Antibiotics susceptibility pattern and prevalence of isolated uropathogens in inpatient and out patients with lower urinary tract infections

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ABSTRACT
Bacterial resistance was the consequence of empirical therapy and poor compliance, and its amelioration was difficult. In both in-patients and out-patients, definitive therapy resulted in better outcomes for urinary tract infections (UTI). Our objective was to identify the prevalence and to assess the susceptibility pattern of uropathogens toward antibiotics. It is a prospective, cross-sectional study conducted during the period of November, 01, 2019 to April, 28, 2020. A total of 200 UTI patient’s antibiotic susceptibility pattern was analyzed of which, 105 were males and 95 were females. Among them, 147 were in-patients and 53 were out-patients. The prevalence of UTI was high between the ages of 40 and 79 years. Less than 20% susceptibility was noticed in 20–39 years of age groups and less than 10% were observed in 80–99 years of age groups. The antibiotic susceptibility was found to be 35%–40% among the 40–59 years of age group, of them
*Ertapenem and Trimethoprime (TMP) & Sulfamethoxazole (SMZ) only has 48.5% and 44.6% of susceptibility and literally poor in 80–99 years. Colistin and Amikacin showed higher susceptibility toward *Escherichia coli* and *Klebsiella pneumoniae* in in-patients. The remaining antibiotics showed 40%–67% of susceptibility. TMP and SMZ, Amoxicillin Clavulanic acid, Cefoperazone, and Amoxicillin plus Clavulanic acid had relatively low susceptibility (16%–27%). Colistin, Amikacin, and Nitrofurantoin showed better susceptibility in in-patients and in out-patients. TMP and SMZ, Amoxicillin Clavulanic acid, Cefoperazone, and Amoxicillin plus Clavulanic acid had relatively low susceptibility. *Escherichia coli* was more prevalent Gram-negative bacteria isolated among the individuals and 57.8% of In-patients (IP) and 67.9% of out-patients (OP) with UTI were isolated with *E. coli*.

INTRODUCTION
Urinary tract infection (UTI) is a common health problem in both community and nosocomial settings, affecting both men and women equally. It is characterized by uropathogens colonization in the urinary tract, resulting in pyouria, dysuria, and urgency (Sarwar *et al.*, 2013). Worldwide, approximately 6 million patients are visiting outpatient clinics each year and 3 million being treated in wards (Akortha *et al.*, 2008; Medina and Castillo-Pino, 2019).

UTI episodes affect 50%–60% of women at some point in their lives (American College of Obstetricians and Gynecologists, 2008; Langer *et al.*, 2021). The dreadful sequels of UTI are pyelonephritis, renal scarring, and renal failure (Foxman, 2010). Gram-negative bacteria such as *Escherichia coli* (*E. coli*), *Proteus* species, *Pseudomonas aeruginosa* (P. aeruginosa), *Acinetobacter* species, *Klebsiella* species, *Enterobacter* species, and *Citrobacter* species are commonly responsible for UTIs. *Staphylococcus saprophyticus*, *Enterococcus* species, and Coagulase-negative bacteria are Gram-positive bacteria, among them, *Staphylococcus* is traditional type of bacteria that cause infections (Momoh *et al.*, 2011). Nearly 75% of uncomplicated UTIs are caused by *E. coli* (Flores *et al.*, 2015).

Antibiotics are the mainstay of UTI treatment. Emerging antimicrobial resistance is the major issue in treating UTIs. However, with increasing reports of bacterial resistance to antibiotics, treating UTIs has become difficult (Sharef *et al.*, 2015). Lack of hygiene, noncompliance with dose, duration of prescribed antibiotic(s), and unjustified prescription writing
by quacks and doctors are all factors that have been shown to influence bacterial resistance. Furthermore, the easy availability of antibiotics as over-the-counter medication will contribute to the development of resistance (Kidwai et al., 2017). Several national and international surveillance systems have been developed to track the susceptibilities of clinically significant uropathogens causing infections (Hertz et al., 2016; Qiao et al., 2013). Another source of concern is the emergence of beta-lactamase producing Gram negative bacteria, such as E. coli and Klebsiella species, that are multirdrug resistant not only to all Cephalosporin generations, but also to fluoroquinolones and beta lactam inhibitor/lactamase inhibitor (Piperacillin/Tazobactam) combinations, leaving only Carbapenem as a treatment option (Mazzulli, 2012).

