

## EMERGENCE OF MULTI DRUG RESISTANT STRAINS OF *E. COLI* ISOLATED FROM URINARY TRACT INFECTION IN NAMAKKAL

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**ABSTRACT:** In this study we evaluated the multi drug resistant strains of *Escherichia coli* isolated from urinary tract infection in different age groups in Namakkal. Totally 153 isolates of *E.coli* were obtained from 700 urine samples. Present study indicates that there is a high prevalence of multidrug resistant *E. coli* in female 24.34% followed by male 18.14%. After confirmation, isolates were performed to resistance patterns of different antibiotics were determined by standard disk diffusion method. The antibiogram patterns of the isolates showed a high percentage of multidrug resistance to cephodoxime, Novobiocin and vancomycin which was repeated to most of the isolates. Fifty resistance patterns were recognized among the MDR strains. This study revealed that *E. coli* was the predominant bacterial pathogen of community acquired UTIs in Aligarh, India. It also demonstrated an increasing resistance to number of antibiotics. This study is useful for clinician in order to improve the empirical treatment.

**Keywords:** E.coli, UTI, MDR, Age groups, Cephodoxime.

## INTRODUCTION

A UTI is an infection in the urinary tract. Infections are caused by microbes. UTIs are the most common of all bacterial infections and can occur at any time in the life of an individual. Urinary tract infections are the second most common type of infection in the body, accounting for about 8.1 million visits to health care providers each year (Schappert *et al.*, 2008). Women are especially prone to UTIs for anatomical reasons. One factor is that a woman's urethra is shorter, allowing bacteria quicker access to the bladder. Also, a woman's urethral opening is near sources of bacteria from the anus and vagina. For women, the lifetime risk of having a UTI is greater than 50 percent (Griebing *et al.*, 2007). UTIs in men are not as common as in women but can be serious when they occur. Almost 95% of cases of UTIs are caused by bacteria. Most UTIs are caused by bacteria that live in the bowel. The bacterium *Escherichia coli* (*E.coli*) cause the vast majority of UTIs (Abu Setta *et al.*, 2008).

Increasing resistance in bacterial pathogens is of world-wide concern. The prevalence of antimicrobial resistance in both out and hospital patients with UTI is increasing and can vary according to geographical and regional location (Asad, 2006). A remarkable increase in antibiotic resistance among the *E. coli* isolates has been observed during the last few years particularly in developing countries. Such rising resistance is due to mechanisms of mutation and then resistance gene transfer by transport means. Since a plasmid or transposon can carry several resistance indexes, resistance to several antimicrobial agents may be acquired simultaneously and results in multiple drug resistant (MDR) organisms (Shohreh *et al.*, 2010). Antimicrobial resistance surveillance is necessary to determine the size of problem and to guide empirical selection of antimicrobial agents for treating infected patients. The goal of this study was to determine the current prevalence and phenotype of multidrug resistant strains among UTI causing *E.coli* isolates against commonly used antimicrobial agents.

## MATERIALS AND METHODS

### Sample collection and isolation of *E.coli*

In this "prospective study" we collected 700 urine samples from local hospitals. The age of patients ranged between 0 to above 60 years old. Samples were collected in sterile plastic containers. Samples were placed on to the Mac Conkey agar plates. Pink colonies were observed. A well-isolated single colony was picked and incubated in nutrient broth, at 37° C for 12-16 hrs. then the inoculum was spread on Eosin methylene blue agar plates and incubated at 37°C for 24 hrs., green colonies having a metallic sheen were observed. A single colony was picked up from this and was then streaked on to the hard agar plates. These plates were then kept in the incubator at 37°C for overnight followed by characterized by battery of biochemical test.

### Antimicrobial resistance

Antimicrobial resistance was determined by the disk diffusion method. The following antimicrobial agents at the indicated concentrations were tested: Gen –Gentamycin (10mcg) CF-Ciprofloxacin (50mcg), A-Ampicillin (30mcg),E-Erythromycin (10mcg), CTX – Cephotaxime ( 30mcg), K –Kanamycin (30mcg), Nv-Novobiocin (30mcg), NF-Nitrofurantoin (300mcg), B-Bacitracin (10U), Co -co-trimaxazole (25mcg), NA -Nalidixic acid (30mcg), T –Tetracycline (30mcg), VA – Vancomycin (10mcg), CZX-Ceftizomine (10mcg),CPD-Cefpodoxime (10mcg). Throughout this study, results were interpreted according to zone of inhibition.

