Original Research Article

Bacteriological profile and antimicrobial susceptibilty of blood culture isolates from the patients of tertiary care hospital, Udaipur

Pratibha Mittal[1*], Anshu Sharma[2]

[1]Sr. Demonstrator, [2]Professor, Department of Microbiology, RNT Medical College, Udaipur-313001, Rajasthan, INDIA.
Email: pratibhamittal1@gmail.com

Abstract

Blood stream infection are major public health problem which leads to high morbidity and mortality of patients. So for on time diagnosis and appropriate treatment, total 214 blood samples received in microbiology laboratory from tertiary care hospital of R.N.T medical college, Udaipur over the duration of one year. Blood samples collected in BHI broth and the growth obtained was identified by colony morphology, grams staining of isolated colonies and by conventional biochemical tests as per the standard protocol followed in our laboratory and then antibiotic susceptibility test done of isolated colony. Positive blood cultures were obtained in 54.6% of total cases of which Gram-positive bacteria accounted for 61% of cases with *staph. aureus* predominance, gram negative bacteria accounted for 28% with the predominance of *Escherichia coli*, *klebseilla* sp. and *pseudomonas* sp. and 11.1 % were fungal isolates. The most sensitive drugs for Gram-positive isolates were Amikacin, Ciprofloxacin, Doxycycline, Levofloxacin, Tetracycline and for Gram-negative were Levofloxacin, Tetracycline, Doxycycline and Ciprofloxacin.

Key Words: Bacteremia,, Coagulase negative staphylococci(CONS), *Escherichia coli, klebseilla* species, Resistance, *Staphylococcus aureus.*

[*]**Address for Correspondence:**
Dr. Pratibha Mittal, Senior demonstrator, Department of Microbiology, R.N.T Medical College, Udaipur-313001, Rajasthan, INDIA.
Email: pratibhamittal1@gmail.com
Received Date: 14/02/2017 Revised Date: 14/03/2017 Accepted Date: 04/04/2017

Access this article online

Quick Response Code:

Website: www.statperson.com

DOI: 15 May 2017

INTRODUCTION

Bacterial bloodstream infections are a major public health problem, which leads to high morbidity and mortality of patients.[1] Bacteremia refers to the presence of bacteria in blood whereas sepsis or septicemia indicates organisms are present in the blood, producing an infection and reproducing within the blood stream.[2] The mortality rate ranges from 20% to 50% in cases of bacteremia.[3,4] There has been an increase in the incidence of bacteremia caused by the members of *Enterobacteriaceae* and other Gram-negative bacilli in recent years. The most common routes of entry for bacteremia are genitourinary tract (25%), respiratory tract (20%), abscesses (10%), surgical wound infections (5%), biliary tract (5%), miscellaneous sites (10%) and uncertain sites (10%). In several studies, fewer than 30 CFU/ml of blood were commonly found in adults patients with clinically significant bacteremia. Blood specimens from septic children may yield fewer than 5CFU/ml of the organism, nevertheless smaller volumes may yield higher level of bacteremia more than 1000CFU/ml detected in some infants.[2] Blood stream infections require urgent and invasive management with antibiotics.[5] Increasing antimicrobial resistance is a worldwide concern. The prevalence of resistance of blood borne isolates is increasing and it also varies in accordance with geographical and regional location.[6] Now days, sensitive strains are being replaced by multi-drug resistant(MDR) strains *Klebsiella*, *Pseudomonas*, *Acinetobacter* and *Citrobacter* species[7] which may prolong the hospital stay, increase the risk of death, and require treatment with more expensive antibiotics. So, on time diagnosis and appropriate medication will be the

How to cite this article: Pratibha Mittal, Anshu Sharma. Bacteriological profile and antimicrobial susceptibilty of blood culture isolates from the patients of tertiary care hospital, Udaipur. *International Journal of Recent Trends in Science and Technology* May 2017; 23(1): 24-28 http://www.statperson.com (accessed 22 May 2017).

best way to save the lives of affected ones.[1] The present study was undertaken to know the bacteriological etiology of blood stream infections and the antibiotic susceptibility pattern of the isolated strains, as it would be a useful guide for clinicians initiating the empiric antibiotic therapy.

