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BACTERIOLOGICAL PROFILE AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF BLOOD ISOLATES FROM A TERTIARY CARE HOSPITAL IN NORTH INDIA

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Abstract

Background and Aims: As the incidence of bacteraemia is on rise and it contributes to significant morbidity and mortality, so early diagnosis and appropriate treatment of blood stream infections is essential. The present study was aimed to know the prevalence and antimicrobial susceptibility pattern of blood isolates. **Material and methods**: A total of 9010 specimens of blood received for culture during one year were processed by Bac T alert method and identified by standard bacteriological techniques. Antimicrobial sensitivity was done by Kirby Bauer's disc diffusion method. 1258 (13.9%) specimens yielded positive results out of which *Staphylococcus aureus* was the commonest (37.2%), followed by *Escherichia coli* (20.3%), *Salmonella* Typhi and Paratyphi A (14.6%) and *Pseudomonas aeruginosa* (8.2%) . For *S. aureus* isolates vancomycin and tetracycline were most effective while penicillin and ampicillin were least effective. 40.8% isolates of *S .aureus* were methicillin resistant (MRSA). For *E.coli* isolates amikacin was the most effective antibiotic. *Salmonella* species were sensitive to all the antimicrobial agents used. **Conclusion**: Present study revealed that both gram positive and gram negative bacteria were responsible for blood stream infections and most of the strains were multidrug resistant. The most commonly isolated bacteria were *S. aureus*, *E. coli* and *Salmonella* species. This study provides information on antibiotic resistance of blood isolates which may be a useful guide for physicians initiating empiric therapy and will help in formulation of antibiotic therapy.

INTRODUCTION

Enhanced detection of blood stream infections needs to be a national priority. Early diagnosis and appropriate treatment of blood stream infections can make the difference between life and death. It would reduce mortality from septicemia, reduce turnaround time and improve patient management.[1] Blood stream infections (BSI) are an important cause of mortality and morbidity and are among the most common health care associated infections.[2] Illness associated with BSI ranges from self limiting infection to life threatening sepsis that require rapid and aggressive antimicrobial treatment.[3] Moreover, fatalities among patients infected with gram negative bacilli are higher than those who have gram positive cocci. [4-7]

BSI are a major cause of mortality in both developed and developing countries.[8,9] Despite important progress in treatment and prevention of infectious diseases, they are considered as leading cause of death and disability. [10]

The isolation of a bacterium from patient's blood stream often indicates a serious infectious illness with potentially severe morbidity and mortality unless early appropriate therapeutic measures are initiated. The gold standard for diagnosis is a positive blood culture. Culturing blood samples to reveal the presence of micro organisms is a highly specific indicator of BSI and results of antimicrobial susceptibility testing may assist in choice of appropriate antimicrobial therapy. [11] Since early 1950's there has been an increase in the incidence of bacteraemia caused by members of enterobacteriacae and other gram negative bacilli. Multi drug resistance from gram negative bacilli like *Salmonella, Klebsiella, Acinetobacter, Pseudomonas* and *Citrobacter* species are gaining importance and have replaced *Escherichia coli* which was reported to be common in the past. [12] The surveillance of blood stream pathogens in a hospital is important in monitoring the spectrum of microorganisms that invade blood stream and types of organisms associated with particular clinical discipline. Such data are often used to determine empiric antibiotic therapy and to alert clinicians to emerging pathogens that may pose a threat to community. [13] As there is high mortality and morbidity associated with septicaemia a right choice of empiric therapy is of utmost importance. Rapid detection and identification of clinically relevant microorganisms in blood culture is very essential and determination of antimicrobial susceptibility pattern for rapid administration of antimicrobial therapy has been shown to reduce mortality and morbidity associated with blood stream infections. [14] Therefore the present study was undertaken to know the bacteriological profile of blood stream infections and their antibiotic susceptibility pattern.

MATERIAL AND METHODS

A total of 9010 blood specimens received in the department of Microbiology from clinically suspected cases of bacteraemia during one year were included in the study.