The purpose of this study was to determine the prevalence and antibiotic sensitivity pattern of the most common uropathogens isolated in both in-patients and out-patients with UTIs who visited the hospital. Several studies have been carried out in India; however, as a part of the continuous surveillance program, antibiotic resistance would need to be treated on a regular basis in order to change the guidelines accordingly.

MATERIALS AND METHODS

Study design

A single center, prospective and cross-sectional study conducted in patients with UTI was referred in the Central Laboratory of Ramesh Hospital, India, to analyze urine cultures from the age more than 18 years were collected in the inpatient and outpatient setting during the period of November, 01, 2019 to April, 28, 2020 (6 months). The mid-stream urine was collected in a suitable sterile container under hygienic conditions. The samples were incubated at 37°C, and then it was investigated after 24–18 hours.

Study population

Inclusion criteria

The data were collected from the patients those who are diagnosed with UTI (both inpatients and out patients) with either sex of age more than 18 years and willing to give informed consent to participate in the study were recruited.

Exclusion criteria

The patients were excluded in the study with the criteria such as, patients with previous history of antibiotic resistance, indiscriminate use of antibiotics such as (use of antibiotics without the supervision of registered medical practitioner or violating his/her direction of use and non-adherent to prescription and use of antibiotics as self medications), immune compromised patients, pyelo-nephritis, known history of drug resistant uropathogens and recurring UTIs, and the patients those who are unwilling to participate are excluded.

Antibiotic susceptibility test

In vitro antibiotic susceptibility tests on urinary pathogens were performed using the National Committee for Clinical Laboratory Standards’ basic agar disc diffusion technique. For the procedure, 11 antibiotics were used, such as Colistin, Amikacin, Imipenem, Meropenem, Gentamicin, Ertapenem, Nitrofurantoin, Cefoperazone and sulbactam, Cefoperazone, Trimethoprim-Sulfamethoxazole (TMP-SMX), and Amoxicillin Clavulanic acid. An automated (disc diffusion technique) for antimicrobial susceptibility testing systems was used for our study.

Statistical analysis

For categorical data, summary statistics were calculated using the frequency and proportion. The Chi-square test was applied to compare difference in uropathogen prevalence and resistance trends between outpatient and inpatient urinary isolates. The p value with <0.05 was considered to be statistically significant. The Statistical Package for the Social Sciences, IBM and version 21 was used for this study to perform the statistics.

RESULTS

In our study (Table 1), UTIs are high in in-patients (55.78%) of males and out-patients females were high (55.77%). There was no statistical gender difference among OP and IP patients (p > 0.05). Table 2 represents the distribution of Gram negative and Gram positive uropathogens among the in and out patients. A total of 121 E. coli (60.5%), 32 (16%) of K. pneumoniae and 18 (9%) were P. aeruginosa were observed among the both IP and OP. A 147 IP and 53 OP patients were recruited in our study. Among them, in-patients 85 (57.8%) were isolated with E. coli and 36 (67.9%) were observed in out-patients. A very smaller number of Gram positive isolates were observed in our study, and the distribution of isolates are statistically significant among IP and OP patients (p < 0.05). Table 3 depicts distribution of Gram-negative uropathogens. A total 181 Gram-negative uropathogens

| Gender | In-patients | Out-patients | p. value |
|--------|-------------|--------------|----------|
| Females | N = 147 (%) | N = 53(%) | 0.1232 |
| N = 95 | 65 (44.22) | 30 (55.77) | |
| Males | N = 105 | 82 (55.78) | 23 (44.23) | |

The p value <0.05 were considered statistically significant.
isolates were observed in our study, of which *E. coli* was the most prevalent bacteria among all the age groups. Fifty-eight isolates (48%) of *E. coli* were observed in the age range of 60–79 years and 45 (35%) was observed in 40–59 years of age. A 32 of *K. pneumoniae*, 18 of *P. aeruginosa*, and 10 of *Staphylococcus aureus* were isolated in our study. All Gram-negative bacteria are significant among all the age groups (*p* < 0.05).