MATERIALS AND METHODS

Sample collection and inoculation

A total 214 Blood samples were received with strict aseptic venipuncture from various clinical departments from patients of clinically diagnosed case of sepsis received in Microbiology laboratory of tertiary care hospital, R.N.T medical college over a duration of 12months from 01.01.2016 to 31.12.2016. Specimens were collected in brain heart infusion broth. Blood samples were collected from the patients before the use of systemic antimicrobials if possible. This needs to be taken into consideration because culture results are interpreted. Reference strains *Escherichia coli* (ATCC 25922) and *Staphylococcus aureus* (ATCC 25923) were used as a control reference strains for identifications and drug susceptibility testing. Between 10ml and 30ml of blood was drawn from each venipuncture of adult patient by nursing personnel or physicians and 1 to 5 ml of blood from infants by using strict aseptic precautions, and inoculated immediately in a ratio of about 1:5 or 1:10 in Brain Heart Infusion (BHI) broth to dilute any inherent antibiotics or other antimicrobial substances. In infant and small children less than 1ml may not be adequate to detect pathogen but smaller volumes should still be cultured, because high levels of bacteremia more than 1000CFU/ml detected in some infants.[2] The culture bottles were incubated at 37°C and periodic subcultures were done on 5% sheep Blood agar and Mac Conkey's agar after overnight incubation on day 2, 4 and day 7. The growth obtained was identified by colony morphology, grams staining of isolated colonies and by conventional biochemical tests as per the standard protocol followed in our laboratory. The final report as negative will dispatched on 10^{th} day by examining the broth daily and doing a final subculture.

Antibiotic susceptibility testing

The antibiotic susceptibility test was done for isolated organisms by modified Kirby-Bauer disc-diffusion method and zone of inhibition were measured and interpreted according to CLSI standard guidelines.[8]

OBSERVATIONS AND RESULTS

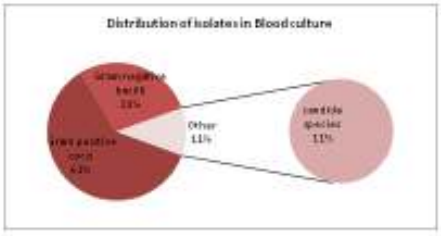
From total 214 Blood culture samples screened during the one year study period, 117 (54.6%) were positive for growth in which Bacterial growth was found 104 (48.5%) and fungal growth were 13 (6.07%) and 97 (45.3%) were sterile [Table 1]. Polymicrobial growths were seen in 3 blood samples out of 104 (2.88%) blood samples with bacterial etiology whereas the remaining 101 (97.1%) blood samples yielded mono-bacterial growth. Three patients whose culture had poly-microbial growth had *Klebsiella* sp. with *Escherichia coli* and two were *Escherichia coli* with *pseudomonas* species. Out of the total positive blood cultures 117(54.6%), the microorganisms recovered were gram positive cocci 71(60.6%), gram negative bacilli 33(28.2%) and 13(11.1%) were *Candida* species respectively [Fig.1]. Among gram positive isolates, Coagulase positive staphylococci (COPS) were predominant organisms as shown in (Table no 3). Coagulase negative staphylococci i.e. 31 (43.6%) approx same as COPS i.e. (45.07%) and least were *Enterococcus* species (11.26%) [Fig.2]. The most common predominant isolated gram negative bacilli were *Escherichia coli, Klebseilla* sp. and *Pseudomonas* sp., 8 (24.24%) each. *Acinetobacter* sp. 6 (18.18%) and *Citrobacter* sp. 2 (6.06%) and least common was *Enterobacter* sp., one case only (3.03%) [Fig.3].