## RESULTS

### Identification of isolates

Identification and confirmation was done on the basis of morphological, biochemical and phenotypic characteristics. All isolates were found to be Gram negative rods. Biochemical tests confirmed that all the isolates were *E. coli*. Of the total collected samples, 153 (21.2%) *E. coli* strains were recovered and detected based on culture characterization. The higher incidences were showed in female 24.34% followed by male 18.14% (Fig.1). In female out off 6 types of age groups the highest prevalence in 21-30 (49.45%). In case of male the highest prevalence in above 60 years (20%) and followed by 41-60 (18.9%).

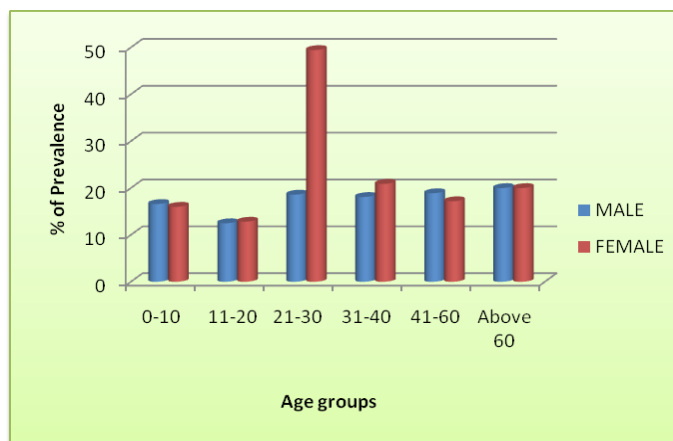
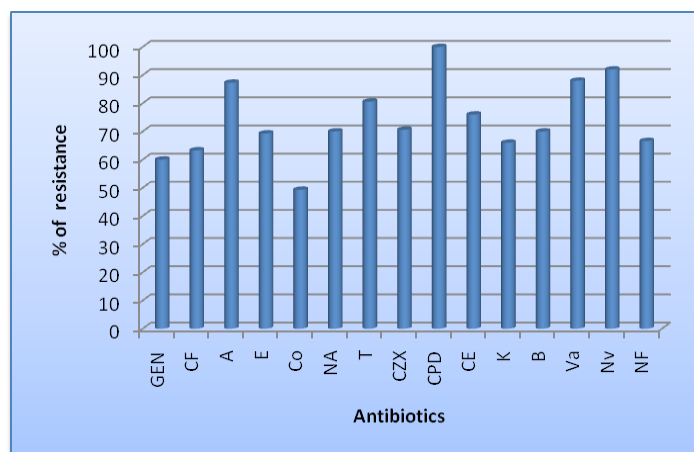
### Antimicrobial resistance patterns

Based on the antibiogram test results, the greatest resistance found in 14 (9.1%) isolates to all 15 antibiotics followed by 8 isolates (5.2%) to 14 antibiotics, 13 isolates (8.4%) to 13 antibiotics. Single antibiotic of cephalosporin was resistant to all isolate of *E.coli*. According to Table 1 those which were resistant to 6 or more antibiotics considered as MDR strains, in lowest pattern, 6 antibiotic resistances in 3.92% of isolates. Fifty resistance patterns were recognized among the MDR strains. In this study no one antibiotic sensitive to all isolates. The most frequent phenotypical pattern that was resistant to cephalosporin, Novobiocin, Vancomycin, Ampicillin, Tetracycline, cephotaxime and Bacitracin. Among the 15 antibiotics, lowest antibiotic resistance was Co-trimaxazole (49%) (Fig.2).

In case of female samples out of 6 age groups highest antibiotic resistance observed from 31-40 (83.7%) followed by 11-20 (77.3%) and above 60(75.8%). The lowest antibiotic resistance was showed in 0-10 age groups (69.9%). In male sample highest resistance were observed in 31-40 (78.8%) followed by 0-10 and 21-30 (76.6%) of age groups. The lowest resistance percentage wear in 41-60 age groups (65.6%). In our overall result highest resistance was obtained in 31-40 age groups peoples in both gender and compared to male female had highest percentage of resistance. In this study we observed 33.3% of isolates sensitive to gentamycin antibiotic.

