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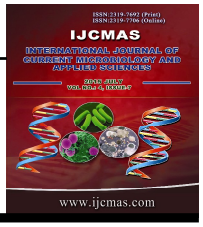

Original Research Article

Bacterial Isolates and antibiotic Susceptibility Pattern in Blood Stream Infections Suspected Patients Attending a Teaching Hospital in Telangana, India

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A B S T R A C T

Bacterial blood stream infection constitutes a significant public health problem and an important cause of morbidity and mortality in hospitalized patients. The aim of this study was to observe the bacterial profile prevalent in our area and the antimicrobial sensitivity pattern. Blood was collected for culture from suspected patients from various wards and ICUs and inoculated Brain Heart Infusion Broth. Isolates were identified and Sensitivity tests were put up on Mueller Hinton Agar by Kirby Bauer's Technique. Out of the total 629 blood samples, 117 (18.6%) were positive for growth. 58.1% of them were Gram Negative bacilli and 41.9% were Gram Positive Cocci among which *Klebsiella* and *Staphylococcus aureus* were predominant respectively. High sensitivity was observed to Carbapenems by Gram Negative Bacilli and to Vancomycin and Linezolid by Gram Positive Cocci. Other drugs showed variable resistant pattern with common drugs like Ampicillin, Penicillin, Amoxycillin, Cephalosporins being highly resistant. Due to increasing resistance pattern of antibiotics regular surveillance of antimicrobial resistance must be done, so that proper timely treatment can be given to the patients.

Keywords

Septicemia, Antibiotic sensitivity tests, Bacterial profile, Gram positive cocci, Gram negative bacilli

Introduction

Septicemia, is a potentially life-threatening infection in which large amounts of bacteria are present in the blood. It is commonly referred to as blood poisoning or Blood Stream Infection. Bloodstream infections have known to cause a significantly high patient morbidity and mortality especially in the ICUs, many a times result in the loss of life (Renu Bharadwaj et al., 2014).

The detection of microorganisms in the patients' blood has a great diagnostic and prognostic significance. The early positive results provide valuable diagnostic information, based on which the appropriate antimicrobial therapy can be initiated. Blood culture is the most significant specimen type which is used for the diagnosis of Blood Stream Infections (BSIs) (Santwana Pandey et al., 2013).

BSIs are one of the main causes of death in hospitalized patients, with mortality rates between 30-70% (Vincent JL et al.,1995).

Blood cultures also provide essential information for the evaluation of a variety of diseases like endocarditis, pneumonia, and pyrexia of unknown origin and particularly, in patients with suspected sepsis. The microorganisms which are present in the circulating blood, whether continuously or intermittently, are a threat to the host (Yagupsky P et al., 1990).

BSI has also known to be one of the most significant causes of mortality among the Hospital Acquired Infections. Advances in medical science have resulted in increased interventions in critically ill patients creating foci from where bacteria can gain access to the blood stream resulting in an increase nosocomial BSI. They represent about 15% of all nosocomial infections and affect approximately 1% of all hospitalized patients [Exline MC et ., 2013]. Community acquired BSIs can also occur. A BSI is primary when the central line is the only probable source of infection and secondary when there is an underlying cause for the BSI (genitourinary/respiratory infection or any other obvious source of infection in the body(Renu Bharadwaj et al., 2014).

Gram negative bacilli such as Enterobacteriaceae and Non fermenters like Pseudomonas and Acinetobacter are regarded as the leading causes of BSI. Among them, Antibiotic Resistant Strains are emerging with great speed, causing a deep concern to the medical fraternity, and present therapeutic challenges (Kang C-Iet al., 2005; Itokazu G S et al., 1996; National Nosocomial Infections Surveillance System. 1999). Such infections result in longer hospital stay, higher costs and death as compared to Antibiotic susceptible bacteria(Kollef MH. 2000). Appropriate antimicrobial therapy has been shown to reduce mortality among patients with gram-negative bacteremia and, when initiated early, in critically ill patients with bacteremia (Hanon, FX.,2002; Leibovici L et al.**,**1998; Leone M et al., 2003). Since the antibiotic resistance pattern can vary with the geographical region, this study was designed to identify the various aerobic bacteria infecting the blood and find out its antibiogram so that an appropriate treatment can be given to these patients.

