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EPIDEMIOLOGICAL PROFILE OF VARIOUS BACTERIAL ISOLATES FROM BLOOD SAMPLE AND THEIR ANTIBIOTIC SUSCEPTIBILITY PATTERN AT A TERTIARY CARE HOSPITAL JAIPUR

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ABSTRACT

Background: Bloodstream Infection (BSI) is a significant and important caused of morbidity and mortality worldwide. Illness associated with the BSIs range from self limiting infection to life threatening sepsis that required rapid intervention and the choice of antimicrobial therapy for BSI is often empirical and based on local antimicrobial therapy of the most common aetiologies.

Objective: Epidemiological profile of various bacterial isolates from blood sample and their antibiotic susceptibility pattern at a tertiary care hospital.

Methodology: A total 680 samples were collected from suspected patient with bloodstream infection from various IPD and OPD wards of NIMS. Blood sample with positive growth by BACTEC automated blood culture system and processed to microbiology standard laboratory method and then antibiotic sensitivity test was performed by using CLSI guidelines.

Result: Out of 680 totals blood sample, 155(22.79%) were blood culture positive. Out of 155 positive sample 97(62.58%) were male while 58(37.42%) were female. Gram negative bacilli were 82(52.90%) while Gram positive cocci were 73(47.09%). Total isolates, the most predominant Gram negative organism was Klebsiella species 30(19.35%), while Gram positive cocci was MSSA (Staphylococcus aureus). Most of the Gram negative cocci were susceptible to Meropenem and Gram positive cocci susceptible to Vancomycin and Linezolid.

Conclusion: Gram negative organism are most predominate in bloodstream infection. Increasing in antibiotic resistance for BSIs causing of all pathogens has been necessitated continuous monitoring of susceptibility pattern of organism to prevent and spread of drug resistance.

KEYWORDS : MSSA- Methicillin Sensitive *Staphylococcus Aureus*, BSIs- Bloodstream infection, Indoor Patient department, Outdoor patient department.

INTRODUCTION

Bloodstream infections (BSI) are associated with significant morbidity and mortality. The condition can be life threatening in critically ill patients especially in intensive care units (ICUs) of the hospitals.[1]
The incidence of blood stream infection either community acquired or hospital acquired has dramatically increased.[2]
Blood is sterile, but bacteria occur transiently in the blood stream after dental surgery or instrumentation of the genitourinary tract or bowel they are infection in blood.[3]

Bacteremia refers to presence of viable bacteria in blood. Septicemia indicates systemic infection caused by bacteria and their toxins in the blood. BSI on the basis of time or setting of acquisition, can be community-acquired or nosocomial BSI. Usually community - acquired BSI is defined as a BSI that is detected within the first 48 hrs after hospitalization while BSI is referred to as nosocomial; if detected more than 48 hour after hospitalization.[4]

These infections require rapid identification or direct sensitivity test and also antibiotic susceptibility testing of the causative agent in order to facilitate specific antimicrobial therapy.[5]

The isolate of bacteriological cultures to the offending pathogens and knowledge about sensitivity pattern of the isolates remains the mainstay of definite diagnosis and management of septicemia.[6] Microbial incursion to the blood stream resulting from any organism can have serious quick consequences including shock, multiple organ failure, DIC(Disseminated Intravascular Coagulation) and death.[7-9]

Bacteria can enter in blood stream through many route but most common sites are genitourinary tract (25% cases), respiratory tract (20% cases) abscesses and miscellaneous sites (20% cases), wound infection (5% cases), uncertain sites (25% cases) and biliary tract (5% cases).[10]

Common organism isolated from blood culture are *Staphylococcus aureus, Escherichia coli, Coagulase Negative Staphylococci (CONS), Enterococcus species, Pseudomonas aeruginosa, Klebsiella pneumoniae, Proteus* species and *β-hemolytic streptococci. CONS* have long been considered mainly as non pathogenic blood culture or contaminant, however as a result of combination of increased use of intravascular devices and an increase in the number of hospitalized immune compromised patients, *CONS* has emerged as a major cause of nosocomial blood stream infection.[11-13]

Early detection of the infectious agent and determination of their susceptibility are essential for the optimization of treatment and represents the best way to reduce hospital stay and improve patient outcomes. Despite recent advances in clinical diagnostics, blood culture remains the gold standard for the detection of Bacteremia and fungal infection.[14]

MATERIAL AND METHOD

Study Design

Study was conducted in the department of microbiology in National institute of medical science Jaipur; Rajasthan from August 2018 to July 2019 to study the Epidemiological profile of various bacterial isolates from blood sample and their antibiotic susceptibility pattern at a tertiary care hospital Jaipur. A total of 680 patients suspected of blood stream infection attending different department such as IPD, OPD and ICU were collected and sent to microbiology department for processing.

