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EXTENDED SPECTRUM BETA LACTAMASE PRODUCING UROPATHOGENIC ESCHERICHIA COLI AND THEIR ANTIMICROBIAL SUSCEPTIBILITY PATTERN IN A TERTIARY CARE HOSPITAL

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ABSTRACT Urinary tract infection (UTI) due to extended spectrum beta-lactamase (ESBL)-producing bacteria including *Escherichia coli* has become widespread and also there is a changing trend in the antibiotic susceptibility pattern to the conventional drugs used in the treatment of urinary tract infections due to the production of extended-spectrum beta-lactamases (ESBLs).This study was done to find the percentage of ESBL producing *Escherichia coli* and to study the antibiotic resistance profile of the ESBL and non-ESBL producing *Escherichia coli*. Urine samples were processed and identification of bacterial growth was confirmed by standard microbiological procedures. Antimicrobial susceptibility and ESBL detection were performed using Clinical and Laboratory Standards Institute (CLSI M100 2022) guidelines, ESBL detection was done by combined disc method using cefotaxime (30 ug) versus cefotaxime plus clavulanate (30+10 ug). Out of total 1238 urine samples 549 were found to be culture positive. *Escherichia coli* (36.2%) was the most frequently isolated uropathogen followed by *Klebsiella pneumoniae* (23.6%). Among the isolated *Escherichia coli* 44.2% were ESBL producers which were found highly resistant in comparison to non-ESBL producers.

KEYWORDS : Uropathogen, Escherichia coli, Antimicrobial susceptibility, Extended spectrum beta lactamases (ESBL)

INTRODUCTION:

Urinary Tract Infections (UTI) is one of the most common community-acquired as well as nosocomial infections [1], *Escherichia coli* being the common causative organism of urinary tract infections (UTI) [2]. Other organisms responsible are *Proteus, Pseudomonas, Salmonella, Staphylococcus saprophyticus, Enterococcus, Staphylococcus aureus* [3]. The introduction of antimicrobial therapy has contributed significantly to the management of UTIs. However the main problem with current antibiotic therapies is the rapid emergence of antimicrobial resistance in hospitals and the community [4].

Extended spectrum beta-lactamases (ESBL's) are enzymes that confer resistance to most common beta-lactam antibiotics such as Penicillins, Cephalosporins and Monobactams but not for Cephamycins and Carbapenems. Detection of ESBL producers from sample is important because it represents an epidemiologic marker of colonization and therefore there is potential for transfer of such organisms to other patients. The rapid increase of resistance to broad spectrum beta lactams among uropathogens has recently become a major problem globally. It leads to antibiotic ineffectiveness, increased severity of illness and cost of treatment. The serious increase in the prevalence of ESBL's worldwide creates a need for effective and easy to perform screening methods for detection [5].

MATERIAL AND METHODS:

The present prospective study was carried out from October 2022 to December 2022 in the Department of Microbiology of a tertiary care hospital. A total of 1238 urine samples were collected in sterile containers. The samples were processed on chromogenic (urochrome) media and the culture plates were incubated at 37°C for 18-24 hrs under aerobic conditions. Identification of bacterial growth was confirmed by standard microbiological and biochemical techniques.

Confirmed *Escherichia coli* identified phenotypically were further tested for antimicrobial susceptibility which was performed using Clinical and Laboratory Standards Institute (CLSI M100 2022) guidelines

Extended spectrum beta-lactamase confirmatory tests as per CLSI M100 2022, was done by using cefotaxime and cefotaxime plus clavulanic acid (30/10 mcg) discs on Muller-Hinton agar using double disc diffusion method. Organism was considered as ESBL producer if there was an increase of ≥5 mm increase in the zone diameter to increase of ≥5 mm in the zone diameter of cefotaxime/ clavulanic acid disc with respect to that of cefotaxime disc alone.