Materials and Methods

A total of 629 blood samples were collected from the various wards (Medical, Surgical, Paediatric) and ICUs of Malla Reddy Hospital including Medical ICU, Surgical ICU and Paediatric ICU and neonatal ICU from the period between Feb 2013 and Feb 2015. 1 ml from neonates, 3 ml from children and 5 ml blood was collected from adults by aseptic measures and were inoculated onto Brain Heart Infusion Biphasic medium (Courtesy HiMedia). All these blood culture bottles were kept in tilted position for 10 minutes after which they were kept upright and incubated at 37°C overnight.

The next day after checking the turbidity in the liquid part of the medium, a loopful of the liquid media was inoculated into MacConkey's Agar and Blood Agar. The bottles were then again tilted for 10 .minutes, turned upright and reincubated along with the plates at 37^0C overnight. If growth was observed, antibiotic sensitivity was put up otherwise, the tilting and incubating procedure was repeated for 5 days.

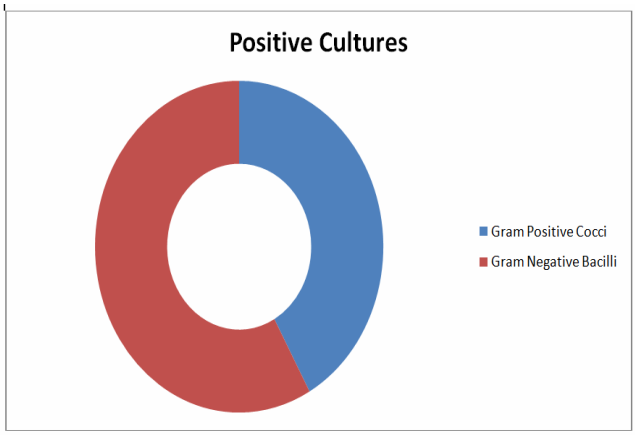
On the 6^{th} day, a loopful of the BHI broth was inoculated on to MacConkey and Blood Agar. If no growth was observed, it was considered negative for septicemia. If growth was observed, the organism was identified by gram staining and biochemical reactions.

They were then subjected to antibiotic sensitivity testing by Kirby Bauer's technique on Mueller Hinton Agar as per CLSI guidelines, using Cefotaxime (30mcg), Ceftazidime (30mcg), Cefuroxime (30mcg), Amikacin (30mcg), Piperacillin-Tazobactum (100/10mcg), Ciprofloxacin (5mcg), Levofloxacin (5mcg), Imipenem (10mcg), Meropenem (10mcg), Ertapenem Tobramycin(10mcg) for Gram Negative Bacilli and Ampicillin(10mcg), Amoxycillin (10mcg), Amoxyclav (20mcg), Erythromycin (15mcg), Clindamycin (2mcg), Penicillin (10mcg), Vancomycin (30mcg), Oxacillin(1mcg) Linezolid (30mcg) among others for Gram Positive Cocci.

Results and Discussion

Out of the total 629 blood samples, 117 (18.6%) were positive for growth. Of them, predominant growth was of Gram Negative Bacilli (58.1%) (Fig 1). Enterobacteriaceae was the most common amongst the Gram Negative Bacilli.

Fig.1 Positive Cultures



The most common organism was Klebsiella spp (24.8%), followed by CONS (20.5%) (Table : 1).

Amongst the gram positive cocci, all the isolates were sensitive to Vancomycin, Linezolid and Clindamycin. Most of them were resistant to ampicillin and Amoxycillin. (Table: 2).

Table.1 No of isolates

Organism	Number	Percentage
CONS	24	20.5%
Staph aureus	14	12%
E. coli	22	18.8%
Klebsiella	29	24.8%
Pseudomonas	11	9.4%
Streptococcus viridans	2	1.7%
Acinetobacter	6	5.1%
Enterococci	9	7.7%

High sensitivity to Carbapenems like Imipenem and Meropenem was observed by the Gram Negative Bacilli. Piperacillin Tazobactum and Amikacin also showed a considerably high rate of sensitivity. Rising resistance was observed to cephalosporins (Table:3).

