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Beta-Lactam Resistance Profile of *E. Coli* Isolated from Urinary Tract Infection Patients in Selected Hospitals within Gusau Metropolis

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ABSTRACT

The emergence of antibiotic resistance, particularly beta-lactam resistance, poses a significant challenge in the management of urinary tract infections (UTIs) caused by *Escherichia coli* (*E. coli*). This study aimed to investigate the beta-lactam resistance profile of *E. coli* isolated from UTI patients in selected hospitals within the Gusau metropolis. A total of 92 urine samples were collected from UTI patients attending selected hospitals in Gusau metropolis. Isolation and identification of *E. coli* were conducted using standard microbiological techniques. Antibiotic susceptibility testing was performed using the disc diffusion method according to Clinical and Laboratory Standards Institute (CLSI) guidelines. Furthermore, phenotypic confirmation of beta-lactam resistance was carried out using double disc synergy testing (DDST). Out of 92 urine samples obtained from patients with UTIs, *E. coli* was identified in 19 samples (20.7%), out of the 19 *E. coli* isolates, ESBL production was detected in 9 (47.36%) based on the results of the DDST, and among the *E. coli* isolates tested, 12 (63.16%) exhibited resistance to beta-lactam (Ceftriaxone), while 5 (26.31%) showed intermediate susceptibility, and 2 (10.53%) were susceptible to this antibiotic. Results revealed a concerning prevalence of beta-lactam resistance among *E. coli* isolates, highlighting the urgent need for effective antimicrobial stewardship and infection control measures in the region.

Keywords: UTIs, ESBL, *E. coli*, beta-lactam, Prevalence

1.0 INTRODUCTION

Urinary Tract Infections (UTIs) arise when bacteria, typically *Escherichia coli* (*E. coli*), invade the urinary system, encompassing the bladder, urethra, ureters, and kidneys. The most prevalent type of UTI affects the bladder and urethra. Symptoms include pelvic discomfort, a burning sensation during urination, cloudy or bloody urine, and a frequent urge to urinate (Foxman, 2014). UTIs are typically addressed with medications customized to suit the specific infection and patient profile, among which beta-lactam drugs are commonly prescribed.

Additionally, supportive measures such as pain management with ibuprofen or acetaminophen, increasing fluid intake, and maintaining basic hygiene practices are recommended (Nicolle, 2014).

Beta-lactam antibiotics, which encompass well-known medications like penicillin, cephalosporins, carbapenems, and monobactams, play a crucial role in combating bacterial infections. They work by disrupting the synthesis of bacterial cell walls, a process essential for their

survival, through binding to penicillin-binding proteins, pivotal enzymes involved in peptidoglycan formation. However, despite their effectiveness, the widespread and often inappropriate use of beta-lactam antibiotics has led to a significant challenge: the emergence of resistance against these drugs. This trend of antibiotic resistance poses a considerable global threat, highlighting the urgent need for judicious antibiotic prescribing practices and the development of alternative treatment strategies (Laxminarayan *et al.*, 2013). Bacterial resistance mechanisms, particularly the production of beta-lactamase enzymes that hydrolyze the beta-lactam ring, compromise the efficacy of these drugs. Notably, extended-spectrum beta-lactamases (ESBLs) and carbapenemases contribute to resistance against a wide spectrum of beta-lactams, including cephalosporins and carbapenems changes in penicillin-binding proteins (PBPs) within bacteria, as observed in methicillin-resistant *Staphylococcus aureus* (MRSA), contribute to decreased susceptibility of these microbes to beta-lactam antibiotics. Moreover, the rise of multidrug-resistant strains such as carbapenem-resistant Enterobacteriaceae (CRE) emphasizes the critical need to tackle beta-lactam resistance through various approaches. These include prudent antibiotic administration, implementation of infection control measures, and the investigation of innovative therapeutic alternatives (Paterson and Harris, 2016).

In Nigeria, as is the case worldwide, the escalating issue of beta-lactam resistance in UTIs is a cause for alarm. Factors such as the inappropriate use and excessive administration of antibiotics, coupled with insufficient infection control protocols, have fostered the emergence and spread of resistant bacterial strains. Research indicates the widespread presence of extended-spectrum beta-lactamase (ESBL)-producing bacteria, notably *Escherichia coli* and *Klebsiella pneumoniae*, in UTIs across Nigeria (Oluduro *et al.*, 2012). However, there is a scarcity of epidemiological data regarding the susceptibility of major bacteria, such as *E. coli*, to beta-lactam antibiotics in certain regions of the country, particularly in the Northwestern area. This research focuses on specific hospitals in the Gusau metropolis, the capital city of Zamfara

State in the Northwestern region. In this region, the burden of antibiotic resistance not only jeopardizes individual patient treatment outcomes but also presents a substantial public health concern. The aim of this study is to investigate the antibiotic susceptibility pattern of beta-lactam antibiotics in *E. coli*, shedding further light on their prevalence and variability.

