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Descriptive Analysis of Drug-Resistant *Enterobacteriaceae* Isolated from Clinical Specimens at Bowen University Teaching Hospital, Ogbomosho

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Abstract

The emergence of drug-resistant *Enterobacteriaceae* is a major concern in clinical care. The aim of this study was to determine the prevalence of drug-resistant *Enterobacteriaceae* among patients at Bowen University Teaching Hospital (BUTH), Ogbomosho. This cross-sectional experimental study received ethical approval from BUTH's Research Ethics Committee (BUTH/REC-1146). Clinical samples including sputum, wound swabs, eye/ear swabs, and urine were collected via convenience sampling from various hospital departments. Standard microbiological culture techniques were used, with biochemical identification performed using API 20E and API 20NE. Antimicrobial susceptibility testing was conducted via E-test, following 2023 Clinical and Laboratory Standards Institute (CLSI) guidelines. Data were analyzed using descriptive statistics. Of the 96 patients enrolled, 71 (73.96%) had *Enterobacteriaceae* infections. Females accounted for 44 cases (61.46%), while males represented 27 (38.54%). The highest infection rate was in the 18-64 age group (49, 68.75%), followed by older adults (12, 16.67%). Traders constituted the largest occupational group affected (24, 28.13%). *Escherichia coli* was the most frequently isolated organism (33, 34.38%), followed by *Klebsiella pneumoniae* (28, 29.17%). Sputum samples yielded the highest pathogen recovery rate (31, 43.66%), followed by urine (23, 32.39%). Antimicrobial susceptibility testing revealed that *Klebsiella pneumoniae* had the highest resistance to amoxicillin (100%), while *Escherichia coli* showed 60.5% resistance to trimethoprim-sulfamethoxazole. The ESBLs

producing strains recovered were 21.7% and 63.6% respectively. The lowest resistance (2.4%) was observed for imipenem against both *Escherichia coli* and *Klebsiella pneumoniae*. The high resistance of *Klebsiella pneumoniae* to amoxicillin particularly in respiratory and urinary tract infections highlights the urgent need for ongoing antimicrobial resistance surveillance and alternative treatment strategies in Ogbomosho.

Keywords: Drug-Resistance, *Enterobacteriaceae*, Descriptive Analysis, Clinical Specimens, Ogbomosho

Introduction

Antimicrobials have been used successfully to treat infections for many years and have made the management of infectious diseases easier thereby decreasing morbidity and mortality (Chukwu *et al.*, 2020). However, microbial organisms have responded through many phenotypic or genotypic evolutionary processes in an attempt to adapt and survive, a phenomenon referred to as antimicrobial resistance (AMR) in both hospital and community settings (Oloso *et al.*, 2019). Antimicrobial Resistance (AMR) occurs when microorganisms including bacteria, viruses, fungi, and parasites become able to adapt and grow in the presence of medications that once impacted them (CDC, 2019). Antimicrobial resistance (AMR) has developed as one of the major urgent threats to public health causing serious issues to successful prevention and treatment of persistent diseases. Despite different actions taken in recent decades to tackle

this issue, the trends of global AMR demonstrate no signs of slowing down (Dadgostar, 2019). It is estimated that globally approximately 700,000 deaths are attributed annually to antimicrobial resistance with the possibility of this already staggering figure rising to 10 million deaths per year by 2050 (Christaki *et al.*, 2020) and it is projected that 40% of these 10 million deaths from Antimicrobial Resistance (AMR) will occur in Africa (Egwuenu *et al.*, 2017).

