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Research Article

Molecular Characterization of Beta-Lactam and Aminoglycoside Antibiotics Resistance Genes Present in *Escherichia coli* Isolated from Human Urine Samples

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ABSTRACT

Understanding the genetic pathways behind antibiotic resistance is critical for developing effective preventative and control methods. Furthermore, identifying and characterizing antibiotic resistance genes (ARG) can aid in the development of novel antimicrobial medicines that target these unique resistance pathways. The goal of this study is to look at the pattern of antibiotic resistance genes in *E. coli* isolated from human urine at the University of Port Harcourt Teaching Hospital, Nigeria. Pathogens were isolated and identified using Eosin methylene blue agar. The disc diffusion test was used to assess antimicrobial sensitivity. The identification of the resistance genes encoding beta-lactamases (MBLs, ESBLs, and AmpC) and aminoglycoside modifying enzymes (AME) was done by Polymerase Chain Reaction (PCR). Out of 31 biochemically identified *E. coli* isolates, 24 (77%) were identified as *E. coli* using PCR and the remaining 7 (23%) as different bacteria. *E. coli* demonstrated 100% resistance to cefotaxime, cefuroxime, and imipenem, 97% resistance to ampicillin and amoxicillin/clavulanic acid, 68% resistance to cefixime and 35% resistance to gentamycin using the disc diffusion technique. Other antimicrobial resistance (AR) traits were seen in fewer than 55% of the *E. coli* isolates. PCR screening revealed the presence of five genes among the isolates, namely blaCTX-M (26.9%), blaSHV (15.4%), blaEBC (23.1%), blaIMP (3.8%), and aac(3)I (30.8%). The aac(3)I gene was the most prevalent significantly (p-value < 0.001). The findings indicate a significant incidence of antimicrobial resistance in *E. coli* isolates, especially against certain antibiotics, indicating a possible issue with the efficacy of these medications in treating *E. coli* infections.

Keywords: Urine, *E. coli*, Beta-Lactam Antibiotics, Aminoglycoside Antibiotics, Resistance Genes

Introduction

The worldwide epidemic of bacterial illnesses has been increasing throughout the years, owing in large part to the development of resistance mechanisms by bacteria (Flores-Mireles *et al.*, 2015; Mehrad *et al.*, 2015). Due to its genomic flexibility, *E. coli* plays a role in the rapid development of antibiotic-resistance genes (Evans *et al.*, 2020). They can acquire, aggregate, and transmit antimicrobial resistance-related mobile genetic elements (MGEs), such as transposons and plasmids (Evans *et al.*, 2020; Xanthopoulou *et al.*, 2020).

To comprehend both the molecular mechanisms of resistance and the origins of their spread, it is imperative to investigate multidrug resistance (MDR) mechanisms and the sequencing of antibiotic resistance genes (ARGs) in this species (Blair *et al.*, 2015).

Since the development of antibiotic-resistant strains, the beta-lactam group of antibiotics has been used more frequently in the therapeutic therapy of *E. coli* infections than any other class of antibiotics (Thakuria and Lahon, 2013). The development of producers of extended-spectrum β-lactamase (ESBL), AmpC β-lactamase (AmpC), and metallo-β-lactamase (MBL) poses a major medical crisis and a threat to the public's health. Notably, the rising prevalence of MDR *E. coli* creates an unfavorable conundrum in treatment choices (Maurya *et al.*, 2016; Oberoi, 2013).

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The increased use or misuse of antibiotics in human health, agriculture, and veterinary medicine, as well as the transmissibility of MGEs, are important factors leading to the emergence of antibiotic-resistant bacterial infections (Cergole-Novella *et al.,* 2011). Specific mutations in genes like blaTEM, blaSHV, and blaCTX-M govern the phenotypic expression of ESBLs. The alterations change the amino acid structure of the encoded proteins, allowing the ESBLs to hydrolyze a broader range of β-lactam antibiotics, including penicillin and oxyimino-cephalosporins (Jena *et al.,* 2017). Another category of therapeutically relevant cephalosporinases, AmpC β-lactamases, cause drug resistance by a particular mutation in the bacterial chromosome or a transmissible material, mostly a plasmid (Oberoi, 2013). Chromosomal genes or heterologous genes acquired through horizontal gene transfer (HGT) encode MBLs. Considering the fact that MBL-mediated resistance in Enterobacteriaceae is a significant therapeutic issue, clinical data on its prevalence and genetic diversity are limited (Frost *et al.,* 2005). Amid many different types of resistance, data from throughout the world is still emerging on resistance to aminoglycoside antibiotics, which had previously been described as extremely effective treatments against life-threatening Gram negative bacterial infections (Shokravi *et al.,* 2015). Aminoglycosides primarily affect bacterial protein synthesis by binding to prokaryotic ribosomes via 16S ribosomal RNA (16S rRNA) and impacting the structure of the bacterial cell membrane (Mingeot-Leclercq *et al.*, 1999). Aminoglycoside resistance has been identified in bacteria that are both Gram-negative and Gram-positive, and the primary mechanisms that influence the potency of aminoglycoside drugs involve reduced drug absorption and/or accumulation in bacteria as well as the synthesis of aminoglycoside modifying enzymes (AMEs), which subsequently inactivate the drugs (Gad *et al.,* 2011).