2.0 MATERIALS AND METHOD

2.1 Sample Collection

This research was conducted among individuals diagnosed with urinary tract infection (UTI) who were receiving treatment at the General Hospital in Gusau, Zamfara State, Nigeria. A total of 92 midstream urine samples were collected using sterile, wide-mouthed, leak-proof universal urine containers. These samples were promptly transported to the Microbiology Laboratory at Federal University Gusau, Nigeria. Prior to sample collection, all patients provided informed consent in accordance with ethical guidelines and institutional protocols. They were comprehensively briefed on the objectives of the study, the procedures involved, as well as any potential risks and benefits. Furthermore, patients were assured of the confidentiality and anonymity of their data, and the voluntary nature of their participation was emphasized. Participation in the study was strictly voluntary, and patients had the freedom to withdraw at any point without any adverse effects on their medical treatment.

2.2 Isolation and Identification of *Escherichia coli*

The isolation and identification of *Escherichia coli* followed the protocol outlined by Abayneh *et al.*, (2018). In brief, each urine sample was gently mixed, and a 10ul (0.001 mL) aliquot was inoculated onto MacConkey agar (Oxoid, UK) before being incubated at 37°C for 24 hours. Colonies were initially screened based on their morphology, pigmentation, and Gram staining reaction. Subsequently, confirmation of colonies was achieved through biochemical tests including indole, methyl red, voges proskeur, citrate, urease, and TSI.

2.3 Preliminary Detection of ESBL Producing *Escherichia coli*

ESBL-producing *E. coli* strains were phenotypically screened following the Clinical Laboratory Standard Institute (CLSI) 2018 guidelines. The screening for ESBL production was conducted using standard disc diffusion methodology with a 30 µg Ceftriaxone disc (Oxoid, UK). Overnight cultures of *E. coli* were suspended in sterile normal saline and adjusted to a turbidity equivalent to 0.5 McFarland standards. Using a sterile cotton swab, the bacterial suspension was spread onto Mueller Hinton agar plates, and antibiotic discs were applied before incubating them at 37°C for 24 hours. ESBL production was suspected in isolates showing susceptibility to Ceftriaxone (≤ 27 mm).

2.4 Phenotypic Confirmation of ESBL Producing *Escherichia coli*

Phenotypically, suspected ESBL-producing *E. coli* strains were validated using the double disc

Table 1: Frequency Distribution of *Escherichia coli*

S/N	Number of Samples	Number of <i>E. coli</i> Positive	Percentage (%)
1.	92	19	20.7

3.2 Phenotypic Confirmation of ESBL Producing *Escherichia coli*

Out of the 19 *E. coli* isolates, ESBL production was detected in 9 (47.36%) based on the results of the DDST, while the remaining 10 (52.63%)

Table 2: Phenotypic result of ESBL production

Isolates ID	Ceftriaxone (30 µg) mm	Ceftriaxone (30 µg) + Sulbactam (10 µg) mm	Differences
EC.6	20	23	3
EC.9	0	0	0
EC.10	0	0	0
EC.18	23	28	5
EC.22	19	24	5

synergy test (DDST) following the CLSI, (2018) recommendations on Mueller Hinton agar. A combination disc of Ceftriaxone and Sulbactam (30 µg / 10 µg) was positioned at the center of the Mueller Hinton agar plate, with a Ceftriaxone disc (30 µg) placed 20 mm apart from the Ceftriaxone and Sulbactam combination disc. Following an incubation period of 24 hours at 37°C, the results were assessed for any increase in the diameter of the inhibition zone (≥ 5 mm) around the Ceftriaxone and Sulbactam combination disc, indicating ESBL positivity.

3.0 RESULTS

3.1 Bacterial Isolation

Out of the 92 urine samples obtained from patients with UTIs, *E. coli* was identified in 19 samples (20.7%) as shown in Table 1. The presence of *E. coli* was not observed in the remaining 73 samples (79.3%).

isolates did not exhibit ESBL production (Table 2). The DDST involves assessing a diameter increase of ≥ 5 mm in the zone of inhibition when Ceftriaxone and Sulbactam are present, indicating ESBL positivity.