Table 4 and Figure 1 indicates the overall antibiotic(s) susceptibility pattern among the different age groups. The antibiotic(s) susceptibility was less than 20% was noticed in 20–39 years of age groups and less than 10% were observed in 80–99 years of age groups. The antibiotic susceptibility was found to be 35%–40% among the 40–59 years of age group, of them Ertapenem and TMP & SMX only has 48.5% and 44.6% of susceptibility. The antibiotic(s) susceptibility was increased (42%–50%) in 60–79 age groups and toward Ertapenem it was reduced to 26.4%. The antibiotic(s) susceptibility was literally poor in 80–99 years. Table 5 and Figure 2 represents the percentage of susceptibility of antibiotic(s) toward the Gram-negative bacteria. The greatest susceptibility of *E. coli* and *K. pneumoniae* toward Colistin (*E. coli*-96.7%, *K. pneumoniae*-100%), followed by Amikacin (*E. coli*-77.7%, *K. pneumoniae*-65.6%), Imipenem (*E. coli*-66.1%, *K. pneumoniae*-53.1%), and Meropenem (*E. coli*-64.5%, *K. pneumoniae*-46.9%). A change in susceptibility patterns was seen in case of *Pseudomonas aeruginosa* which have shown greater susceptibility to Colistin (83.3%), followed by Amikacin (27.8%), Meropenem (16.7%), and Gentamicin (16.7%). TMP and SMZ and Amoxicillin Clavulanic acid were found to be very less susceptibility, i.e., *E. coli* and *K. pneumoniae* are more resistant (70%) with those antibiotics. The pattern of antibiotic(s) susceptibility was not significant among the Gram-negative bacteria (*p* > 0.05).

In-patients, the susceptibility of *E. coli* to Colistin (96.5%), Amikacin (81.2%) and *K. pneumoniae* to Colistin (100%), Amikacin (85.4%) were observed, respectively. The remaining antibiotics showed 40%–67% of susceptibility. TMP and SMZ and Amoxicillin Clavulanic acid had very poor (less than 30%) susceptibility toward the uropathogens (Table 6). In out-patients, Colistin was proved to be very good susceptibility (84.6%–100%) toward the Gram-negative bacteria (Table 7). The remaining antibiotics showed less than 70% of susceptibility toward *E. coli* and very poor susceptibility toward *K. pneumoniae* (16%–33%). In contrast to remaining antibiotics such as TMP and SMZ, Amoxicillin Clavulanic acid, Cefoperazone, and Amoxicillin plus Clavulanic acid had relatively low susceptibility (16%–27%).

**DISCUSSION**

Antibiotic resistance is the major challenges in the treatment of UTIs. In our study, we examined the susceptibility of uropathogens toward the antibiotics in IP and OP patients visited with UTIs. The study was conducted during 6 months

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**Table 2.** Distribution of pathogens isolated in urine cultures among in-patients and out-patients.

| Pathogen                  | Total number (%) | IP N = 147 (%) | OP N = 53 (%) | *p* value |
|---------------------------|------------------|----------------|--------------|-----------|
| Gram negative             |                  |                |              |           |
| *Escherichia coli*        | 121 (60.5)       | 85 (57.8)      | 36 (67.9)    |           |
| *Klebsiella pneumoniae*   | 32 (16)          | 26 (17.7)      | 6 (11.3)     |           |
| *Pseudomonas aeruginosa*  | 18 (9)           | 13 (8.8)       | 5 (9.4)      |           |
| *Enterococcus*            | 4 (2)            | 3 (2)          | 1 (1.9)      |           |
| *Acinetobacter junii*     | 2 (1)            | 2 (1.4)        | 0 (0)        | 0.007*    |
| *Enterococcus avium*      | 2 (1)            | 1 (0.7)        | 1 (1.9)      |           |
| *Enterococcus fecalis*    | 2 (1)            | 2 (1.4)        | 0 (0)        |           |
| Gram positive             |                  |                |              |           |
| *Staphylococcus aureus*   | 10 (5)           | 8 (5.4)        | 2 (3.8)      |           |
| *Candida albicans*        | 5 (2.5)          | 3 (2)          | 2 (3.8)      |           |
| *Candida*                 | 4 (2)            | 4 (2.7)        | 0 (0)        |           |