All specimens were inoculated aseptically into Fan aerobic bottles, incubated in Bac T alert automated blood culture system (BioMerieux). From all positive blood culture bottles, subcultures were done on blood agar and Mac Conkey's agar plates. After 18-24 hours of incubation isolates were identified by Gram staining, colony characters, biochemical and serological reactions. [15] All negative bottles were kept for 7 days then discarded. Antimicrobial susceptibility testing was done on Mueller Hinton agar by Kirby Bauer's disc diffusion method as per NCCLS guidelines. [16] Control strains of *E.coli* ATCC 25922, *P. aeruginosa* ATCC 27853 and *S. aureus* ATCC 25923 were used. After 18-24 hours of incubation results were read.

RESULTS

During one year study period, out of 9010 blood cultures studied 1258 were found to be positive (13.9%). Majority(40%) of blood culture bottles were detected positive in 13-24 hours, followed by 7-12 hours(27%), 0-6 hours(17%), 25-36 hours(12%) and 37-48 hours(4%) of incubation respectively. None of the bottle was detected positive after 48 hours of incubation.

Gram positive (49.3%) and gram negative (50.6%) isolates were obtained in almost equal frequency. Among gram positive cocci , *Staphylococcus aureus* was the commonest isolate (37.2%) followed by *Staphylococcus epidermidis* (7.1%), *Enterococcus faecalis* (1.8%).The most commonly isolated gram negative bacilli were *Escherichia coli* (20.3%), *Salmonella*

Typhi and Paratyphi A (14.6%), *Pseudomonas aeruginosa* (8.2%) and *Acinetobacter* species (4.3%).(Table-1)

All isolates of *Salmonella* and *Streptococcus pneumoniae* were sensitive to all the antimicrobial agents used. In *S. aureus* isolates vancomycin and tetracycline were most effective while penicillin, ampicillin & gentamicin were least effective. Methicillin resistant *Staphylococcus aureus* (MRSA) were isolated in 40.8% cases. No vancomycin resistant *S.aureus* and *Enterococci* were detected. (Table-2, Figure-1)

A-ampicillin, T-tetracycline, G-gentamicin, Ak- amikacin, Of- Ofloxacin, Cip – ciprofloxacin, P- penicillin, E- erythromycin, Met- methicillin, Van- vancomycin

For *E. coli* isolates cefoperazone +sulbactum (1.4%) and amikacin (8.9%) were the most effective antimicrobial agents. High percentage of resistance was seen to ampicillin (95%), piperacillin (92.5%), cephalexin (86.5%), ofloxacin (83.5%) and gentamicin (80.5%). For *P. aeruginosa* isolates amikacin, piperacillin and ciprofloxacin were most effective. (Table-3, Figure-2)

A-ampicillin, T-tetracycline, G-gentamicin, Ak-amikacin, Cip- ciprofloxacin, Ceph-cephalexin, Cex- cefotaxime, Cez-ceftazidime, Cpz- cefoperazone, Pip-piperacillin, Cpz+Sb-cefoperazone+sulbactum

DISCUSSION

Study of bacteriological profile with antibiotic susceptibility pattern plays an important role in effective management of bacteraemia cases. [17] This study revealed that majority of the blood culture bottles were detected within 13-24 hours of incubation. So the report can be given in 48 hours, more effective than manual blood culture methods.

The blood culture positivity rate in our study was 13.9%which is similar to reported by other workers like 9.9% by Mehta *et al*, [14] 20.02% in a study by Arora and Devi [18] 19.3%byAyobola *et al* [19] and 20.5 % by Garg et al [20] . Low culture positivity of 7.9% [17] 5.17% [21] and 5.6% [22] was reported in studies on bacteriological profile of blood isolates. In contrast to our study high culture positivity of 56%, [23] 47.5%, [24] 33.9%, [25] 32%, [26] 31.4% [27] and 28.3% [28] was reported by various authors.

Gram positive (49.3 %) and gram negative (50.6%) isolates were obtained in almost equal frequency similar to reported by Chaudhary *et al*. [29] It also corresponds with study by Anbumani *et al*, [17] as they reported 54.3% of gram negative pathogens and 45.7% of gram positive pathogens from blood cultures of hospitalized patients. But in contrast to our study, predominance of gram negative bacteria were reported by Mehta et al (80.96%), [14] Mehdinejad *et al* (86.5%) [22] Barati M *et al* (91.8%), [21]Ayobola *et al* (77.1%) [19], Garg *et al* (67.5%) [20] and predominance of gram positive bacteria in various studies. [27, 28]

Among gram positive organisms in our study *S. aureus* was the commonest (37.2%) followed by *S. epidermidis* (7.1%) and *Streptococcus* species (3.9%). This is in accordance to a study by Anbumani et al [17] where 36.4% of *S. aureus*, 3.36 % of *Streptococcus* species and coagulase negative staphylococci in 1.12 % were isolated. In other studies low isolation rate of 14.6% [19], 13.86%, [14], 6.9% [21] and 8.3% [20] of *S. aureus* was reported.