Table-1: Antibiotic resistant pattern of *E. coli*

S.NO	ANTIBIOTICS NAME	TOTAL ISOLATES	ISOLATES PERCENTAGE
1	GEN,NA,CPD,K,VA,NV	3	1.96
2	A,E,NA,CPD,VA,NV	3	1.96
3	E,CZX,CPD,K,B,VA,NV	2	1.30
4	GEN,CE,K,B,CPD,VA,NV	2	1.30
5	A,CPD,CE,K,B,VA,NV,NF	2	1.30
6	NA,T,CZX,CPD,B,VA,NV,NF	2	1.30
7	CF,A,E,T,CPD,K,B,VA,NV	3	1.96
8	A,T,CPD,CE,K,B,VA,NV,NF	5	3.26
9	GEN,NA,CF,CZX,CE,K,CPD,VA,NV	3	1.96
10	A,NA,T,CPD,K,B,VA,NV,NF	3	1.96
11	A,T,CZX,CPD,K,B,VA,NV,NF	2	1.30
12	A,CO,T,CPD,CE,B,VA,NV,NF	2	1.30
13	A,E,T,CZX,CPD,K,B,VA,NV	2	1.30
14	A,NA,CPD,CE,K,B,VA,NV,NF	2	1.30
15	GEN,A,T,CPD,CE,K,B,VA,NV	3	1.96
16	CF,A,E,T,CPD,K,B,VA,NV,NF	3	1.96
17	CF,A,E,CO,NA,T,CZX,CPD,CE,VA	3	1.96
18	GEN,A,E,NA,T,CPD,CE,K,B,VA	3	1.96
19	A,E,CO,T,CZX,CPD,CE,VA,B,NV	2	1.30
20	A,T,CZX,CPD,CE,K,B,VA,NV,NF	3	1.96
21	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE	4	2.61
22	CF,A,E,CO,NA,T,CZX,CPD,CE,VA,NV	3	1.96
23	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE,NF	3	1.96
24	A,E,NA,T,CPD,CE,K,B,VA,NV,NF	3	1.96
25	CF,A,E,T,CPD,CE,K,B,VA,NV,NF	2	1.30
26	CF,A,E,NA,T,CZX,CPD,K,B,VA,NV,NF	2	1.30
27	GEN,CF,A,CO,NA,CZX,CPD,CE,NF,K,B,VA,NV	3	1.96
28	GEN,CF,A,T,CPD,CE,NF,K,B,VA,NV	3	1.96
29	E,CO,NA,T,CZX,CPD,K,B,VA,NV,NF	2	1.30
30	GEN,CF,A,E,CO,T,NA,CZX,CPD,CE,NV	9	5.88
31	A,E,CO,T,CZX,CPD,CE,K,VA,B,NV,NF	4	2.61
32	GEN,CF,A,E,CO,T,NA,CZX,CPD,CE,VA,NV	3	1.96
33	GEN,CF,A,T,CZX,CPD,CE,K,B,VA,NV,NF	3	1.96
34	GEN,CF,A,E,CO,NA,T,CZX,CPD,VA,NV,NF	3	1.96
35	CF,A,E,NA,CZX,CPD,CE,VA,NV,K,B,NF	4	2.61
36	GEN,A,E,NA,CZX,CPD,CE,K,VA,B,NF,NV	3	1.96
37	GEN,A,E,NA,T,CZX,CPD,CE,NF,K,B,VA,NV	1	0.65
38	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE,B,NV	3	1.96
39	GEN,CF,NA,T,CZX,CPD,CE,K,B,VA,NV,NF	2	1.30
40	GEN,A,NA,T,CZX,CPD,CE,NF,K,B,VA,NV	2	1.30
41	A,CO,NA,T,CZX,CPD,CE,K,VA,B,NV,NF	2	1.30
42	GEN,CF,E,CO,T,CZX,CPD,CE,K,B,VA,NV	2	1.30
43	CF,A,E,CO,T,CZX,CPD,CE,K,B,VA,NV,NF	2	1.30
44	GEN,A,E,CO,T,CZX,CPD,CE,K,B,VA,NV,NF	2	1.30
45	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE,VA,NV,NF	6	3.92
46	GEN,CF,A,E,NA,T,CZX,CPD,CE,K,VA,NV,NF	3	1.96
47	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE,B,VA,NV,NF	3	1.96
48	GEN,CF,A,E,NA,T,CZX,CPD,CE,B,VA,K,NV,NF	3	1.96
49	GEN,CF,A,E,CO,NA,CZX,CPD,CE,K,B,VA,NV,NF	2	1.30
50	GEN,CF,A,E,CO,NA,T,CZX,CPD,CE,K,B,VA,NV,NF	14	9.15