Blood stream infections are one of the main reasons of morbidity and mortality in ICUs. Very few of them are diagnosed based on physical signs and symptoms. It is mainly through bacteriological culture of these blood samples that definitive diagnosis is made and the antibiotic sensitivity is established (Tariq Mahmud Tariq et al., 2014). The correct treatment depends upon the knowledge of the organisms causing the infection and their sensitivity pattern.

We studied 629 blood cultures of which 18.6% were positive, which was similar to studies done by Santwana Pandey et al (12.6%), 22.4% by PP Mishra et al (17%), 22% by Tsering et al. This study showed a predominance of Gram Negative Bacilli compared to Gram Positive Cocci with Enterobacteriaceae being most common. This was very similar to a study conducted by Tsering et al who showed 61% of gram negative septicemia and 59% was reported by Jyothi et al.

Table.2 Antibiotic susceptibility of Gram Positive Cocci

Antibiotic	*Staph aureus (n=14)*		*CONS (n=24)*		*Strept viridians (n= 2)*		*Enterococcus (n = 9)*	
	Sensitive %	Resistant %	Sensitive %	Resistant %	Sensitive %	Resistant %	Sensitive %	Resistant %
Penicillin	3 (21.4%)	11 (78.6%)	9 (37.5%)	15 (62.5%)	2 (100%)	0		
Ampicilin	2 (14.3%)	12 (85.7%)	9 (37.5%)	15 (62.5%)	2 (100%)	0		
Amoxycillin	2 (14.3%)	12 (85.7%)	8 (33.3%)	16 (66.7%)	2 (100%)	0	9 (100%)	0
Erythromycin	4 (28.6%)	10 (71.4%)	13 (54.2%)	11 (45.8%)	2 (100%)	0		
Oxacillin	10 (71.4%)	4 (28.6%)	18 (75%)	6 (25%)	2 (100%)	0		
Ofloxacin	12 (85.7%)	2 (14.3%)	4 (16.7%)	20 (83.3%)	2 (100%)	0	9 (100%)	0
Amikacin	13 (92.9%)	1 (7.2%)	23 (95.8%)	1 (4.2%)	2 (100%)	0	9 (100%)	0
Ceftriaxone	13 (92.9%)	1 (7.1%)	22 (91.7%)	2 (8.3%)	2 (100%)	0	9 (100%)	0
Clindamycin	14 (100%)	0	24 (100%)	0	2 (100%)	0	9 (100%)	0
Linezolid	14 (100%)	0	24 (100%)	0	2 (100%)	0	9 (100%)	0
Vancomycin	14 (100%)	0	24 (100%)	0	2 (100%)	0	9 (100%)	0
Amoxyclav	7 (50%)	7 (50%)	14 (58.3%)	10 (41.7%)	2 (100%)	0	9 (100%)	0

