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# ANTIBIOTIC RESISTANCE PROFILE OF UROPATHOGENIC *ESCHERICIA COLI* ISOLATED FROM URINARY TRACT INFECTED PATIENTS AT ESUT TEACHING HOSPITAL, PARKLANE, ENUGU STATE.

## ABSTRACT

Antibiotic resistance in Escherichia coli (E. coli), a predominant pathogen in urinary tract infections (UTIs), represents a critical public health concern, particularly for both immunocompromised and asymptomatic individuals. This study aimed to assess the antibiotic resistance profile of E. coli isolates obtained from UTI patients. A cross-sectional study design was employed, with ethical clearance obtained. Data were collected through structured questionnaires that assessed sociodemographic factors, UTI prevalence rates, and patient knowledge regarding UTIs. The study was conducted over a 6-month period, from June to November, 2024, at ESUT Teaching Hospital Parklane, Enugu State. A total of 250 urine samples were collected from consenting patients, of which 177 (70.8%) were diagnosed with UTIs. Uropathogenic Escherichia coli was identified using standard microbiological techniques. Antibiotic susceptibility testing was conducted using the Kirby-Bauer disk diffusion method, assessing sensitivity to commonly prescribed antibiotics. The age group 61-70 years (76, 30.4%) exhibited the highest proportion of UTI cases, while patients aged 41-50 years (35, 14%) and  $\leq$ 40 years (33, 13.2%) showed lower resistance to UTIs. Significant resistance was observed to third-generation cephalosporins, such as ceftriaxone (93.8%), and trimethoprimsulfamethoxazole (89.3%). Resistance was also noted to ciprofloxacin (53.4%) and gentamicin (54.6%). However, tetracycline demonstrated relatively lower resistance (41.0%). A notable

proportion of multidrug-resistant (MDR) strains (14.1%) was identified. This raises concerns about the available treatment options for UTI patients. The findings underscore the rising prevalence of antibiotic-resistant E. coli in UTIs, highlighting the urgent need for continuous surveillance, prudent antibiotic use, and the development of alternative treatment approaches to combat resistance in clinical settings.

Keywords: Antibiotics, resistance, Eschericia coli, urinary tract infection.

### **1.0 INTRODUCTION**

#### **1.1 BACKGROUND OF STUDY:**

Urinary Tract Infection (UTI) is one of the most common infections described among patients attending tertiary hospitals and is caused by the presence and growth of micro-organism within the genito-urinary tract system. It is a major cause of kidney damages with high morbidity and economic loss in both the hospital and community settings, affecting all age groups. This infection involves the lower urinary tract and could either be symptomatic or asymptomatic (Ochada et al., 2014). Patients who are said to be symptomatic are infected at the lower tract (acute cystitis) and upper tract (acute pyelonephritis) with significant bacteriuria affecting the bladder mucosa or pelvis. And asymptomatic bacteriuria, is a condition characterized by presence of bacteria in two consecutive clear voided urine specimens both yielding positive culture ( $\geq 105$  cfu/ml) of same uro-pathogen in the patient without classical symptoms. Majority of this infection are caused by retrograde ascent of bacteria from fecal flora to bladder and kidney through the urethra especially in females whom their urethra is shorter, wider and more susceptible to trauma during sexual intercourse example Escherichia coli, klebsiella spp, Proteus spp. This bacteria tends to navigate its way up the urethra then to the bladder especially during pregnancy and delivery. Studies have shown that despite the presence of some antibacterial factors in urine, like the pH, and urea concentration, the uro-pathogenic bacteria example the Eschericia coli are able to adhere, grow and resist effects against host immune defense mechanism resulting to colonization and infection of the uro-genitals. These symptoms of UTI are vague and may lead to muzzy diagnosis (Abejew et al., 2014). It includes; Dysuria (pain or

burning during urination), Increased frequency and urgency of urination, hematuria (blood in urine), Lower abdominal discomfort, Fever or chills, especially in severe cases (Nicolle, 2014).

