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PREVALENCE OF AMPC BETA LACTAMASE PRODUCING ORGANISMS AMONG ADMITTED CAUTI PATIENTS IN A TERTIARY CARE MEDICAL COLLEGE OF EASTERN INDIA.

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ABSTRACT AmpC Beta lactamases are class C or group I cephalosporinases that confer resistance to a wide variety of beta lactam antibiotics including cefoxitin, cephalosporins, aztreonam, and are poorly inhibited by beta lactamase inhibitors such as clavulanic acid. Several epidemiological studies have shown that AmpC enzyme producing bacteria are recovered from hospitalized patients after several days of admission to the hospital. Organisms producing these types of beta lactamases often go undetected and therefore have been responsible for several nosocomial outbreak. Hence a study was conducted to see the prevelance of Amp C beta lactamases mainly in CAUTI patients along with the detection method in a tertiary care hospital setup of eastern India among 100 Catheter associated urinary tract infection (CAUTI) cases. The prevalence rate of AmpC beta lactamases was found to be 78.33% by cefoxitin/ cefepime detection method. 81.4% of Ecoli strains and 75.75 % of klebsiellas were AmpC producers in our setup. Study reveals the prevalence of AmpC beta lactamase among CAUTI was relatively high hence antibiotic resistance is an important concern for clinicians treating patients with suspected or confirmed bacterial infections mainly in CAUTI .

KEYWORDS : Catheter associated urinary tract infection (CAUTI), AmpC Beta lactamases, cefoxitin, cefepime.

INTRODUCTION :

Extended-spectrum β-lactamases (ESBLs) and AmpC β-lactamases are of increasing clinical concern. The predominant mechanism for resistance to beta lactam antibiotics in Gram-negative bacteria is by the synthesis of beta lactamases [1]. Among the beta lactamases the production of ESBLs and AmpC beta lactamases are the most common [2]. AmpC Beta lactamases are class C or group I cephalosporinases that confer resistance to a wide variety of beta lactam antibiotics including cefoxitin, narrow and broad spectrum cephalosporins, aztreonam, and are poorly inhibited by beta lactamase inhibitors such as clavulanic acid. Chromosome-mediated AmpC β-lactamases have been described in a wide variety of Gram-negative bacilli, such as *Pseudomonas aeruginosa* and *Enterobacter spp., Acinetobacter spp., Aeromonas spp., C. freundii, E. coli, S. marcescens*, etc. Overproduction of their chromosomal AmpC beta lactamase by mutation is probably responsible for the resistance in these organisms [3,4]. Several epidemiological studies have shown that AmpC enzyme producing bacteria are recovered from hospitalized patients after several days of admission to the hospital. Affected patients have often had prolonged stay. A majority of patients had been treated with beta lactam antibiotics including cefoxitin[5]. Although it has been over a decade since plasmid-mediated AmpC β lactamases were discovered, most clinical laboratories and physicians remain unaware of their clinical importance[6]. As a result, organisms producing these types of beta lactamases often go undetected and therefore have been responsible for several nosocomial outbreak [4].Without accurate laboratory detection and reporting of such resistant phenotypes and strains producing plasmid-mediated AmpC, treatment of Gram negative bacterial infection may remain suboptimal. There is a paucity of information on the documentation of AmpC beta lactamases among Gram negative isolates in this region. Hence, this study was undertaken to determine the occurrence of AmpC beta lactamases in Gram negative clinical isolates at this centre to increase awareness and demonstrate the need to detect the AmpC enzymes in clinical strains.

MATERIALS AND METHODS:

This study was carried out in a urban, tertiary care hospital of eastern India in the year of 2017. A total of 100 patients with indwelling catheter for more than 3 days with suspected urinary tract infection , who did not receive any antibiotics were included in this study.

Urine sample was collected from the catheter in sterile wide mount container of 50 ml with a chemical preservative i.e. buffered boric acid. Urine sample was cultured on MacConkey's agar plate and incubation was done at 35º C for 48 hrs. Biochemical tests was performed for bacterial identification. Gram straining and motility testing was also done for the reconfirmation. Only gram negative bacilli were included in this study. Screening of AmpC Beta lactamase was done by using cefoxitin disc (30µg) on Muller Hinton Agar plate [7] . Zone diameter of < 18mm are suspected as AmpC β lactamase producer[7]. Inducible and non-inducible AmpC β lactamase producing isolates were detected using cefoxitin (30µg), cefotaxime (30µg), ceftazidime (30µg) with and without boronic acid (400µg) according to standard guidelines[8]. A >5 mm increase in zone diameter of antimicrobial tested in combination with boronic acid versus its zone when tested alone confirms AmpC β lactamases production. All the procedures were performed after getting approval from the institutional ethical committee.