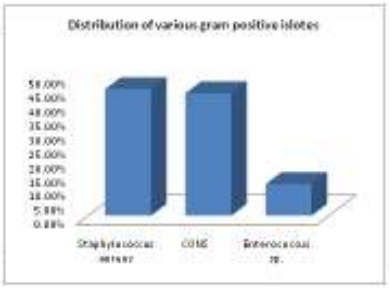
Antibiotic sensitivity pattern

Among the gram positive isolates, the predominant isolate was *Staphylococcus aureus* showed high resistance towards Amoxicillin. All *Staphylococcus aureus* were highly sensitive to Amikacin (59.3%) and then CONS which exhibited highly resistant to Erythromycin, Amoxicillin and Amoxyclav and 80.64% highly sensitive to Amikacin [Table 2]. It shows the various sensitivity pattern of gram positive isolates towards various antibiotics. Among all the gram positive isolates, Amoxicillin showed high degree of resistance for all gram positive isolates as shown in table no 2 respectively. Other gram positive cocci such as *Enterococcus* sp. (11.26%) showed highly sensitivity to Ciprofloxacin, Doxycyclin, Tetracycline and Levofloxacin. In gram negative bacilli the most common resistance were seen for third generation Cephalosphorins and Cephalexin in all isolated bacteria of *Enterobacteriacae* and highly sensitive to levofloxacin in *Escherichia coli* and for *klebseilla* sp. were Tetracycline and Doxycycline [Table 3].

Table 1: Total no of Isolates in Blood culture

Total samples	Positive growth (%)	Sterile (%)
214	117(54.6)	97(45.3)





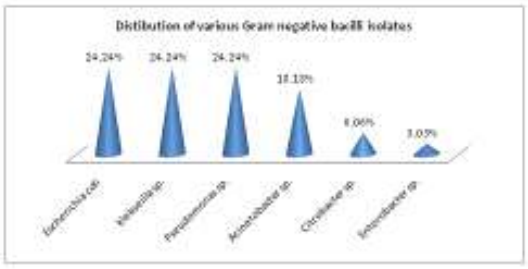


Figure 1 **Figure 2** **Figure 3**

Legend

Figure 1: Distribution and percentage of various isolates among positive Blood culture GPC: Gram positive cocci, GNB: Gram negative bacilli and Candida species.

Figure 2: Distribution and Percentage of various Gram positive Bacterial isolates in blood culture.

Figure 3: Distribution and percentage of various Gram negative Bacterial Isolates in blood culture.

Table 1: Drug sensitivity pattern of various Gram positive isolates in blood culture

Antibiotics	CONS*(n=31)*	COPS *(n=32)*	Enterococci *(n=8)*
Amikacin	80.64%	59.3%	-
Gentamicin	74.1%	50%	-
Cephalexin	35%	25%	-
Cefotaxime	35%	12.5%	-
Erythromycin	12.9%	12.5%	33.3%
Ceftazidime	22.5%	9.3%	-
Ceftriaxone	35%	25%	-
Ampicillin	29%	12.5%	37.5%
Amoxicillin	12.9%	3.1%	37.5%
Amoxyclav	12.9%	12.5%	25%
Ciprofloxacin	45%	37.5%	62.5%
Cotrimoxazole	35%	40.6%	-
Doxycycline	61%	46.8%	62.5%
Nalidixic acid	38.7%	25%	25%
Tetracycline	48%	53%	62.5%
Ceftaclav	25.8%	12.5%	-
Methicillin	19%	6.25%	33.3%
Oxacillin	22.5%	12.5%	33.3%
Vancomycin	19%	53.1%	37.5%
Levofloxacin	51.6%	43.7%	62.5%

Note: -=intrinsic resistance

Table 3: Drug sensitivity pattern of major Gram negative isolates in blood culture

Antibiotic	Escherichia coli (*n=8)*	Klebseilla sp. *(n=8)*	Pseudomonas sp.(*n=8)*	Acinetobacter sp.*(n=6)*	Citrobacter sp.*(n=2)*	Enterobacter sp.*(n=1)*
Amikacin	37.5	12.5	37.5	50	0	0
Gentamicin	12.5	0	25	66.6	0	0
Cephalexin	0	0	0	16.6%	0	0
Cefotaxime	0	0	0	0	0	0
Ceftriaxone	0	0	0	0	0	0
Tetracycline	12.5	50	-	33.3	50	100
Ceftaclav	12.5	12.5	50	16.6	0	0
Ampicillin	0	-	0	0	0	0
Augmentin	0	0	0	0	0	0
Ciprofloxacin	25	25	62.5	50	50	100
Carbenicillin	37.5	12.5	25	33.3	0	0
Cotrimoxazole	12.5	37.5	-	16.6	0	0
Doxycycline	37.5	50	0	50	0	100
Levofloxacin	62.5	12.5	62.5	50	0	0

