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"DANGER IN THE BLOOD" BSI AND CURRENT TREND IN ANTIMICROBIAL RESISTANCE

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ABSTRACT

It's a leading cause of death. It's a major source of health care costs, adding days or weeks to the hospital stays, no one fully understands how best to fight it. Bacteremia may be self limiting or life threatening. So, correct and accurate antibiotics to be employed to understand the common pathogens and their resistance pattern time -to-time. The present study was undertaken to know the pathogens causing Blood stream infections and incidence of multidrug resistance (MDR). Altogether 776 blood samples were investigated during the working period between January to December, 2012 at microbiology laboratory, ASRAM hospital, Eluru. A total of 191 (24.6 %) blood samples were found to be culture positive. *CoNS* (50 %) were the predominant isolate, followed by *Staphylococcus aureus* (44.56 %) and *Klebsiella pneumoniae* (38.38 %). Analysis of the samples showed that Bacteremia was common in males when compared to females. Antimicrobial drug resistance is a major problem in India. During the last few years a lot of attention was being paid to microbial resistance. There have been many debates but the outcomes are negative. This study shows that a good percentage of people were infested by multi-drug resistant bacterial agents.

KEYWORDS: Blood stream infections, Gram positive bacteria, Gram negative bacteria, Multidrug resistance.

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INTRODUCTION

Blood is a body fluid that delivers necessary substances such as nutrients and oxygen to the cells and transports metabolic waste products away from those same cells. In vertebrates, it is composed of blood cells suspended in plasma. Blood is normally sterile, but if invaded by foreigners, then they pose a major threat to our health. So there presence in blood makes it a danger. Bacterial infection of the blood is bacteremia and it's a major part of the very serious illness called sepsis-"blood poisoning." Bloodstream infections (BSI) are potentially life-threatening and require rapid identification and also antibiotic susceptibility testing of the causative agent in order to facilitate specific antimicrobial therapy,(Berit E C et al[1]). Despite considerable progress in hygiene, antimicrobial therapy, and supportive treatment, blood stream infections remain important causes of morbidity and mortality. In developing countries, more than 14 million deaths of children under five years of age,(UNICEF[2]) occur during the childhood. Bacteremia has an increasing trend in some regions of the world (Madsen et al[3]). The isolated bacteria are numerous (Reacher et al[4], Cohen et al[5]). The organisms responsible for bacteremia vary across geographical boundaries. Organisms like *Staphylococcus aureus,* Coagulase negative *staphylococci* (CoNS), *Pseudomonas spp.,E. coli , Klebsiella* spp.*, Salmonella* spp. and *Acinetobacter* spp. are potential pathogens in bacteremia because of their frequent isolation and multi-drug resistance which has reached worrying levels (Castagnola et al [6],Wise et al [7]).

Antibiotic resistance among bacteria is becoming more and more serious problem throughout the world. It is said that evolution of bacteria towards resistance to antimicrobial drugs, including multidrug resistance, is unavoidable because it represents a particular aspect of the general evolution of bacteria that is un-stoppable (Courvalin et al [8]). Antibiotic resistance emerges commonly when patients are treated with empiric antimicrobial drugs. The widespread use of antibiotics both inside and outside of medicine is playing a significant role in the emergence of resistant bacteria.(Bacon, D.J. *et al* [9]) For practicing physicians, clinical microbiologists and public health officials, knowledge of local antimicrobial resistance patterns is essential to guide empirical and pathogen-specific therapy. Many of the greatest challenges in medicine and public health involve the evolution of drug resistance by pathogens. Unfortunately, data regarding endemic antimicrobial resistance are unavailable in many parts of the world, especially from areas where over-the-counter antibiotic use is common. In the USA alone, 10–20% of nosocomial infections are estimated to involve the bloodstream, resulting in 90 000 fatal cases each year. Appropriate antimicrobial treatment of BSIs is critical in decreasing morbidity and mortality due to BSIs (Bacon, D.J. Reimer LG et al [10]). So, this study was undertaken to investigate bacteria which cause blood stream infection and determination of their resistance pattern to antimicrobial agents since drug resistance is increasing day by day because just as we humans change our habits the microbes are also prone to change and become resistance to the once used drugs - "adaptation is a must if one has to survive".

AIMS

To investigate frequency of bacterial pathogens causing BSI and their antimicrobial resistant pattern – a study undertaken at tertiary care teaching hospital in west Godavari district, Andhra Pradesh.

SETTINGS AND DESIGN

It was a study, conducted during the period between January to December 2012 at Alluri Sita Ramaraju Academy of Medical Sciences- a tertiary care teaching hospital, Eluru, West Godavari district, AP, India.

SAMPLE COLLECTION

10 ml venous blood for adults and 1 ml for infants collected following all the strict aseptic precautions. By using 70% ethyl alcohol, the skin over the puncture site was cleansed before the procedure.

