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Profile of Bacterial Isolates from Blood Cultures and their Antimicrobial Susceptibility Pattern

M. Saleem[1]*, R. Gopal[1], T. Mangaiyarkarsi[1], S. Sunil[1], J. Krishnapriya[1] and R. Nagma[2]

[1]Department of Microbiology, Sri Manakula Vinayagar Medical College and Hospital, Puducherry, India
[2]Department of Microbiology, SMVMCH, Puducherry, India

**Corresponding author*

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A B S T R A C T

Blood stream infections (BSI) are an important cause of morbidity and mortality all over the world. Bacteremia is an important and common cause of septicaemia in all age groups. The incidence of bacteremia and the causative bacteria and their antimicrobial susceptibility pattern varies with time and place. The present work is a retrospective study of bacteria isolated from blood cultures of patients with suspected bacteremia and their antimicrobial susceptibility pattern. *Escherichia coli* was the most common isolate among gram negative bacilli (GNB) followed by Staphylococcus aureus among gram positive cocci (GPC). The incidence was high among male children in the age group of 1-10 years. Imipenem was the most effective drug against *Escherichia coli* and Vancomycin against *Staphylococcus aureus*.

Introduction

Septicemia due to bacteria is an important cause of morbidity and mortality among patients hospitalized in critical care units all over the world. The severity of blood stream infections (BSI) varies from a self limiting to life threatening sepsis (Young *et al*., 1995). The spectrum of bacteria that causes B.S.I varies widely from place to place (Fuselier *et al*., 2002; Trevini, 2008; Elhag *et al*., 1985; Crowe *et al*., 1985) with time and can involve all age groups and both sexes.

Many infections in neonatal and pediatric age group can only be established on the basis of etiological agent recovered from blood. Neonatal septicaemia refers to systemic infection in the new born confirmed by a positive blood culture (Klein *et al*., 1983). It remains a major cause of morbidity and mortality among newborn especially in developing countries, where its incidence is higher than in the developed world. Bacteremia has many possible

causes, including catheterization of an infected lower urinary tract, Surgical treatment of an abscess or infected wound and colonization of indwelling devices, especially IV and intracardiac catheters, urethral catheters, and ostomy devices and tubes. Gram-negative bacteremia secondary to infection usually originates in the GU or GI tract or in the skin of patients with decubitus ulcers. Chronically ill and immunocompromised patients have an increased risk of gram-negative bacteremia. They may also develop bacteremia with gram-positive cocci and anaerobes, and are at risk of fungemia. Staphylococcal bacteremia is common among injection drug users and patients with IV catheters. Bacteremia may develop in patients with infections of the abdomen and the pelvis, particularly the female genital tract. If an infection in the abdomen causes bacteremia, the organism is most likely a gram-negative bacillus. If an infection above the diaphragm causes bacteremia, the organism is most likely gram-positive. Bacteremia may cause endocarditis, most commonly with enterococcal, streptococcal, or staphylococcal bacteremia and less commonly with gram-negative bacteremia or fungemia. Patients with structural heart disease (eg, valvular disease, certain congenital anomalies), prosthetic heart valves, or other intravascular prostheses are predisposed to endocarditis. Staphylococci can cause bacterial endocarditis, particularly in injection drug users, and usually involving the tricuspid valve. 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. The infection caused by MDR organisms is more likely to prolong the hospital stay, increase the risk of death, and require treatment with more expensive antibiotics. Periodic monitoring of organisms responsible for sepsis and their antimicrobial susceptibility pattern helps in formulating the empirical treatment for cases presenting with sepsis which helps in reducing the mortality associated with these infections particularly in the context of rapidly evolving multidrug resistant strains.

Materials and Methods

This study was carried out at the department of Microbiology, in a rural tertiary care hospital, Puducherry, India. A total of 476 samples from clinically suspected cases of bacteremia were collected during the period of one year i.e. from Jan 2015 to Dec 2015. Blood sample were collected aseptically from patients. The quantity varied from 1ml to 5ml depending on the age of the patient and inoculated into "BHI" broth aseptically with a dilution of approximately 1in 5. The inoculated bottles were incubated immediately at 37^{o}c. Blood culture broths were checked for sign of bacterial growth (turbidity, haemolysis, clot formation) daily up to 7 days. Samples were incubated for a longer period when indicated, as from suspected cases of endocarditis. Before being reported as a negative result blood culture broths with no bacterial growth after 7 days were sub-cultured. Bottles which showed signs of growth were further processed by gram stain and sub-culture was made onto blood agar, MacConkey agar and chocolate agar and incubated at 37°C for 24 h. The CA and BA plates were incubated in a candle jar providing 5 – 10 % CO_2. Bacterial isolates were identified by colony morphology, gram staining reaction and biochemical tests (Collee *et al*., 1996).

