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**ORIGINAL ARTICLE****Antibacterial Susceptibility Pattern of Uropathogenic Enterobacter Species from a Tertiary Care Hospital**

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**Abstract:**

**Background:** Urinary tract infection (UTI) caused by Enterobacter species have been increased now a days. Emergence of antibiotic-resistance of these microorganisms are complicating the treatment. Since resistance rates differ from place to place, knowledge on prevailing patterns of antimicrobial resistance becomes essential. **Aim and Objectives:** This study was carried out to investigate the antimicrobial susceptibility pattern of Enterobacter species, an opportunistic pathogen isolated from patients with urinary tract infection at a tertiary care hospital. **Material and Methods:** Sixty eight strains of Enterobacter species were isolated from the culture media. Identification of the isolates was done using Matrix Assisted Laser Desorption Ionization – Time of Flight (MALDI-TOF) and antibacterial susceptibility testing was done using Vitex automated system. **Results:** We observed that, males were the most predominantly affected gender groups (57%) compared to that of females (43%). Out of 68 strains, *Enterobacter cloacae* (53) were identified as the most prevalent organisms followed by *Enterobacter aerogenes* (15). About 79% of Enterobacter species were found to be resistant to cefuroxime and 84% of the strains were found to be showing resistance to amoxicillin/clavulanic acid. One multidrug resistant strain susceptible to antibiotic colistin, was also identified. **Conclusion:** From our study, we can conclude that emergence of this unusually rare organism as uropathogen, which is resistant to commonly available antibiotics is alarming. Prompt starting of empirical antimicrobials based on susceptibility pattern is required to significantly reduce the mortality rate caused by these strains.

**Keywords:** Enterobacter, Antibiotic Susceptibility Testing, Urinary Tract Infections

**Introduction:**

Urinary Tract Infections (UTIs) are the most commonly encountered disease especially in the developing countries, annually affects at least 250 million people [1], with a significant financial burden in managing this condition [2]. Uncomplicated cases generally occur in pregnant women where as complicated cases may occur in all age groups eventually leading to serious long term complications like chronic kidney disease[3]. Gram negative organisms such as *Escherichia coli*, *Klebsiella pneumoniae*, Enterobacter species and Proteus species are the most common pathogens causing urinary tract infections. *Staphylococcus aureus*, *Staphylococcus saprophyticus*, *Enterococcus faecalis* are some of the gram positive cocci constantly reported as the causative agents of UTI [4]. Frequency of these pathogens causing disease depends upon gender, age, pregnancy; indwelling catheters and long term hospital stay [5]. During the last three decades, Enterobacter species, the enteric gram-negative bacilli have been reported as important nosocomial pathogens for human infections [3]. Enterobacter species such as *Enterobacter aerogenes* and *Enterobacter cloacae* have been found to be associated not only with urinary tract infections but

also cause meningitis, respiratory tract infections with significant mortality and morbidity [6].

Antimicrobial therapy is indicated in all complicated urinary tract infections. However, the emergence of antibiotic-resistance of these microorganisms is complicating the treatment. Research showed that the hyperproduction of ESBL and AmpC  $\beta$ -lactamase, encoded by both chromosomal and plasmid genes of *E. cloacae* are the key responsible factors for antibacterial resistance to ampicillin, amoxicillin, first-generation cephalosporins and cefoxitin [7-9]. Similarly, resistance to ampicillin, amoxicillin-clavulanic acid, cephalothin and cefoxitin of *E. aerogenes* and *E. cloacae* are due to the Cephalosporinase synthesis [10, 11].

The increased antibiotic resistance seen in recent years with these organisms suggest that the choice of antibiotic should be guided by culture and sensitivity assays. Monitoring of antibiotic resistance pattern is a rational way to reduce the risk of antibiotic treatment failure. Hence, the present study was aimed to determine the frequency and antibiotic resistance patterns of Enterobacter species isolated from urine samples collected from patients attending a tertiary care hospital in coastal Karnataka.

