

## PREVALENCE AND ANTIBIOTIC SUSCEPTIBILITY PATTERN OF GRAM NEGATIVE BACILLI ISOLATED FROM URINARY TRACT INFECTIONS

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**Abstract:** The study aimed to determine the prevalence and antibiotic susceptibility pattern of Gram-negative bacilli isolated from urinary tract infections. This cross-sectional study was conducted at the Department of Urology Lady Reading Hospital Peshawar from February 2021 to August 2023. Mid-stream urine samples were collected from individuals with urinary tract infection symptoms and processed and identified through various biochemical tests. Following CLSI recommendations, the isolated bacteria were screened for sensitivity to antibiotics. The Kirby-Bauer disc diffusion method was used in the current study. Through MS Excel, data were analyzed. The Chi-square test was applied to calculate the p-value (significant, i.e., <0.05) and antibiotic-resistant profiles of GNB isolated from urine samples. A total of 60000 urine samples were collected, of which 65% (n=3900) patients were outdoors, and 35% (n=2100) were hospitalized patients. Among them, 20 % (n=1200) were culture-positive. Among these, 58% of urine samples were from outdoor patients, and 42% were from hospitalized patients. *E. coli*, *Klebsiella pneumonia*, *P. aeruginosa*, and *Acinetobacter baumannii* were the Gram-negative bacilli (GNB) isolated in our study. The most prevalent (72%) GNB was *E. coli*. *Pseudomonas aeruginosa* isolated from indoor patients' urine samples showed high resistance. The antibiotic sensitivity pattern of *K. pneumonia* was the same as that of *E. coli* in outpatient and indoor patients. The *Acinetobacter baumannii* resistance pattern was relatively lower, observed at 85% in Nitrofurantoin (F), 80% in Ampicillin (AMP), and 11% in Fosfomycin (FOS), respectively, in urine samples of indoor as compared to outdoor patients. The most prevalent GNB observed in our study were *E. coli*, *Klebsiella pneumonia*, *P. aeruginosa*, and *Acinetobacter baumannii*, which were involved in UTIS. Antibiotics were chosen for its treatment: Fosfomycin, Piperacillin tazobactam, Cefoperazone-sulbactam, Amikacin, and Nitrofurantoin.

**Keywords:** Prevalence; Antibiotic Susceptibility Pattern; Gram-Negative Bacilli; Urinary Tract Infections

### Introduction

Urinary tract infections (UTIs) are among the most prevalent and life-threatening illnesses in the world, with over one hundred and fifty million cases of infection reported annually (Stamm and Norrby, 2001). Urinary tract infections, septicemia, and even noticeable kidney damage are among the fatal consequences and severe health issues that it can cause due to its effects on the prostate and urinary system (Flores-Mireles et al., 2015; Mathur et al., 2006). Since the urinary system has several mechanisms that prevent long-term colonization, it is usually clear of all bacteria. The defense mechanisms might be physiological (such as host immunity, mucus formation, and increased urea production) or mechanical (such as bladder emptying during urination, which washes out any present bacteria). Still, the inflammation interferes with regular urinary system function, which results in insufficient clearance of germs and, ultimately, the formation of UTIs. (Yenehun Worku et al., 2021). However, several risk variables, including gender, age, time spent in the hospital, being pregnant, insulin resistance, use of catheters for urinary purposes, and immunological or genitourinary tract

anomalies, influence the probability of getting the infection (Emiru et al., 2013; Odoki et al., 2019; Storme et al., 2019)—various pathogens, including viruses, fungi, and bacteria, frequently cause UTIs. The more frequent cause of this kind of illness is bacteria [1]. Previous data from studies suggests that Gram-positive bacilli cause ten percent of cases, whereas nearly ninety percent of cases are caused by Gram-negative bacteria. *E. coli* is commonly observed in the majority of cases of UTIs. However, the occurrence of infections such as urinary may also be aided by other urinary pathogens, such as *spp* of *Klebsiella*, *Staphylococcus* and *Streptococcus*, *Proteus*, and *Pseudomonas* (Karlowsky et al., 2006; Rowe & Juthani-Mehta, 2013). It is generally known that uropathogens are prevalent across the world (Odoki et al., 2019). Hospitals and communities are now at risk due to the rising prevalence of multi-drug resistance gram-negative bacilli related to UTI in Pakistan. This issue causes a daily increase in disease rates and fatalities—data on antibiotic susceptibility and resistance point to suitable management choices and tactics for MDR-GNB management. Reputable strains can transfer resistance genes to susceptible bacteria. Resistance may, therefore,