Antimicrobial susceptibility test

Antibiogram of all suspected pathogens was performed by Kirby bauer disc diffusion method as per CLSI guidelines [(9)]. The

antibiotic discs and their concentrations were: Amoxicillin-clavulanic acid; (30 µg), Ceftriaxone, (30 µg) ,Vancomycin (30 µg) (Oxacillin (1 µg) , Ciprofloxacin (5 µg) , Gentamicin (120 µg), Norfloxacin (10 µg), Doxycycline (30 µg), Erythromycin (15 µg), and Trimethoprim- sulphamethoxazole (25 µg). These are the antibiotics which are being frequently used for the routine antimicrobial susceptibility test in our hospital.

Results and Discussion

The percentage of bacteraemia in males (57.4%) was higher than females (42.6%). Children below 10years had a higher incidence of bacteraemia (table.1). Gram negative bacilli were more common than gram positive cocci. *Staphylococcus aureus* was the predominant organism found among gram positive cocci and *Escherichia coli* the most common among gram negative bacilli (table.2 and 3). Imipenem was the most effective antimicrobial against gram negative bacilli(table.4), whereas Vancomycin was the most effevtive antimicrobial against gram positive cocci(table.5).

Sepsis is a systemic inflammatory response to an underlying infection of the blood stream. Blood stream infections (BSI) remain one of the most important causes of morbidity and mortality all over the world (Zenebe *et al*., 2011). Around 13 million people have sepsis and as many as 4 million die of sepsis each year across the globe. A variety of bacteria have been reported to cause bacteraemia with variation in distribution from place to place (Kaul *et al*., 2006).

Table.1 Age & sex distribution of isolates

Age (Yr)	Male	Female	Total
0-10	64 (23.4%)	57 (28.0%)	121(51.4%)
11-20	29 (10.6%)	9 (4.4%)	38(15%)
21-30	20 (7.3%)	16 (7.8%)	36 (15.1%)
31-40	27 (9.8%)	30 (14.7%)	57(24.5%)
41-50	24 (8.7%)	23 (11.3%)	47(20%)
51-60	41 (15%)	25 (12.3%)	66(27.3%)
>60	68 (24.9%)	43 (21.1%)	111(46%)
TOTAL	273(57.4%)	203(42.6%)	476(100%)

Table.2 Distribution of Gram Positive Cocci

Organisms (GPC)	Total
Staph. Aureus	59 (66 %)
CONS	21 (23.5%)
Enterococcus	5 (5.6%)
Streptococcus	4 (4.4 %)
TOTAL	89

Table.3 Distribution of Gram negative bacilli

Organisms(GNB)	Total
E.coli	67 (33.3%)
Pseudomonas species	43 (21.3%)
N.f.GNB	39 (19.4%)
Klebsiella pneumoniae	26 (12.9%)
Proteus species	9 (4.4%)
Salmonella typhii	9 (4.4%)
Citrobacter	5 (2.4%)
Acinitobacter	3 (1.4%)
Total	201

Table.4 Sensitivity pattern of Gram negative bacilli (% Susceptible)