The surveillance strategy comprises rigorous antibiotic resistance monitoring at the local, national, and global levels. The limited treatment options associated with infections caused by these ESBL-, AmpC-, MBL-, and AME-generating bacteria continue to lead to higher rates of mortality and morbidity as well as higher treatment costs (Zilahi *et al.*, 2016). We identified and characterized the chromosomal genetic determinants of ESBL, AmpC, MBL, and AME resistance in *E. coli* isolates from human urine.

Materials and Methods

Non-hospitalized patients with asymptomatic bacterumia visiting the clinical laboratory at the University of Port Harcourt Teaching Hospital (UPTH), Rivers State, Nigeria, were included in the research. Between October and December 2021, 100 consecutive urine samples were taken at random. The clean catch midstream urine samples were collected in sterile containers and transferred to the laboratory within half an hour of collection. The Ethical Committee at UPTH authorized this research. Following informed verbal consent, urine samples were taken from patients. Following that, the samples were examined for numerous indicators and parameters.

Identification of *Escherichia coli* and antimicrobial susceptibility testing

The bacteria were isolated as the first step using eosin methylene blue (EMB) agar. Isolates were identified primarily through Gram staining and standard biochemical tests such as citrate utilization, glucose and lactose fermentation in Kligler iron agar tubes, urease, and indole (Wauters and Vaneechoutte, 2015), as well as their colors and growth characteristics on chromogenic media. Phenotypic antimicrobial susceptibility testing (AST) was performed using the disc diffusion method against an antibiotic panel including amoxicillin-clavulanate (30 µg), ampicillin (10 µg), cefuroxime (30 µg), ceftriaxone (45 µg), cefotaxime (25 µg), cefixime (5 µg), imipenem/ cilastatin (10 µg), nalidixic acid (30 µg), gentamicin (10 µg), ofloxacin (5 µg), levofloxacin (5 µg), and nitrofurantoin (300 µg). Strains from the American Type Culture Collection (ATCC), including K. pneumoniae ATCC® 700603 and *E. coli* ATCC 25922, were used as quality controls. Results were interpreted according to the Clinical and Laboratory Standards Institute (CLSI) guidelines (Wayne, 2010).

Genotypic detection of *E. coli* isolates.

Extraction of DNA - The DNA extraction kit approach was successful and effective in the extraction of pure and intact DNA. The isolated DNA was electrophoresed on a 1% Agarose gel, stained with ethidium bromide, electrophoresed at 70 volts for 1 hour, and photographed using an ultraviolet (UV) transilluminator.

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PCR reaction

The 16S rRNA, blaIMP, blaVIM, blaNDM, blaOXA-48, blaCTX-M, blaSHV, blaEBC, blaCIT, and aac (3)I genes were amplified using specific primers in a PCR reaction mix (Table 1). The PCR assay was carried out in a total volume of 10 µl of mixture, which included 2 µl master mix, 2 µl DNA sample, 0.3 µl forward primer, 0.3 µl reverse primer, and 5.4 µl ddH2O. Table 2 shows the PCR timetable program for the test genes. The PCR products were then electrophoresed in a 2% agarose gel stained with ethidium bromide and viewed under a UV trans-illuminator (Ajuga *et al.*, 2021).

Polymerase Chain Reaction (PCR) Mix

PCR reaction mix of Phase, temperature, time and runs is as follows Initial denaturation**,** 94ºC**,** 5 mins and 1Run; denaturation, 94ºC**,** 30 sec., and 35Run; annealing, 56ºC**,** 30 sec., extension 72ºC**,** 1 mins., Final extension, 72ºC**,** 5 mins and 1 Run.

Primer sequences and Polymerase chain reaction (PCR) condition for detection of antimicrobial resistance

The resistance enzymes of isolated *E. coli* and their respective resistance in parenthesis ( ) are as follows; MBLs **(**blaNDM**,** blaIMP**,** blaVIM**,** blaOXA-48); ESBL **(***blaCTX-M***,** blaSHV); AmpC **(**blaCIT**,** blaEBC), and AME **(***aac(3)-1*). The resistance genes of the specific resistance enzymes for antimicrobial resistance, were amplified by employing the primer sequences, specific annealing temperatures and predicted product sizes in base pairs (bp) of the amplified products of Liang *et al.* (2018), Sadeghi *et al.* (2021), Rahman, *et al.* (2020), Sadeghi *et al.* (2021), Sadeghi *et al.* (2021), and Zhao *et al.* (2018), respectively, for the targeted genes.

Statistical Analysis

For the purposes of descriptive statistical analysis, all data regarding the presence or absence of the investigated factors in each strain were imported into the SPSS (SPSS 19; IBM) program as binomial variables, and prevalence, 95 % confidence level, and P value were calculated.

Result

In the present study, 54 (54%) positive growth strains were isolated from 100 urine samples evaluated (Figure 1). Of the 54 positive bacterial growths, 31 (57%) were biochemically confirmed *E. coli* (Figure 2). A total of 24 (77%) were molecularly confirmed as *E. coli* via PCR (Figure 3). Of this, 14 (58%) of the *E. coli* isolates possessed at least one of the test resistance genes (Figure 4).