Fig- 1 Prevalence of *E.coli* in UTIFig- 2 Multidrug resistance of *E.coli*

## DISCUSSION

UTI are also the most common infection in acute and long term care hospital patients. *Escherichia coli* are the most frequently found bacteria in both communities acquired and hospitalized UTI patients (Asad, 2006). A totally 153 *E. coli* strains were recovered from 700 urine samples. *E. coli* are the most common cause of urinary tract infections (UTIs) in women and because of its high incidence, have been studied during numerous epidemiological studies (Nasreen, 2006). In our current studies highest incidence were observed in female (24.34%) urine samples followed by male (18.14%). Among the 6 types of age groups, the highest prevalence was 21-30 (49.45%).

Similar observations were made by Obi *et al.*, 1996; Shahab *et al.*, 1997. Even higher incidence of UTI causing *E.coli* was observed in young women who were sexually active of age group 20-29 years (Thomas *et al.*, 1996). Our reports correlated with the above findings. Many investigators in India also showed that women were the usual victims of the urinary pathogens (Asad, 2006; Nasreen, 2006; Nancy, 2012). In case of male the highest incidence was occur in above 60 years age groups (20%). There was a steady raise in the cases of UTI's among men with the advanced age due to obstructive complications like enlarged prostate (Dromigny *et al.*, 2002). In this present study 16.3% of isolates observed between 0-10 age groups. This could be because of wet bedding, unhygienic condition and toilet training problems. These UT infections result in significant financial and personal costs for both male and female patients and the health care system (Nancy *et al.*, 2012).

The present study was conducted to achieve resistance profile of urine isolates from our local area against commonly prescribed antibiotics. Our results show very high degree of resistance to almost all antibiotics as compared to previously reported studies. All the isolates were resistant to at least six drugs. Resistance was very high against cephodoxime (100%), Novobiocin (92.1%), Vancomycin (88.2%), Ampicillin (87.5%), Tetracycline (81.0%), cephotaxime (75.81%) and Bacitracin (70.5%), among the 15 antibiotics lowest antibiotic resistance was Co-trimoxazole (49%). In recent study of Nancy *et al.*, (2012) observed hundred percentage of cephotaxime resistance isolates. Our result was increasing incidence compared from Kausar *et al.*, (2009) observation. Who was obtained 40.5% of cephotaxime resistance isolates from UTI.

The greatest resistance in the present study was to ampicillin and tetracycline which were introduced by WHO in 2007 as the drugs of choice. Reports from developed and developing nations such as Turkey, Senegal, Brazil, Slovenia, southern India and Australia are consistent with the present findings. The growing antimicrobial resistance may be due to unrational use of antibiotics and the transfer of resistance genes by transport means including antibiotic resistant plasmids, bacterio-phages, transposons and integrons (Shohreh *et al.*, 2010).

In the present study when the data were visualized in different age groups for both genders, it appeared that in the case of males *E.coli* isolates were highly resistance to Co-trimoxazole in age groups 31-40, while in age groups 11-20 low percent resistance was recorded. *E.coli* in females isolates, the highest resistance in age groups above 60, while it showed 40% resistance in age groups 11-20. Such varying pattern of resistance also appeared in different age and gender groups to other antibiotics in this study. These findings are difficult to explain. McLoughlin and Joseph have indicated that prevalence of resistant organisms vary by geography, patient age and hospitalization status. In previous studies from India, Resistance patterns were not characterized according to age, so no local data is available for comparison. However, in a study reported from Spain, age more than 64 was found to be a risk factor, along with other factors, for higher antibiotic resistance against *E. coli*.

The resistant pattern of uropathogenic *E.coli* to common antimicrobial agent is changing in India and must be taken into account while selecting treatment strategies. Therefore antibiotic policy should be available according to local surveillance data for UTI and there is also a need to improve on infection control methods.

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