*The *p* value <0.05 were considered statistically significant.*
of period, from 2019 November to 2020 April. There are nearly equal numbers of both genders were recruited in our study. There are more number of in-patients as compared to out-patients with UTI. Out of 200 UTI patients, highest prevalence of bacteria was related to Gram-negative bacteria (E. coli and Klebsiella) (90.5%) causing UTI and 9.5% were Gram-positive bacteria. Nitrofurantoin, fosfomycin, sulferniamides, fluoroquinolones, and beta-lactams are generally recommended antibiotics to treat UTIs (Gupta et al., 2011). The prevalence of isolates was similar in Italian study (Magliano et al., 2011) However, their susceptibility was questionable. The susceptibility was found to be 35%–40% in 40–59 years of age group, among them Ertaopenem and TMP & SMX had 48.5% and 44.6%, respectively. The susceptibility was increased to 42%–50% in 60–79 years of age group and notably, the low susceptibility (26.4%) was observed in 60–79 years of age group patients. The antibiotic(s) susceptibility was literally poor in age 80–99; it could be attributed to a weaker immune system, usually seen in this older age group. The results also showed that the highest prevalence of bacteria was related to E. coli in 121 (60.5%) and Klebsiella in 32 (16%) of the cases, respectively, and which was consistent with other study (Mohammadi et al., 2010). Among in-patients E. coli, 96.5%, 81.2%, and 58.8% isolates were susceptible to Colistin, Amikacin and Nitrofurantoin, while K. pneumoniae, 100%, 85.4%, and 26.9% isolates were susceptible to Colistin, Amikacin, and Nitrofurantoin, respectively. Likewise, in Zambian hospital-based study was nearly correlated with our present study. In Zambia site, in-patients with K. pneumoniae and Streptococcus species are resistant to chloramphenical, Nitrofurantoin, and ceftazidime than out patients (Chanda et al., 2019). In out-patients E. coli, 97.2%, 69.4%, and 61.1% isolates were susceptible to Colistin, Amikacin, and Nitrofurantoin and toward K. pneumoniae it was observed to be 100%, 66.7%, and 33.2%. Our findings were quite opposite to the study conducted in Odisha, India, where the E. coli had very low resistant profile toward Amikacin 5.8% and 9.8% with Nitrofurantoin (Dash et al., 2013). The similar study conducted in India (Amladi et al., 2019), noticed that, fosfomycin, Nitrofurantoin and Colistin are susceptible to E. coli, 98.9%, 56% and 95%, while fosfomycin and Colistin are susceptible to 94% and 85% toward Klebsiella species respectively. Nitrofurantoin and fosfomycin had better susceptibility profile and more effective toward Gram negative bacteria causing uncomplicated UTIs (Gardiner et al., 2019). The similar study conducted in China, reported, ampicillin, cotrimoxazole, ciprofloxacin, amoxicillin, and Nitrofurantoin resistance values in E. coli were 59.8%, 31.8%, 23.4%, 1.9%, and 0.9% respectively. The results were quite opposite to our study (Wong et al., 2017). Similarly, nalidixic acid and ceftazidime are

### Table 3. Age Wise Distribution of Gram negative uropathogens.

| Age (Years) | N = 181 | *Escherichia coli* (N = 121%) | *Klebsiella pneumoniae* (N = 32%) | *Pseudomonas aeruginosa* (N = 18%) | *Staphylococcus aureus* (N = 10%) | p value |
|-------------|--------|-------------------------------|----------------------------------|----------------------------------|----------------------------------|---------|
| 20–39       | 25     | 15 (12.4)                     | 5 (15.6)                         | 1 (5.5)                          | 4 (4)                            |         |
| 40–59       | 67     | 45 (37)                       | 15 (47)                          | 6 (33.3)                         | 1 (1)                            |         |
| 60–79       | 81     | 58 (48)                       | 10 (31)                          | 8 (44.5)                         | 5 (5)                            | 0.03*   |
| 80–99       | 8      | 3 (2.5)                       | 2 (6.3)                          | 3 (16.6)                         | 0 (0)                            |         |

*The p value <0.05 were considered statistically significant.