The most prevalent antimicrobial resistance phenotype was against most of the antibiotics, such as 100% resistance to cefotaxime, cefuroxime, and imipenem; 97% resistance to ampicillin and amoxicillin/ clavulanic acid; 68% resistance to cefixime; and 35% resistance to gentamycin. The other antimicrobial resistance (AR) phenotypes were observed in less than 55 % of the *E. coli* isolates (Table 1). Five out of the nine tested resistance genes were detected in these molecularly confirmed isolates (Figure 5).

These isolates exhibited MDR to at least 3 classes of the tested antibiotics (Figure 7) and a minimum MAR index of 0.3 (Figure 6), meaning they were resistant to a minimum of 3 out of the 12 antibiotics used. Furthermore, the presence of the blaCTX-M and blaEBC genes was also correlated with a higher rate of β-lactam resistance in the *E. coli* isolates. In addition, other resistance genes were detected in some isolates, indicating a complex pattern of resistance in *E. coli*.

Twelve phenotypic AR patterns were recognized in this study. No significant difference was observed in the prevalence of AR patterns (p-value > 0.05); the frequency of the patterns was in the range of 1 and 2 (Table 2). All the RG-positive isolates (n = 14), five of which were phenotypically multidrug resistant.

Also, all of the RG-positives harbored at least one of the screened resistance genes; twelve different AR gene profiles were identified, of which aac (3)I was the most prevalent significantly (p-value < 0.001) (Table 3).

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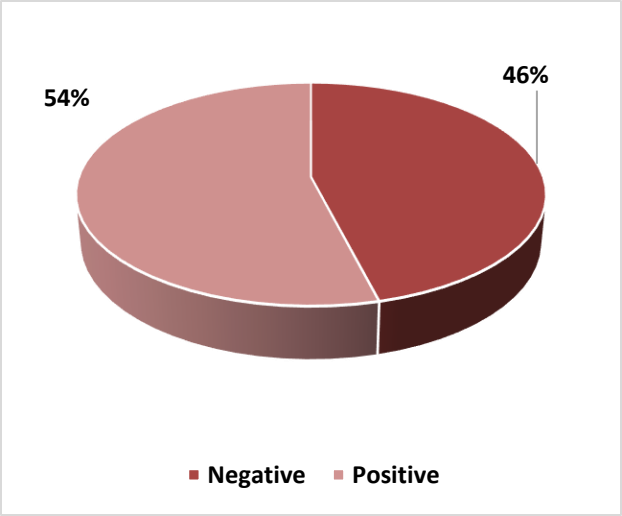

Figure 1: Presence and absence of bacterial growth

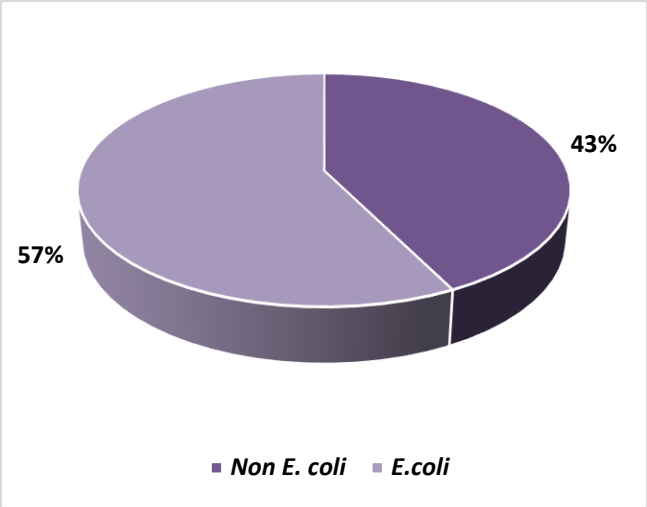

Figure 2: Biochemically confirmed *E. coli*

Table 1: Prevalence of phenotypic antibiotic resistance (n = 31)

Class of antibiotics	Number	%
Cephalosporins		
Ceftriaxone	15	48
Cefotaxime	31	100
Cefuroxime	31	100
Cefixime	21	68
Penicillin		
Ampicillin	30	97
Amoxicillin clavulanate	30	97
Carbapenem		
Imipenem/cilastatin	31	100
Aminoglycoside		
Gentamycin	11	35
Fluoroquinolones		
Levofloxacin	9	29
Ofloxacin	6	19
Nalidixic acids	17	55

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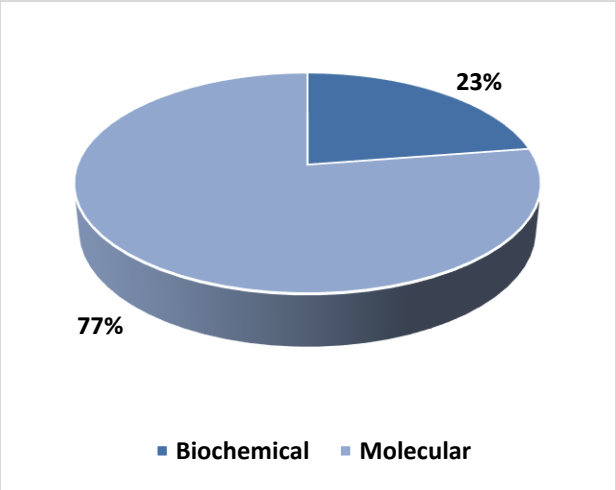

