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Original Research Article.

Spectrum of Microorganisms Causing Blood Stream Infection and the Antibiotic Sensitivity Pattern among Gram Negative Organisms: A Study from a Tertiary Care Centre in Bihar

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ABSTRACT

Introduction: Incidence of Blood stream infection (BSI) is on the rise and so is the increasing drug resistance among different organisms causing BSI. The trend of such infection and the antibiotic sensitivity pattern may vary from region to region.

Materials and Methods: This study was conducted with the same aim in which about 240 blood culture and sensitivity results were noted from the records and were analysed to see the current trend in this region. Our focus was mainly on Gram negative organisms, as they have been found to cause increased number of BSI.

Results: *Escherichia coli* was the main Gram negative organism, and *Staphylococcus aureus* along with Coagulase negative Staphylococcus (CoNS) were the main gram positive organisms isolated. *Candida albicans* was also found in four of the cases. The antibiotic sensitivity pattern of gram negative isolates showed more than 90% susceptibility to higher antibiotics, whereas the susceptibility to other antibiotics were found to be variable.

Conclusion: *E. coli* is the main GNB causing the infection followed by *K. pneumoniae* and *P. aeruginosa.* Increasing trend of drug resistance is being noted by GNBs against most commonly used antibiotics.

Keywords: Blood Stream Infection, Sepsis, Culture and Sensitivity, Gram Negative Organisms.

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INTRODUCTION

Sepsis is one of the leading causes of death worldwide with an estimated incidence of up to 19 million people every year.[1] Blood stream infection (BSI) has often been associated with hospitalization, insertion of catheters into blood vessels and other predisposing factors like lapses in hand washing and non-adherence to infection control practices of medical staff in intensive care units and in high risk areas. Respiratory tract, genitourinary tract and intra-abdominal foci are the common sources of blood stream infections.[2]

Septicemia is a life threatening condition which requires rapid and aggressive antimicrobial treatment.[3] But the fact that the organisms isolated from blood cultures vary according to different geographical distribution and has higher tendency of developing multidrug resistance in their prolonged course, is of great concern.[2] Blood culture still remains the gold standard for diagnosing septicemia.[4] Gram negative bacteria have been shown to cause more incidence of sepsis as compared to gram positive bacteria in many studies.[2,5,6] The culture and sensitivity pattern of the organisms isolated from blood culture may vary from place to place and from time to time depending upon the geographic variations.[7] So, frequent studies are needed from time to time to know the changing trends of the culture and sensitivity pattern in different regions.

AIMS AND OBJECTIVES

a. To study the spectrum of microorganisms causing blood stream infection in suspected sepsis patients.
b. To study the antibiotic sensitivity pattern of gram negative organisms causing blood stream infection.

MATERIALS AND METHODS

This retrospective study was carried out in the department of microbiology of Nalanda Medical College and Hospital, Patna, Bihar, from July 2017 to May 2018. A total of 240 blood samples from clinically diagnosed cases of sepsis, received in the microbiology laboratory were included in the study. The

demographic details of the patients like name, age and sex were noted down as per the maintained records in the department. Samples received were from the inpatient population of the hospital. Blood samples were collected aseptically from the patients before the administration of any antibiotic. Blood culture bottles inoculated with the sample were incubated at 37°C aerobically, and periodic subcultures were done on blood agar and MacConkey agar on day 2, 3 and day 7 respectively, and in between, if there was visible turbidity. The growth, if obtained, was identified by colony morphology, Gram stain of the isolated colonies, and conventional biochemical identification tests as per the standard protocol followed in our laboratory.[8,9] The antibiotic susceptibility test of the isolated Gram negative organisms were performed by Kirby–Bauer disc diffusion method on Mueller–Hinton agar media as per the Clinical and Laboratory Standards Institute (CLSI) guidelines[9] and their sensitivity pattern was noted down. The sensitivity to colistin was seen using the E-test method.[9] Data were entered into Microsoft excel sheet and different percentages were calculated and presented in tabular form and in pie charts.