DISCUSSION

Enhanced detection of blood stream infection and appropriate treatment can make the difference between life and death. It will reduce mortality from septicaemia, reduce turnaround time and improve patient management. So study of bacteriological profile with antibiotic susceptibility pattern plays an important role in effective management of bacteremia cases. In our study, shows 117 (54.6%) were positive out of 214 total samples screened in microbiology laboratory. Similar results were observed in study conducted by Tiwari *et al*[10]. Oza *et al* reported 18.62% low blood culture positivity rate in Gujarat[11]. The variation in blood culture positivity is related to different factors such as the number and amount of blood cultures taken for screen as reported by Lee *et al*[12]. Our study showed that the rate of isolation of Gram positive isolates were higher (60.6%) than Gram negative bacteria (28.2%) quite similar to others studies[13,14] and has been observed by Sharma *et al*[15] While other studies like Mehta *et al*[16] observed that gram negative organisms higher than gram positive organisms. Among gram positive isolates, *Staphylococcus aureus* were the predominant isolates similar to findings reported in earlier studies Roy *et al*[17] while some other studies observed that the most common isolated organisms in blood among gram positive isolates were CONS[18]. The prevalence of CONS was approximately same as COPS in present study this may be due to as it is major skin contaminant in blood culture. Among Enterobacteriacae, Escherichia coli and Klebsiella sp. (24.24%) were the predominant Gram-negative isolates which is in accordance with other studies[16,13,14,19]. We also observed similar frequency of Pseudomonas sp. (24.24%) and Acinetobacter sp.(18.18%) as in these studies but we did not observe any isolate of Salmonella which is isolated in the frequency of 10 to 20% in these studies. In our study the antibiotic susceptibility pattern of Staphylococcus aureus and CONS showed maximum susceptibility for Amikacin were 59.3% and 80.64% followed by Gentamicin while *Enterococus* sp. much more sensitive to Ciprofloxacin, Doxycycline Llevofloxacin and Tetracycline (62.5%)each. In contrast, *Staphylococcus aureus* showed high resistant to Amoxicillin while CONS for Erythromycin, Amoxicillin, Amoxyclav. In our study only two were sensitive to Methicillin in staphylococcus aureus out of 32. Oza *et al* revealed that *Staphylococcus aureus* were 80% resistance towards Ampicillin[11]. All *Staphylococcus aureus* were highly sensitive to Vancomycin, Linezolid, Quinolones, Clindamycin and *Enterococcus* sp. was highly resistant to Penicillins, Cephalosporins and Aminoglycosides. Most of the gram negative isolates were resistant to first and second generation Cephalosporins and resistance to third generation Cephalosporins is now emerging which is due to Beta lactamase activity by the bacteria[20,21] and third or fourth generation Cephalosporins and Amoxiclav were most misused antibiotics.

In present study, the most common resistance of gram negative bacilli were seen for third generation Cephalosphorins and Cephalexin in all isolated bacteria of *Enterobacteriacae* and highly sensitive to Levofloxacin in *Escherichia coli* and for *klebseilla* sp. were Tetracycline and Doxycycline. *Sharma et al*[15] *revealed that Escherichia coli* isolates showed least resistance to Aminoglycosides and moderate resistance to beta-lactam beta-lactamase inhibitors. *Klebsiella sp.* showed moderate resistance to Aminoglycosides, and beta-lactam beta-lactamase inhibitor combination.

CONCLUSIONS

Routine and regular surveillance of BSI etiology is important in examine the spectrum of microorganisms associated with septicaemia and antibiotic susceptibility surveillance of blood culture isolates helps in prevention of spread of antibiotic resistance by initiating empiric antibiotic therapy.