Figure 3: Molecularly confirmed *E. coli* using PCR technique

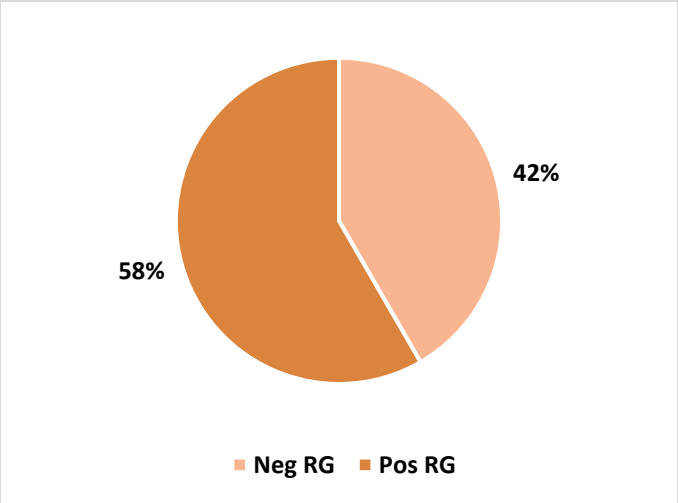

Figure 4: Confirmed *E. coli* isolates having the tested resistance genes (RG)

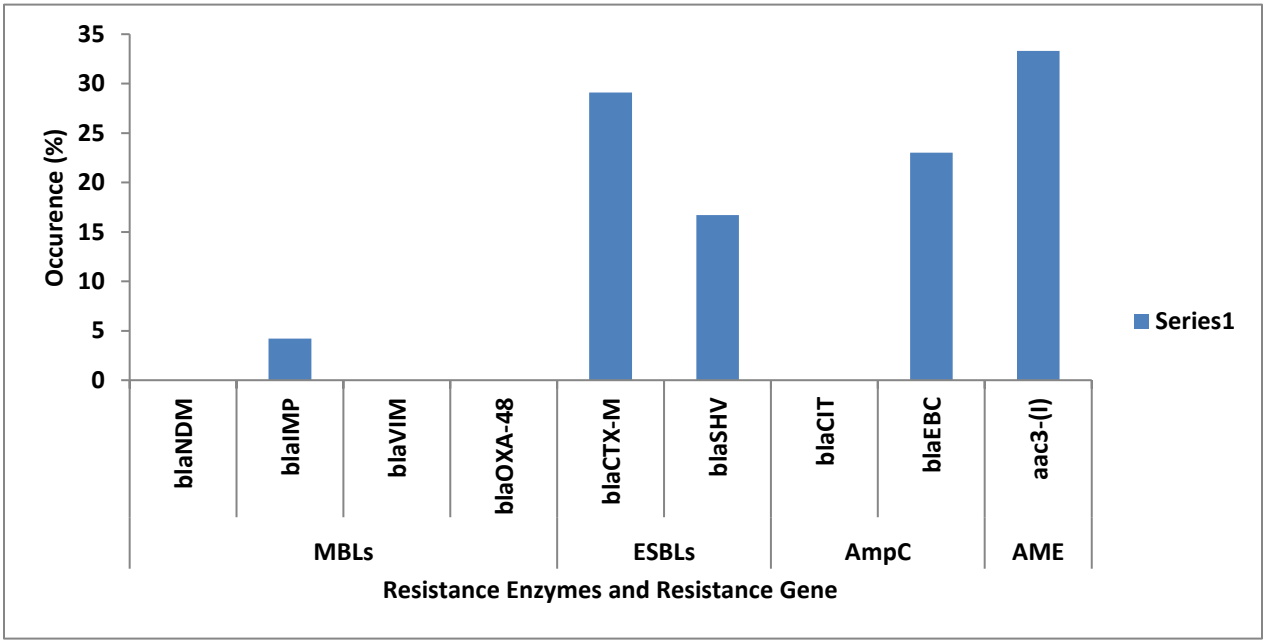

Figure 5: Prevalence of molecular antibiotic resistance genes in PCR confirmed *E. coli* (n = 24)

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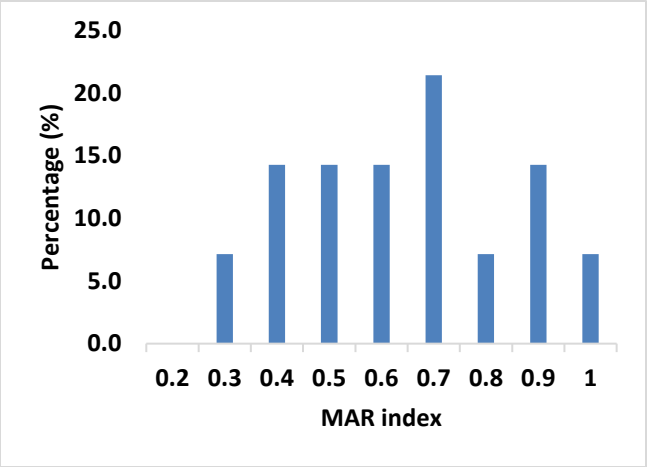

Figure 6: MAR index of the isolates having resistance genes (n = 14)

