ANTIMICROBIAL SUSCEPTIBILITY PATTERN IN ESCHERICHIA COLI ISOLATES CAUSING URINARY TRACT INFECTIONS AMONG THE PATIENTS AT A TERTIARY CARE HOSPITAL AT KANPUR

Nilam Nigam¹, Shalini Gupta², Sanjay Kumar Nigam³, Niraj Kumar Nigam⁴

¹Professor and HOD, Department of Pharmacology, Rama Medical College Hospital and Research Centre, Kanpur.

²Assistant Professor, Department of Pharmacology, Rama Medical College Hospital and Research Centre, Kanpur.

³Professor, Department of Pathology, Rama Medical College Hospital and Research Centre, Kanpur.

⁴M. Sc. Medical Pharmacology Student, Rama Medical College Hospital and Research Centre, Kanpur.

ABSTRACT

BACKGROUND

Urinary Tract Infections (UTI), defined as the persistent presence of actively multiplying microorganisms within the urinary tract. It is the most common infectious disease cause of morbidity and mortality in human. The most common causes of urinary tract infections are bacteria, and less often viruses, yeast or other intracellular microorganism. Many different microorganisms can cause UTIs, though the most common pathogens causing the simple ones in the community are *Escherichia coli* and other Enterobacteriaceae, which accounts for approximately 75% of the isolates.

Objectives- The aim of this study was to determine the types of bacteria isolated from the urinary tract infections and their susceptibility pattern from patients reporting in Tertiary Care Urology Centre in order to rationalise the use of antibiotics.

MATERIALS AND METHODS

To study antimicrobial susceptibility pattern of *E. coli* culture positive urine sample from Rama Medical College, Hospital and Research, Kanpur. A total of 100 *E. coli* culture positive mid-stream urine samples were collected from UTI patients.

RESULTS

A study on antibiotic susceptibility pattern of 100 samples of *E. coli* culture positive urinary samples showing that according to sex, female (70%) are more susceptible to infection than male (30%). As according to age group above 50 years most commonly showing urinary tract infection. After testing with Kirby-Bauer disc diffusion method on Mueller-Hinton agar with antibiotics like Amikacin, Netilmicin, Nitrofurantoin, Levofloxacin, Norfloxacin, Ciprofloxacin, Amoxiclav, CFS, PIT, Imipenem. In this study, Antibiotic Susceptibility Pattern of *E. coli* (Total 100 isolates) more susceptibility to Imipenem (96%), followed by Nitrofurantoin 90%, Amikacin 88%, Piperacillin/Tazobactam 82%, Netilmicin 78%, Cefoperazone/Sulbactam 71%, lower susceptibility Ciprofloxacin 40%, Norfloxacin 44% and Amoxiclav 23%.

CONCLUSION

Majority of the isolates *E. coli* were sensitive to Injectable Imipenem (IMP) with disc concentration of 10 µg showing 96% susceptibility and oral antimicrobials Nitrofurantoin (NIT) with disc concentration of 300 µg showing 90%. Susceptibility are commonly used to treat urinary tract infections.

KEYWORDS

Urinary Tract Infections, *Escherichia coli*.

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BACKGROUND

Urinary Tract Infections (UTI) defined as the persistent presence of actively multiplying microorganisms within the urinary tract. It is the most common infectious diseases ranking next to Upper Respiratory Tract Infections (RTI) and an important cause of morbidity and mortality in human.^[1] It is an infection with more than 105 organisms per millilitres in the mid-stream urine samples. The most common causes of urinary tract infections are bacteria and less often viruses,

yeast or other intracellular microorganisms.^[2] Many different microorganisms can cause UTIs, though the most common pathogens causing the simple ones in the community are *Escherichia coli* and other Enterobacteriaceae, which accounts for approximately 75% of the isolates.^[3]

Neonates, girls, young women and older men are most susceptible to UTIs. In women, bacterial cystitis is the most common bacterial infection. Every woman has a 60% lifetime risk of developing bacterial cystitis, which develops mostly before the age of 24 years. By contrast men have a lifetime risk of only 13%,^[4] in children approximately 5% of girls and 1% of boys have UTI by 11 years of age.^[5]

Urinary tract infection is prevalent among men and women, but is more common among women, especially during pregnancy. Unrecognised patients may have serious long-term effect and chronic pyelonephritis may occur in adults.^[2]

Urinary tract infection occurs in 17% to 20% of pregnancies resulting in premature rupture of membranes, chorioamnionitis, premature delivery and postpartum

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Corresponding Author:

Dr. Nilam Nigam,

Professor and HOD,

Department of Pharmacology,

Rama Medical College Hospital & Research Centre,
Kanpur.