#### Material and Methods:

Institutional research committee approval was taken for the study. Sixty eight urine samples received in the Microbiology Laboratory from January to December 2016, having significant bacteriuria with Enterobacter species, were tested for antimicrobial activity. Urine samples were received from both inpatients and patients attending as outpatients to a tertiary care hospital. Age and gender of the patients were noted. Midstream urine samples were inoculated using a

standard calibrated wire loop into blood agar and MacConkey agar following semi quantitative technique using Kass concept. Streaking of the specimens was performed using four quadrant streaking method without intermittent heating, on blood agar and MacConkey agar. Organism with the growth of  $\geq 10^5$  colony forming unit per milliliter of the urine sample was considered significant. Matrix Assisted Laser Desorption Ionization – Time of Flight (MALDI-TOF) (Biomerio, ElToile, France) was used for the identification of the organism and Vitex automated system (Biomerio, ElToile, France) was used for antibacterial susceptibility testing. Trimethoprim-sulfamethoxazole (co-trimoxazole), amoxicillin/clavulanic acid, gentamicin, amikacin, cefuroxime, norfloxacin and imipenem were employed for antibiotic susceptibility testing. Organism showing resistance to multidrugs was then tested for colistin antibiotic susceptibility. *Escherichia coli* (ATCC 8739) was used as control organism.

#### Results:

A total of 68 strains of Enterobacter species were isolated from the culture plates with significant bacteriuria. Out of them, 53 (77.94%) strains were identified as *Enterobacter cloacae* and 15 (22.05%) as *Enterobacter aerogenes*. Thirty nine males and 29 females had significant growth of Enterobacter species in their urine samples respectively (Fig.1). This indicated that uropathogenic Enterobacter species causing significant growth in culture plates were more from male patients than in female patients.

Fig. 2 shows age wise distribution of UTI caused by Enterobacter species in our study group. Except for the age group 20-39 years, UTI was found to be more prevalent in males than in females. It was observed, higher incidence of UTI

cases in males after 60 years of age when compared to other age groups.

The antibiotic susceptibility results of Enterobacter species (Fig.3) revealed the susceptibility of 73.68% for Trimothoprim-sulphamethoxazole (co-trimoxazole), 91.22% for aminoglycosides and 92.98% for norfloxacin. Very high susceptibility

was observed to imipenem (98%). Organisms were found to be resistant to commonly used antibiotics like amoxicillin/clavulanic acid 84% and cefuroxime 79%. One multidrug resistant strain was isolated from a male patient, but it was susceptible to antibiotic colistin.