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spread from hospitals to local populations. Thus, this study aimed to identify GNB as the cause of UTIs based on its prevalence and in vitro antibiotic resistance pattern.

**Methodology**

This cross-sectional study was conducted at the Department of Urology Lady Reading Hospital Peshawar from February 2021 to August 2023. Mid-stream urine samples were collected aseptically from each outdoor and hospitalized individual with urinary tract infection symptoms (Graham & Galloway, 2001). All the samples were taken in sterilized bottles. With the help of rectified (1 µl) disposable plastic loops, all the samples were inoculated on Cystine Lactose Electrolyte Deficient (CLED) agar by using the streak plate method. These plates were incubated aerobically at 37°C for 18 to 24 hours. After bacterial growth, the colonies were counted as colony-forming units (cfu). 10<sup>4</sup> CFU/ml or more were included, and the count below was excluded from the study (Scarparo et al., 2002).

Colonies were selected from CLED media, and after gram staining, these were inoculated on various selective and deferential media for conformation on conventional methods (O'hara, 2005; Rasool et al., 2019). Based on CLSI recommendations, the isolated bacteria were screened for antibiotic sensitivity. The Kirby-Bauer disc diffusion method was used in the current study. The different antibiotics used in our study were Ampicillin (AMP), imimacin (AK), Amoxicillin-clavulanate (AMC), piperacillin-tazobactam (TZP), cefotaxime (CTX), cefoperazone+sulbactam (SCF), trimethoprim/sulfamethoxazole (SXT), nalidixic acid (NA), aztreonam (ATM), ciprofloxacin (CIP), fosfomycin (FOS), imipenem (IPM), ceftazidime (CAZ), nitrogenfurantoin (F), and polymyxin B (PB) similar to previous study (uk, 2001). MS Excel data were analyzed in percentages by determining the incidence of bacterial pathogens in the urine samples and their resistance. The Chi-square test was applied to calculate the p-value (significant, i.e., <0.05) and antibiotic-resistant profiles of GNB isolated from urine samples.

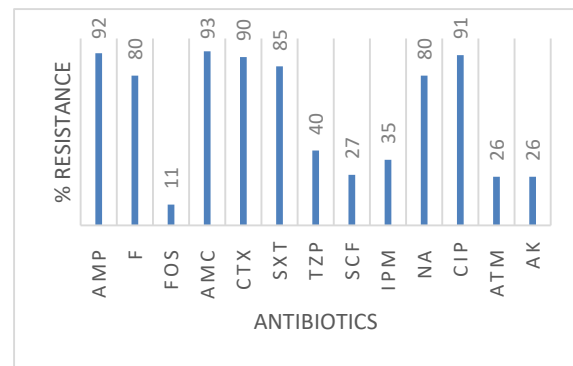
**Results**

A total of 60000 urine samples were collected, of which 65% (n=3900) patients were outdoors, and 35% (n=2100) were hospitalized patients. Among them, 20 % (n=1200) were culture-positive. Among these, 58% of urine samples were from outdoor patients, and 42% were from hospitalized patients. *E. coli*, *Klebsiella pneumoniae*, *P. aeruginosa*, and *Acinetobacter baumannii* were the Gram-negative bacilli (GNB) isolated in our study. The most prevalent (72%) GNB was *E. coli*. The prevalence of *E. coli* was 60% in outdoor patients and 40% in hospitalized patients. The prevalence of *K. pneumoniae* was similar to *E. coli*. *P. aeruginosa* was observed in 45% of outdoor and 55% of indoor patients. *Acinetobacter baumannii* was observed in 55% of outdoor and 45% of indoor patients (table 1).