However, the emergence of antimicrobial resistance in the management of urinary tract infections poses a serious challenge to public health in resource-limited countries due to lack of infrastructures, circulation of fake and use of un-prescribed antibiotics (Ayukekbong *et al.*, 2017). Also, this has made it pertinent to research into susceptibility profile of commonly used antibiotic at various intervals so to guide in clinical management of the infection. Hence the aim of this study was to determine the prevalence, and assess the patterns of antibiotic resistance in Escherichia coli (E. coli) strains isolated from patients with urinary tract infections (UTIs) at ESUT Teaching Hospital Parklane, Enugu State.

## **MATERIALS AND METHODS**

## 3.1: Study area and population:

This research utilized a cross-sectional study whereby a total of 250 urine samples were collected from consenting patients diagnosed with urinary tract infections at ESUT Teaching Hospital (ESUTH), Parklane, Enugu State. Ethical clearance was obtained from the ethics committee of Enugu State University Teaching Hospital before the commencement of the study, also a well structured questionnaire was used to collect demographic and clinical data from participants, including: Age, gender, their history in to urinary tract infection etc.

### **3.2 Sample collection method**

Freshly voided midstream urine samples, about 10ml were collected aseptically using wide mouthed sterile universal containers and was labeled with a reference code, age and sex of the patients. The samples were placed in a cold box for transportation to the laboratory, where they were stored for analysis. Each urine sample (10ml) were allowed to rest for 5 minutes, which their supernatants were discarded and their sediments used in microbial culture.

## 3.3: Laboratory procedures

Initially, MacConkey Agar was prepared and sterilized at 121 degree Celsius for 15 minutes and allowed to cool at 45-50 degrees. This sterilized already prepared Agar was poured onto a sterile culture plate (20ml per plate) and was allowed to solidify at room temperature. During this process, isolation of *Eschericia coli* was conducted following the method described by Prescott *et al.*, (2005). Thus, a sterile wire loop was deep into the sediments of the urine samples and inoculated onto the surface of the plate containing agar. This was incubated at 37 degree Celsius for 24hours and observed for growth. The presumptive colony of *Eschericia coli* from each cultured plates was further sub-cultured as isolates onto a nutrient Agar in a sterile culture plate, and identified using biochemical tests like catalase, lactose fermentation test etc, so as to obtain a pure culture for susceptibility.

## 3.3.1 The Antibiotic Susceptibility Test:

The principle of this states that bacterial growth culture on media can be inhibited, killed or not by exposure to specific antibiotics. This was performed using the standardized method (example Kirby-Bauer disc diffusion method) from the Clinical and Laboratory Standard Institute (CLSI) criteria on Mueller-Hinton Agar (MHA). The defining criteria for multi-drug resistance in the study was resistance to more than two antimicrobial agents of different chemical structure. The concentration of antibiotics used on disc were Ciprofloxacin (CPR, 5mcg), Trimethoprim-Sulphamethoxazole (TMP-SMX, 25mcg), Tetracycline (TE, 30mcg), Ceftriaxone (CTR, 30mcg), Gentamicin (GEN, 30mcg). However, the zones of inhibition on culture plate were measured and recorded using a standard calibrator as to determine the resistant, intermediate and sensitive isolate.

## RESULT

Variables	Frequency	Percentage (%)
Age Years		
-≤ 40	33	13.2
-41-50	35	14.0
-51-60	52	20.8
-61-70	76	30.4
->70	54	21.6
Gender		
-Female	115	46.0
-Male	135	54.0
Marital Status		
-Single	57	22.8
-Married	82	32.8
-Widowed	111	44.4
Educational Level		
-No formal Education	83	33.2
-Primary	76	30.4
-Secondary	53	21.2
-Tertiary	38	15.2
Occupation		
-Unemployed	98	39.2
-Employed	60	24.0
-Self-employed	92	36.8
Residence		
-Rural	103	41.2
-Urban	147	58.8
Sexual Activity (Last 10		
days)		
-Yes	67	26.8
-No	183	73.2

# Table 4.1: Sociodemographic Data (n = 250) Description

Table 4.1 presents the demographic characteristics of the respondents. Among the participants, the majority were male, comprising 135 (54%), while females accounted for 115 (46%). The highest proportion of respondents were aged 61–70 years (76 or 30.4%), followed by those aged >70 years (54 or 21.6%) and 51–60 years (52 or 20.8%). Respondents aged 41–50 years and  $\leq$ 40 years constituted smaller proportions, with 35 (14%) and 33 (13.2%), respectively.