Among gram negative isolates, *E. coli* (20.3%) was commonest followed by *Salmonella* species (14.6%),*Pseudomonas aeruginosa* (8.2%) and *Acinetobacter* species (4.3%) similar to a study by Mehta et al [14] reporting *Pseudomonas aeruginosa* (19.75%), *E.coli* (15.17%), Klebsiella pneumoniae (14.99%), *Salmonella typhi* (12.87%) as common gram negative isolates. *S aureus* and *P. aeruginosa* comprised 45.4% of isolates similar to study by Chaudhary et al [29] where they were 36.8% of the total isolates. In our study *Salmonella* species were isolated in 14.6% cases in accordance to Garg et al (14.2%) [20] and Mehta et al (12.8%).[14] An increasing incidence of *Salmonella* species has also been reported by Sharma et al. [23]Most of the gram negative isolates were multidrug resistant. Other studies have also reported similar multidrug resistance in gram negative isolates. [18, 21, 30] Ampicillin was the least effective drug in all the isolates. An increased ampicillin resistance has also been reported by Guha et al[31] and Khatua et al. [32] Amikacin and cefoperazone +sulbactum were the most effective drugs similar to other studies. [14,20] In our study *Pseudomonas* isolates were sensitive to amikacin, piperacillin, cefoperazone+ sulbactum and cefoperazone+ sulbactum similar to study by Mehta et al where cefoperazone+ sulbactum(82.66%), ciprofloxacin (65.17%) and amikacin (62.50%) showed higher activity. [14] Majority of *S. aureus* strains were resistant to penicillin and ampicillin similar to other reports. [17, 20, 22] All the isolates were sensitive to vancomycin similar to as reported by other workers. [14, 17, 22] In our study MRSA were isolated in 40.8% of isolates while 15% [21], 30% [17] and 75.6% [20] of MRSA were reported by other workers.

CONCLUSION

BacT alert automated system is rapid and sensitive method for diagnosis of bacteraemia Resistant bacteria are emerging world wide as real threat to favorable outcome of common infections. A greater caution is required in selection of antibiotic therapy to avoid development of resistant strains and treatment failure.

The use of automated blood culture equipment, direct sensitivity and identification testing from positive blood culture and urgent clinical liaison between microbiology department and clinical team are essential to make a difference in the in the struggle between life and death. [33]

Early initiation of appropriate antimicrobial treatment is critical in decreasing morbidity and mortality among patients with blood stream infections due to gram negative organisms. [34] The initiation of such therapy is decided upon knowledge of likely pathogens and their usual antimicrobial susceptibility pattern. [35]

To conclude this study provided information on prevalence and antimicrobial resistance pattern among pathogens causing blood stream infections. The most commonly isolated bacteria from blood culture were *S. aureus, E. coli, Salmonella* species. Amikacin and cefoperazone + sulbactum were the most effective antibiotics against gram negative bacilli and vancomycin for gram positive cocci. Most of the Gram negative isolates were multidrug resistant. The rise in antibiotic resistance emphasizes the importance of sound hospital infection control, rational prescribing policies and need for new antimicrobial drugs & vaccines. [22] To reduce the incidence of BSI appropriate use of antibiotics along with standard antimicrobial susceptibility testing is essential. [23]

Table-1: FREQUENCY OF BLOOD CULTURE ISOLATES

ISOLATES	NUMBER (%)
Staphylococcus aureus	469 (37.2%)
Escherichia coli	256 (20.3%)
Salmonella Typhi & Paratyphi A	185 (14.6%)
Pseudomonas aeruginosa	104 (8.2%)
Staphylococcus epidermidis	90 (7.1%)
Acinetobacter species	55 (4.3%)
Enterococcus faecalis	23 (1.8)
Klebsiella pneumoniae	21 (1.6%)
Non haemolytic streptococci	20 (1.5%)
α haemolytic streptococci	16 (1.2%)
Enterobacter species	16 (1.2%)
Streptococcus pneumoniae	*3 (0.17%)*