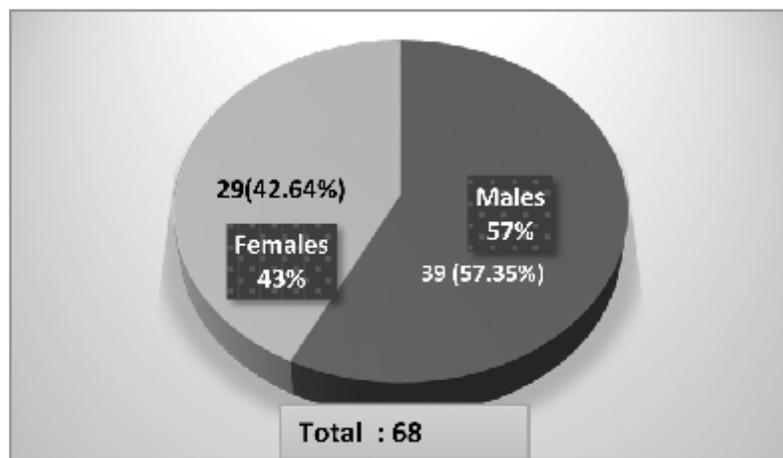


Fig. 1: Enterobacter Species Isolated in Different Genders

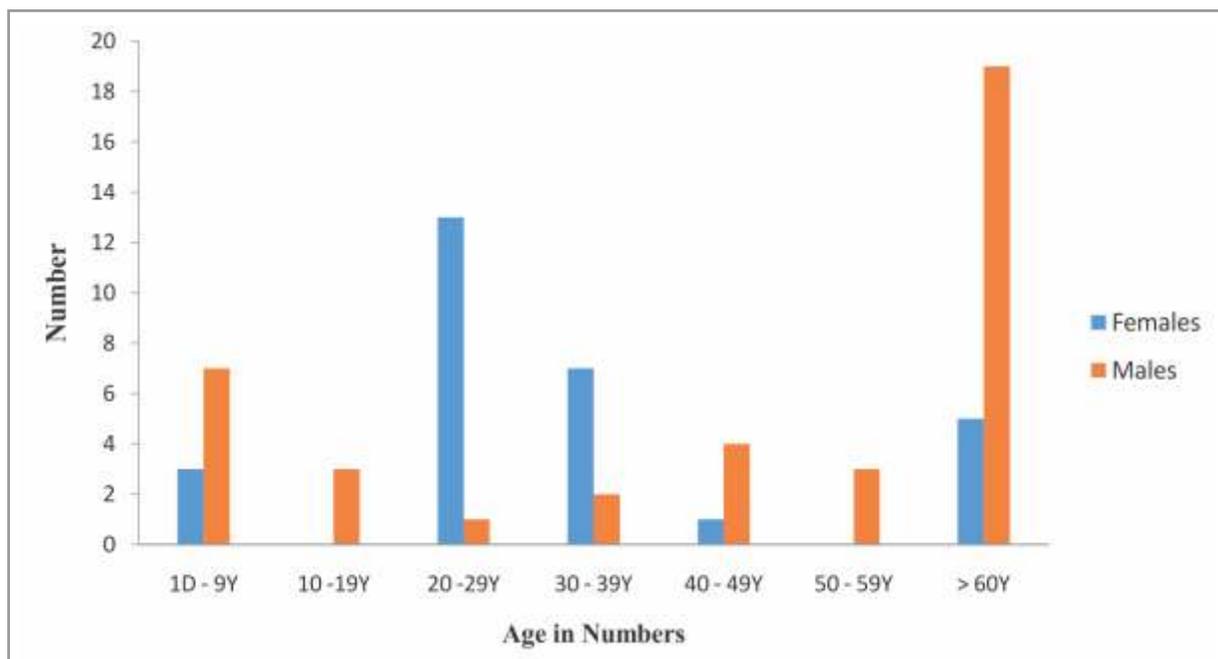
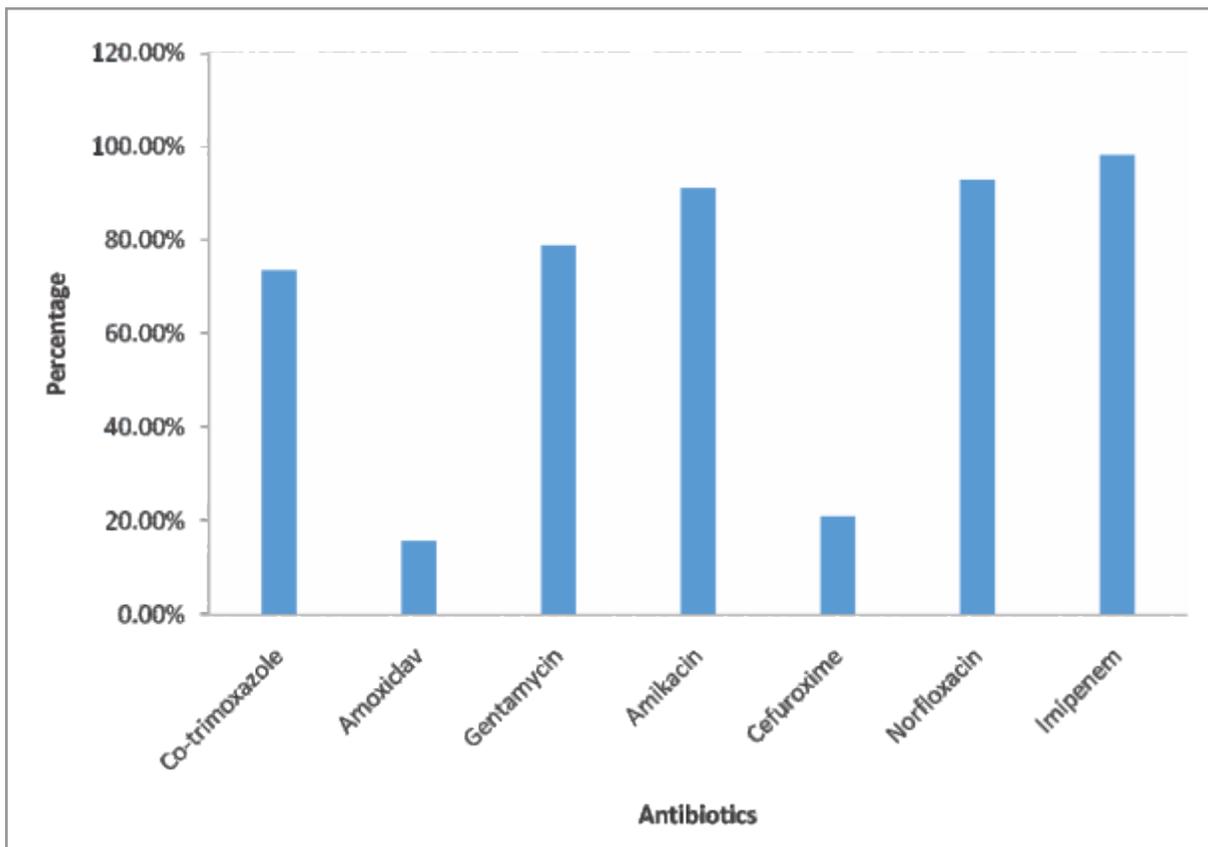