E-mail: drnilamnigam@gmail.com

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maternal and neonatal infections. The majority of patients present during the first year of life. Up to 1% of term neonates and 4% - 25% of premature may be involved; it is more common during neonatal period and declines afterwards.^[6] About 8% of girls and 2% of boys experience at least one episode of urinary tract infection up to the age of 7. It occurs in 0.1 to 0.4% of infant girls and increase up to 1.4% during 1-5 years and 0.7% to 2.3% in school age. Close to 0.2% of circumcised and 0.7% of uncircumcised boys are at risk.^[6]

It has been estimated that more than 6 million outpatient visit and 300000 hospital stays every year are due to urinary tract infections. Approximately, 10% of human will have urinary tract infections. The exact prevalence of urinary tract infection is age and sex dependent. Antibiotic resistance may develop in uropathogens due to frequent misuse of antibiotics.^[7]

In addition to Cephalosporins, the uropathogens are also exhibiting increasing resistance to antibiotics like Cotrimoxazole, quinolones and nitrofurantoin. The wide array of resistance mechanism has jeopardised the empirical use of quinolones and cephalosporins. The therapeutic options to treat UTIs caused by multidrug resistant bacteria have forced the clinicians to resort to carbapenems, colistin and fosfomycin.^[8,9]

The aim of this study was to determine the types of bacteria isolated from the urinary tract infections and their susceptibility pattern from patients reporting in tertiary care urology centre in order to rationalise the use of antibiotics.

MATERIALS AND METHODS

Study Setting

The present study was conducted in the Department of Pharmacology, Medicine and Microbiology at Rama Medical College Hospital and Research Centre - Mandhana, Kanpur.

Study Design

Descriptive Study

During the 12-month period, a total of 587 urine samples were culture and sensitivity testing. Of the total urine samples, 23 (3.91%) showed mixed bacterial growth may be due to improper specimen collection or delay in transporting the sample to the laboratory. A total of 188 different organisms were isolated, thus the culture positivity was 32.02%. Out of the total 188 different organisms, E. coli was the most isolated organism accounting for 100 (53.19%). The plates were examined after 18 hours of incubation. The zone of inhibition around the disc was measured with the help of millimetre ruler and the results were interpreted according to the sensitive, intermediate or resistant.

Study Population

E. coli culture positive UTI patients from the Department of Medicine and Microbiology at Rama Medical College Hospital were included in the study.

Inclusion Criteria

Urine culture samples, testing positive for E. coli isolates from the microbiology laboratory were included in the study.

Exclusion Criteria

1. Pregnant women.

2. Patients already diagnosed with UTI and on treatment.
3. Patients with renal disease.
4. Patients already on antibiotics.
5. Relapse patients who are already included in the study.
6. Patients suffering from STDs.
7. Immunocompromised patients, bedridden patients/ paraplegics/neurogenic bladder cases.

Collections

Specimen Collection- urine samples collected.

Study Duration

Study conducted over a period of one year.

Sampling and Sample Size

No sampling done. All UTI E. coli culture positive isolates was taken from microbiology laboratory, Rama Medical College, Kanpur, for this study. We have assumed that sensitivity in 90% cases can be safely accepted in claiming sensitivity. The sample size has been calculated using the formula $n \geq p \times q \times 1.96 / d^2$,

Where $p = 0.9$, $q = 0.1$.

1.96 is the value of 'z' for 95% confidence.

D is the allowable error= 7%.

The sample size comes out to be 69.

Therefore, we have included 100 patients in our study.

Method of Data Collection

- **Male-** Glans penis was wiped clean with sterile cotton wool and sterile mid-stream urine was collected by the clean catch technique in a sterile container.
- **Females-** Vulva was cleaned with water and mid-stream urine was collected as above.
- **Indwelling Catheter-** Catheter tubing should be clamped off above the port to allow collection of freshly voided urine. The catheter port or wall of the tubing should then be cleaned vigorously with 70% ethanol and urine aspirated via a needle and syringe. The integrity of the closed drainage system must be maintained to prevent the introduction of organisms into the bladder.^[10]
- **Specimen Transportation-** Urine samples were put up for culture within an hour of receiving the sample. In case of any delay it was kept in refrigerator at 4°C, because urine is an excellent supportive medium for growth of most bacteria. Bacterial count in refrigerated 4°C urine remains constant for as long as 24 hours.^[10]
- **Culture-** Urine sample was mixed thoroughly. A standard calibrated loop was inserted vertically into urine sample and inoculated on Blood Agar, MacConkey agar by streaking method incubate the inoculated plates at 37°C for 18 - 24 hours. Any growth that took place was examined and the number of colonies were counted. Development of approximately 500 colonies corresponds to 10,000 bacteria/mi.^[10]

The isolates were tested by Kirby-Bauer disc diffusion method on Mueller-Hinton Agar.