EC.25	0	14	14
EC.26	0	13	13
EC.27	0	0	0
EC.28	11	22	11
EC.55	12	14	2
EC.58	0	9	9
EC.62	14	18	4
EC.63	0	13	13
EC.69	18	19	1
EC.70	15	20	5
EC.72	13	15	2
EC.77	0	0	0
EC.88	0	14	14
EC.92	15	8	3

3.3 Antibiotic Susceptibility Pattern of Ceftriaxone on *E. coli*

During the study, the antibiotic susceptibility of *E. coli* to Ceftriaxone alone was assessed, and the results are presented in Table 3. Among the *E. coli* isolates tested, 12 (63.16%) exhibited resistance to this antibiotic, while 5 (26.31%) showed intermediate susceptibility, and 2 (10.53%) were susceptible.

Table 3. Frequency Distribution of Antimicrobial Susceptibility Test for Ceftriaxone (30 µg) on *E. coli*

Organism	Susceptible	Intermediate	Resistance
<i>E. coli</i>	2 (10.53%)	5 (26.31 %)	12 (63.16%)

4.0 Discussion

The prevalence of ESBL-producing *E. coli* among UTI patients in Gusau, Nigeria, as revealed in this study, raises concerns about the escalating trend of antibiotic resistance in this region. The high prevalence of ESBL producers emphasizes the urgent need for robust antibiotic stewardship programs to optimize the use of antibiotics and prevent the further development and spread of resistant strains.

This study found *E. coli* in the urine samples of UTI patients at a rate of 20.7%. The findings are consistent with those of Simba *et al.* (2022), who reported a prevalence rate of 23.5% in Kenya. In

a separate investigation Oladeinde *et al.* (2011), the prevalence rate was found to be 39.69%, which is greater than what was revealed in the current study. Similarly, the findings of Odongo *et al.* (2020) in Uganda suggest a 10% prevalence rate of *E. coli* in UTI patients. The variance in prevalence rates can be attributable to changes in sample size Faber and Fonseca, (2014). They used a bigger sample size of 514 samples in their analysis.

The ability of ESBL-producing *E. coli* to hydrolyze extended-spectrum antibiotics like cephalosporins makes them challenging to treat and necessitates the use of alternative antibiotics,

often with higher costs and potential side effects Rawat and Nair, (2010). The results of the study reveal a significant prevalence of ESBL-producing *E. coli* among UTI patients, accounting for 47.36 % of the isolates. This finding is consistent with prior research in the field. For instance, a study conducted by Abalkhail *et al.* (2022) observed a similar prevalence of ESBL-producing *E. coli* in urinary tract infections among hospitalized patients in a tertiary care center. The congruence between our findings and those of Abalkhail *et al.* (2022) underscores the persistence of ESBL resistance mechanisms within *E. coli* populations, especially in clinical settings. An explanation for the high prevalence of ESBL-producing *E. coli* could result in the evolutionary dynamics of the bacterium. *E. coli* has the intrinsic ability to acquire and exchange genetic material, including plasmids carrying ESBL genes. The selective pressure exerted by the widespread use of antibiotics, particularly beta-lactams, has favored the survival and dissemination of ESBL-producing strains Ruotsalainen *et al.* (2020). Comparisons with other studies indicate that the prevalence of ESBL-producing *E. coli* can vary based on geographic location, healthcare practices, and patient demographics. Investigations by Perera *et al.* (2022) demonstrated a lower prevalence of ESBL-producing *E. coli* in community-acquired UTIs compared to hospital-acquired infections, highlighting the complex interplay between bacterial ecology and patient exposure.

This study highlights the concerning prevalence of ESBL-producing *E. coli* in UTI patients in Gusau, Nigeria, indicative of the escalating trend of antibiotic resistance in the region. The high proportion of ESBL producers underscores the urgent need for robust antibiotic stewardship programs to optimize antibiotic use and prevent

further spread of resistant strains. Our findings align with previous research, emphasizing the persistent challenge of ESBL resistance in clinical settings. Moving forward, continued surveillance and stringent infection control measures are essential to mitigate the spread of ESBL-producing strains and preserve the effectiveness of available treatment options. Additionally, further research into the molecular mechanisms of ESBL resistance and the development of novel antimicrobial strategies are warranted to address this growing public health concern effectively.

5.0 Conclusion

In conclusion, our research underscores the alarming dissemination of ESBL-producing *E. coli* among UTI patients in Gusau Metropolis, emphasizing the critical need for ongoing molecular surveillance efforts. These findings not only elucidate the evolving epidemiology and genetic diversity of ESBL genes but also carry significant implications for infection control practices, antibiotic therapy optimization, and the broader challenge of antibiotic resistance in our region. To effectively combat this growing public health threat, it is imperative that healthcare providers, policymakers, and researchers collaborate closely, implementing targeted interventions, stringent infection control measures, prudent antibiotic prescribing practices, and adopting a comprehensive "One Health" approach to safeguard the efficacy of antibiotics for both current and future generations.

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