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BLOOD STREAM INFECTIONS CAUSED BY ESBL AND AMPC PRODUCING GRAM NEGATIVE BACILLI IN A TERTIARY CARE HOSPITAL

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ABSTRACT:

Objective: Knowledge of the spectrum of micro-organisms involved in bloodstream infections is needed to guide the selection of empirical antibiotics and their drug resistant mechanism. To find out the susceptibility patterns, prevalence of ESBL and AmpC β-lactamase producing gram-negative bacilli isolated from Blood samples obtained from IPD, OPD and ICU's of SMS and allied hospitals, Jaipur. **Material And Methods:** Total 872 Blood culture samples were received from different wards, intensive care units and outpatient departments of SMS Hospital, J.K.Lon Hospital, Mahila Chikitsalaya, Zanana Hospital and Mental hospital in the Department of Microbiology, SMS Medical College, Jaipur from May 2007 to December 2007. These strains (total no. 63) were tested for Antibiotic susceptibility testing, ESBL production and AmpC production as per CLSI guidelines. **Result:** The maximum isolated strains were Klebsiella pneumoniae, followed by Enterobacter spp., Escherichia coli, Acinetobacter spp etc. Most sensitive antibiotic was meropenam followed by cefoperazone + sulbactum and piperacillin + tazobactum. Most resistant antibiotic was ceftriaxone. The overall production of ESBL was 82.5% (M/C in Escherichia coli) and of AmpC β lactamase was 26.9% (M/C in Enterobacter spp.).**Conclusion:**As newer bacterial pathogens are originating with different mechanism of action on drugs, therefore training must be done to prevent spread of resistance mechanism. If laboratories continue to lag behind in detecting pathogens with the help of advanced techniques, new pathogens will spread resulting in increasing problems and increase cost for patients and institutions. To prevent the emergence of ESBL and AmpC producing strains a strategy is to be formed for effective control and treatment.

Keywords: Blood stream infection, ESBl, AmpC, antibiotic resistance.

INTRODUCTION:

Nosocomial Infections such as blood stream infection occur worldwide affecting both developed and developing countries, is a major causes of death, result in substantial morbidity, mortality and increased financial burden among hospitalized patients, also an important public health problem. The socio-economic impact, i.e., prolongation of hospitalization, mortality and cost, of these infections adversely affects patients and nation's economy. (1, 2)

Blood culture is the gold standard technique used for the diagnosis of Blood Stream Infections. Bacteriological culture is done to isolate and to know the prevalent antibiotic resistance pattern in the offending pathogens. It always remains the mainstay of definitive diagnosis and the management of Blood Stream Infections (3). Respiratory tract infection, genitourinary tract infection and intra-abdominal foci are often the bacterial sources which invade the blood stream. (3) Blood stream infections are one of the main and serious causes of mortality as well as morbidity in hospitalized patients, with mortality rates between 30-70% (5).

Gram negative bacterial infection in blood can result in septicaemia that finally leads to septic shock and the chances of mortality is much more with the involvement of highly virulent and polymicrobial etiology . That's why the presence of multidrug resistant organism leads to therapeutic failure along with longer duration of hospital stay, use of toxic and costly drugs and a higher mortality rate. The potential for antimicrobial resistance is also one of the considerations of the physicians when they select a regimen to treat the patients, especially those with bacteremia and septicaemia. Hence this study was done to know the pattern of the bacterial isolates in the blood and their antibiograms with the resistance mechanism, in a tertiary care hospital at Jaipur.

MATERIAL AND METHODS

Total 872 Blood culture samples were received from different wards, intensive care units and outpatient departments of SMS Hospital, J.K.Lon Hospital, Mahila Chikitsalaya, Zanana Hospital and Mental hospital in the Department of Microbiology, SMS Medical College, Jaipur from May 2007 to December 2007. Of these 63 strains i.e. gram negative bacilli were isolated and identified, were processed for Antibiotic susceptibility testing, using antibiotics (in µg) such as Amikacin (30), Gentamicin (10), Cefoperazone + Sulbactum (75/30), Piperacillin + Tazobactum (100/10), Cefuroxime (30), Cefpodoxime (10), Gatifloxacin (5), Cefixime (5), Cefepime (30), and Meropenam (10) as per CLSI guidelines. (6)

Phenotypic Confirmatory Disc Diffusion Test (PCDDT) for ESBL-

All the strains were subjected to confirmation for ESBL production using the PCDDT as given by the CLSI. (6) This test requires use of both cefotaxime (30 µg) and ceftazidime (30 mcg) discs alone and in combination with clavulanic Acid (30 µg/ 10 µg) applied onto a plate of Mueller Hinton Agar (MHA) inoculated with the test strain. An increase of ≥ 5mm in the zone diameter of the combination discs (ceftazidime + clavulanic Acid & cefotaxime + clavulanic Acid), in comparison to that seen around individual test antibiotic was considered to be a marker for ESBL production.

AmpC Disc Test (for confirmation of AmpC β-lactamase)-

On Muller Hinton Agar plate a lawn culture of *Escherichia coli* ATCC 25922 was prepared. A sterile disc (6 mm in diameter) was moistened with sterile saline and inoculated with several colonies of test organism, was placed besides a cefoxitin disc (almost touching) on the inoculated plate. The plate was incubated overnight at 37^{o} C aerobically. Flattening or indentation of cefoxitin inhibition zone near to the test disc indicated a positive test. A negative test had an undistorted zone.