Ethical Issues

The study was started after approval from the Institutional Medical Ethical Committee.

RESULTS

The present study was conducted with the objectives to study antimicrobial susceptibility pattern of bacterial isolates (*E. coli*) causing UTI to know which antibiotic was better choice for UTI patients.

A total of 100 *Escherichia coli*, culture positive urine samples were collected from microbiology laboratory during a period of one year for studies.

The isolates were tested by Kirby-Bauer disc diffusion method on Mueller-Hinton Agar according to guidelines of the National Committee for Clinical Laboratory Standards.

Sex	Total Number of Cases (<i>E. coli</i>)	No. of Infected Cases (<i>E. coli</i>)	Percentage
Female	70	70	70%
Male	30	30	30%
Total	100	100	100%

Table 1. Distribution of Cases according to Sex

Graph 1

Out of 100 Urine

- Culture positive samples, 30 were male patients (30%) and 70 were (70%) female patients having urinary tract infection.
- From the Table 1, it is seen that urinary tract infection is more common in females (70%) than males (30%).

Distribution of Case according to Age

Graph 2

Graph 2 shows distribution of cases according to age. The urinary tract infection rate was high in higher age groups. Out of 100 urinary tract infected case, 37 were from patients above 50 years of age and 21% cases having urinary tract infection between 21 - 30 years of age.

Antibiotics	Disc Conc.	Sensitive Isolates	
		No. of Sensitive Cases	(%)
Amikacin	30 µg	88	88
Netilmicin	30 µg	78	78
Nitrofurantoin	300 µg	90	90
Levofloxacin	5 µg	67	67
Norfloxacin	10 µg	44	44
Ciprofloxacin	5 µg	40	40
Amoxiclav	30 µg (20/10)	23	23
CFS	75/10 µg	71	71
Piperacillin/Tazobactam	100/10 µg	82	82
Imipenem	10 µg	96	96

Table 2. Antibiotic Susceptibility Pattern of *E. coli* (Total 100 Isolates) in Urine Culture Positive Samples

Graph 3

Amikacin Imipenem has maximum susceptibility (96%) for *E. coli* followed by nitrofurantoin (90%), Norfloxacin (44%), ciprofloxacin (40%) and Amikacin (88%).

DISCUSSION

The present study was undertaken in the Department of pharmacology, Rama Medical College, Hospital and Research Centre, Kanpur, U.P.

The study was conducted over a period of one year. To explore the antimicrobial susceptibility pattern of *E. coli*, culture positive urine sample from hospital isolate admitted to different wards and those attending OPD of our hospital.

During the 12-month period, a total of 587 urine samples were processed for culture and sensitivity testing. Of the total urine samples studied, 23 (3.91%) showed mixed bacterial growth may be due to improper specimen collection or delay in transporting the sample to the laboratory. A total of 188 different organisms were isolated, thus the culture positivity was 32.02%. Out of the total 188 different organisms, *E. coli* was the most isolated organism accounting for 100 (53.19%). The plates were examined after 18 hours of incubation. The zone of inhibition around the disc was measured with the help of millimetre ruler and the results were interpreted according to the sensitive, intermediate or resistant.

Urinary Tract Infections (UTIs) could be defined as the persistent presence of actively multiplying microorganisms within the urinary tract. It is an infection with more than 10⁵ organisms per millilitres in the mid-stream urine samples. The most common causes of urinary tract infections are bacteria and less often viruses, yeast or other intracellular microorganism.^[2]

Many different microorganisms can cause UTIs, though the most common pathogens causing the simple ones in the community are *Escherichia coli* and other Enterobacteriaceae, which accounts for approximately 75% of the isolates.^[3]

Members of the family Enterobacteriaceae is a well-known cause of urinary tract infections. *E. coli* is reported to be the most common bacteria with prevalence as high as 71% and resistance to commonly used antimicrobials.^[11]

About 150 million people developed a urinary tract infection each year.^[12] It is more common in women than men.^[13] In women, UTI is the most common form of bacterial infection.^[14] Up to 10% of women have a urinary tract infection in a given year and half of the women having at least one infection at some point in their lives.^[13,15] They occur most frequently between the ages of 16 and 35 years.

We had observed that out of 100 total cases, 70 (70%) cases were found to be urinary tract infection caused by *E. coli* in females and 30 (30%) were found UTI positive in males (Table No. 01, Fig. No. 02).