Regarding marital status, the majority of respondents were widowed (111 or 44.4%), followed by married individuals (82 or 32.8%), while 57 (22.8%) were single. In terms of education, most respondents had no formal education (83 or 33.2%), followed by those with primary education (76 or 30.4%), secondary education (53 or 21.2%), and tertiary education (38 or 15.2%).

Occupationally, the majority were unemployed (98 or 39.2%), while 92 (36.8%) were selfemployed, and 60 (24.0%) were employed. Regarding residence, most respondents resided in urban areas (147 or 58.8%), while 103 (41.2%) were from rural areas. Lastly, concerning sexual activity in the last 10 days, a significant majority (183 or 73.2%) reported no sexual activity, while only 67 (26.8%) were sexually active.

Variables	Prevalence	
	N	%
Urinary Tract infected patients examined	250	100.0
Presence of Escherichia coli	177	70.8
Absence of Escherichia coli	73	29.2

## Table 4.2: Prevalence of *Escherichia coli* from Urinary Tract infected patients.

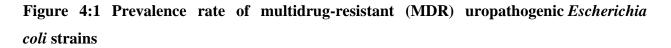
Table 4.2 shows the prevalence of *Escherichia coli* among urinary tract-infected (UTI) patients in the study population. Out of the 250 UTI patients examined, 177 (70.8%) had the presence of *E. coli*, while 73 (29.2%) showed its absence. These results underscore the significant role of *E. coli* as a predominant causative agent of UTIs in the study group.

Antibiotics	Susceptible	Resistance
Tetracycline	108 (61.0)	69 (41.0)
Ciprofloxacin	86 (48.6)	91 (53.4)
Gentamicin	81 (45.8)	96 (54.6)
Ceftriaxone	11 (6.2)	166 (93.8)
Sulfamethoxazole	19 (10.7)	158 (89.3)

 Table 4.2: Antibiotic Susceptibility Pattern of the Isolated Escherichia coli from Urinary

 Tract infected patients.

Table 4.2 shows the antibiotic susceptibility pattern of *Escherichia coli* isolates. The majority of isolates were resistant to ceftriaxone (93.8%) and sulfamethoxazole (89.3%), with very low susceptibility rates of 6.2% and 10.7%, respectively. Gentamicin and ciprofloxacin showed relatively higher resistance rates (54.6% and 53.4%, respectively), while their susceptibility rates were moderate at 45.8% and 48.6%. Tetracycline demonstrated the highest susceptibility among the antibiotics tested (61.0%), though 41.0% of isolates were resistant. These findings highlight significant resistance in *E. coli* isolates, particularly to ceftriaxone and sulfamethoxazole, which are commonly used antibiotics.



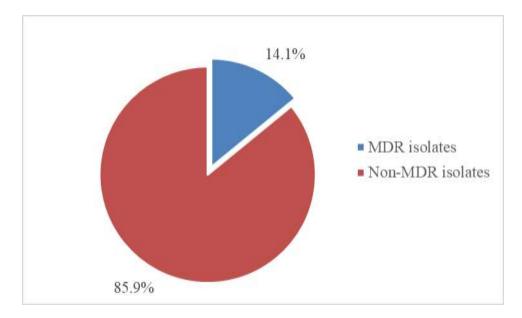


Figure 4.1 shows the prevalence rate of multidrug-resistant (MDR) uropathogenic *Escherichia coli* strains. The chart illustrates that 14.1% of the *E. coli* isolates were MDR, while the remaining 85.9% were non-MDR. This indicates a relatively low prevalence of MDR strains among the *E. coli* isolates from urinary tract infection (UTI) patients.