BLOOD CULTURE

Blood drawn and inoculated aseptically into blood culture bottle containing 10ml of brain heart infusion broth (Hi-media laboratories Pvt. Ltd, Mumbai, India). After blood was inoculated into blood culture bottle, it was incubated at 37^{o}C under aerobic conditions for 7 days. The first subculture was done on 1st day. The second and third was on 2nd, 3rd day and last subculture on 7th day. The subculture was done onto Blood agar (M001- Hi media laboratories, Mumbai) and Mac Conkey agar (M082- Hi media laboratories) plates which were incubated at 37^{o}C for overnight. If any growth was seen on the plates, it was processed according to standard. Isolated colonies were initially Gram stained. By using Bergey's Manual of Determinative Bacteriology (9th edition), the isolates were biochemically characterized and identified.

RESULTS

Frequency of occurrence among blood culture microorganisms

Of the total 776 blood samples screened, 191 (24.61%) were positive for bacterial growth. They belonged to 167 adults, mean age was 65 with 88 males (46.07%) and 79 females (41.36%) and also 24 (12.56%) infants < than 12 months.

Table 1
Distribution of positive blood cultures

	No. of patients (ή = 191)	
Category	**Total**	**Percentage (%)**
Males	88	46.07
Females	79	41.36
Infants < 12 months	24	12.56

Microorganisms which were recovered from blood cultures were: 99 (51.83%) gram negative bacilli and 92 (48.16%) gram positive cocci. The most common isolated gram negative bacilli were *Klebsiella* spp 38 (38.38 %), *Pseudomonas aeruginosa* 32 (32.32%) and E.coli 19 (19.19%) and the least common were *Enterobacter cloacoa* 4 (4.04%), *Acinetobacter* spp 4 (4.04%) and *Proteus mirabilis* 2 (2.02%).

Table 2
Distribution of Gram Negative Organisms

	No. of isolates (ή=99)	
Organisms	**N**	**Percentage (%)**
Escherichia coli	19	19.19
Klebsiella pneumoniae	38	38.38
Pseudomonas aeruginosa	32	32.32
Enterobacter cloacae	4	4.04
Acinetobacter spp	4	4.04
Proteus mirabilis	2	2.02

Among gram positive cocci, *Coagulase negative staphylococci* 46 (50 %) was predominant, followed by *Staphylococcus aureus* 41 (44.56%)

Table 3
Distribution of Gram positive organisms

	No. of isolates (ή=92)	
Organisms	N	Percentage (%)
CONS	46	50
CPS	41	44.56
Others	5	5.43

All the isolates were multidrug resistant. Gram negative isolates were resistant to third generation cephalosporins and penicillin.

Table 4
Antibiotic resistance pattern of Gram Positive organisms

	Organisms	
Antibiotic	**CONS (ή = 46)**	**CPS (ή = 41)**
Vancomycin	2 (4.37%)	1(2.43%)
Erythromycin	22 (47.82%)	20(48.78%)
Gentamicin	3(6.52%)	2(4.87%)
Cefotaxime	23 (50%)	27(65.85%)
Ceftazidime	32(69.56%)	24(58.53%)
Cefazolin	30(65.21%)	26(68.29%)
Ciprofloxacin	3(6.52%)	3(7.31%)
Penicillin	14(30.43%)	21(51.21%)
Methicillin	19(41.30%)	25(60.97%)
Ampicillin	32(69.56%)	29(70.73%)
Oxacillin	12(26.08%)	27(65.85%)
Novobiocin	12(26.08%)	NOT DONE
Clindamycin	5(10.86%)	4(9.75%)

Gram negative isolates showed resistance to Ampicillin, imipenim and third generation cephalosporins.

TABLE 5
Antibiotic resistance pattern of Gram negative organisms

	Organisms		
Antibiotic	**Klebsiella (ή = 38)**	**Pseudomonas (ή = 32)**	**E.coli (ή = 19)**
Ampicillin	26(68.42%)	19(59.37%)	13(68.42%)
Amikacin	3(7.89%)	2(6.25%)	1(5.26%)
Ceftazidime	25(65.78%)	23(71.87%)	10(52.63%)
Cefotaxime	26(76.31%)	28(87.5%)	15(78.94%)
Ciprofloxacin	17(44.73%)	5(15.65%)	9(47.36%)
Co-trimoxazole	2(5.26%)	2(6.25%)	1(5.26%)
Imipenim	24(63.15%)	22(68.75%)	10(52.63%)
Cefoperazone-sulbactum	3(7.89%)	3(9.37%)	2(10.52%)
Piperacillin+tazobactum	3(7.89%)	-	4(21.05%)
Gentamicin	24(63.15%)	25(78.12%)	11(57.89%)

DISCUSSION

Bacteremia is defined as the presence of viable microorganisms in the bloodstream and can be categorized as transient, intermittent, or persistent (Fluit A, et al [11]). According to global surveillance reports, bloodstream isolates are the best candidates for the study of antimicrobial susceptibility of human bacterial pathogens (Von Eiff et al[12], Khanal et al [13]). In

our study 24.61 % Blood cultures were positive. In contrast to the above reports, the studies done in India, by Khanal et al [13], Sharma[14], Mehta et al[15], Arora and Devi et al[16] , have reported frequency of positive blood cultures accounting for 44%, 33.9%, 16.4%, 9.94%, and 20.2% respectively. So, our study correlates with Arora and Devi et al [16.] Our study shows that BSI was common in males (46.07%) than females (41.36%), which correlates with the study of Mehdinejad et al[17.] In India, variation might be due to the reason that most of the patients are given antibiotics before they come to the tertiary care hospital and other reason is that in most of the cases self medication is very common as the medicines are available at the counter due to wide spread of pharmacies. The growth in the present study was monomicrobial. *Coagulase-negative staphylococcus* was the most common followed by *Staphylococcus aureus* and the study correlates with that of Mamishi et al [18], Ammita et al[19.]