Antibiotic Susceptibility Pattern of *Escherichia*

Amikacin

- In present study, the amikacin was found 88% susceptible for *E. coli*.
- In previous study done by (C. Manikandan et al 2015)^[16] showed 92.77% susceptible for *E. coli*.
- Our finding is slightly lower than (C. Manikandan et al 2015),^[16] which is 90%.

Netilmicin

- In our present study, Netilmicin was found 78% susceptibility for *E. coli*.
- The previous study done by (Akhtar N et al 2016)^[17] showed 62.70% susceptibility for *E. coli*. Our finding is comparatively higher than (Akhtar N et al, 2016).^[17]
- Netilmicin is a member of aminoglycosides family of antibiotics. These antibiotics have ability to kill a wide variety of bacteria.
- Nitrofurantoin.
- In our present study, nitrofurantoin was found 90% susceptible for *E. coli*.
- In study in India by (Shalini et al 2011)^[18] showed 93.48% susceptibility for *E. coli* and our finding is slightly lower than (Shalini et al 2011)^[18] which is 90%.
- Currently, bacterial resistance to nitrofurantoin is more frequent than the resistance to fluoroquinolones and cotrimoxazole making nitrofurantoin 2-line agents for treatment of UTIs.

Fluoroquinolones

These are the potent bactericidal drugs.

Levofloxacin

- In the present study, it was found to have 67% susceptibility to *E. coli*. The study done by (Shalini et al, 2011)^[18] showed 75% susceptibility to *E. coli*. Our finding is comparatively lower than (Shalini et al, 2011).^[18]

Imipenem

- In our present study, Imipenem was found 96% susceptible for *E. coli*.
- In previous study by (Niranjan V. and Malini A. 2014)^[19] showed 98.9%; susceptibility for *E. coli* in our finding is comparatively lower than (Niranjan V. and Malini A. 2014).^[19]
- In previous study by (C. Manikandan and A. Amsath)^[16] showed 92.1%; susceptibility for *E. coli* in our finding is comparatively higher than (C. Manikandan and A. Amsath).^[16]
- Cefoperazone/Sulbactam.
- In our present study, Cefoperazone/Sulbactam was found 71% susceptible for *E. coli*.
- In previous study by (Abdul Ghafur, 2012)^[20] showed 80.3%; susceptibility for *E. coli* in our finding is comparatively lower than (Abdul Ghafur, 2012).^[20]

Piperacillin/Tazobactam

- In our present study, Piperacillin/Tazobactam was found 82% susceptible for *E. coli*.

In previous study by (InamUllah Khan 2014)^[21] showed 80.7%; susceptibility for *E. coli* in our finding is comparatively slightly higher with the study of (InamUllah Khan 2014).^[21]

In this study, Antibiotic Susceptibility Pattern of *E. coli* (Total 100 isolates) had more susceptibility to Imipenem (96%), followed by Nitrofurantoin 90%, Amikacin 88%, Piperacillin/Tazobactam 82%, Netilmicin 78%, Cefoperazone/Sulbactam 71%, lower susceptibility Ciprofloxacin 40%, Norfloxacin 44% and Amoxiclav 23%.

CONCLUSION

E. coli was the predominant pathogen causing UTIs in our population presenting at tertiary care Rama Medical College, Hospital and Research Centre, Kanpur. Study on antibiotic susceptibility pattern of 100 samples of *E. coli* culture positive urinary samples showing that according to sex, female (70%) are more susceptible to infection than male (30%). As according to age group of above 50 years is most commonly showing urinary tract infections, i.e. 37%; next commonly infected age group is 21 - 30 years was 21% showing urinary tract infection. After testing with Kirby-Bauer disc diffusion method on Mueller-Hinton agar with antibiotics like Amikacin, Netilmicin, Nitrofurantoin, Levofloxacin, Norfloxacin, Ciprofloxacin, Amoxiclav, CFS, PIT and Imipenem. Majority of the isolates of *E. coli* were sensitive to Injectable Imipenem (IMP) with disc concentration of 10 µg showing 96% susceptibility and oral antimicrobials Nitrofurantoin (NIT) with disc concentration of 300 µg showing 90% susceptibility are commonly used to treat urinary tract infections.

As far as the oral antibiotics are concerned, Nitrofurantoin revealed encouraging results proving to be the only effective oral antibiotic in this study. As drug resistance among bacterial pathogens is an evolving process, routine surveillance and monitoring studies should be conducted in different parts of the country to provide physicians an effective knowledge regarding the empirical treatment of UTIs in that particular area.

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