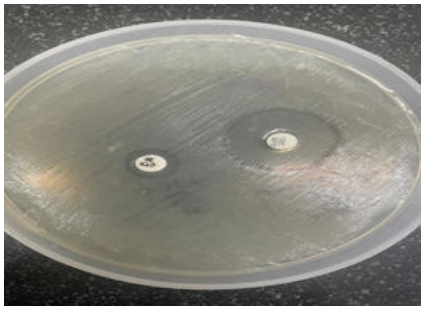

Figure 1: Phenotypic expression of extended spectrum beta-lactamase production (zone sizes of cefotaxime (CTX)/ clavulanic acid(CEC) ≥5 mm. than the zone sizes of cefotaxime

RESULT AND DISCUSSION:

A total of 1238 urine samples were collected from patients visiting the outpatient departments (OPD) and inpatient departments (IPD) of a tertiary care hospital out of them 638 were sterile and only 549 sample revealed uropathogens. Among them 376 (69.0%) isolates were Gram-negative, 128

(23%) were Gram-positive while 45 (9%) were identified as *Candida* species. Out of the total 549 uropathogenic isolates *Escherichia coli* (199,36.2%) was the predominant uropathogen, followed by *Klebsiella spp.* (130,23.6%), *Enterococcus spp.* (120,21.8%), *P. aeruginosa* (20,3.6%) and other pathogens like *Proteus spp, Acinetobacter baumanii, Enterobacter spp.* (26,4.84%) were also isolated (Table 1).

Among 199 strains of *Escherichia coli* isolate, 88 were shown to be ESBL producers.

In the current study, out of total 199 *Escherichia coli* 88 (44.2%) were ESBL producers (Table 2)which is similar to the studies done by Bajpai et *al,* Kumar et *al*,Taneja et *al and* Tankhiwala *et al*. (49.8%), (41.1%,39.0% and 40.2%)[5,6,7,8] but much less than the studies done by Ramesh *et al*. (60.7%), Singhal *et al*. (62%)[9,10].

Table 1: Distribution Of Organisms Isolated

ORGANISM ISOLATED	NUMBER	PERCENT-AGE(%)	ORGANISM	PERCEN-TAGE
GRAM NGATIVE BACILLI	357	65.0(%)	Escherichia coli	36.2%
			Klebsiella spp	23.6%
			Pseudomonas aeruginosa	3.6%
			Acinetobacter baumanii	3.4%
			Proteus spp	0.9%
			Enterobacter spp	0.54%
GRAM POSITIVE COCCI	128	23.0	Enterococcus spp	21.8%
			Staphylococcus aureus	1.45%
BUDDING YEAST CELL	45	9.8%	Candida spp	9.8%