CoNS is usually treated as a contaminant but is also an increasingly important pathogen, which is a significant clinical problem because there is no true "gold standard" for determining contamination from relevant pathogens (Aronson et al[20], Lee et al[21],Weinstein[22]) Unlike isolates such as *Escherichia coli, Klebsiella pneumoniae, Pseudomonas* and *Staphylococcus aureus*, which are encountered often as pathogens and diphtheroids and bacillus species which invariably are contaminants, *Coagulase negative staphylococci* could be either pathogens or contaminants when encountered in blood cultures. Furthermore, because of their low virulence, they may not evoke sufficient inflammatory response and thus a number of patients with Coagulase negative staphylococcal bloodstream infection may not have typical clinical manifestations and laboratory indices of infection (Hirakata et al[23]). Cultures which were positive for CoNS were all from different ICU units. The patients were of extreme age group with hematological malignancies, venous catheters and undergoing invasive procedures. The growth of *CoNS* was monomicrobial with MRD pattern. In *CPS* high degree of resistance was shown towards cephalosporins, methecillin, Ampicillin and Oxacillin. Oxacillin - resistant S*. aureus* are extremely important causes of bloodstream infections and evidence has been presented that Oxacillin-resistant *S. aureus* are increasing globally among bloodstream isolates and among isolates from other anatomical sites (Diekema 2000[24],Diekema 1997-1999[25]). Penicillin resistance for staphylococci was found to be 30.43% and 51.21% of them were also resistant to Methicillin. Best drugs for treating GPC BSI were Vancomycin, Gentamicin and ciprofloxacin. Among all these vancomycin was found to be the most effective drug for gram positive isolates. Life saving drug Vancomycin is already being used indiscriminately. In the present scenario, where bacterial resistance is being reported continuously to traditional antibiotics, there is a great urgency to save such antibiotics which can take care of almost all pathogenic bacteria known to medical science.

Among gram negative isolates, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* dominated *Escherichia coli* and all of them were multi drug resistant. Although *Escherichia coli* is the most common etiologic agent of Gram-negative bacteremia (Wagner et al[26]), our results revealed that *Klebsiella* was the predominant. There are also earlier studies in agreement to present findings which reports Klebsiella as the most common cause of gram negative bacteremia (Kumar et al[27], Kang et al[28]). Bacteremia due to *Pseudomonas aeruginosa* is associated with grave clinical outcomes. Our study shows that *Pseudomonas aeruginosa* is the second common isolate among gram negative bacilli. The most common resistance was seen to Ampicillin in all isolated gram negative bacteria. They were also resistant to third generation cephalosporins, aminoglycoside and Gentamicin. The best antimicrobial agents for treating BSI due to GNB were Amikacin, Co-Trimoxazole, cefoperazone-sulbactum in our study. However, Indian data on antibiogram for isolates from bacteremia remains scanty over the recent years. The earlier reports have been mainly about the methodologies. (Koshi .G et al[29], Ganguli et al[30]). The combination of antibiotics is preferred

and has to have the following advantages, (Anderson et al [31])1. Both the drugs should be bactericidal. 2. The effect of the combination preferably should be synergistic and at least additive but not antagonistic. 3. The drug concentrations in the blood should be above the MIC levels. However it has to be noted that, microbial pathogens, as well as their antibiotic sensitivity patterns may change from time to time and place to place. The isolated organisms and their susceptibility of our place may be same or different when compared to others. The discovery of antibiotics revolutionized the management of infectious diseases. However, the overuse and misuse of antibiotics is leading to the emergence of resistance to these life saving drugs. Hospital antibiogram are commonly used to help guide empiric antimicrobial treatment and are an important component of detecting and monitoring trends in antimicrobial resistance (Pharmacotherapy 2007.[32])

CONCLUSION

Clinically defined BSIs are always significant. Danger invaders must be tackled. Determining whether these are primary or are associated with an infection at another site is a priority in selecting the most appropriate management strategy and requires a thorough examination, including microbiological, pathological, and imaging studies. Rather than a single drug, combinations of drugs were far more effective. But we have only few such drugs in India. We can affirm that the choice of drugs in the treatment of BSI today is narrow , no surety to say that a particular drug will always at effective at the same dose due to widespread resistance & ongoing changes of the resistance patterns. So, regular antimicrobial susceptibility surveillance is essential. This should to be the national strategy to control the spread of resistance in India.

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