The inappropriate use of antibiotics is a major driver of antimicrobial resistance (AMR) and is a considerable public health concern across many continents and countries including sub-Saharan Africa (Godman *et al.*, 2022). As a result, the Global Action Plan on Antimicrobial Resistance was adopted in May 2015 by the World Health Assembly and endorsed by Heads of States of many countries, including Nigeria, at the United Nations General Assembly in New York in September 2016 to try and reduce AMR rates (Babatola *et al.*, 2020). Despite this, in a review of antimicrobial resistance in Africa, it was reported that resistance to commonly used antibiotics such as fluoroquinolones, tetracyclines, and penicillins among others was already widespread (Aliyu *et al.*, 2021). There are three types of antimicrobial resistance-exhibiting microorganisms: multidrug-resistant (MDR), extensively drug-resistant (XDR), and pan-drug-resistant (PDR) microorganisms (Alkofide *et al.*, 2020). MDR microorganisms acquire non-susceptibility to at least one agent in three or more antimicrobial classes (Alkofide *et al.*, 2020). XDR microorganisms are non-susceptible to at least one agent in all but two or fewer antimicrobial classes (i.e., bacterial isolates remain susceptible to only one or two categories). Microorganisms with non-susceptibility to all agents in all antimicrobial classes are referred to as PDR (Abushaheen *et al.*, 2020).

The family *Enterobacteriaceae* consists of a large heterogeneous group of Gram-negative rod-shaped bacteria that are naturally found in the mammalian gut although can also be found in other environments (Uzeh *et al.*, 2021). They are useful indicators of food quality, hygiene, and contamination. Examples include *Escherichia coli*, *Salmonella* spp. and *Shigella* spp.

Enterobacteriaceae is responsible for a range of enteric infections such as diarrhea and endocarditis, to infections of the respiratory tract, skin, soft-tissues, urinary tract, joints, bones, eyes and central nervous system (Uzeh *et al.*, 2021). This study seeks to analyze the prevalence and characteristics of drug-resistant *Enterobacteriaceae* strains isolated from clinical samples in Ogbomoso, Oyo State.

Materials and Methods

Ethical Approval

This study was approved by the Bowen University Teaching Hospital (BUTH) Research Ethics Committee with approval number BUTH/REC-1146. It was carried out in accordance with guidelines for obtaining informed consent from patients. Written consent and assents were obtained from each participant who signed the consent form.

Study area

The bacteria isolates were obtained from clinical samples collected from the various departments of the Bowen University Teaching Hospital, Ogbomoso in Oyo State, Nigeria. The laboratory procedures were done at the Teaching laboratory, Bowen University Teaching Hospital, Ogbomoso, Oyo State.

Study design

This study employed a prospective, cross-sectional, and experimental design, focusing on patients diagnosed with infections caused by drug-resistant *Enterobacteriaceae*.

Collection of Clinical Samples

Clinical samples, including sputum, wound swabs, eye swabs, ear swabs and urine, were collected from various hospital units. These units included the male and female surgical wards, wound dressing room, accident and emergency department, intensive care unit, private wards, gynecological ward, surgical heart patient ward, outpatient department, female medical ward, male medical ward, and general outpatient department.

Laboratory Procedures

Microbiological Culture

Specimens were cultured on Cetrimide Agar,

Blood Agar, Chocolate Agar, and MacConkey Agar. Following inoculation, the plates were incubated at 37°C for 20 hours. Growth characteristics and colony morphology were documented post-incubation.

Biochemical Analysis

All the Enterobacterales were identified by using API 20E while all the oxidase-positive isolates underwent further identification using the API 20 NE system (BioMérieux, USA), adhering strictly to the manufacturer's protocol.

Antimicrobial Susceptibility Testing (AST)

Phenotypic antimicrobial susceptibility testing was conducted via the E-Test method, with inoculum preparation standardized using the Kirby-Bauer disk diffusion technique. Sterile forceps were employed to position the E-Test strips on Mueller Hinton agar, followed by incubation at 37°C for 24 hours. Zone of inhibition measurements and interpretations as resistant, intermediate, or susceptible were based on the 2023 guidelines of the Clinical Laboratory Standards Institute (CLSI) with specific attention to detecting resistance mechanisms like ESBL and carbapenem resistance.