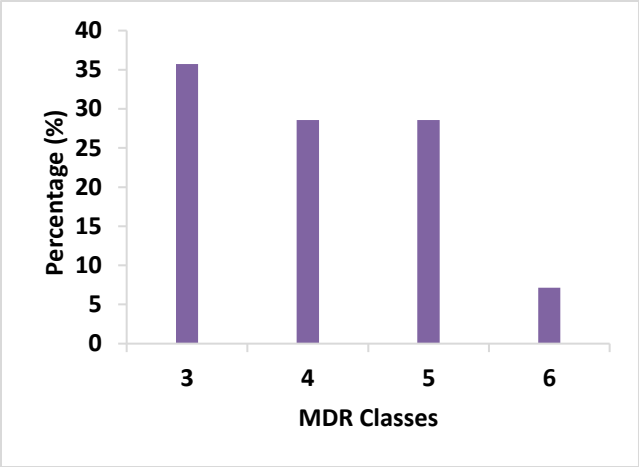

Figure 7: Number of MDR classes of *E. coli* isolates having the test resistance genes (n = 14)

Key: 3 (Cephalosporins-Penicillin-Carbapenem); **4** (Cephalosporins-Penicillin-Carbapenem-Aminoglycoside/Fluoroquinolone/ Nitrofurantoin); **5** (Cephalosporins-Penicillin-Carbapenem-Fluoroquinolones-Aminoglycoside); **6** (Cephalosporins-Penicillin-Carbapenem- Fluoroquinolones-Aminoglycoside- Nitrofurantoin)

Table 2: Phenotypic antibiotic resistance profile among *E. coli* isolates

	Classes of Drugs	Resistance pattern	Frequency	Percentage (%)
1	CEPH-CARB-PEN	CTX-CXM-AUG-IMP	1	7.1
2		CTX-CXM-AUG-ACX-IMP	1	7.1
3		CTX-CXM-ZEM-AUG-ACX-IMP	2	14.3
4	CEPH-CARB-PEN-FLUORO-	CTX-CXM-AUG-ACX-IMP-NA	1	7.1
5		CRO-CTX-CXM-ZEM-AUG-ACX-IMP-NA	2	14.3
6		CTX-CXM-ZEM-AUG-ACX-IMP-NA-LBC	1	7.1
7		CTX-CXM-AUG-ACX-IMP-NA	1	7.1
8	CEPH-CARB-PEN-AMIN-FLUORO	CRO-CTX-CXM-ZEM-AUG-ACX-IMP-OFX- NA-LBC-GN	2	14.3
9		CTX-CXM-AUG-ACX-IMP-NA-LBC-GN	1	7.1
10		CRO-CTX-CXM-ZEM-AUG-ACX-IMP-OFX- NA-LBC-GN	1	7.1
11	CEPH-CARB-PEN-FLUORO-NF	CRO-CTX-CXM-ZEM-AUG-ACX-IMP-OFX-LBC-NF	1	7.1
12	CEPH-CARB-PEN-FLUORO-AMINO-NF	CRO-CTX-CXM-ZEM-AUG-ACX-IMP-OFX- NA-LBC-GN-NF	1	7.1

Key: CRO-ceftriaxone sulbactam; **CTX**-cefotaxime; **IMP**-imipenem/cilastatin; **AUG**-augmentin; **OFX**-ofloxacin; **GN**-gentamycin; **NA**-nalidixic acid; **NF**-nitrofurantoin; **CXM**-cefuroxime; **ACX**-ampiclox; **ZEM**-cefixime; **LBC** -levofloxacin; **CEPH**-cephalosporins; **PEN**-penicillin; **FLUORO**-fluoroquinolones; **AMINO**-aminoglycoside; **NF**-nitrofuran; **CARB**-carbapenem.

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Table 3: Distribution and co-occurrence of resistance genes profile

	Resistance Enzymes	ARG	No	%
1	MBLs	blaIMP	0	0.0
2	ESBLs	blaSHV	1	7.1
3		blaCTX-M	2	14.3
4	AmpC	blaEBC	1	7.1
5	AME	aac (3)I	1	7.1
6	ESBL + AME	BlaCTX-M + aac (3) I	1	7.1
7		blaSHV + aac (3)-I	2	14.3
8	AmpC + AME	blaEBC + aac (3) -I	2	14.3
9	ESBL + AmpC	BlaCTX-M + blaEBC	1	7.1
10		BlaCTX-M + blaSHV + blaEBC	1	7.1
11	ESBL + AmpC + AME	BlaCTX-M + blaEBC + aac (3)-I	1	7.1
12	MBLS + ESBL + AME	BlaIMP +blaCTX-M + aac (3)-I	1	7.1

Discussion

Antibiotic resistance is becoming a worldwide concern with far-reaching economic effects. A common source in the population can lead to the widespread diffusion of resistant *E. coli* strains in community and hospital settings. The rate of *E. coli* antibiotic resistance is thought to fluctuate based on sample type and geographic location, making it critical for healthcare workers to monitor. Urinary tract infections (UTIs) caused by *E. coli* are one of the most prevalent reasons for hospitalization (Flores-Mireles *et al.,* 2015). Most illnesses, including cholecystitis, bacteremia, cholangitis, and diarrhea, have been linked to this bacterium (Dong *et al.,* 2018).