Fig. 2: Agewise Distribution of UTI Cohort



**Fig. 3: Antimicrobial Susceptibility Testing of Enterobacter Species**

### Discussion:

This study describes the relationship between sex, age and the antibacterial resistance pattern of isolated Enterobacter species from UTI. Significant by higher cases of bacteriuria was observed in males (57.35%) than in females (42.64%) and more number of males were infected at the age group of > 60 years. This study is comparable with the research findings of Stothers *et al.* [12] and Griebing *et al.* [13] on demographics and economic burden of male urinary incontinence in the United States of America. Both concluded that the incidence of UTI increases with age and UTI in men aged 65-74 years increases to 0.05 per person-year [12, 13]. Reason for the number of

males more than females in our study could be number of Enterobacter species isolates were few in number. Our study showed more number of UTI in females with the age group of 20-39 years. Our study was in concordance with the earlier study conducted [14].

Trimthoprim-sulfamethoxazole inhibits the folic acid production which is required for DNA synthesis in bacteria. This is used against a wide spectrum of bacteria including pathogens causing urinary tract infections [15]. In our study, we observed the antibiogram of Enterobacter species for trimothoprim-sulphamethoxazole susceptibility was 73.68% and for gentamycin 78.94%. A study

conducted by Mansour *et al.* [16] from Iran on the isolation and susceptibility of uropathogens showed Trimethoprim-sulphamethoxazole susceptibility of 66.7% to Enterobacter species. In a recent study, Maraki *et al.* [17] isolated 939 Enterobacter species for the period of six years (2010-2015). They have reported 83.3% sensitivity of these strains to Trimethoprim/sulfamethoxazole. Susceptibility of Enterobacter species was high for amikacin (91.22%), norfloxacin (92.98%) and imipenem (98%) in our study. Mansour *et al.* [15] also reported imipenem (susceptibility 96.1%) and gentamicin (susceptibility 95.7%) and were the two most active antimicrobial agents. In contrast to our findings, a similar study conducted in Asia - Pacific countries during the period 2013-2015, by Karlowsky *et al.* [18] on 154 isolates of Enterobacter spp., from urinary tract infections, reported resistance to amikacin (8.4%), and imipenem (6.5%). Thiolas *et al.* [19] also reported imipenem resistance in *Enterobacter aerogenes* strains.

Our study showed the resistance pattern of organisms to commonly used antibiotics like

amoxicillin/clavulanic acid (84%) and cefuroxime (79%). Researchers showed that production of ESBL and AmpC- $\beta$  lactamase [7-9] are the key factor for such resistance in Enterobacteriaceae group of organisms. The high rates of resistance to these antibiotics is concerning and our study indicated that empirical treatment with Amoxicillin/Clavulanic acid or Cefuroxime in suspected urinary tract infection caused by Enterobacter species may not work.

### Conclusion:

The emergence of Enterobacter species, usually the rare organisms as uropathogens resistant to commonly available antibiotics is alarming. Our study showed that amikacin, imipenem and norfloxacin can be the choice in the management of urinary tract infection. Amoxiclav and cefuroxime should be excluded from the empirical treatment for the UTI caused by Enterobacter strains. Since the drug resistance is common in these strains, periodic revisions in antibiotic policy should be promptly and strictly adapted to reduce the mortality and morbidity rates from urinary tract infections.

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