Data collection and analysis

A structured questionnaire was issued to participants who expressed willingness to participate. The data collected includes demographic data, socio-demographic data, family history, medical history, and environmental factors. The data collected was subjected to statistical analysis using Microsoft Excel. This analysis incorporated descriptive

statistics, along with various other fundamental statistical analysis. An alpha level of 0.05 was employed, which corresponds to a 95% confidence interval, ensuring the results are statistically significant while allowing for a balanced consideration of Type I error risk.

Results

A total of 96 clinical samples were obtained from an initial sample size of 100 (Figure 1). Demographic analysis indicated that adults aged 18 to 64 constituted the majority of participants, while children aged 0 to 17 accounted for the smallest proportion. Gender distribution revealed a higher representation of females compared to males (Table 1). Occupational categorization identified students as the largest group, followed by traders (Table 2). Analysis of bacterial isolates showed that *Escherichia coli* was the most frequently isolated organism, followed by *Klebsiella pneumoniae*, while *Klebsiella oxytoca* was the least commonly isolated (Table 3).

The study identified Enterobacteriaceae isolates in 71 participants, with *Escherichia coli* being the most prevalent, detected in 33 samples (46.48%). This was followed closely by *Klebsiella pneumoniae*, found in 28 samples (39.44%), indicating that these two species were the dominant pathogens in the sampled population. *Proteus mirabilis* was present in 8 samples (11.27%), while *Klebsiella oxytoca* was the least common, isolated from only 2 samples (2.82%) (Table 4). This was correlated with the leucocytes counts to distinguish between colonization and infection.

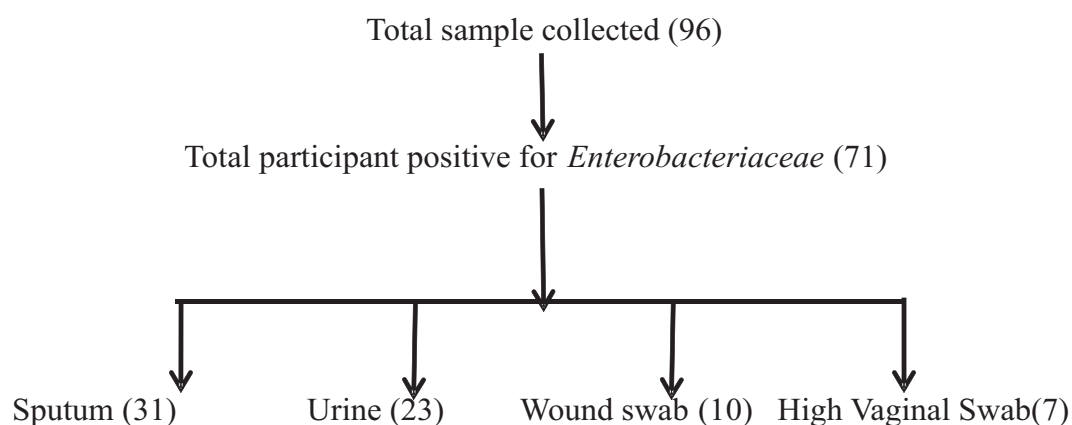


Figure 1: Distribution of Enterobacteriaceae among the clinical samples isolated

Table 1: Demographic characteristics of study participants (N = 96)

Demographic Data	Frequency (%)
Gender	
Males	37 (38.54)
Females	59 (61.46)
Age Group	
0-17 years	18 (18.75)
18-64 years	66 (68.75)
65 years and above	12 (12.50)

Table 2: Socio-demographic data of all study participants (N = 96)

Occupation	Frequency (%)
Driver	8 (8.33)
Meat seller	1 (1.04)
Banker	1 (1.04)
Trader	24 (25)
Carpenter	2 (2.08)
Tailor	5 (5.20)
Student	18 (18.75)
Nurse	1 (1.04)
Filling Station attendant	1 (1.04)
Shoemaker	1 (1.04)
Unemployed	6 (6.25)
Drycleaner	1 (1.04)
Security guard	1 (1.04)
Teacher	9 (9.38)
Business	11 (11.46)
Islamic Cleric	2 (2.08)
Pastor	4 (4.16)

