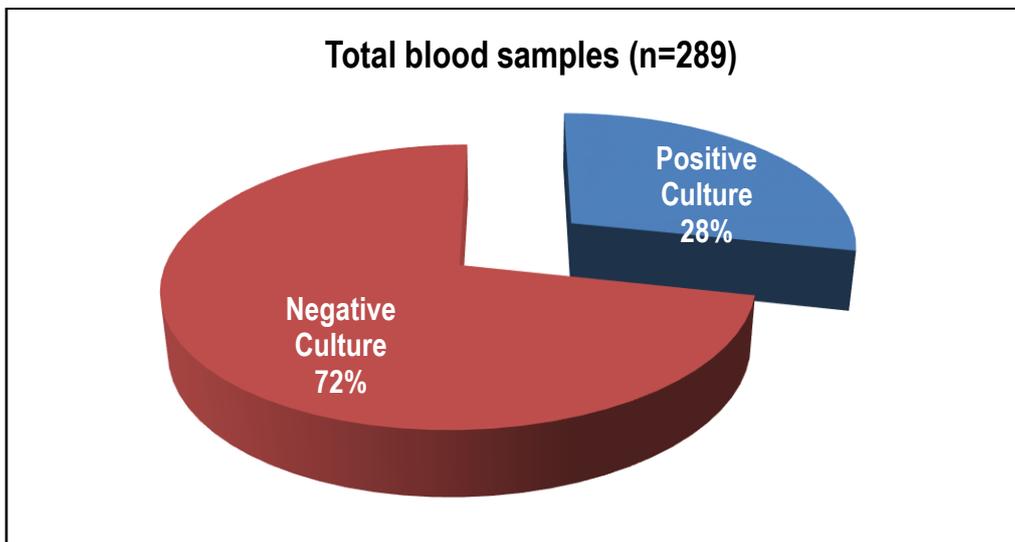


Figure 1: Total positive blood culture

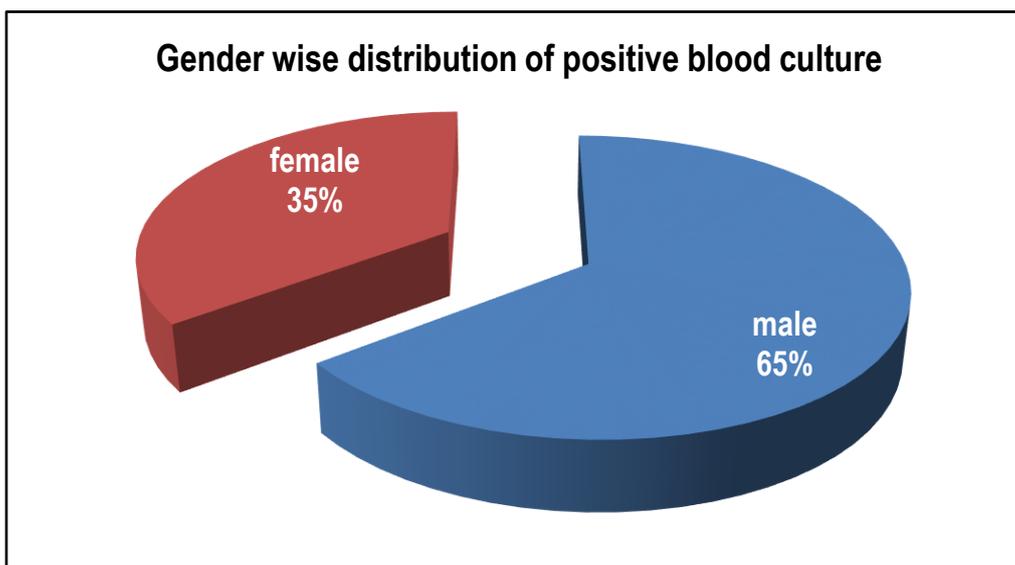


Figure 2: Gender wise distribution of positive blood culture

## RESULTS

Total 289 samples were collected from suspected blood stream infection from different departments. Out of 289 samples, 82 (28%) were shown positive growth in culture, while 207 (72%) samples had no growth in culture (fig 1). Sex wise distribution of positive samples revealed that 53 (65%) patients were male, while 29 (35%) were female (fig 2). Age wise distribution of cases shows that children were 250 (86.50%) whereas adult were 39 (13.49%). (Table 1) Among 82 positive cases, Pediatrics (medicine + surgery) were 75 (91.46%) whereas medicine adult and surgery adult were 6 (7.31%) and 1 (1.21%) respectively (table 2). Study showing nosocomial blood stream infection by Gram positive organisms were 54 (65.85%) while by Gram negative organisms were 28 (34.15%) (table 3 & 4). Staphylococcus aureus was the predominant organism 38 (46.34%) followed by CONS 12 (14.63%). (Table 4) The antimicrobial resistance pattern of gram negative and gram positive organisms is shown in the table 3 & 4 respectively. Different Bacterial species isolated from blood cultures are shown in table 5.