Fig 1: Sex distribution of positive blood culture samples

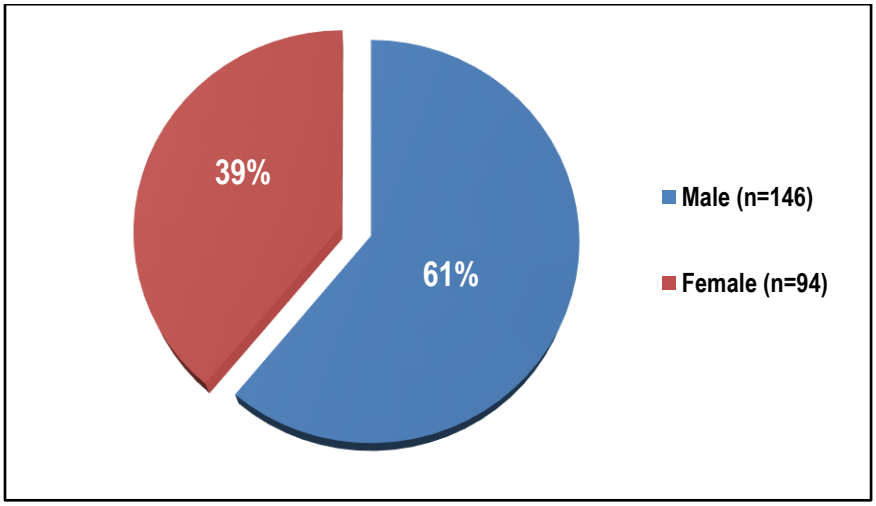

Table 1: Age distribution of Blood culture sample

Age distribution	Growth n=51	Sterile n=189	Total n=240	Positivity %
Infant (<1 year)	13	59	72	18.05 %
Children and adolescents (up to 18 yrs)	20	78	98	20.40 %
Adult (>18 yrs)	18	52	70	25.70 %

Table 2: Bacterial isolates from positive Blood culture

Bacterial Isolates	No. of Isolates (n=51)	Percentage (100%)
Escherichia coli	14	28.0%
Klebsiella pneumoniae	10	20.5%
Pseudomonas aeruginosa	07	13.5%
Staphylococcus aureus	07	13.5%
Coagulage negative Staphylococcus	07	13.5%
Enterococcus sp.	01	2.0%
Candida albicans	04	7.1%
Yeast other than Candida albicans	01	1.9%

RESULTS

Out of 240 blood cultures, growth was observed in 51 (21%) samples, whereas 189 (79%) samples were sterile. BSI was found to be more common among male patients as compared to female patients [Fig-1]. As regards to the age group of the patients having septicemia, adults were found to be mostly infected in about 25.7% cases, followed by the children and adolescents taken together and the infants in 20.4% and 18% cases, respectively [Table 1]. Bacterial growth was seen in 91% positive blood samples and fungal growth was obtained in 9% of the samples (Table 2). Gram negative bacteria was found to be in around 31 (60.8%) culture positive samples and gram positive bacteria was found in 20 (39.2%) samples. Most common bacteria isolated

were *Escherichia coli* (28%), followed by *Klebsiella pneumoniae* (20.5%), *Pseudomonas aeruginosa* (13.5%), *Staphylococcus aureus* (13.5%), *Coagulase negative Staphylococcus, CoNS* (13.5%) and *Enterococcus sp* (2%). Among the fungal growth, only *Candida* was isolated in around 9% of the positive samples. 4 of the 5 candida isolates were identified as *Candida albicans*, whereas 1 was yeast other than *Candida albicans* [Table 2]. The antibiotic sensitive pattern of gram negative isolates showed more than 90% susceptibility to higher antibiotics, whereas susceptibility to other antibiotics were found to be variable.[Table 3]

Table 3: Antibiotic sensitivity pattern of isolated gram negative organisms (n=31)

Antibiotics	*E. coli* (n=14) (%)	*K. pneumoniae* (n=10) (%)	*P. aeruginosa* (n=07) (%)
Ampicillin	02 (14.3)	01 (10)	01 (14.2)
Ceftriaxone	05 (35.7)	02 (20)	NT
Cefixime	03 (21.4)	02 (20)	NT
Ceftazidime	NT	NT	02 (28.5)
Cotrimoxazole	08 (57.1)	04 (40)	NT
Ciprofloxacin	07 (50.0)	04 (40)	03 (42.8)
Piperacillin-Tazobactam	12 (85.7)	08 (80)	05 (71.4)
Amikacin	13 (92.8)	08 (80)	06 (85.7)
Gentamicin	11 (78.5)	06 (60)	03 (42.8)
Meropenem	13 (92.8)	09 (90)	06 (85.7)
Imipenem	12 (85.7)	08 (80)	07 (100)
Colistin	14 (100)	10 (100)	07 (100)