Table 3: Occurrence of Organisms detected (N = 96)

Organism	Frequency (%)
<i>Escherichia coli</i>	33 (34.38)
<i>Klebsiella pneumoniae</i>	28 (29.17)
<i>Pseudomonas aeruginosa</i>	12 (12.5)
<i>Proteus mirabilis</i>	8 (8.33)
<i>Staphylococcus aureus</i>	9 (9.37)
<i>Streptococcus pyogenes</i>	4 (4.17)
<i>Klebsiella oxytoca</i>	2 (2.08)

Table 4: Occurrence of Enterobacteriaceae isolated from study participants (N = 71)

Organism	Number of Samples	Percentage (%)
<i>Escherichia coli</i>	33	46.48
<i>Klebsiella pneumoniae</i>	28	39.44
<i>Proteus mirabilis</i>	8	11.27
<i>Klebsiella oxytoca</i>	2	2.82

Overall, *Escherichia coli* and *Klebsiella pneumoniae* exhibit substantial resistance to multiple classes of antibiotics, underscoring the escalating threat of antimicrobial resistance (AMR) among Enterobacterales. *E. coli* demonstrates high levels of resistance to trimethoprim (60.5%), tetracycline (53.1%), cefotaxime (45.7%), and ciprofloxacin (42.6%), with 21.7% of strains identified as β -lactamase producers (Figure 2). Similarly, *K. pneumoniae* shows pronounced resistance to ampicillin (100%), cefotaxime (36.4%) and

ciprofloxacin (35.9%) while 63.6% ESBL strains were also reported (Figure 3). These findings indicate the reduced therapeutic efficacy of commonly used antibiotics against these pathogens. However, *E. coli* maintains relatively low resistance (2.4%) to imipenem, making this drug more viable treatment options. Likewise, *K. pneumoniae* exhibits comparatively lower resistance (2.4%) to carbapenems such as imipenem, suggesting they remain effective choices in managing infections caused by this organism.