### Table 4. Age wise distribution of antibiotic susceptibility.

| Antibiotics                | Number | 20–39 N (%) | 40–59 N (%) | 60–79 N (%) | 80–99 N (%) | p value |
|----------------------------|--------|-------------|-------------|-------------|-------------|---------|
| Colistin                   | 170    | 19 (11.17)  | 65 (38.2)   | 78 (45.8)   | 8 (4.7)     |         |
| Amikacin                   | 124    | 19 (15.3)   | 45 (36.2)   | 55 (44.3)   | 5 (4)       |         |
| Imipenem                   | 98     | 18 (18.3)   | 35 (35.3)   | 42 (42.8)   | 3 (3)       |         |
| Meropenem                  | 84     | 20 (23.8)   | 35 (41.67)  | 27 (32.13)  | 2 (2.3)     |         |
| Gentamicin                 | 94     | 16 (17)     | 35 (37.2)   | 40 (42.5)   | 3 (3)       | 0.142*  |
| Ertaopenem                 | 68     | 14 (20.5)   | 33 (48.5)   | 18 (26.4)   | 3 (4.4)     |         |
| Nitrofurantoin             | 91     | 17 (18.6)   | 26 (28.5)   | 46 (50.5)   | 2 (2.1)     |         |
| Cefoperazone and sulbactam | 87     | 18 (20.6)   | 32 (36.7)   | 34 (39)     | 3 (3.4)     |         |
| Cefoperazone               | 58     | 13 (22.4)   | 22 (37.9)   | 20 (34.4)   | 3 (5.1)     |         |
| TMP-SMX                    | 47     | 6 (12.)     | 21 (44.6)   | 18 (38.2)   | 2 (4.2)     |         |
| Amoxicillin Clavulanic acid| 45     | 9 (20)      | 17 (37.7)   | 17 (37.7)   | 2 (4.4)     |         |

*The p value <0.05 were considered statistically significant.
Table 5. Percentage susceptibility of uropathogens toward different antibiotics.

| Antibiotics          | %  | EC % | KP % | PA % | p value |
|----------------------|----|------|------|------|--------|
| Colistin             | 85 | 96.7 | 100  | 83.3 |        |
| Amikacin             | 62 | 77.7 | 65.6 | 27.8 |        |
| Imipenem             | 49 | 66.1 | 53.1 | 5.6  |        |
| Meropenem            | 48 | 64.5 | 46.9 | 16.7 | 0.58   |
| Gentamicin           | 47.5 | 62 | 40.6 | 16.7 |        |
| Ertapenem            | 45.5 | 61.2 | 53.1 | -    |        |
| Nitrofurantoin       | 45.5 | 59.5 | 28.1 | -    |        |
| Cefoperazone and sulbactam | 43.5 | 58.7 | 40.6 | 5.6  |        |
| Cefoperazone         | 29 | 38   | 28.1 | 5.6  |        |
| TMP-SMX              | 23.5 | 25.6 | 31.3 | -    |        |
| Amoxicillin Clavulanic acid | 22.5 | 29.8 | 28.1 | -    |        |

EC: *Escherichia coli*; KP: *Klebsiella pneumonia*; PA: *Pseudomonas aeruginosa*.

