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## RESEARCH ARTICLE

# MICROBIOLOGICAL PROFILE OF GRAM NEGATIVE BACILLI ISOLATED FROM BLOOD CULTURE AMONG HOSPITALIZED PATIENTS IN A TERTIARY CARE HOSPITAL (LAB BASED STUDY)

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#### **ABSTRACT**

**Introduction:** Blood stream infections are a major cause of morbidity and mortality despite the availability of antimicrobial therapy. Bacteremia due to Gram negative bacilli is a significant problem in hospitalized patients and frequent cause of sepsis.

**Aim:** The aim of this study was to identify the distribution of Gram negative bacterial isolates and their antibiogram from blood cultures.

Material and methods: A retrospective analysis was conducted from January 2014- June 2014 to identify the microbial profile in the blood culture isolates and their antibiotic susceptibility patterns.

Results: A total of 996 blood cultures were processed during the study period from which 317 Gram negative bacilli were isolated. Among the Gram negative bacteria, highest was Escherichia coli 101(32%) followed by Salmonella Typhi 73(23%), Klebsiella 48(15%), Salmonella Paratyphi A 25 (8%), Gram negative Non fermenter 42(13%), Pseudomonas species 16(5%), Enterobacter species 6(2%), Citrobacter species 3(1%), Aeromonas species 3 (1%). Antibiotic resistance pattern of the Non fermenters were totally (100% resistance) resistant to the all antibiotics, the Salmonella species isolated in our study were all susceptible to Ciprofloxacin and Ceftriaxone. The maximum resistance in Escherichia coli was to the Aminoglycoside group of antibiotics while Klebsiella showed maximum resistance to Cephalosporins.

**Conclusion:** Early and appropriate identification and treatment of gram negative sepsis is necessary for reducing the mortality and morbidity among the hospitalized patients.

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## INTRODUCTION

Blood stream infections are an important cause of mortality and morbidity and are common among the health-care associated and community acquired infections. The spectrum of illness associated with blood stream infection ranges from self-limiting infections to life-threatening sepsis, multiorgan failure to disseminated intra vascular coagulation that require rapid antimicrobial treatment. (Adugna *et al.*, 2015) The risk factors in hospital acquired infections in patients with the increasing use of central venous catheters and other pre disposing factors including intensive care unit (ICU) stay, lapses in hand washing and non-adherence to infection control practices by medical staff. Respiratory, genitourinary, and intra-abdominal foci are often identifiable sources of blood streaminfections (Yatin Mehta *et al.*, 2014). Rational and

correct use of the drugs requires understanding of common pathogensand their drug resistance pattern (Shobani *et al.*, 2004). In developing countries infections with Gram negativebacteria are more common in blood stream infections. Escherichia coli bacteremia is one of themost common blood stream infection (Cheol-In Kang *et al.*, 2005). This study was conducted to identify the bacteriologicalprofile and their antibiotic susceptibility patterns by analyzing the data on the blood cultureisolates at a tertiary care teaching hospital.

## Aim of the study

The aim of this study was to identify the distribution of Gram negative bacterial isolates and their antibiogram from blood cultures.

## **MATERIALS AND METHODS**

A retrospective analysis was conducted on all blood culture reports during the period from January 2014 to June 2014 in the Department of Microbiology.

**Study design:** This study was conducted as a retrospective cross sectional study

**Inclusion criteria:** Blood cultures from adult patients, were in single type of Gram negative bacterial growth wasidentified (first isolate per patient) were included.

**Exclusion criteria:** Blood cultures which yielded mixed growth were excluded.

Methodology: Blood cultures were done by BacT/Alert Microbial Detection System and isolate identification done by standard phenotypic methods. Antimicrobial susceptibility testing was performed for all isolates by Kirby-Bauer disc diffusion method as recommended in the Clinical and Laboratory Standards Institute guidelines (CLSI) (Clinical and laboratory standards Institute M100-S25, 2015). The antibiotics tested by disc diffusion in the following concentrations: Ampicillin (10 µg), Ciprofloxacin (5 µg), Gentamicin (10 µg), Amikacin (30 µg), Netlimycin (30 µg), Co-trimoxazole (1.25)Trimethoprim/23.75µg μg Sulfamethoxazole), Meropenem (10 μg), Piperacillin/ Tazobactam (100/10 µg). Additional anti-biotics Salmonella including Ceftriaxone (30 µg) was included.

**Statistical analysis:** All categorical variables have been expressed in numerical and percentages.

## **RESULTS**

During the study period from January 2014 to June 2014, a total of 996 blood cultures of adult patients of more than 18 years of age were studied of which 603 were culture positive. Among them 317 were Gram negative bacilli and 286 were Gram positive organisms. Of the 317, 206 were from males (65%) and 111 were from females subjects (35%). 174(55%) isolates were from the patients admitted in the ICU and 143(45%) were from the wards. Among the Gram negative bacteria, highest was Escherichia coli 101(32%) followed by SalmonellaTyphi 73(23%), Klebsiella 48(15%), Salmonella ParatyphiA 25 (8%), Gram negative Non fermenter 42(13%), Pseudomonas species 16(5%), Enterobacter species 6(2%), Citrobacter species 3(1%), Aeromonas species 3 (1%).

In our study Escherichia Coli showed resistance to Carbapenem (10%), Cephalosporins (36%), Aminoglycosides (39%), Fluoroquinolone (0%). Klebsiella species showed resistance to Carbapenem (15%), Cephalosporins (37%), Aminoglycosides (15%), Fluoroquinolone (0%). Non fermenters showed multi drug resistance.