Blood sample collection, organism identification and susceptibility testing:

Blood sample were collected after cleaning area of the venous site with 70% alcohol and subsequently followed by povidone iodine. Blood sample were collected from the patient before the administration of any antibiotic treatment. Blood sample was collected from adult(5-10ml) while 1-5ml blood was collected from pediatric patient, and 1-2ml from neonates for blood culture and immediately incubated into in BactAlert a fully automated blood culture system BACTEC culture system vials containing 70 ml to Brain Heart Infusion (BHI) broth with 0.05% Polyanethol Sulfonate (SPS) as anticoagulant (Soybean-casein digest broth with resins) and incubated in BACTEC blood culture system. Growth when detected by flag and an audible beep of the

system was subculture on 5% sheep blood containing blood agar and MacConkey agar plates and incubated 37° C overnight for bacterial isolation and identification with specific biochemical reaction. If no growth observed on plate by next day subculture repeated on day 3, day 4 and finally day 7.

Antimicrobial Susceptibility Testing:

Antimicrobial Sensitivity was determined by Kirby Bauer's disk diffusion method in Mueller Hinton agar (MHA) as per CLSI guidelines. Antibiotic used for sensitivity testing were Amikacin, Ampicillin, Cefotaxime, Cefoxitin, Ceftazidime, Ceftriaxone, Chloramphenicol, Cotrimoxazole, Erythromycin, Gentamycin, Imipenem, Meropenem, Linezolid, Penicillin-G, Tetracycline, Vancomycin, Cefepime, Levofloxacin, Aztreonam, Piperacillin-Tazobactam, Teicoplanin. *Staphylococcus aureus* (25923), *Escherichia coli* (25922), *Pseudomonas aeruginosa* (27853) were used as quality control throughout the study for culture and antimicrobial susceptibility testing.

RESULT

A total of 600 suspected patient were collected in present study among which 155 (22.79%) werr positive (fig.1). Out of total positive culture 97 (62.58%) were male while 58 (37.42%) were female (fig.2). The culture positive patient age ranged from 1 day to 90 years old patients. Among positive culture are 18 (11.61) isolate were from 1-15 years age, 44 (28.39) from 16-30 years age, 29 (18.71) from 31-45 years age, 26 (16.77) from 46-60 years age, 20 (12.90) from 61-75 years age, 18 (11.61) from 76-90 years age group patients respectively (fig.3). Gram negative organism were more than gram positive organism, Gram negative bacilli constituting about 82 (52.90) and Gram positive cocci were 73 (47.09) of total isolates (fig.4). Among the 155 isolates, the most predominant gram negative organism was *Klebsiella* species 30 (19.35), followed by *Escherichia coli* were 23 (14.84), *Pseudomonas aeruginosa* 15 (9.68) and where as gram positive organism was MSSA 26 (16.77), MRSA 19 (12.27), *CONS* 16 (10.32) (fig.5). Meropenem is more sensitive to other drug in gram negative bacilli, where as *Klebsiella spp*. is 93.33%, *E.coli* 82.6%, *Pseudomonas aeruginosa* 86.67%, and in gram positive cocci whereas, Vancomycin and Linezolid are more than sensitive to other drugs likely 100%, but *Enterococcus spp*. is intrinsic resistance so 25% are sensitive that is described in (fig.6-7 & table no. 1&2).