NT: Not tested, E. coli: Escherichia coli, K. pneumoniae: Klebsiella pneumoniae, P. aeruginosa: Pseudomonas aeruginosa

DISCUSSION

The present study shows us the distribution of bacterial isolates causing BSI and the antibiotic susceptibility pattern of the gram negative bacteria causing such infection. Out of 240 blood culture samples, about 51 (21%) samples were positive, which is slightly higher as reported by some other studies but is in accordance with the study done by Pal N et al.[2] Some studies have shown lower rate of positivity varying from 14 to 17% of the total blood samples.[6,10] This variation may be due to the varying number of samples in different studies or due to the varying geographical conditions. Males were affected more as compared to female patients, which were also shown by many studies.[2,6,10-13] Infection was more prevalent in the adult age group as compared to others as has also been shown by a previous study.[2] Another study by Banik A et al has shown more prevalence in the pediatric age group.[10] This age wise differences may be attributed to the regional variations which is known to occur.[2,5]

Among the organisms isolated, gram negative bacteria exceeded the gram positive bacteria as has previously been shown by many studies.[2,5,6] Some studies have even shown the changing pattern in which gram positive organisms dominated the list.[10] Among the gram negative organisms, *E. coli* was the most common isolate, followed by *Klebsiella* and *Pseudomonas*. Enterobacteriacae as a group with predominant *E. coli*, followed by *Klebsiella* has also been shown as the main GNB in other studies.[2,6] *Pseudomonas* along with other non-fermenter group of organisms has also been shown as the agents causing septicemia in many other studies[2,5,6,10,12,13] but in our study only *Pseudomonas aeruginosa* was isolated in 7 cases. Similarly, other studies have also shown the isolation of Gram negative organisms like *Enterobacter, Citrobacter, Samonella* and others, which were not found in our study.[5,6,12,13] This may be attributed to the low sample size in our study, since sample size in other studies showing their isolation is large. But still their isolation percentage in those studies is very low, suggesting that they cause sepsis in fewer cases. Among the Gram positive organisms, *Staphylococcus aureus* and *CoNS*, both were equally isolated. In some studies *S. aureus* has been the predominant isolate among all gram positive ones[5,6,10], whereas *Coagulase Negative Staphylococcus* predominates the list in some.[2] For last few decades, CoNS, considered as the usual skin commensals, are increasingly being considered as bloodstream pathogens in select settings. Improper methods of blood collection and the presence of long standing intravascular catheters are recognized as possible modes of spread of blood stream infection (BSI) by CoNS.[10] Few studies have reported CoNS as the most common isolate causing BSIs in ICU patients.[14,15]

Among the fungal isolates, *Candida albicans* was the main agent causing BSI in our study, which is in accordance with most other studies.[16,17] In a study, *Candida glabrata* and *Candida krusei* was shown to cause more infection as compared to *Candida albicans*.[2] There was a single non albicans Candida in our study, the identification of which could not be done.

Resistance pattern among the gram negative isolates were studied, in which they showed more than 90% susceptibility to higher antibiotics, whereas the susceptibility to other antibiotics were found to be variable [Table 3]. More than 85% sensitivity was seen with imipenem, meropenem and colistin. Amikacin and piperacillin-tazobactam also showed good sensitivity against all the three groups of organisms. Less than 50% sensitivity was seen with other antibiotics like ampicillin, gentamicin, ceftriaxone, cefixime, ceftazidime, ciprofloxacin and cotrimoxazole. Similar results have been shown by some studies.[2,5,6,10] *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* exhibited lower sensitivities against these antibiotics as compared to *Escherichia coli*. These findings show that there is an increasing trend of acquisition of resistance by Gram negative organisms, mainly *K. pneumoniae* and *P. aeruginosa*.

CONCLUSION

We can conclude that gram negative bacteria are predominantly causing the BSI, mainly in adult population. *E. coli* is the main GNB causing the infection followed by *K. pneumoniae* and *P. aeruginosa.* Increasing trend of drug resistance is being noted by GNBs against most commonly used antibiotics like ampicillin, gentamicin, ceftriaxone, cefixime, ceftazidime, ciprofloxacin and cotrimoxazole. Imipenem, meropenem, amikacin, piperacillin-tazobactam and colistin can be used effectively against these GNBs causing BSI. Higher antibiotics, though seems to be a promising option, should be used judiciously.

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