Figure 4.2: Prevalence Rate of *Escherichia coli* Among Urinary Tract Infected Patients by Gender

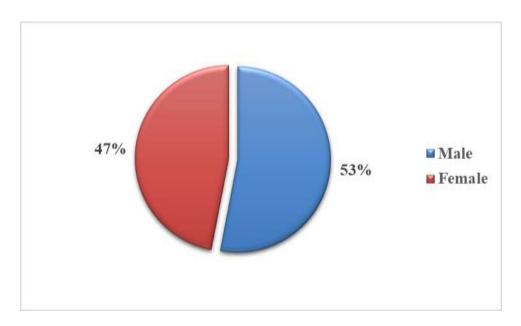


Figure 4.1 shows the prevalence rate of *Escherichia coli*-positive UTI cases by gender. The chart illustrates that 53% of *Escherichia coli*-positive UTI cases were males, while the remaining 47% were females. This indicates a higher prevalence of *E. coli* infections among males compared to females.

## DISCUSSION

The antibiotic resistance profile of E. coli in UTI patients is a dynamic and evolving issue. Escherichia coli (E. coli) on the other hand is the most predominant pathogen responsible for uncomplicated and complicated UTIs thus complicating treatment options and leading to increased morbidity and healthcare costs, responsible for approximately 70-90% of infections (Foxman, 2022; Sokhandan *et al.*, 2023). The rise of antibiotic-resistant uropathogenic *Eschericia coli* can be attributed to various factors, including the overuse and misuse of antibiotics, which selects for resistant strains. Mechanisms of resistance include the production of  $\beta$ -lactamases, particularly Extended Spectrum Beta-Lactamases (ESBLs).

This study determined the prevalence also evaluated the antibiotic resistance profile of E. coli from UTI patients at ESUT Teaching Hospital Parklane. The isolates which gave an overall prevalence of 70.8%, yielded 177 significant growth after culture in about 24 hours. However previous research by Farouk *et al.*, (2019) indicated higher prevalence rate, 88% and by Ajide *et al.*, (2016), lower prevalence at 62.67%. This research results also indicated that upec was high in age group 61-70 years by 30.4%, and low in patients less than or equal to 40 years old by 13.2%. Also, that there was a higher prevalence rate in males by 54.8%, than in females, 45.2%. This might be that because males may have a relatively weaker immune system, have an underlying health issues like diabetes, prostate cancer etc, possess a longer urethra compared to females, which might make it more difficult for antibiotics to reach the site of infection effectively or that the testosterone may suppress certain immune functions unlike the females. Though It has always been noted that mainly the females are highly prone to this infection probably because the female's urethra is short and that their rectum lies close to the urine outlet allowing bacteria to penetrate easily.

However, this study observed that the *E.coli* isolates were highly resistant to ceftriaxone (93.8%) and sulfamethoxazole (89.3%), with very low susceptibility rates at 6.2% and 10.7%, respectively. Gentamicin and ciprofloxacin also showed relatively higher resistance rates (54.6% and 53.4%, respectively), while their susceptibility rates were moderate at 45.8% and 48.6%. Tetracycline demonstrated the highest susceptibility among the antibiotics tested (61.0%), though 41.0% of isolates were resistant. Reasons for such an alarming resistance might be due to the role of patient to medicine store thus overuse and misuse of antibiotics, antibiotic use with

laboratory diagnosis, lack of policy commitment, poverty and infrastructural challenges. The limitations to this study were that only a limited range of antibiotics was tested. Also failure to account for underlying conditions like kidney stones and damages. Therefore these findings highlighted significant resistance in E. coli isolates, particularly to ceftriaxone and sulfamethoxazole, which are commonly used antibiotics.