Antibiotic	*E.coli*	*Klebsiella*	*Pseudo-Monas*	*Nf GNB*	*Salmonella*	*Citro-Bacter*	*Proteus*	*Aceinit-Obacter*
AMIKACIN (AK)	33(49.2%)	16(61.5%)	29(67.4%)	18(46.1%)	3(33.3%)	3(60%)	2 (22.2%)	2(66.6%)
CIPROFLOXACIN(CIP)	12(17.9%)	11(42.3%)	31(72.0%)	16(41.0%)	4 (44.4%)	3(60%)	3 (33.3%)	3(100%)
GENTAMICIN(GM)	19(28.3%)	13 (50%)	19(44.1%)	28(71.7%)	5 (55.5%)	2(40%)	2 (22.2%)	2 (66.6%)
IMIPENEM (IPM)	36(53.7%)	19(73.0%)	35(81.3%)	30(76.9%)	9 (100%)	5(100%)	6 (66.6%)	3(100%)
CEFTRIAXONE(CTR)	22(32.8%)	7 (26.9%)	40(93.0%)	22(56.4%)	5 (55.5%)	2 (40%)	4 (44.4%)	2 (66.6%)
CEFOTAXIME(CTX)	14(20.8%)	20(76.9%)	35(81.3%)	23(58.9%)	8 (88.8%)	3 (60%)	2 (22.2%)	1 (33.3%)
AMOXYCLAV(AMC)	15(22.3%)	14(53.8%)	22(51.1%)	9 (23.0%)	8 (88.8%)	5(100%)	2 (22.2%)	3(100%)
CAFTAZIDIME(CAZ)	15(22.3%)	11(42.3%)	38(88.3%)	34(87.1%)	9 (100%)	3 (60%)	3 (33.3%)	2 (66.6%)
PIPERACILLIN (PI)	27(40.2%)	5 (19.2%)	36(83.7%)	28(71.7%)	6 (66.6%)	3 (60%)	2 (22.2%)	1(33.3%)
CEFUROXIME(CXM)	5 (7.4%)	6 (23.0%)	15(34.8%)	16(41.0%)	4 (44.4%)	3 (60%)	1 (11.1%)	2 (66.6%)
LEVOFLOXACIN(LE)	17(25.3%)	5 (19.2%)	11(25.5%)	6 (15.3%)	5 (55.5%)	3 (60%)	6 (66.6%)	2 (66.6%)
TOBRAMYCIN (TOB)	13(19.4%)	4 (15.3%)	7 (16.2%)	2 (5.1%)	5 (55.5%)	3 (60%)	2 (22.2%)	2 (66.6%)
CLAVULANIC ACID(CCC)/ CEFTAZIDIME	17(25.3%)	2 (7.6%)	4 (9.3%)	3 (7.6%)	2 (22.2%)	3 (60%)	2 (22.2%)	2 (66.6%)

Table.5 Sensitivity pattern of Gram positive cocci (% Susceptible)

Antibiotic	CONS	*Staph.aureus*	*Entero*	*Strepto*
AMIKACIN (AK)	4(19.0%)	49 (83.0%)	2 (40%)	2 (50%)
CIPROFLOXACIN (CIP)	10(47.6%)	40 (67.7%)	4 (80%)	2 (50%)
GENTAMICIN (GEN)	13(61.9%)	46 (77.9%)	3 (60%)	2 (50%)
VANCOMYCIN(VA)	16(76.1%)	59 (100%)	5 (100%)	2(50%)
COTRIMOXAZOLE (COT)	13(61.9%)	17(28.8%)	3(60%)	2(50%)
PENICILLIN(P)	5(23.8%)	17(28.8%)	4(80%)	2 (50%)
OXACILLIN(OX)	9(42.8%)	19(32.2%)	1(20%)	2 (50%)
ERYTHROMYCIN(E)	11(52.3%)	24(40.6%)	2(40%)	2 (50%)
CLINDAMYCIN(CD)	12(57.1%)	54(91.5%)	3(60%)	2 (50%)
CEPHOXITIN(CX)	4(19.0%)	18(30.5%)	2 (40%)	2 (50%)
TEICOPLANIN(TEI)	6(28.5%)	18(30.5%)	-	2 (50%)
LINEZOLID(LZ)	4(19.0%)	2(3.3%)	-	2 (50%)

The isolation and identification of bacteria in the blood stream caries a significant therapeutic value for initiation of appropriate antimicrobial therapy in a variety of diseases like endocarditis, pneumonia and pyrexia of unknown origin (Weinstein *et al*., 1997). In the present study the highest incidence of bacteraemia was in the age group of 0-10 and relatively high among male than female in all age groups. A similar high positivity among males was noticed in the study by Maimoona and Meenakshi Kante. Gram negative bacilli constituted the majority of the isolates with *Escherichia coli* being the most frequent isolate. Among the gram positive isolates *Staphylococcus aureus* was the most common similar to a study by Meenakshi *et al*., (2014). Imipenem was the most effective drug against *Escherichia coli* and Vancomycin was the most effective against *Staphylococcus aureus*.

The present study shows that a gram negative bacillus (*E.coli*) was the most common cause of bacteraemia among all study population. However the distribution and susceptibility varies with geographical location. Regular surveillance studies help in initiation of appropriate antimicrobial drug for empirical therapy and treatment which is essential for a favorable outcome of these cases.

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