



## Microbiological Enumeration of Pathogens Associated with Urinary Tract Infection: Current Scenario and Its Medical Relevance

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### Authors' contributions

This work was carried out in collaboration among all authors. Authors PS and MSS designed the study and performed the analysis. Authors KG and SS managed the analysis of the study. Author NS managed the literature searches and guided in paper writing. All authors read and approved the final manuscript.

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### ABSTRACT

**Background:** Urinary tract infection (UTI) is the most common among community acquired infections worldwide affecting men as well as women, many reports suggested that uropathogens are becoming more resistant towards antibiotics, which can cause various problems during treatment.

**Methods:** In this study, 48 samples were collected from the suspected patients and these samples were subjected to various clinical evaluations.

**Results:** Among them 29 were females and 19 were males, in the studied cases *E. coli* (48.3%) was most frequently observed pathogens, *S. aureus* (29.03%). Among antibiotics, Tigecyclin (TGC) was the most effective antibiotic against all the three isolated bacteria, about 100% of bacteria

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showed the sensitivity towards it. *E. coli* was found to be the most resistant bacteria against Tetracycline (TET) (66.6%) followed by Amoxiclav (AMC) (60%), Fosfomycin (Fo) (53.3%), Piperacillin (PIP) (53.3%), Cefepime (CPM) (53.3%). Among *K. pneumonia* isolates had shown resistance against Tetracycline (TET) (57.1%), Fosfomycin (FO) (57.1%), Piperacillin (PI) (57.1%), Cephalexin (CZ) (57.1%) and Amikacin (AK) (57.1%). *Staphylococcus aureus* had shown maximum resistance against Tetracycline (TET) (66.5%), Co-amoxiclav (AMC) (66.5%) followed by Chloramphenicol (55.4%) and Cefepime (CPM) (55.4%).

**Conclusion:** In conclusion UTI infection rates are higher among females, *E. coli* was the most causative agents of UTI and the emergence of resistance in bacteria is probably due to the overuse of antibiotics. Antibiotic Tigecyclin could be the drug of choice for the treatment of UTI.

**Keywords:** Urinary tract infection; uropathogens; clinical evaluation; antibiotics; emergence.

## 1. INTRODUCTION

Urinary tract infection (UTI) is major cause of morbidity in humans and one of the most frequently encountered infections that needs to be detected at early stage, delay in treatment can cause chronic kidney damage. Nearly 10% of population experience urine tract infection during their life time [1]. Urinary tract infection may involve an upper and lower part of urinary tract. If infection occurs within the lower urinary tract, then this condition is known as cystitis, and when the infection occur within an upper urinary tract, the condition tends to be medically termed as pyelonephritis in which infection has already spread into the kidneys. Symptoms of lower UTI include painful and frequent urination cloudy or foul smelly urine, pain within the lower abdomen [2]. However, the symptoms of upper UTI include flank pain, high fever, chills, nausea and vomiting. It has been reported that UTI affects both genders but women of the age between 15 years to 44 years are more prone to this infection [3]. UTI infection is a global infection and in United States of America (USA) more than 150 million cases are reported annually which approximately cost around 6 billion US Dollars [4]. The major causative agent responsible for most cases of UTI are bacteria [5], mainly gram negative bacterial species which accounts for 80% to 85% reported cases, out of which 80% of cases registered are due to *E. coli*, followed by *Klebsiella pneumonia* and *Proteus mirabilis* [6]. Clinical evaluation that tends to be performed to have an initial suspicion of urinary tract infection does entail urine analysis (urinalysis) in which evidence of infection tends to be based upon presence of bacteria in the urine, nitrites in the urine, as well as white blood cells (WBCs) within the sample of urine. The antibiotic sensitivity pattern of the cultured organism and the allergy

status of each individual patient, the availability of the antibiotics and the local costs of the various antibiotics guide the clinician to decide upon the choice of treatment. The proposed study determined the causing Urinary tract infection (UTI) causing microorganisms, their distribution among genders and different age groups, antimicrobial susceptibility was also determined in this study.

## 2. MATERIALS AND METHODS

### 2.1 Specimen Collection and Bacterial Isolation

This is a cross sectional study conducted in DNA-Labs A Centre for Applied Sciences, Dehradun, Uttarakhand, India. In this study a total of 48 midstream specimens of urine were collected from the patients that showed the symptoms of the patients associated with the urinary tract infection. The samples were obtained from DNA Labs- A Centre for applied sciences. Mid-stream urine samples were collected in sterile, dry, leak-proof disposable 20 ml containers. The samples were subjected to further physical, chemical and microscopic analysis. The Urine was analyzed on parameters such as transparency and odor of the urine. For the chemical testing, reagent test strips were used to determine the pH, specific gravity, glucose and protein amount within the urine specimen. The urine sample was examined under a microscope for the detection of casts, crystals, cells including, RBCs, WBCs, Epithelial cells and microbes such as bacteria, yeast, and trichomonas. The samples were then samples were inoculated on 5% blood agar, McConkey agar incubated for 24 hours at 37°C. The samples that had a colony count  $\geq 10^5$  cfu/mL were considered positive isolates.