**Table 1: Percentage of Gram-negative bacilli in hospitalized and outdoor patients of UTI**

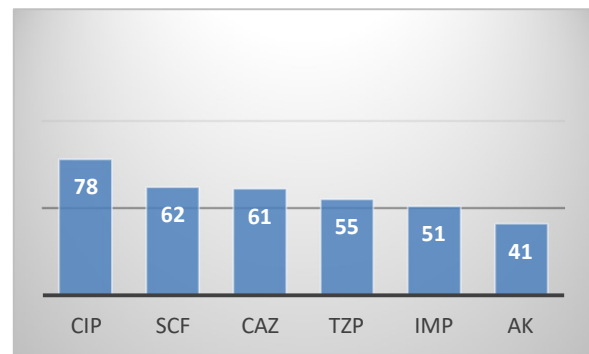
Gram -ve rod	Indoor patients %	In outdoor patients%	Total %
<i>E. coli</i>	40	60	100
<i>K. pneumoniae</i>	40	60	100
<i>P. aeruginosa</i>	55	45	100
<i>A. baumannii</i>	45	55	100

Antibiotic resistance was almost identical in outdoor and indoor UTI patients, excluding Cefotaxime (CTX). 78% of indoor patients' isolates of *E. coli* were resistant to cefotaxime (CTX), and 60% were outdoor patients. Resistance to Ampicillin was observed in 91% of *E. coli* isolates from indoor and outdoor patient samples. The antibiogram assay of *E. coli* is shown in Fig 1.



**Fig.1 Antibiotic susceptibility pattern of E.coli**

*Pseudomonas aeruginosa* isolated from indoor patients' urine showed high resistance. Ciprofloxacin 78%, Cefoperazone-sulbactam 62%, Ceftazidime 61%, Piperacillin-Tazobactam 55%, Imipenem 51% and Amikacin 41% in contrast to that of outdoor patients (fig 2).

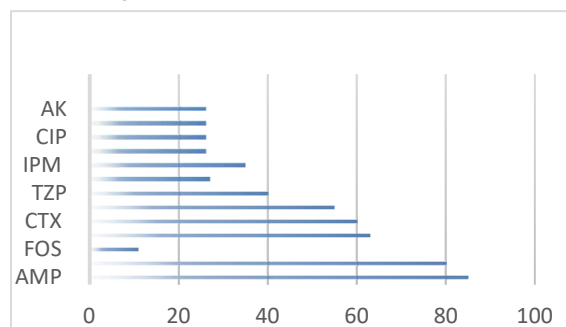


**Fig 2: Antibiogram of P.aeruginosa**

The antibiotic sensitivity pattern of *K. pneumoniae* was the same as that of *E. coli* in both outpatient and indoor patients. The *Acinetobacter baumannii* resistance pattern was relatively lower 85%, 80% 11% Ampicillin, Nitrofurantoin and Fosfomycin respectively in indoor urine

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samples compared to outdoor patient. Indoor urinary isolates were more resistant to Amoxicillin-clavulanate 63%, Cefotaxime 60%, Trimethoprim/sulfamethoxazole 55%, Piperacillin-tazobactam 40%, Cefoperazone-sulbactam 27% and Imipenem 35%, whereas around equal rates of resistance were noted in Nalidixic acid 26%, Ciprofloxacin 26%, Aztreonam 26% and Amikacin 26 % both in indoor and outdoor isolates from urine samples as shown in fig 3



**Fig no 3: Antibiotic resistance profiles of *Acinetobacter baumannii***

### Discussion

The high (20%) prevalence of urinary tract infections was explored in this study both in the out and indoor patients; therefore proper health protocols is the need of the day .This may be due to various factors specially due to stones in the kidneys or diabetes mellitus (Al-Badr and Al-Shaikh, 2013; Perla and Carifio, 2005).