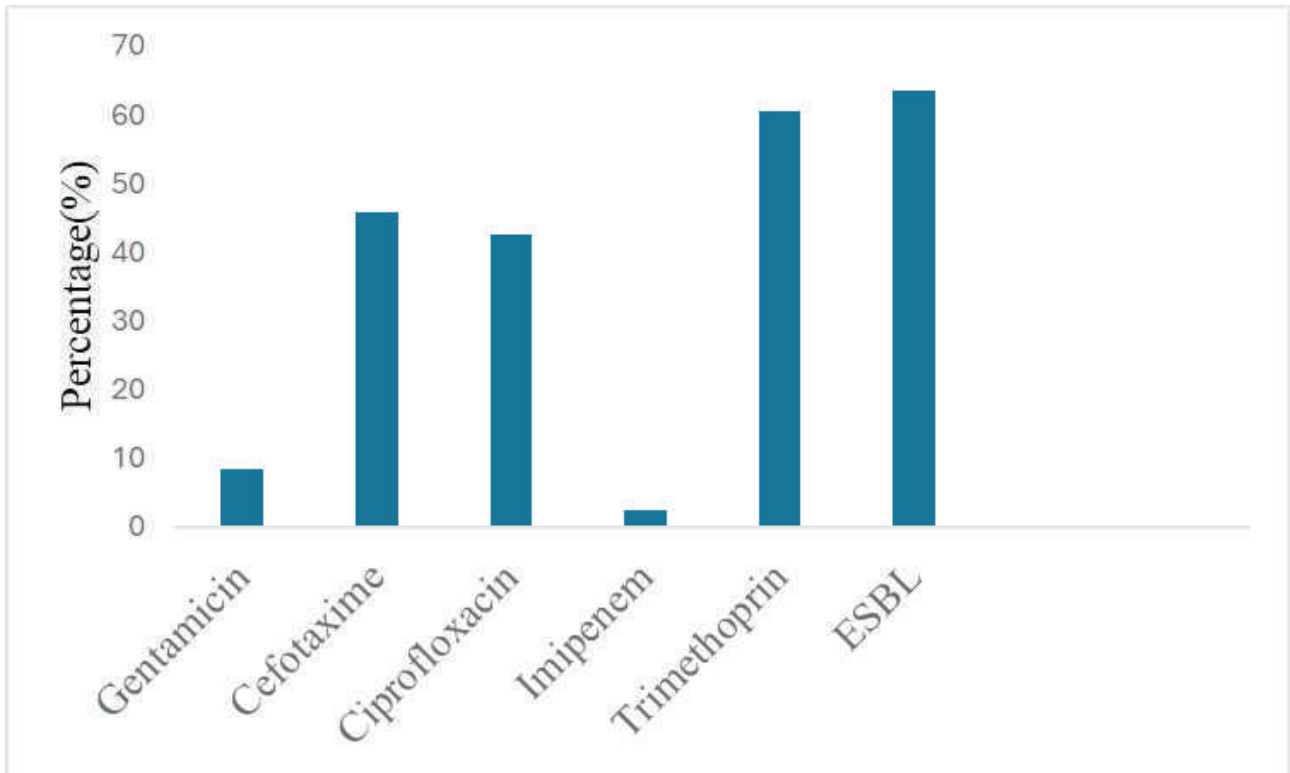


Figure 2: Percentage of Resistance of Escherichia coli

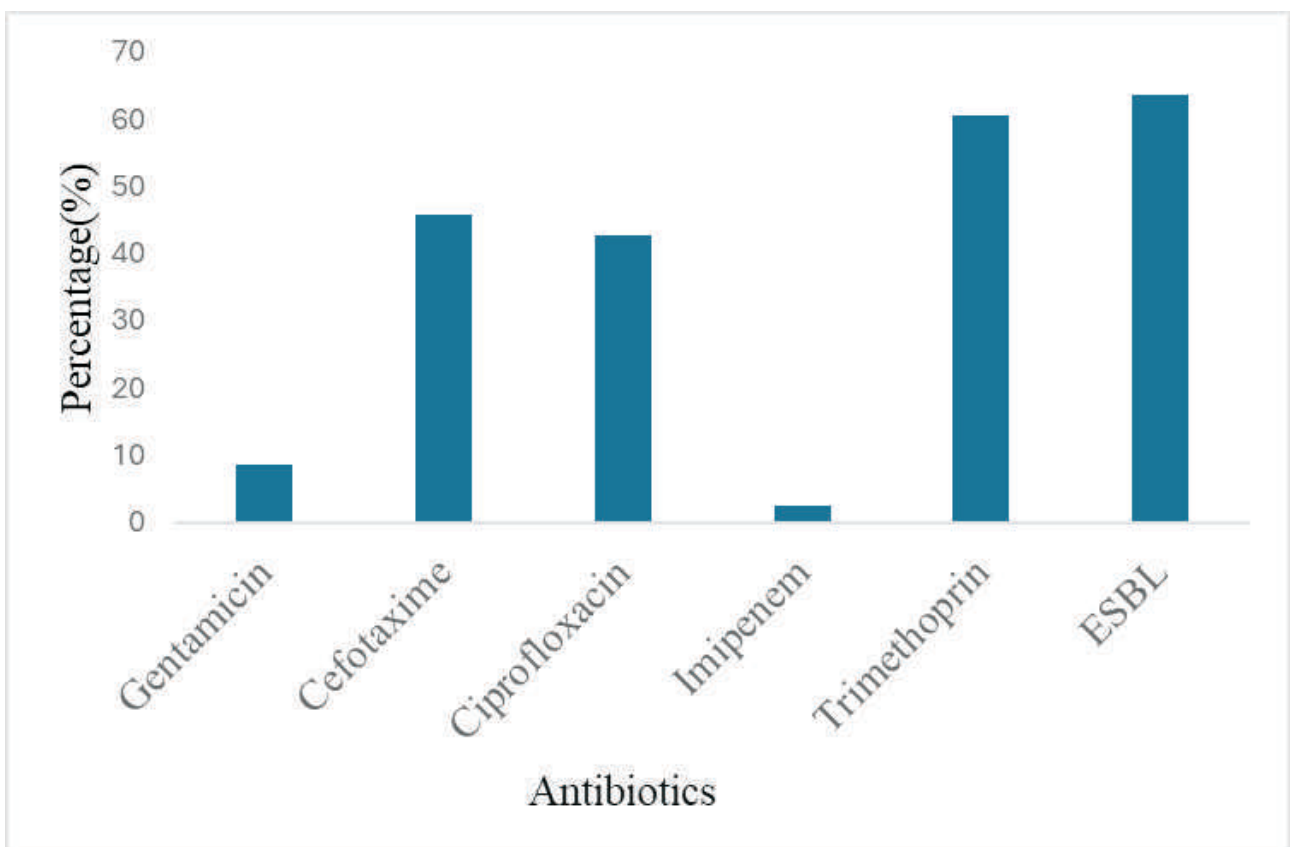


Figure 3: Percentage of Resistance of Klebsiella pneumoniae

Discussion

Escherichia coli was identified as the predominant causative agent, accounting for 34.38% of bacterial isolates from clinical samples in a tertiary healthcare institution in Ogbomosho. This finding aligns with the observations of Banga *et al.* (2019), which highlighted regional variations in bacterial populations, differences in hospital infection control protocols, and variability in clinical sample types. According to the Study for Monitoring Antimicrobial Resistance Trends (SMART) surveillance program, *E. coli* constituted 16.2% of gram-negative bacterial isolates in hospital environments. The significantly higher prevalence (34.38%) observed in this study suggests that *E. coli* and other *Enterobacteriaceae* may pose a greater infection burden in Nigerian hospital settings compared to industrialized nations. This disparity could be attributed to factors such as limitations in local healthcare infrastructure, resource constraints, and challenges in implementing effective infection control measures.

Resistance to carbapenems within the *Enterobacteriaceae* family, particularly to imipenem, was observed at a rate of approximately 2.4%, different from the reported 11.2% resistance to imipenem in Nigeria, as highlighted by Irekola *et al.* (2024) in Nigeria. Carbapenems, often regarded as the antibiotics of last resort for treating multidrug-resistant bacterial infections, are becoming increasingly compromised in their efficacy against *Enterobacteriaceae*.

This convergence in resistance rates underscores the emerging threat posed by carbapenem resistance, signaling a critical public health concern. The growing ineffectiveness of these critical antimicrobials highlights the urgent need for enhanced antimicrobial stewardship, robust surveillance systems, and innovative therapeutic approaches to mitigate the spread of resistant strains within clinical and community settings.