Table.3 Antibiotic sensitivity of Gram Negative Bacilli

Antibiotic	*E. coli* (n=22)		*Klebsiella* (n=29)		*Pseudomonas* (n=11)		*Acinetobacter* (n=9)	
	Sensitive %	Resistant %	Sensitive %	Resistant %	Sensitive %	Resistant %	Sensitive %	Resistant%
Imipenem	21 (95.5%)	1 (4.5%)	29 (100%)	0	11 (100%)	0	8 (88.9%)	1 (11.1%)
Meropenem	21 (95.5%)	1 (4.5%)	29 (100%)	0	11 (100%)	0	8 (88.9%)	1 (11.1%)
Ertapenem	21 (95.5%)	1 (4.5%)	29 (100%)	0	11 (100%)	0	8 (88.9%)	1 (11.1%)
Doripenem	20 (90.9%)	2 (9.1%)	29 (100%)	0	11 (100%)	0	7 (77.8%)	2 (22.2%)
Tobramycin	22 (100%)	0	25(86.2%)	4(13.8%)	11(100%)	0	8(88.9%)	1(11.1%)
Amikacin	15 (68.2%)	7 (31.8%)	22 (75.9%)	7 24.1%)	1 0 (90.9%)	1 (9.1%)	7 (77.8%)	2 (22.2%)
Gentamycin	18 (81.2%)	4 18.2%)	18 (62.1%)	11 (37.9%)	8 (72.7%)	3 (27.3%)	6 (66.7%)	3 (33.3%)
Levofloxacin	19 (86.4%)	3 (13.6%)	23 (79.3%)	6 (20.7%)	11 (100%)	0	4 (44.4%)	5 (55.6%)
Ciprofloxacin	13 (59.1%)	9 40.9%)	20 (69%)	9 (31%)	9 (81.8%)	2 (18.2%)	5 (55.6%)	4 (44.4%)
Cefuroxime	4 18.2%)	18 (81.2%)	4 (13.8%)	25 (86.2%)	5 (45.5%)	6 (54.4%)	4 (44.4%)	5 (55.6%)
Cefotaxime	8 (36.4%)	14 (63.6%)	9 (31%)	20 (69%)	7 (63.6%)	4 (36.4%)	3 (33.3%)	6 (66.7%)
Ceftriaxone	8 (36.4%)	14 (63.6%)	10 (34.5%)	19 (65.5%)	8 (72.7%)	3 (27.3%)	4 (44.4%)	5 (55.6%)
Ceftazidime	9 40.9%)	13 (59.1%)	13 (44.8%)	16 (55.2%)	8 (72.7%)	3 (27.3%)	4 (44.4%)	5 (55.6%)
Pip-taz	20 (90.9%)	2 (9.1%)	28 96.6%)	1 (3.7)	11 (100%)	0	9 (100%)	0
Cotrimoxazole	6 (27.3%)	16 (72.7%)	9 (31%)	20 (69%)	7 (63.6%)	4 (36.4%)	3 (33.3%)	6 (66.7%)

In contrast, in some studies conducted in Africa, prevalence of Gram positive was more than Gram negative, 69% by Mulat et al, 60.9% in Jimma Ethiopia, 70.2% in Gondar, 71.9% in Zimbabwe, 62.6% in Addis a Baba (Mulat Dagnew et al. 2013; Zenebe T et al., 2011; Shitaye D et al., 2010; Ali J et al., 2008; Obi CLet al., 1996).

The present study showed that Gram Positive Cocci were highly sensitive to Linezolid, Vancomycin, Clindamycin, Cephalosporins. Increased sensitivity was also seen for Ofloxacin and Amikacin but high resistance pattern were seen for Erythromycin, Penicillin, Amoxycillin.

The high penicillin resistance was seen in many parts of the world, probably due to indiscriminate use of antibiotics. Tsering et al, in a study in Gangtok, Sikkim for 70% of Staph aureus trains to be resistant to penicillin, 95 % was seen by Roy et al from Lucknow, 90% by P. Jyothi et al.

Amongst the Gram Negative Bacilli, high sensitivity was seen for carbapenems and combination drugs like Piperacillin-Tazobactum. Cephalosporins and Cotrimoxazole showed high level of resistance. Among the aminoglycosides, Amikacin seemed to be more sensitive than Gentamicin. This was observed by many other investigators in India and abroad (P. Jyothi et al., 2013; Bhat YR et al., 2011).
Although Imipenem, Linezolid and Vancomycin were the most sensitive drugs as per our study, these cannot be used on regular basis as they are high end drugs and should kept in reserve. If used indiscriminatively, there are high chances of resistance being developed for these drugs also.

It is clear form our study that the bacterial profile and the antibiotic susceptibility pattern vary from hospital to hospital. We found Staph aureus in Gram Positive Cocci and Klebsiella spp in Gram Negative Cocci to be the most common isolates in patients with Bacterial Septicaemia and most of these isolates were multidrug resistant. It is therefore imminent to continually conduct surveillance of antimicrobial resistance, so that proper treatment can be given depending on the sensitivity pattern. Since common drugs like penicillin and amoxicillin are showing high resistance, we recommend health education to common man on hazards of misuse of antibiotics which can be obtained over the counter.

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