Table 6. Percentage of antibiotic susceptibility of pathogens in in-patients.

| Antibiotics          | %  | EC % | KP % | PA % | p value |
|----------------------|----|------|------|------|--------|
| Colistin             | 84.4 | 96.5 | 100  | 84.6 |        |
| Amikacin             | 63.9 | 81.2 | 85.4 | 30.8 |        |
| Gentamicin           | 50.3 | 65.9 | 46.2 | 23.1 |        |
| Imipenem             | 49.7 | 67.1 | 57.7 | 7.7  |        |
| Meropenem            | 49.7 | 65.9 | 53.8 | 23.1 | 0.118  |
| Ertapenem            | 45.6 | 61.2 | 57.7 | -    |        |
| Nitrofurantoin       | 44.2 | 58.8 | 26.9 | -    |        |
| Cefoperazone and sulbactam | 44.2 | 58.8 | 46.2 | 7.7  |        |
| Cefoperazone         | 32   | 42.4 | 30.8 | 7.7  |        |
| TMP-SMX              | 24.5 | 25.9 | 34.6 | -    |        |
| Amoxicillin Clavulanic acid | 23.1 | 30.6 | 30.8 | -    |        |

EC: *Escherichia coli*; KP: *Klebsiella pneumonia*; PA: *Pseudomonas aeruginosa*.

The in-patients and out-patients resistance to TMP and SMX, Cefoperazone, and Amoxicillin plus Clavulanic acid was higher. Colistin, Amikacin, and Nitrofurantoin showed better susceptibility in in-patients and in out-patients. TMP and SMX, Amoxicillin Clavulanic acid, Cefoperazone, and Amoxicillin plus Clavulanic acid had relatively low susceptibility.

More resistant to *Enterobacter* and *Proteus* species in females than male patients, in Zambia (Chanda et al., 2019).

In the Iranian settings of both IP and OP departments, the *E. coli* shows low antibiotic resistant toward Nitrofurantoin (IP-16.5% and OP-16.3%), Gentamicin (IP-52.4% and OP-31.7%), and ciprofloxacin (IP-60.1% and OP-34.3%). High resistant with cotrimoxazole (IP-80% and OP-62.7%) and cephalotin (IP-78.3% and OP-54.3%) (Davoodabadi et al., 2012). In pediatrics of IP developed 24% and 30% in out-patients resistance toward TMP/SMX with *E. coli* (Saperston et al., 2014), where in our study we did not have pediatrics. Utility of TMP-SMX was limited in China because of poor sensitivity with *E. coli* (Zhao et al., 2010). A survey was conducted in Australia; they evaluated the resistance profile of *E. coli* since 2015–17. They observed that, *E. coli* had 7% for ciprofloxacin, 43% for ampicillin, 9% for amoxicillin plus clavulanic acid, 16% for cefazolin, and 22% for TMP. These findings of TMP were similar to Indian context (Australian Commission on Safety and Quality in Health Care, 2017). In our study, IP *E. coli* was (IP-67.1% and OP-63.9%) *K. pneumoniae* was (IP-57.7% and OP-33.2%) sensitive toward Imipenem. Likewise, in the India, Karala state, the majority of Gram-negative bacteria is susceptible to Imipenem (Thattil et al., 2018). The findings of this study indicate that selecting drugs for empiric treatment would be difficult as no single popular medication can be prescribed for UTI.

### CONCLUSION

Overall, our investigation reports that the resistance of uropathogens to antibiotics was common in both genders of UTI patients. *Escherichia coli* was more prevalent in 40–79 years of age group. The poor susceptibility was observed in 80–99 years of age groups. Colistin, Amikacin, and Nitrofurantoin showed better susceptibility in in-patients and in out-patients. TMP and SMX, Amoxicillin Clavulanic acid, Cefoperazone, and Amoxicillin plus Clavulanic acid had relatively low susceptibility.

### Limitations

Periodic monitoring of antibiotic susceptibility patterns in clinical settings is vital to determine the potency as well as re-establishing empirical therapy. It is a single center study, hence using this data we cannot generalize the results to whole population. Therefore, the compilation of all the data from different sites are required to establish empirical therapy.
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CONFLICT OF INTERESTS
None.

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ETHICAL APPROVAL
The study procedure was approved by the Institutional Ethics Committee, Ramesh Hospitals, (ECR/81/Inst/AP/2019, Dated: 15-10-2019) Vijayawada, India; this research was carried out in conjunction with good clinical practice guidelines.

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