## CONCLUSION

The findings of this study revealed that Escherichia coli remains the most common and prominent uropathogenic bacterial isolate in urinary tract infections (UTIs). Additionally, there is a mild increase in multidrug resistance among the isolates. Therefore, it is recommended that antibiotic susceptibility testing be routinely used to guide the selection of antibiotics for UTI management. The data from this study could serve as a baseline for ongoing surveillance of uropathogens. Furthermore, the development of novel antibiotics, alternative therapies, adjuncts, and the promotion of antimicrobial stewardship and education in both hospital and public settings should be prioritized.

## **ABBREVIATIONS**

CLSI - Clinical and Laboratory Standard Institute Protocol.

CFU - Colony Forming Unit.

E.coli - Eschericia coli.

ML - Milliliter.

MCG - Microgram.

UPEC - Uropathogenic Eschericia coli.

UTI - Urinary Tract Infection.

## REFERENCES

- Abejew, A.A., Denboba, A.A. and Mekonnen, A.G., 2014. Prevalence and antibiotic resistance pattern of urinary tract infections in Dessie Area, North East Ethiopia. *BMC Research Notes*, 7(1), 687.
- Abubakar, E.M., (2009). Antimicrobial susceptibility pattern of pathogenic bacteria causing urinary tract infections at the specialist hospital, Yola Adamawa State, Nigeria. *Journal of Clinical Medical Research*, 1(1), 1-8.
- Alabi, O.O. and Odukoya, O.O., (2020). Sample size determination in clinical research: Cochran's approach. *African Journal of Clinical and Experimental Microbiology*, 21(2), 122-130.
- Ayukekbong, J.A., Ntemgwa, M. and Atabe, A.T., (2017). The threat of antimicrobial resistance in developing countries: Causes and control strategies. *Antibiotics*, 6(1), 5.
- Aslam, B., (2023). Emerging trends in antibiotic resistance among uropathogenic *E. coli*: A global perspective. *Antibiotics*, 12(4), 1-12.
- Bello, M.B., (2022). Antimicrobial resistance patterns of *Escherichia coli* in urinary tract infections in Nigeria. *BMC Infectious Diseases*, 22(1), 211.
- Cohen, S.P., (2019). Plasmid-mediated antibiotic resistance in *Escherichia coli*. Journal of Antimicrobial Chemotherapy, 74(6), 1704-1715.
- Cosgrove, S.E., (2006). The impact of antimicrobial resistance on health and economic outcomes. *Clinical Infectious Diseases*, 42(1), 82-89.
- Dixon, D.M., (2018). Current trends in antibiotic resistance: A review. *Journal of Antibiotics*, 71, 452-469.
- Dulawa, J., (2003). Urinary tract infections. Annals of Academic Medical Bialostoc, 49,182-184.
- Foxman, B., (2022). Urinary tract infections: An overview of the epidemiology, diagnosis, and management. *Clinical Microbiology Reviews*, 35(1), 1-23.
- Gao, X., (2023). Molecular mechanisms of enterotoxigenic *E. coli* pathogenesis. *Microbial Pathogenesis*, 179, 105038.
- Gordon, D.M., (2022). The role of biofilm formation in uropathogenic *Escherichia coli* infections. *Clinical Microbiology Reviews*, 35(1), e00129-21.
- Hannan, T.J., (2010). Uropathogenic *Escherichia coli* and bladder infection. *Nature Reviews* Urology, 7(8), 389-396.
- Hernandez, C., (2018). Mechanisms of resistance in uropathogenic *Escherichia coli*. Journal of Clinical Microbiology, 56(6), e00045-18.
- Hooton, T.M., (2012). Urinary tract infections in women. *New England Journal of Medicine*, 366(1), 32-43.