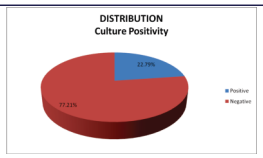

Fig.1 Distribution of Positive and Negative samples

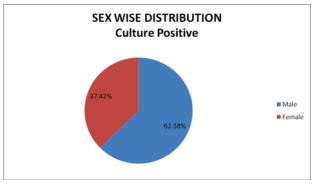

Fig.2:-Distribution of Male & Female

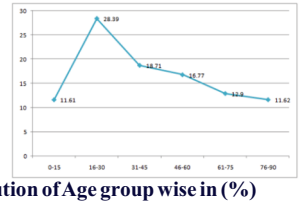

Fig. 3:- Distribution of Age group wise in (%)

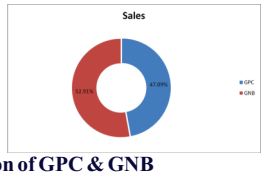

Fig.4:- Distribution of GPC & GNB

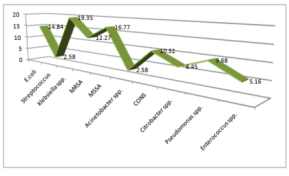

Fig.5:-Distribution of organism in (%)

Table no.1:- Gram negative bacilli sensitivity susceptibility Pattern

S.N.	ANTIBIOTIC	E.coli (14.84%)	Klebsiella spp.(19.35%)	Pseudomonas spp.(9.68%)	Citrobacter spp.(6.45%)	Acinetobacter spp.(2.58%)
1	Meropenem(10µg)	82.6	93.33	86.67	60	100
2	Gentamycin(10µg)	30.43	60	60	50	50
3	Tetracycline(30µg)	17.39	63.33	60	50	50
4	Cefotaxime(30µg)	8.69	50	26.66	40	25
5	Ampicillin(10µg)	4.35	46.66	40	40	50
6	Levofloxacin(5µg)	17.39	26.66	66.67	50	50
7	Piperacillin-Tezobactam (100/10µg)	56.52	63.33	66.67	70	75
8	Ceftazidime(30µg)	26.08	60	80	40	25
9	Amikacin(30µg)	39.13	60	46.67	60	50
10	Ciprofloxacin(5µg)	13.04	50	53.33	40	50
11	Aztreonam(30µg)	8.69	26.66	13.33	40	50
12	Imipenem(10µg)	78.26	86.66	86.67	80	75
13	Cefepime(30µg)	13.04	50	53.33	30	50
14	Chloramphenicol(30µg)	26.08	33.33	33.33	40	25

Fig.6:-Gram negative bacilli sensitivity susceptibility pattern

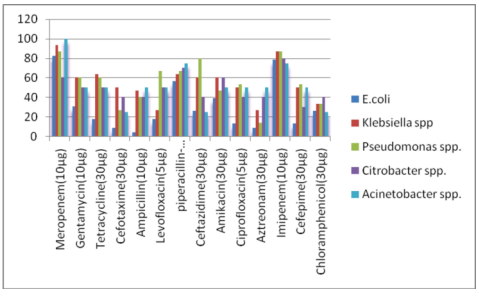

Fig.7:- Gram positive cocci Sensitivity susceptibility pattern

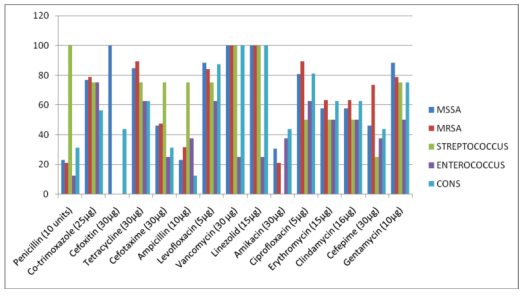

Table No.2:- Gram Positive cocci sensitivity susceptibility pattern

S.N.	ANTIBIOTIC	MSSA (16.77%)	MRSA (12.27%)	*STREPTOCOCCUS* (2.58%)	*ENTEROCOCCUS* (5.16%)	*CONS* (10.32%)
1	Penicillin(10 Unit)	23.07	21.05	100	12.5	31.25

2	Co-trimoxazole (25µg)	76.92	78.94	75	75	56.25
3	Cefoxitin(30µg)	100	0	ND	ND	43.75
4	Tetracycline(30µg)	84.61	89.48	75	62.5	62.5
5	Cefotaxime(30µg)	46.15	47.36	75	25	31.25
6	Ampicillin(10µg)	23.07	31.58	75	37.5	12.5
7	Levofloxacin(5µg)	88.46	84.21	75	62.5	87.5
8	Vancomycin(30µg)	100	100	100	25	100
9	Linezolid(15µg)	100	100	100	25	100
10	Amikacin(30µg)	30.77	21.05	ND	37.5	43.75
11	Ciprofloxacin(5µg)	80.77	89.47	50	62.5	81.25
12	Erythromycin(15µ)	57.69	63.16	50	50	62.5
13	Clindamycin(16µg)	57.69	63.16	50	50	62.5
14	Cefepime(30µg)	46.15	73.68	25	37.5	43.75
15	Gentamycin(10µg)	88.46	78.94	75	50	75

DISCUSSION:-

In this study an attempt was made to blood stream infection from transient Bacteremia to septic shock and it's a challenging problem and more often they may be life threatening. Therefore, timely detection, identification, and antimicrobial susceptibility testing of blood borne pathogens.