## 2.2 Identification of Uropathogens and Antibiotic Susceptibility Testing

Different biochemical tests were performed for the conformation of bacteria that are present within the urine sample, such as Gram's staining, Triple sugar iron agar test (TSI), Simmon citrate agar test, Catalase test, Coagulase test, Indole test, Oxidase test, and MR-VP test. The Kriby-Bauer disk method was used to determine the Antibiotic susceptibility of the isolated bacteria. Müller-Hinton agar plates were used for the identification of sensitivity pattern [7]. After 24 hours of incubation at 37°C measurement of zone of inhibition of bacterial growth was performed and comparison was done with the guidelines of Clinical and Laboratory Standards Institute [8]. Antibiotics used for identification of Resistance pattern were, Fosfomyin (FO), Ofloxacin (OF), Norfloxacin (NX), Nitrofurantoin (NIT), Colistin (CL), Piperacillin (PI), Ceftriaxone (CTR), Cefepime (CPM), Cefazoline (CZ), Amoxyclav (AMC), Meropenem (MRP), Ertapenem (ETP), Gentamycin (GEN), Amikacin (AK), Levofloxacin (LE), Tetracycline (DO), Tigecycline (TGC), Chloramphenicol, Erythromycin (ERY), Clindamycin (CLI), Vancomycin (VAN) Doxycyclin (DO).

## 2.3 Statistical Method

Simple percentage method was used for the analysis of the observed data.

## 3. RESULTS

A total of 48 samples were screened out of which 31 (64.5%) cases were found to be positive for bacterial infection. Among them 60.4% (29/48) cases were found in women, and 39.5% (19/48) were found in men. The ages of the patients ranged between 13 years and 70 years and the average age of the with the patients were 41.5 years. For the age distribution of study sample, samples were arranged into four different groups. First group represents patients of ages were between 1 year to 20 years in this group of

patients 14.5% of cases that were reported, among them 10.4% cases were females and 4.16% were male. In the 21 years to 40 years age group, maximum cases of UTI were reported that amounted to about 57.8% out of all the cases in total, in this group, 43.75% were females and 14.58% were males (Table 1). Patients who were within the age group between 41 years and 60 years that amounted to about 16.7% cases were reported and among them 12.5% were males and 4.16 % were females. The elderly group age of patients who were aged older than 60 years amounted to 12.4% of cases and out of this age group 8.3% were males and 4.1% were females.

In this study out of 48 cases, about 31 (64.58%) cases were found to be infected from bacterial pathogen 70.9% of isolated bacteria were found to be Gram negative organisms, while 29.1% were Gram positive organisms. Among them 49.3% of the infections were caused by *E. coli*, followed by *S. aureus* (30.7%) and *K. pneumonia* (20.3%). Out of all the cases that were found to have bacterial infections, among whom about 77.1% females were infected in which 46.1% infections were caused by *E. coli*, 17.1% infections were reported to be caused by *S. aureus* and 17.1% were caused by *K. Pneumonia* in females (Table 2). Among males, the percentage rate of infection was very low in comparison with females in that about 19.3% of infection was observed, in which 3.2% were infected by *E. coli*, 3.2% by *K pneumonia* and 13.6% by *S. aureus*.

Strength of antibiotics was tested against three isolated bacteria in Table 3. Among antibiotics, Tigecycline (TGC) was the most effective antibiotic against all three isolated bacteria, in that about 100% of bacteria had shown the sensitivity towards it. *E. coli* was found to be the most resistant against tetracycline (TET) (66.6%) followed by Co-amoxiclav (AMC) (60%), Fosfomycin (Fo) (53.3%), Piperacillin (PIP) (53.3%), Cefepime (CPM) (53.3%).

**Table 1. Frequency distribution of patients according age and gender**

Age (year)	Total No. (%)	Female (%)	Male (%)
0-20 yrs	7 (14.5)	5 (10.4)	2 (14.1)
21-40 yrs	28 (57.4)	21(43.4)	7 (1.4)
41-60 yrs	8 (16.6)	2 (4.1)	6 (12.5)
> 60 yrs	5 (12.4)	2 (4.1)	4 (8.3)
Total	48 (100)	29 (60.4)	19 (39.5)

**Table 2. Frequency distribution of pathogen in patients according to gender**

Bacteria isolated	Total (%)	Female (%)	Males (%)
<i>E. coli</i>	15 (49.3)	14 (46.1)	1 (3.2)
<i>K. pneumonia</i>	6 (20.3)	5 (17.1)	1 (3.2)
<i>Staph. aureus</i>	9 (30.7)	5 (17.1)	4 (13.6)