The prevalence of *E. coli* isolated from urine in the current investigation was 31 (62%). This prevalence is higher than the reported prevalence's of 25.5% in Ondo (Oladoja and Onifade, 2015) and 23.5% in Minna (Iseghohi *et al.,* 2020), but lower than the reports of Idakwo *et al.* (2015), who recorded 32.5% in Minna, Niger State, and 33.3% in Lagos (Adenipekun *et al.,* 2016). Although *E. coli* is a typical part of the human body's flora, the incidence in this study is alarmingly high, which is concerning because *E. coli* is a primary source of community and opportunistic infections in people (Allocati *et al.,* 2013). This means that the risk of infection may be increased among individuals in the study population.

The presence of *E. coli* in the urine was found in 24/100 (24%) of these healthy individuals (HI). Asymptomatic bacteriuria (ABU) is a symbiotic condition in which people carry more than 105 CFU/ml of bacteria without experiencing any symptoms (Salvador *et al.,* 2012). The virulent uropathogenic *E. coli* strains that gave rise to its prototype (ABU strain *Escherichia coli* 83972) possessed substantial mechanisms of bacterial adaptation that allowed them to develop a smaller genome than uropathogenic *E. coli* (UPEC) strains. This UTI *E. coli* strain is thought to have originated from these virulent uropathogenic *E. coli* strains. An analysis of the bacterium revealed point mutations or deletions in the majority of the bacteria's virulence genes. This demonstrates that ABU strains undergo evolutionary modification inside their hosts (Zdziarski *et al.,* 2008). ABU strains may develop in 2–20% of the population within a period of weeks to years, and they are impacted by variables including gender and age (Salvador *et al.,* 2012).

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The urine samples contained biochemically verified *E. coli* isolates that were 100% resistant to cefotaxime, imipenem, cefuroxime, and augmentin; 97% to ampiclox; 68% to cefixime; 55% to nalidixic acid; 49% to ceftriaxone; 35% to gentamycin; 29% to levofloxacin; 19% to ofloxacin; and 16% to nitrofurantoin. According to Okafor and Nweze (2020), *E. coli* has limited resistance to ofloxacin (22%), nitrofurantoin (6%), and cefuroxime (100%), and significant resistance to augmentin (100%).

Additionally, the isolates in this study's analysis had gentamicin resistance rates of 35%, which is close to the 34% reported by Okafor and Nweze (2020). Adenipekun *et al.* (2016) found poor resistance of *E. coli* to cefuroxime (8.1%) and gentamycin (7.7%), which is in contrast to our findings. Contrary to this study, Isegohi *et al.* (2020) found resistance patterns to gentamicin (25%), cefuroxime (80%), and ofloxacin (17.5%), but a high sensitivity to cefotaxime (80%).

PCR verified 24 (77%) of the 31 (62%) biochemically confirmed *E. coli*. Other bacteria made up the remaining 7 (23%) of the microorganisms. 14 (58%) of the 24 isolates were positive for at least one of the screening RGs. The remaining ten (42%) had none of the screening RGs. One of these isolates was resistant to 11 of the 12 antibiotics tested, and all of them had a multiple antibiotic resistance index (MARI) of 0.3. In 100% of the isolates, there was consistent resistance to three kinds of antibiotics (cephalosporins, penicillin, and imipenem). This study demonstrates a large resistant pool in the study environment as well as the likelihood of beta-lactamase synthesis in the tested *E. coli* isolates. The work done by Ochada *et al.* (2015) adds to the evidence that *E. coli* from urine exhibits a pattern of resistance based on the drugs examined. Resistance to all six antibiotic classes assessed in two isolates from separate individuals could suggest a plasmid profile expressing diverse resistance genes or biofilms. ESBLs and AmpC-producing isolates, on the other hand, have been shown to develop resistance to a wide range of antibiotics widely used in the environment.

As previously noted, drug resistant isolates are generally evaluated using molecular genotyping and phenotyping techniques, which are used in screening and to validate the expression of antimicrobial drug resistance genes within a population (Alyamani *et al.*, 2017).

This was done on verified *E. coli* to determine the presence of MBLs, ESBLs, AmpC, and AME resistance enzymes, which are known to be among the top causes of mortality globally among UTI patients due to the presence of multidrug resistance and virulent genes (Ramadan *et al.*, 2019). During this investigation, 100% of the urine samples (24% of the total) demonstrated the phenotypic capacity to manufacture beta-lactamase enzymes and amino-glycoside modifying enzymes. An examination of these isolates reveals that 50% (7) [i.e., 29.1% of the total *E. coli*] have the blaCTX-M gene; 29% (4) [i.e., 16.7%] have the blaSHV gene; 57% (8) [i.e., 33.3%] have the aac (3)I gene; 42% (6) [i.e., 23%] have the blaEBC gene; 7.1% (1) [i.e., 4.2%] had the blaIMP gene, while none of the bacteria had the blaNDM, blaVIM, blaOXA-48, and blaCIT genes. Further empirical research revealed that all 14 isolates expressing these enzymes were multidrug resistant and had MAR indices greater than 0.3. In total, 3.8% (1/24) MBLs, 42.3% (11/24) ESBLs, 23% (6/24) AmpC, and 33.3% (8/24) AME enzyme producers were found. The coexistence of these RGs was also detected in several isolates: 35.7% (5) of the isolates had a combination of two RGs, and 21.4% (3) of the isolates had a combination of three RGs. The coexistence of multiple types of resistance genes is a concerning issue in terms of antibiotic resistance, particularly in seemingly healthy individuals, because it can result in fewer treatment options, an increased risk of infection, antibiotic resistance transmission, complications in routine procedures, diagnostic challenges, and overall public health concerns. Overall, the presence and persistence of several resistance mechanisms in E. coli from our research population indicates the possibility of highly resistant bacteria spreading throughout the community and emphasizes the critical need for effective ways to restrict their transmission.