Table -2: Antimicrobial resistance pattern of Gram positive isolates (%age)

	A	T	G	Ak	Of	Cip	P	E	Met	Van
S. aureus	46.7	7.3	38.53	11.9	28.4	17	59.6	31.1	40.8	0
S. epidermidis	9.5	0	9.52	0	4.7	0	19	9.5	0	0
αHS	40	30	50	30	20	10	30	20	-	-
NHS	20	0	80	40	20	50	40	60	-	0
E. faecalis	25	0	62.5	50	25	38	50	75	-	0

Table-3: Antimicrobial resistance pattern of Gram negative isolates (%age)

Isolates	A	T	G	Ak	Cip	Ceph	Cex	Cez	Cpz	Pip	Cpz+Sb
E. coli	95.5	43.2	80.6	8.9	74.6	86.5	67.1	46.2	80.6	93	**1.4**
P. aeruginosa	38.4	38.4	30.8	0	7.6	53.8	30.7	38.4	23.1	7.7	**15.3**
Acinetobacter spp	72.7	9	81.8	27.2	45.4	81.8	81.8	72.7	63.6	73	**9**
K. *pneumoniae*	100	75	100	50	50	100	100	100	100	100	**0**
Enterobacter spp	**80**	**40**	**80**	**40**	**0**	**80**	**60**	**60**	**80**	**60**	**0**

Figure-1 Antimicrobial resistance of Gram +ve isolates

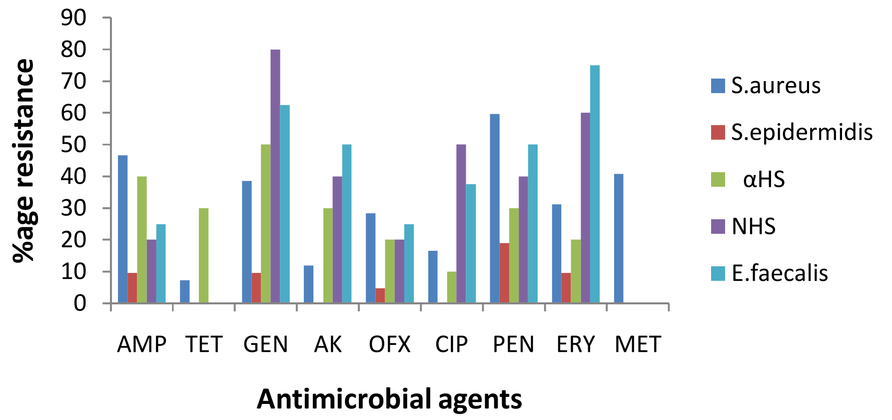

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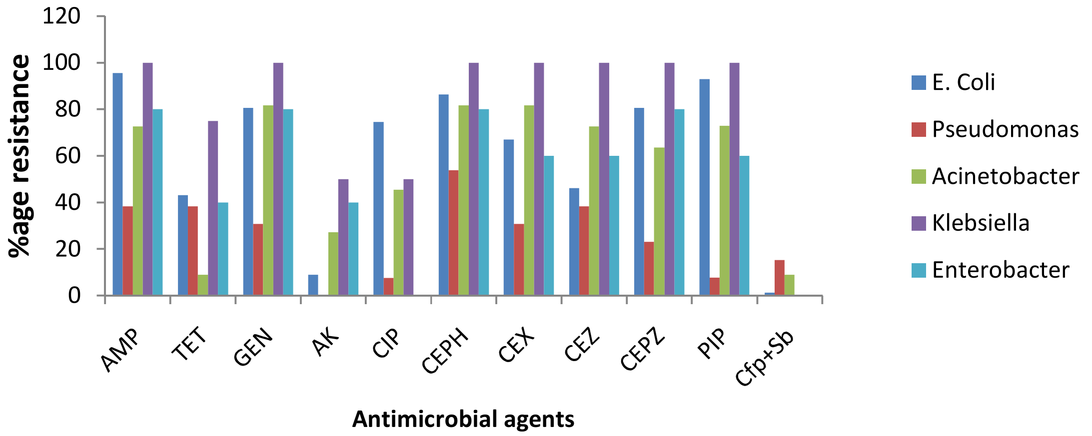
Figure-2 Antimicrobial resistance of Gram -ve isolates



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