REFERENCES

1. Wasihun AG, Wlekidan LN, Gebremariam SA, Dejene TA, Welderufael AL, Dejenie TD, and Muthupandian S:Bacteriological profile and antimicrobial susceptibility patterns of blood culture isolates among febrile patients in Mekelle Hospital, Northern Ethiopia: SpringerPlus. 2015; 4:314.
2. Patricia M. Tille: Bailey and Scott's Diagnostic Microbiology Fourteenth Edition.
3. Diekema DJ, Beekmann SE, Chapin KC, Morel KA, Munson E, Doern GV:Epidemiology and outcome of nosocomial and community-onset bloodstream infection:J Clin Microbiol 2003;41:3655-60.
4. Mehdinejad M, Khosravi AD, Morvaridi A: Study of prevalence and antimicrobial susceptibility pattern of bacteria isolated from blood cultures: J Biologic Sci. 2009; 9:249-53.
5. Shrestha S, Amatya R: Frequency of blood culture isolates and their antibiogram in a teaching hospital:J Nepal Med Assoc.2014;52(193):692-6.
6. Garg A, Anupurba S, and Garg J: Bacteriological profile and antimicrobial resistance of blood culture isolates from a university hospital:J Ind Acad Clin Med.2007; 8(2):139–143.
7. Vanitha RN, Kannan G, Venkata NM, Vishwakanth D, Nagesh VR, Yogitha M et al:A retrospective study on blood stream infections and antibiotic susceptibility patterns in a tertiary care teaching hospital:Int J Pharm Sci.2012;4:543-8.
8. Clinical and Laboratory Standards Institute.Performance Standards for Antimicrobial

Susceptibility Testing. Twenty-Second Informational Supplement.Wayne, PA, USA: Clinical and Laboratory Standards Institute; 2012. M100-S22.
9. Tiwari P, Kaur S: Profile and sensitivity pattern of bacteria isolated from various cultures in a Tertiary Care Hospital in Delhi: Indian J Public Health. 2010; 54:213-5.
10. Oza SS, Mehta JS, kikani KM, Oza GS:Bacteriological profile and antibiogram of blood culture isolates from patients of rural tertiary care hospital: Int J Microbiol Myco.2016;4(3): 1-7.
11. 12.Lee,A,S.Mirrett,L.B.Reller and M.P.Weinstein:Detection of bloodstream infections in adults,How many blood cultures are needed?:J Clin Microbiol.2007;45:3546-3548.
12. China and Gupta V: Bacteriological profile and antimicrobial susceptibility pattern of blood isolates from a tertiary care hospital in North India:Int J Pharm Sci Res.2013;2: 24–35, 2013.
13. Kamga HLF, Njunda AL and Nde PF:Prevalence of septicemia and antibiotic sensitivity pattern of bacterial isolates at the University Teaching Hospital:Afr J Cln Exper Microbiol.2011;12(1):2–8.
14. Sharma R, Sharma R and Gupta S: Bacteriological Analysis of Blood Culture Isolates with their Antibiogram from a Tertiary Care Hospital:Int J Pharm Sci Res.2015;6(11): 4847-5.
15. Mehta M, Dutta P, and Gupta V: Antimicrobial susceptibility pattern of blood isolates from a teaching hospital in North India: J Infect Dis.2005; 58(3):174–176.
16. Roy, Jain A, Kumar M, and Agarwal SK:Bacteriology of neonatal septicemia in a tertiary care hospital of Northern India:Indian J Med Microbiol.2002;20:156–159.
17. Devi V, Sahoo B, Damrolien S, Praveen S, Lungran P, Devi M:A study on the bacterial profile of bloodstream infections in Rims Hospital:J Dent Med Sci.2015;14:18-23.
18. Karlowsky A, Jones ME, Draghi DC, Thornsberry C, Sahm DF, and Volturo GA: Prevalence and antimicrobial susceptibilities of bacteria isolated from blood cultures of hospitalized patients in the United States: Ann Clin Microbiol Antimicrob.2004; 3(7).
19. Radji M, Fathni R, Fauziyah S: Evaluation of surgical antibiotic prophylaxis in tertiary care hospital in jakarta Indonesia:Experi J.2014;18(4):1292-6.
20. Vera VP, Igor F, Goran P, Novak S, Abram M, Ulf Bergman:Antimicrobial use at a university hospital: appropriate or misused? A qualitative study:Int J Clin Pharmacol Ther. 2007;45(3):169-74.

Source of Support: None Declared
Conflict of Interest: None Declared