RESULTS:

A total of 100 suspected CAUTI (Catheter associated urinary tract infection) patients were screened and 60 number of isolates were identified by conventional culture methods. Culture isolation rate was found to be 60% on MacConkey agar . 55 % were found to be Klebsiella followed by Escherichia coli 45% among the total isolates. Both isolated organisms were screened first for AmpC beta lactamases by using cefoxitin and cefepime disk of 30µg and 78.33% came positive however all the organisms were found to be real AmpC producers when retested using cefoxitin + boronic acid or ceftazadime + boronic acid or Cefotaxime + boronic acid. 81.4% of Ecoli and 75.75 % of klebsiella isolates were found to be AmpC producers in our set up and 21- 30 yrs of age group of patients (78.33 %) were found to be predominant AmpC producers followed by 31-40 yrs.

Table 1: Distribution Of Ampc Strains Among Bacterial Isolates:

Isolates	AmpC screening positive	AmpC confirmation positive
Escherichia coli n= 27	22	22
Klebsiella sp n=33	25	25
Total= 60	47	47

Table 2: Age Wise Distribution Of Ampc Isolates:

Age group (yrs)	Number of patients
0-10	1
11-20	3
21-30	19
31-40	8
41-50	5
51-60	6
61-70	3
71-80	2
Total	47

DISCUSSION AND CONCLUSION:

Despite the discovery of ESBLs and AmpC β-lactamases at least a

decade ago, there remains a low level of awareness of their importance and many clinical laboratories have problems in detecting ESBLs and AmpC β-lactamases. Confusion exists about the importance of these resistance mechanisms, optimal test methods, and appropriate reporting conventions. Failure to detect these enzymes has contributed to their uncontrolled spread and sometimes to therapeutic failures[9]. The objective of this work was to obtain some experimentally based prediction on the possible emergence of AmpC β-lactamases and the best detection method in a tertiary care teaching hospital of eastern India though detection and reporting isolates producing AmpC β lactamases are more difficult issues than those associated with ESBLs. *E. coli*, *Klebsiella pneumoniae* and *Proteus mirabilis* are the species in family *Enterobacteriaceae* most commonly isolated in the clinical laboratory[10]. In our study the prevalence of AmpC beta lactamase in catheter associated urinary tract infection (CAUTI) was observed from the hospitalized patients. Urine samples were taken from all suspected patients accordingly. A total of 100 suspected cases of UTI were selected during the study period of 2015-2017 and urine culture positivity was detected in 60 (n=60) cases. 47 no of cases 47 (n=47) cases were detected AmpC beta lactamase producers by cefoxitin and cefepime screening method . However, different study reports have shown that screening test positivity rate ranging between 40-60%. In our study there is high positivity rate of 78.33%. It has been reported that at present in India AmpC harbouring isolates are largely restricted to the hospitalized patients only[11]. In this study klebsiella spp and E. coli was isolated predominantly in CAUTI patients. In our study cefoxitin and cefepime were used as screening antibiotics. Among 33 isolates of klebsiella spp., 25 (75.75%) were AmpC producers and among 27 isolates of E. coli, 22 (81.48%) were AmpC producers by screening test however using cefoxitin + boronic acid or ceftazadime + boronic acid or Cefotaxime + boronic acid confirmation of AmpC strains were found to be same. Though there are scarcity of data but from North India, 20 % of *P. aeruginosa* (Delhi) and 20.7 % of Gram-negative organisms (Aligarh) and 47.8% *E.coli*, 17.3% *P. aeruginosa, 13% K. pneumoniae* (Kolkata) were reported as AmpC beta lactamase producers [12, 13,14]. From South Indian states, 24.1 % of *Klebsiella* spp. and 37.5 % of *E. coli* were AmpC producers from Chennai*;* In Karnataka, 3.3% of *E. coli*, 2.2 % of *K. pneumoniae*, 5 % of *C. freundii*, and 5.5 % *E. aerogenes* were found to harbour AmpC enzymes[15,16.] .In this study prevalence of AmpC beta lactamase among CAUTI was relatively high in comparison to other studies . This clearly indicates that awareness about the potential risk factors could help in early suspicion followed by early diagnosis of UTI. Antibiotic resistance was always an important concern for clinicians, treating patients with suspected or confirmed bacterial infections. So early detection of microorganism in urine culture should be considered as an indicator of CAUTI. In conclusion, this study has revealed the occurrence of AmpC _ -lactamase producing strains of Gram negative isolates for the first time in our region which need to be evaluated properly in near future.

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