In our study, a total of 60000 urine samples were collected in which 65% (n=3900) patients were outdoor and 35% (n=2100) were hospitalized patients. Among them 20 % (n=1200) were culture positive. Among which 58% urine samples were from outdoor patients and 42% samples were from hospitalized patients. *E.coli* (72%), *Klebsiella pneumonia* (13%), *P. aeruginosa* (12%) and *Acinetobacter baumannii* (2%) were the Gram negative bacilli (GNB) isolated in our study. The most prevalent (72%) GNB was *E. coli*. The prevalence of *E. coli* was 60% in outdoor patients and 40% in hospitalized patients. The prevalence of *K. pneumoniae* was similar to *E. coli*. *P. aeruginosa* was observed in 45% outdoor patients and was observed in 55% indoor patients. *Acinetobacter baumannii* was observed in 55% outdoor patients and in 45% of indoor patients. Same frequency of GNB in UTIs was detected in a study conducted in Khyber Pakhtunkhwa, (Ullah et al., 2018) in which the most prevalent was as *E. coli* followed by *Klebsiella* and *Pseudomonas. Aeruginosa* another carried out on UTIs reported *E. coli* (76.6%) (Shabbir et al., 1970). *Klebsiella pneumonia* was placed in the 2nd position of bacteria involved in UTIS as (40%) indoor and (60%) outdoor patients (Nizami et al., 1997). Among all four GNB *P. aeruginosa* was more prevalent in hospitalized patients of UTI but its prevalence was low in community these results were similar with the previous studies carried out in Iran and Pakistan (Noor et al., 2004; Shabbir et al., 1970). *E.coli* 91 % was resistance to Ampicillin, Cefotaxime,

Ciprofloxacin, Trimethoprim sulfamethoxazole and Aztreonam both in out and indoor patients. It may be due to misuse of antibiotics and this resistance is similar to previous findings (70-80%) (Noor et al., 2004; Shabbir et al., 1970; Ullah et al., 2018). *Klebsiella pneumonia* exhibited extraordinary resistance contours against imipenem, cefotaxime, trimethoprim-sulfamethoxazole, aztreonam, amoxicillin-clavulanate, and piperacillin-tazobactam. This might be the result of using some antibiotics frequently and expending them cautiously to cover *Klebsiella pneumoniae* in hospitals. These consequences were revealed too in the earlier reports (Abdullah et al., 2013; Shabbir et al., 1970). For *Pseudomonas aeruginosa*, Imipenem, Fosfomycin, Amikacin, Cefoperazone-sulbactam, and Piperacillin tazobactam are effective treatments for UTIs caused by these GNB. These results are in accordance with other studies (Livaditis and Gougoutas, 2011).

### Conclusion

The most prevalent GNB observed in our study were *E. coli*, *Klebsiella pneumonia*, *P. aeruginosa* and *Acinetobacter baumannii* involved in UTIS. The choice of antibiotics for its treatment were Fosfomycin, Piperacillin-tazobactam, Cefoperazone-sulbactam , Amikacin, and Nitrofurantoin.

### Declarations

#### Data Availability statement

All data generated or analyzed during the study are included in the manuscript.

#### Ethics approval and consent to participate

Approved by the department Concerned.

#### Consent for publication

Approved

#### Funding

Not applicable

### Conflict of interest

The authors declared absence of conflict of interest.

### Author Contribution

#### JAMIL AHMED KHAN (Assistant professor)

Conception of Study, Final approval of manuscript. Manuscript revisions, critical input.

#### KHUSHBOO QAMAR (Lecturer)

Coordination of collaborative efforts.

Data acquisition, analysis.

#### FARYAL GOHAR

Manuscript drafting.

#### JAMIL AHMAD

Data entry and Data analysis, drafting article.

#### SAIRA BANO

Data acquisition, analysis.

Coordination of collaborative efforts.

#### KHALID FAROOQ (Assistant professor)

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Coordination of collaborative efforts.  
 Study Design, Review of Literature.  
 The conception of the Study, Development of Research  
 Methodology Design, Study Design, manuscript Review,  
 and final approval of the manuscript.

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