- Hooton, T.M., (1996). A prospective study of risk factors for symptomatic urinary tract infection in young women. New England Journal of Medicine, 335, 468-474. American Family Physician, 96(7), 464-474.
- Johnson, J.R., (2023). Virulence factors of Uropathogenic *E. coli* and their role in urinary tract infections. *Infection and Immunity*, 91(7), e00145-23.
- Kahlmeter, G., Brown, D.F.J. and Goldstein, F.W., (2021). The European antimicrobial resistance surveillance network: Surveillance of antibiotic resistance in *E. coli* isolates from urinary tract infections. *Journal of Antimicrobial Chemotherapy*, 76(2), 424-431.
- Kumar, A., (2023). Mechanisms of antibiotic resistance in uropathogenic *E. coli*: Implications for treatment. *Journal of Medical Microbiology*, 72(2), 213-225.
- Kuhlmann, E.C., (2021). Antimicrobial stewardship in practice: The importance of antibiotic use. *Clinical Microbiology Reviews*, 34(3), e00013-21.
- Liu, Y., (2023). Virulence mechanisms of enterohemorrhagic *E. coli* and their implications for human health. *Frontiers in Microbiology*, 14, 1076241.
- Laxminarayan, R., (2013). Antibiotic resistance—the need for global solutions. *The Lancet Infectious Diseases*, 13(12), 1057-1098.
- Mulvey, M.A., (1998). Adherence of uropathogenic *Escherichia coli* to bladder epithelial cells is mediated by type 1 pili. *Proceedings of the National Academy of Sciences*, 95(9), 5105-5110.
- Nwanze, I.A., (2007). Urinary tract infection in Okada village: Prevalence and antimicrobial susceptibility pattern. *Science Research Essays*, 2, 112-116.
- Nicolle, L.E., (2014). Urinary tract infections in adults. *New England Journal of Medicine*, 370(4), 389-392.
- Nicolle, L.E. and Gupta, K., (2022). Antimicrobial resistance in *E. coli* and its role in urinary tract infections. *Current Opinion in Infectious Diseases*, 35(1), 23-31.
- O'Neill, J., (2014). Antimicrobial Resistance: Tackling a Crisis for the Health and Wealth of Nations. *Review on Antimicrobial Resistance*.
- Ofek, I., (2013). Adhesion of *Escherichia coli* to the urinary tract. *Nature Reviews Microbiology*, 11(2), 131-144.
- Patel, P.M., Boubetra, M. and Sharma, R., (2023). Emergence of carbapenem-resistant E. coli in healthcare-associated urinary tract infections: A rising threat. *International Journal of Antimicrobial Agents*, 61(3), 105307.
- Paterson, D.L. and Bonomo, R.A., (2005). Extended-spectrum beta-lactamases: A clinical update. *Clinical Microbiology Reviews*, 18(4), 657-686.
- Scholes, D., (2000). The incidence of urinary tract infection in women: A population-based study. *Archives of Internal Medicine*, 160(11), 1477-1482.

- Smith, J., (2022). Enteropathogenic *Escherichia coli* (EPEC): Mechanisms of intestinal infection. *Microbial Pathogenesis*, 158, 104274.
- Smith, J. and Doe, A., (2023). The prevalence and impact of antibiotic-resistant Uropathogenic *E. coli* in urinary tract infections. *Journal of Infectious Diseases*, 58(2), 134-142.
- Smith, S.W. and Johnson, A.P., (2022). Trends in antibiotic resistance in *E. coli* and their implications for UTI management. *Infection Control & Hospital Epidemiology*, 43(9), 1231-1238.
- Sokhandan, S., (2023). Antibiotic resistance patterns in uropathogenic *E. coli*: A review of recent studies. *Infection and Drug Resistance*, 16, 341-355.
- Sokhandan, S., Zeynali, M. and Moradi, S., (2023). Prevalence of fluoroquinolone resistance among E. coli isolates from outpatient and inpatient UTI cases in Iran. *Infection and Drug Resistance*, 16, 2349-2357.
- Van Gerven, N., (2013). The role of curli fimbriae in *Escherichia coli* biofilm formation. *Microbiology*, 159(4), 623-631.
- World Health Organization (WHO), (2019). Antimicrobial resistance: A global report on surveillance. *World Health Organization*.
- World Health Organization (WHO), (2021). Global antimicrobial resistance and use surveillance system (GLASS). *World Health Organization*.
- Zhang, L., (2023). Enteroaggregative *E. coli*: Pathogenesis and its role in chronic diarrhea. *Clinical Microbiology Reviews*, 36(2), e00268-22.