## DISCUSSION

BSI is one of the major reasons of morbidity and mortality worldwide. Antimicrobial therapy is the mainstay of management of BSI along with treatment of severe sepsis and septic shock.<sup>9</sup> The incidence of BSIs along with resistance against commonly used antimicrobials are on increasing trend during last few years.<sup>10</sup> We found 28% prevalence rate of BSIs in our hospital. The prevalence rate of BSIs found from studies conducted in various parts of India demonstrate a wide variation like Dash M et al [17.2%]<sup>11</sup>, Parihar et al [28.29%]<sup>12</sup>, Ramana et al [42%]<sup>13</sup>, Mehdinejad M et al [33.9%]<sup>14</sup>, Sharma M et al [52.10%]<sup>15</sup>, Alam et al. [20.9%]<sup>16</sup>, Arora et al. [20.02%]<sup>17</sup>, Sharma et al [33.9%]<sup>18</sup>, Roy et al [16.4%]<sup>19</sup> and Gohel K et al [9.2%].<sup>20</sup> These variation in prevalence rate can be explained by practice of prescribing antibiotics to the patients by local health practitioners before the patients reach the tertiary care hospital as well as by volume or the number of blood culture samples obtained for study.<sup>21</sup> In addition the patients those were admitted to emergency at times usually received antimicrobials prior to collection of blood for culture.

In the present study, age wise distribution of cases shows that children were 250 (86.50%), whereas adult were 39 (13.49%). Since pediatric patients are more susceptible to infection owing to immature innate and adaptive immune functions large number of pediatric patients tested for BSIs can be explained in our study.<sup>22</sup> In this study 65% patients were male, while 29 (35%) were female. In India, men are involved in more physical activities for living which prejudice them for BSI as well as they are more fortunate to visit doctor for treatment may explain that the majority of blood samples belong to men.

In our study, bacteremia due to Gram positive pathogens was 65.85% whereas due to Gram negative pathogens were 34.15%. Gram positive organisms preponderance are corroborated by other studies as well.<sup>23-25</sup> Although in few studies gram negative predominance are also found.<sup>26-28</sup>

In present study, among gram positive organisms, 39% was *S.aureus* whereas 12% was CoNS. Prominence of *S. aureus* as a blood stream pathogen has been recognized by other study also.<sup>24</sup> Although CoNS is normal skin flora, the high prevalence of CoNS as bloodstream pathogens can be explained by improper methods of blood collection and the presence of long standing intravascular catheters in few sittings.

In our study, among gram negative organisms, Enterobacteriaceae as a group responsible for maximum positive cases with a preponderance of *Klebsiella* (19.51%) followed by *E.coli* (4.87%). The similar finding was found in other study also.<sup>23</sup> An assessment of the in vitro sensitivity pattern of gram positive culture was attempted.

Among *Staphylococcus* spp, apart from vancomycin and linezolid, that were 100% sensitive, increased susceptibility was seen with piperacillin-tazobactam, gentamycin, ciprofloxacin, erythromycin and cotrimoxazole. CoNS infections were sensitive to levofloxacin, gentamicin, linezolid, vancomycin, imipenem, piperacillin-tazobactam and levofloxacin. Enterococcus was found to be sensitive with linezolid, amikacin, gentamycin. Among gram negative organisms, *Klebsiella* was found to be sensitive with imipenem, cotrimoxazole, levofloxacin and ciprofloxacin. Few possible limitations of this study are less sample size due to short study period, only bacterial agents were included in the study and aerobic blood cultures were done.

## CONCLUSION

It is vital for physicians to renew themselves with up to date data regarding the causative agents of frequent bacterial infections in a particular geographical area in addition to its antimicrobial susceptibility and resistance pattern. The information of etiological prototype and their antibiogram pattern can be useful while framing the antibiotic guidelines for any medical institution. Strict infection control methods as well as well-judged antibiotic guideline for antibiotic therapy should be executed in the health care institution as control measures in opposition to BSIs.

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