**Table 3. Resistance shown by bacteria against antibiotics**

Antibiotics	<i>Escherichia coli</i> (n=15)	<i>Klebsiella pneumonia</i> (n=7)	<i>Staphylococcus aureus</i> (n=9)
Fosfomycin (FO)	8 (53.3%)	4 (57.1%)	2 (22.1%)
Norfloxacin (NX)	5 (33.3%)	2 (28.5%)	4 (44.3%)
Nitrofurantion (NIT)	5 (33.3%)	2 (28.5%)	4 (44.3%)
Colistin (CL)	7 (46.6%)	2 (28.5%)	4 (44.3%)
Piperacillin (PIP)	8 (53.3%)	4 (57.1%)	4 (44.3%)
Ceftriaxone (CTR)	7 (46.6%)	3 (42.8%)	3 (33.2%)
Cefepime (CPM)	8 (53.3%)	3 (42.8%)	5 (55.4%)
Cefazoline (CZ)	7 (46.6%)	4 (57.1%)	6 (66.5%)
Amoxiclav (AMC)	9 (60%)	2 (28.5%)	4 (44.3%)
Meropenem (MRP)	8 (53.3%)	1 (14.2%)	-
Etrapanem (ETP)	8 (53.3%)	2 (28.5%)	-
Gentamycin (GEN)	6 (40%)	4 (57.1%)	-
Amikacin (AK)	5 (33.3%)	2 (28.5%)	-
Levofloxacin (LE)	6 (40%)	1 (14.2%)	-
Tetracyclin (TET)	10 (66.6%)	4 (57.1%)	6 (66.5%)
Doxycyclin (DO)	7 (46.6%)	3 (42.8%)	3 (33.2%)
Tigecycline (TGC)	0 (0%)	0 (0%)	0 (0%)
Ethromycin (ERY)	-	-	2 (22.1%)
Chloremphenacol	-	-	3 (33.2%)
Clindmycin (CLI)	-	-	5 (55.4%)
Vancomycin (VAN)	-	-	2 (22.1%)
Ofloxacin (OF)	4 (26.6%)	4 (57.1%)	3 (33.2%)

(- Indicates that antibiotic was not used against that particular bacteria)

Among *Klebsiella pneumonia* isolates had shown resistance against tetracycline (TET) (57.1%), Fosfomycin (FO) (57.1%), Piperacillin (PI) (57.1%), Cephalexin (CZ) (57.1%) and Amikacin (AK) (57.1%). *Staphylococcus aureus* had shown maximum resistance against Tetracycline (TET) (66.5%), Amoxiclav (AMC) (66.5%) followed by Chloramphenicol (55.4%) and Cefepime (CPM) (55.4%).

#### 4. DISCUSSION

UTI is one of the most common bacterial infections these days, although other microbes such as fungal, viral pathogens can cause urinary tract infections but bacteria predominate over fungal and viral pathogens [9]. UTI encompasses a broad range of clinical fields that are associated with common findings of positive urine culture [10]. The resistance to antimicrobial has increased over the years resistance may vary from country to country [11]. In our study the

results showed that, out of total cases infection in females (60.4%) are found more as compared to, males (39.5%) anatomical and physical factor is the main reason for the higher infections rate in females [12]. Females that come under the age group 21 years to 40 years are at higher risk of being infected about 58% cases were reported in this study, which is in concordance with the findings of similar studies which were done by Khadri et al. [2], Oladeinde B H et al. [13] and Manjunath et al. [14].

The samples taken from urinary tract infected with microbes showed the presence of three types of bacteria which were *E. coli*, *Staph. aureus*, *K. pneumonia*. *E. coli* was the most common pathogen that cause for the urinary tract infection, which is in concordance with the findings of similar studies which were done by Kumar et al. [15]. It is important for the clinician to be aware of regional antibiotic resistance rates before initiating experimental antimicrobial

therapy for UTI treatment, as it well described that urinary infection with resistant pathogen leads to failure in treatment [16]. According to the Table 3 highest antibiotic resistance in *E. coli* against TET (66.6%) and AMC (60%). Antibiotics such as TGC (0%), OF (26.6%) found to be effective against *E. coli* isolates. As for *Staph. aureus* maximum resistance was found to be against TET (66.5%), CZ (66.5%), CPM (55.4%), Chloremphenicol (55.4%). Resistance by *K. pneumonia* was highest in TET (57.1%), GEN (57.1), CZ (57.1%) and FO (57.1%). We have found the Tigecyclin is the best drug for the treatment of UTI as shown in Table 3.

## 5. CONCLUSION

To conclude the prevalence rate of UTI in general practice is high among the females in reproductive age group. *E. coli* was found to be most common cause of urinary tract infection in all the age groups. Isolates showed the resistance towards commonly prescribed antibiotics such as Tetracyclin, Cefepime. More over study concludes that all the isolated bacterias were sensitive towards Tigecyclin as compared to other antibiotics tested and therefore this maybe the drug of choice for the treatment of UTI.

## CONSENT AND ETHICAL APPROVAL

All the authours here by declare that this study is approved by ethical clearance body of the organization, DNA-Labs a Centre for Applied Sciences, Dehradun, Utrakhand, India. This study has maintained strict standards for protecting the privacy and confidentiality of respondents during sample collection and data processing. As per international standard or university standard, patient's written consent has been collected and preserved by the authors.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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