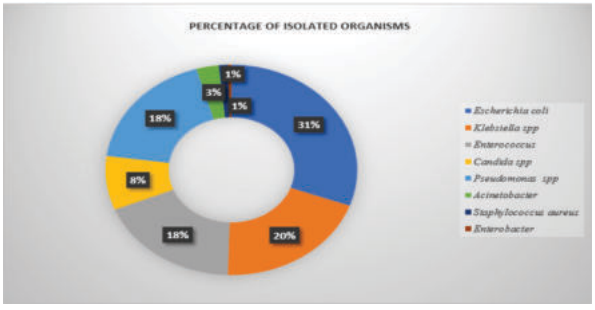

Figure 2: Distribution Of Organisms Isolated

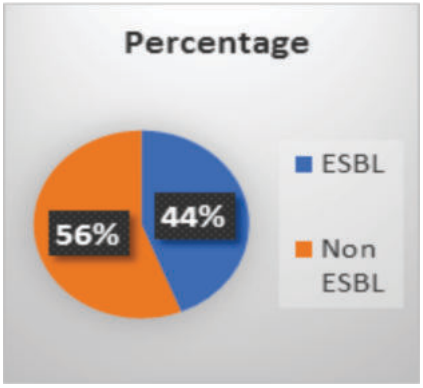

Figure 3: Disribution Of *Escherichia Coli* As Esbl And Non Esbl Producers

The antibiogram (Figure 3) of the *Escherichia coli* isolates shows that the ESBL producers have high degree of resistance not only towards penicillin and cephalosporin group of antibiotics but also towards other group of antimicrobials which are routinely prescribed for UTI's as compared to non-ESBL producers , These findings are similar to those reported by other authors like Bajpai *et al,* Sasirekha, Ndugulile *et al* and Mehrgan *et al* [5,11,12,13], this can be explained by the fact that ESBL are plasmid-coded enzymes, which are transferable between one bacterium to another and such transferable plasmids also code for resistance determinants to antimicrobial agents other than beta-lactams [14].

Table 2: Comparison Of Resistance Pattern Of Esbl And Non Esbl Producers

DRUGS	RESISTANCE PATTERN (%)		
	ESCHERICHIA COLI	ESBL PRODUCERS	NON ESBL PRODUCERS
Gentamicin	35.3%	29.4%	41.6%
Ciprofloxacin	83.1%	81.3%	68.7%
Norfloxacin	74.2%	84.8%	75.3%
Cotrimoxazole	53%	59.1%	43.0%
Imipenam	48.5%	39%	33.3%
Nitrofurantoin	6%	8%	3%
Cefipime	47.4%	58.9%	49.0%
Meropenem	38.4%	42.3%	24%
Ampicillin	90%	100%	85%

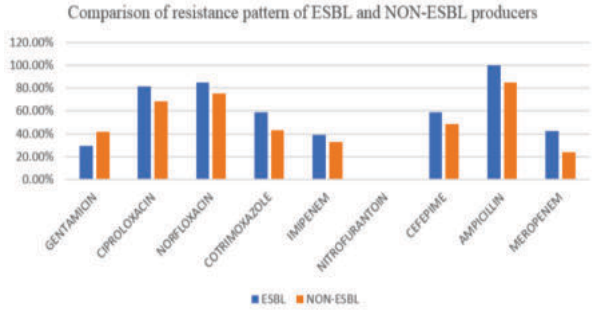

Figure 4: Comparison Of Resistance Pattern Of Esbl And Non Esbl Producers

Highest resistance in our study is seen with ampicillin 100% and 85% among ESBL and non ESBL producers it may be due their widespread use [5,15], high level of resistance is also seen with Norfloxacin (84%,75%) and Ciprofloxacin (81.3%,68.7) which may be due to an increased Quinolone resistance in our institute. This finding is consistent with previous studies [5,15,16]. These drugs are ubiquitously prescribed which may be the reason for the emergence of resistance against them [5].

In case of Gentamycin (29.4% and 41.6%) resistance was observed for both ESBL and non-ESBL producers respectively. In our study resistance to Imipenem and Meropenem is in range of 25%-45% but it is 100% sensitive in most of the previous studies [1,17,18,19] and 6.3% and 11% resistance is observed in the study of Bajpai et *al* and Rajeshwari et *al* respectively [5,3]. The resistance exhibited in our case is due to existence of carbapenemase producing isolates in our setting. The increasing resistance for Carbapenems is alarming and gives rise to an increasing concern over the judicious use of carbapenems in our health facilities[20,21].

In our study lowest degree of resistance is with Nitrofurantoin (8% and 3.7%) it can be attributed to its localized action on urinary tract and not being exposed outside urinary tract [15], and thus Nitrofurantoin is recommended as an appropriate agent for first line treatment of community-acquired UTI [14].

CONCLUSION:

Resistance to commonly used antibiotics is increasing day by day and it also vary over time and geographical distribution. Therefore, continuous monitoring of the antibiotic resistance pattern guides the clinician to initiate the empirical treatment

of UTI. Antimicrobial stewardship knowledge helps in avoiding the treatment failure and rapid dissemination of the antibiotic resistance and its mechanism can be prevented.

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