In conclusion, the genetic determinants of β-lactam and aminoglycoside resistance in *E. coli* from human urine samples are prevalent. The occurrence of various AR gene profiles in the population implies a varied spectrum of resistance mechanisms.

These resistance mechanisms may contribute to the population's high prevalence of aac (3)I, blaEBC, and blaCTX-M. The analysis calls for continual genomic monitoring of resistance trends and improved infection management of these pathogens in hospital settings.

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References

Adenipekun, E. O., Jackson, C. R., Ramadan, H., Iwalokun, B. A., Oyedeji, K. S., Frye, J. G., Barrett, J. B., Hiott, L. M., Woodley, T. A., and Oluwadun, A. (2016). Prevalence and multidrug resistance of *Escherichia coli* from community-acquired infections in Lagos, Nigeria. *Journal of infection in developing countries*. *10(9)*: 920–931.

Ajuga, M. U., Otokunefor, K., and Agbagwa, O. E. (2021). Antibiotic resistance and ESBL production in *Escherichia coli* from various sources in Aba metropolis, Nigeria. *Bulletin of the National Research Centre*. *45*(1):173.

Allocati, N., Masulli, M., Alexeyev, M. F., and Di Ilio, C. (2013). *Escherichia coli* in Europe: an overview. *International journal of environmental research and public health*. *10(12)*: 6235–6254

Alyamani E. J., Khiyami A. M., Booq R. Y., Majrashi A., Bahwerth F. S., and Rechkina E. (2017). "The occurrence of ESBLproducing *Escherichia coli* carrying aminoglycoside resistance genes in urinary tract infections in Saudi Arabia." *Annals of Clinical Microb. and Ant*. *16(1)*:1–13.

Blair, J. M., Webber, M. A., Baylay, A. J., Ogbolu, D. O., and Piddock, L. J. (2015). Molecular mechanisms of antibiotic resistance. *Nature reviews microbiology*. *13(1)*: 42-51.

Cergole-Novella, M. C., Pignatari, A. C. C., Castanheira, M., and Guth, B. E. C. (2011). Molecular typing of antimicrobial-resistant Shiga-toxin-producing *Escherichia coli* strains (STEC) in Brazil. *Research in microbiology*, *162(2)*: 117-123.

Dong, S.L., Seung-Ju, L. and Hyun-Sop Choe (2018). Community-Acquired Urinary Tract Infection by *Escherichia coli* in the Era of Antibiotic Resistance. Biomed Research International, *1*: 1-15.

Evans, D. R., Griffith, M. P., Sundermann, A. J., Shutt, K. A., Saul, M. I., Mustapha, M. M., ... and Van Tyne, D. (2020). Systematic detection of horizontal gene transfer across genera among multidrug-resistant bacteria in a single hospital. *Elife*. *9*: e53886.

Flores-Mireles, A. L., Walker, J. N., Caparon, M., and Hultgren, S. J. (2015). Urinary tract infections: epidemiology, mechanisms of infection and treatment options. *Nature reviews microbiology*. *13(5)*: 269-284.

Frost, L. S., Leplae, R., Summers, A. O., and Toussaint, A. (2005). Mobile genetic elements: the agents of open-source evolution. *Nature Reviews Microbiology*. *3*(9): 722-732.

Gad, G. F., Mohamed, H. A., and Ashour, H. M. (2011). Aminoglycoside resistance rates, phenotypes, and mechanisms of Gram-negative bacteria from infected patients in upper Egypt. *PloS one*. *6(2)*: e17224.

Idakwo S.O, Mawak J.D, and Abalaka M.E (2015). Prevalence and Antibiogram of Urinary Tract Infections in Patients Attending Hospitals in Minna, Niger State. International Journal of Microbiology, Biochemistry, Molecular Biology. *1(1)*: 8-12.

Iseghohi, F., Igwe, J. C., Galadima, M., Kuta, A. F., Abdullahi, A. M., and Chukwunwejim, C. R. (2021). Prevalence Of Extended Spectrum Beta-Lactamases (ESBLs)-Producing *Escherichia coli* Isolated from UTI Patients Attending some Selected Hospitals in Minna, Nigeria. *Nigerian Journal of Biotechnology*. *37(2)*: 56–73.

Islam, S., Aldstadt, J., and Aga, D. (2019). Global antimicrobial resistance: a complex and dire threat with few definite answers. *Tropical Medicine and International Health*. *24(6)*: 658–662.

Jena, J., Sahoo, R. K., Debata, N. K., and Subudhi, E. (2017). Prevalence of TEM, SHV, and CTX-M genes of extended-spectrum β-lactamase-producing *Escherichia coli* strains isolated from urinary tract infections in adults. *Biotech*. *7(3)*: 1-7.