5 - 10 ml of blood is collected in blood culture bottle

 $\downarrow$ 

Blood culture bottle is incubated in BacT Alert/Vitek 2 automated system

1

Automated machine signals when there is growth of microorganism

1

Microorganism were identified by Gram staining, culture and biochemical reactions

1

Antibiotic susceptibility was done by Kirby Bauer method

Table 1. Antibiotic resistance pattern among Gram negative isolates

Isolates	Carbapenems	Cephalosporins	Aminoglycosides	Fluoroquinolone
E.coli (32%)	10%	36%	39%	0%
Klebsiella species (23%)	15%	37%	15%	0%
NFGNB (13%)	100 %	100 %	100 %	100 %

Table 2. Antibiotic resistance pattern in Salmonella

Ciprofloxacin	Ceftriaxone	
0%	0%	

Table 3. Distribution of isolates

Isolates	Present study	Vanitha et al. (2012)	Waghmare et al. (2015)	Lakshmi et al. (2015)
Escherichia coli	32%	35.6%	18.8%	14.5%
Klebsiella	23%	14%	24.8%	11.2%
Non fermenters	13%	20.9%	14.5%	12.8%
Salmonella	23%	25.7%	-	3%

Table 4. Resistance pattern of Escherichia coli

Antibiotics	Present study	Vanitha et al. (2012)	Waghmare et al. (2015)	Lakshmi <i>et al.</i> (2015)
Carbapenems	10%	19.6%	4.5%	22.2%
Cephalosporins	36%	80.4%	36.4%	71.4%
Aminoglycosides	39%	1.63%	31.8%	11.1%
Fluroquinolones	25%	47.5%	40.9%	66.4%

Table 5. Resistance pattern of Klebsiella

Antibiotics	Present study	Vanitha et al. (2012)	Waghmare et al. (2015)	Lakshmi <i>et al.</i> (2015)
Carbapenems	15%	0%	0%	28.5%
Cephalosporins	37%	44.8%	55.2%	71.4%
Aminoglycosides	15%	20.8%	24.1%	28.5%
Fluroquinolones	27%	62.5%	31%	57.1%

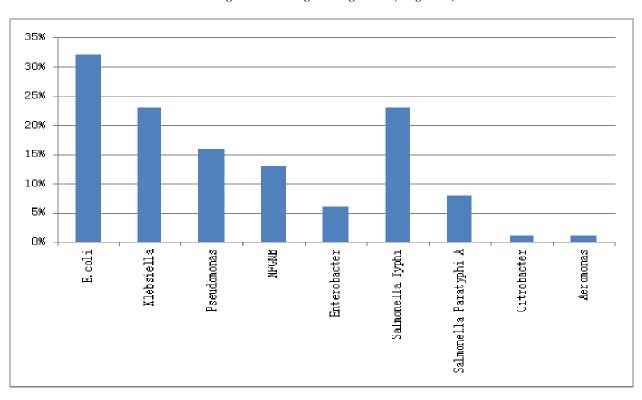
Table 6. Resistance pattern of non fermenters

Antibiotics	Present study	Vanitha et al. (2012)	Waghmare et al. (2015)	Lakshmi et al. (2015)
Carbapenems	100%	28.5%	11.1%	50%
Cephalosporins	100%	52.3%	55.6%	100%
Aminoglycosides	100%	38.9%	22.2%	100%
Fluroquinolones	100%	19%	44.4%	100%

Table 7. Resistance pattern of Salmonella

Antibiotics	Present study	Vanitha et al. (2012)	Garg et al. (2007)	Lakshmi <i>et al.</i> (2015)
Ciprofloxacin	0%	18.1%	7.6%	0%
Ceftriaxone	0%	0%	30.3%	33.3%

## Percentages of Gram negative organisms (Diagram 1)



The pattern of distribution of isolates of our study correlated with the study of Vanitha *et al*.

## **DISCUSSION**

Six months data of positive blood cultures at our clinical microbiology laboratory was reviewed to know the distribution of Gram negative bacteria and their antibiotic susceptibility pattern. Ofthe gram-negative bacillus isolates, the most common strains identified from the were Escherichiacoli 101(32%) followed by Salmonella Typhi 73(23%), Klebsiella 48(15%), Salmonella Paratyphi A 25 (8%), Gram negative Non fermenter 42(13%), Pseudomonas species 16(5%), Enterobacterspecies 6(2%), Citrobacter species 3(1%), Aeromonas species 3 (1%). In a study conducted by Garg et al. in the year Oct 2002- Sep 2004, among 2400 blood samples 493 yielded growth. Pseudomonas species were the most common organism isolated followed by and S. Paratyphi (14.2%), Acinetobacter species (2.6%), E.coli (11%), Klebsiella species (7.3%), Citrobacterspecies (5%). The distribution of isolates in other studies were also similar. (Table 3) Analysing the resistance patterns of the microbes, while the non fermenters were totally (100% resistance) resistant to all antibiotics. The salmonella species isolated in our study wereall susceptible to Ciprofloxacin and Ceftriaxone. The maximum resistance in Escherichia coli was to aminoglycoside group of antibiotics while Klebsiella showed maximum resistance to Cephalosporins. These patterns were in concordance with the other similar studies. The results of the antibiotic susceptibility testing could have been influenced by physical andchemical parameters like temperature, pH and ionic concentration.

#### Limitations

The study was a retrospective study and prior antibiotic use, risk factors and outcomes were not studied. MIC testing for the antibiotics among the resistant strains was not performed which is another limitation of this study.

## Conclusion

The burden of Gram negative bacterial infection is estimated to be very high in developing countries. Patient suffering from comorbidities requiring ICU admission are susceptible to thesemulti drug resistant organisms. This data helps us to generate an antibiotic policy for entericinfections and infections in ICU patients and other Gram negative infections.

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