In our study the isolation rate are positive blood culture was 22.79 % which is similar to studies conducted by Anita Jhajhria *et al*[15] and Asifa Nazir *et al*.[16]Who reported a culture positive rate 18.75% and 25.3%. The low rate of isolation in one study and second study are high rate compare to our study that many of patient probably received antibiotic therapy before they come to the tertiary care hospital.[15-16]

In this study isolation rate are blood culture positive male is high 62.58% while 37.42% were female. The result consistence with the study done by Vanitha Rani *et al*.[17] Who reported high culture positive 60.2% in male and & 36.74% in female. A similar study was done by with Kour and Singh.[18]Who is reported 65.22% male.

In this study the highest prevalence rate was found in the age group of 16-30 years.28.39% which is in according to compared Aroop Mohanty *et al*.[19] Who is reported is 29%.

In this study we found that the rate of isolation Gram Negative bacilli was higher 52.90% than Gram positive cocci were 47.09% which is consistent with the studies conducted by Anbumani *et al*[20] Gram negative were 56% and Gram positive 46%.However in some studies are different as reported by Mehta M *et al* [21] 80.96% was Gram negative and 18% were Gram positive.

We found most common organism and higher than other Gram negative bacteria isolated from *Klebsiella* species 19.35%. This is found in all age group. This study was conduct by Sanjay D Rathor *et al*[22], Mustafa *et al*[23] and Anita Jhajhria.[15] And Gram positive cocci was *Staphylococcus aureus* 16.77% again similar result were reported from studies done by Ghashyam D. Kumar *et al* 24%,[24] Mehta *et al* 13.86%[21]and Anita Jhajhria 18.67%.[15]

In our study, *Coagulase Negative Staphylococci (CONS)* were reported as 10.32%. This is in agreement with studies conducted by Anita Jhajhria 16%[15], and R.Sharma *et al*. (13.3%).[25] However Meenakshi Kante *et al*.[26] had reported very low occurrence of *CONS* 7.1% & 5.6% respectively. This variation in occurrence of *CONS* as blood pathogen is due to fact that they are considered as most common skin commensal and their presence in blood may be result of contamination due to non follow of proper aseptic technique during collection of blood sample.[27-28]

Antibiotic sensitivity pattern of micro-organisms is always changing. In our observed Meropenem is more sensitivity to other drug in 93.33% cases of *Klebsiella spp*. sepsis but today it is no cases due to development of resistance to Meropenem. In Gram positive is more sensitive to *Streptococcus spp*. In this study, among the antibiotics used for susceptibility testing for gram positive isolates, Vancomycin (100%) & Linezolid (100%) showed highest activity.

This correlates with other studies conducted by Mehta M. *et al*.[21] Sharma M *et al*.[29] Atul G *et al*.[30] Mustafa M *et al*.[23]

In this study we found that among the Gram negative isolates Imipenem is highest sensitivity after Meropenem drug 86.66% which is consistent with the studies conducted by Sanjay D Rathod *et al*.[22] and Mustafa M *et al*.[23] *Klebsiella spp*. showed 86.66% sensitivity for Imipenem. Resistance to Ampicillin was 60% This high level of resistance to commonly used drugs is comparable with study conducted by Anumani *et al*.[20] Mustafa M *et al*.[23]and Anita Jhajhria[15] show a significantly high percentage of resistance among gram negative bacilli to Aztreonam, Ciprofloxacin, Cefepime, Levofloxacin but in comparison Imipenem and Meropenem was found to be more effective.

CONCLUSIONS:

Bloodstream infection is one of the main agent causing mortality and morbidity worldwide. The most predominate organism was *Klebsiella Spp*. followed by MSSA. Meropenem was most effective drug for Gram negative bacilli while Vancomycin & Linezolid were effective against Gram positive cocci. The resistance rate of an antibiotic for bloodstream pathogens is high, and in order to monitoring to susceptibility of the organism towards antibiotic has mandatory.

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