RESULT

Of the 63 isolates K. pneumoniae was the most common organism found followed by Enterobacter spp., Escherichia coli, Acinetobacter spp etc. Most sensitive antibiotic was meropenam followed by cefoperazone + sulbactum and piperacillin + tazobactum. Most

resistant antibiotic was ceftriaxone. The overall production of ESBL was 82.5% (M/C in Escherichia coli) and of AmpC β lactamase was 26.9% (M/C in Enterobacter spp.).

Table 1: Percentage of organism isolated from Blood samples

Organism	Blood (%age)
Esch. coli	**15 (23.8%)**
K.pneumoniae	**22 (34.9%)**
Enterobacter spp.	**17 (26.9%)**
Citrobacter spp.	**1 (1.6%)**
Acinetobacter spp.	**6 (9.5%)**
Burkhodelia cepacia	**1 (1.6%)**
Proteus mirabilis	**1 (1.6%)**
TOTAL	**63(100%)**

Table 2: Antibiotic susceptibility pattern of isolated organism

Antibiotic	Sensitive		Resistant	
	Number	% age	Number	% age
Amikacin	39	61.9	24	38.1
Gentamycin	14	22.2	49	77.8
Cefoperazone + Sulbactum	55	87.3	8	12.7
Piperacillin + Tazobactum	55	87.3	8	12.7
Cefuroxime	7	11.1	56	88.9
Cefpodoxime	7	11.1	56	88.9
Gatifloxacin	45	71.4	18	28.6
Cefixime	3	4.8	60	95.2
Cefepime	26	41.3	37	58.7
Meropenam	60	95.2	3	4.8
Cefotaxime	22	34.9	41	65.1
Ceftriaxone	6	9.5	57	90.5
Ceftazidime	12	19.0	51	81.0

Table 3: Percentage of ESBL and AmpC producing organism isolated from Blood samples

Organism	Blood		
	Total	ESBL %	AmpC %
Esch. coli	15	93.3	**33.3**
K.pneumoniae	22	86.4	**13.6**
Enterobacter spp.	17	88.2	**35.3**
Citrobacter spp.	1	100	**100**
Acinetobacter spp.	6	33.3	**33.3**
Burkhodelia cepacia	1	0	**0**
Proteus mirabilis	1	100	**0**
TOTAL	**63 (100%)**	**82.5**	**26.9**

DISCUSSION

Most of the studies have found a definitive role of antimicrobial therapy in patients with blood stream infection as they are serious in nature and can threaten life of the patients, that's why rapid implication of antibiotics in the treatment of these infections can reduce the morbidity and mortality. (7) Rise in temperature with or without the associated symptoms may be useful in identifying the cause of these infections. Blood stream infection can be diagnosed through various methods but blood culture is found to be the most important or gold standard technique for identifying the offending microorganism with their antibiotic susceptibility profile.

We have reported a 7.23% blood culture positivity rate, while a study done in Kavre had slightly lower rate of blood culture positivity 6.9% (8) as well in a study done in Iran also showed a lower positivity rate of 5.6% (9) because of intermittent secretion of microorganism in blood or it can be due to low count or previous antibiotic therapy has been taken by the patient before blood culture. Slightly higher rate of 8.39% culture positive samples was found in a study done in south India (10) as well as in an Ethiopian study a higher rate of 8.8% blood culture positivity was found (11), in a study done in Nepal a positivity rate of 10.28% was found. (12) Studies which were conducted in Mangalore (13) & Delhi (14) and Pakistan (15, 16) showed markedly increased rates of blood culture positivity (of more than 20%).

In comparison, the blood culture positivity rate was found to be especially low in this study. The variation in the blood culture positivity can also be related to different factors such as previous h/o antibiotic consumption before blood sample is taken, the numbers of blood culture samples taken at a definitive point, volume of the blood taken and type of culture broth used, etc. (14)

Majority of strains were found to be resistant to antibiotics such as third generation cephalosporin's and aminoglycosides. The organisms were most sensitive to Meropenam. A very high rate of antibiotic resistance was found in this study. This is a very alarming development as the mortality is much higher with the ESBL and Amp C β-lactamase producing *Enterobacteriaceae* (17).

The Antimicrobial Availability Task Force of the Infectious Diseases Society of America listed various microorganism producing ESBL among Enterobacteriaceae in 2006 (most common being Klebsiella species and E. coli) as one of six problematic drug-resistant pathogens and suggested an urgent need for newer and more effective therapeutics. (18)

Therefore, strategies for the control of emergence of antimicrobial resistance applied to ESBL and Amp C β-lactamase producing organisms are to be made to stop the spread of infection such as (A) Optimal use of all antimicrobials in general, this will decrease resistance towards first and second-line antibiotics; thus decreasing the need to use more broad-spectrum antibiotics. (B) Rotational or cyclic antimicrobial use to reduce resistance to certain antibiotics. (C) Specific for ESBL and Amp C β-lactamase elective removal, control or restriction of antimicrobials or class of antimicrobials like removal, control and restriction of third generation cephalosporins and making use of available alternatives.

CONCLUSION

As newer bacterial pathogens are originating with different mechanism of action on drugs, therefore training must be done to prevent spread of resistance mechanism. If laboratories continue to lag behind in detecting pathogens with the help of advanced techniques, new pathogens will spread resulting in increasing problems and increase cost for patients and institutions. To prevent the emergence of ESBL and AmpC

producing strains a strategy is to be formed for effective control and treatment.

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