Liang, W. J., Liu, H. Y., Duan, G. C., Zhao, Y. X., Chen, S. Y., Yang, H. Y., and Xi, Y. L. (2018). Emergence and mechanism of carbapenem-resistant *Escherichia coli* in Henan, China, 2014. *Journal of infection and public health*. *11(3)*: 347–351.

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Maurya, A. P., Talukdar, A. D., Chanda, D. D., Chakravarty, A., and Bhattacharjee, A. (2016). First description of SHV-148 mediated extended-spectrum cephalosporin resistance among clinical isolates of *Escherichia coli* from India. *Indian Journal of Medical Microbiology*. *34(1)*: 33-37.

Mehrad, B., Clark, N. M., Zhanel, G. G., and Lynch III, J. P. (2015). Antimicrobial resistance in hospital-acquired gram-negative bacterial infections. *Chest*. *147(5)*: 1413-1421.

Mingeot-Leclercq, M. P., Glupczynski, Y., and Tulkens, P. M. (1999). Aminoglycosides: activity and resistance. *Antimicrobial agents and chemotherapy*. *43(4)*: 727-737.

Oberoi, L. (2013). ESBL, MBL and AmpC Lactamases Producing Superbugs – Havoc in the Intensive Care Units of Punjab India. *Journal of clinical and diagnostic research*.

Ochada, NS., Nasiru, IA., Thairu, Y., Okanlowan M B., and Abdulakeem, YO (2015). Antimicrobial Susceptibility Pattern of Urinary Pathogens Isolated from Two Tertiary Hospitals in Southwestern Nigeria. *Afr. J. Cln. Exper. Microbiol*. *16(1)*: 12-22.

Okafor, J., and Nweze, E. (2020). Antibiotic susceptibility of *Escherichia coli* isolated in cases of urinary tract infection in Nsukka, Nigeria. *Journal of Pre-Clinical and Clinical Research*. *14(1)*: 1–7.

Oladoja, M.A and Onifade, A.K. (2015). Dissemination of Multi-Drug Resistance in *Escherichia coli* Isolated from Apparently Healthy School Children in Ondo State, Nigeria. *Clinical Microbiology: Open Access, 04(05)*.

Sadeghi, M., Sedigh Ebrahim-Saraie, H., and Mojtahedi, A. (2022). Prevalence of ESBL and AmpC genes in *E. coli* isolates from urinary tract infections in the north of Iran. *New Microbes and New Infections*. *45*: 100947.

Salvador, E., Wagenlehner, F., Köhler, C. D., Mellmann, A., Hacker, J., Svanborg, C., and Dobrindt, U. (2012). Comparison of asymptomatic bacteriuria *Escherichia coli* isolates from healthy individuals versus those from hospital patients shows that long-term bladder colonization selects for attenuated virulence phenotypes. *Infec. and Imm*. *80(2)*: 668–678.

Shokravi, Z., Mehrad, L., and Ramazani, A. (2015). Detecting the frequency of aminoglycoside modifying enzyme encoding genes among clinical isolates of methicillin-resistant Staphylococcus aureus. *BioImpacts: BI*. *5(2)*: 87.

Thakuria, B., and Lahon, K. (2013). The beta lactam antibiotics as an empirical therapy in a developing country: an update on their current status and recommendations to counter the resistance against them. *Journal of clinical and diagnostic research: JCDR*. *7(6)*: 1207.

Wauters, G., and Vaneechoutte, M. (2015). Approaches to the identification of aerobic Gram-negative bacteria. *In*: J. H. Jorgensen, M. A Pfaller, K. C. Caroll, G. Funke, M. L. Landry, S.S. Ritcher, and D.W. Warnock [Eds], *Manual of clinical microbiology [11th edition. Pp*. 613-634].

Wayne, P. A. (2010). Clinical and Laboratory Standards Institute: Performance standards for antimicrobial susceptibility testing: 20th informational supplement. *CLSI document M100-S20*.

Xanthopoulou, K., Carattoli, A., Wille, J., Biehl, L. M., Rohde, H., Farowski, F., Krut, O., Villa, L., Feudi, C., Seifert, H., and Higgins, P. G. (2020). Antibiotic Resistance and Mobile Genetic Elements in Extensively Drug-Resistant Klebsiella pneumoniae Sequence Type 147 Recovered from Germany. *Antibiotics*. *9(10)*: 675.

Zdziarski J, Svanborg C, Wullt B, Hacker J, and Dobrindt U (2008). Molecular basis of commensalism in the urinary tract: low virulence or virulence attenuation? *Infect Immun. 76(2)*: 695-703.

Zhao, X., Yang, J., Ju, Z., Chang, W., and Sun, S. (2018). Molecular Characterization of Antimicrobial Resistance in *Escherichia coli* from Rabbit Farms in Tai'an, China. *BioMed Research International*. *2018*: 1-7.

Zilahi, G., Artigas, A., and Martin-Loeches, I. (2016). What's new in multidrug-resistant pathogens in the ICU? *Annals of intensive care*. *6(1)*: 1-11.

Citation: Adewuyi-Oseni *et al*. (2023). Molecular Characterization of Beta-Lactam and Aminoglycoside Antibiotics Resistance Genes Present in *Escherichia coli* Isolated from Human Urine Samples. *International Journal of Microbiology and Applied Sciences*. *2*: 145 - 154.