Conclusion

The findings of this study indicate a significant prevalence of *Enterobacteriaceae* infections at Bowen University Teaching Hospital, with many

isolates exhibiting multidrug resistance. Notably, high levels of resistance were observed against amoxicillin and trimethoprim-sulfamethoxazole, as well as an alarming resistance to tetracycline and cefotaxime. These patterns emphasize the critical need for continued surveillance of antimicrobial resistance, particularly among patients with respiratory tract infections, who appear to be significantly impacted.

Recommendations

To deepen the understanding of the genetic basis of resistance in *Enterobacteriaceae*, future research should aim to expand the range of antibiotics tested and incorporate advanced molecular techniques, such as whole-genome sequencing (WGS) and polymerase chain reaction (PCR). These methods could provide detailed insights into the specific genes and mutations driving antimicrobial resistance. Additionally, longitudinal studies would be valuable for monitoring trends in resistance across different time frames and therapeutic environments.

Enhanced funding and resource allocation are crucial for facilitating comprehensive investigations and establishing robust surveillance systems. Collaborative efforts among research institutions could enable the generation of larger, more diverse data sets, offering a broader perspective on resistance patterns. Such cooperative initiatives would be instrumental in devising effective national strategies to combat the growing threat of drug-resistant infections.

Conflict of Interest Declaration

The authors declared no conflict of interest.

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