



Prevalence and Antibiotic Susceptibility Pattern of *Escherichia coli* 0157:H7 Associated with Gastroenteritis in Minna, Niger State, Nigeria

P. E. Omebije¹, L. Y. Adogo^{2*} and B. Ajide²

¹Department of Microbiology, Faculty of Natural Sciences, Kogi State University, Ayingba, Nigeria.

²Department of Biological Sciences, Faculty of Science and Technology, Bingham University, Karu, Nasarawa State, Nigeria.

Authors' contributions

This work was carried out in collaboration between all authors. Author PEO designed the study, performed the statistical analysis, wrote the protocol, wrote the first draft of the manuscript and managed literature searches. Authors LYA and BA managed the analyses of the study and literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aim: To determine the prevalence and antibiotic susceptibility pattern of *Escherichia coli* 0157:H7 associated with gastroenteritis in Minna, Niger state.

Place and Duration of Study: The study was carried out in the Department of Microbiology, Federal University of technology, Minna, between the months of August 2013 to April 2014.

Study Design: The study was a cross sectional study.

Materials and Methods: Four hundred and twenty eight stool specimens were collected using random sampling method. Stool specimens were obtained from outpatients who visited five different hospitals and clinics in Minna. These patients presented signs and symptoms suggestive of gastroenteritis. Isolates were examined for enteropathogens using standard bacteriological

*Corresponding author: E-mail: adogolillian@gmail.com;

techniques. The physiological and biochemical characteristics of recovered isolates were compared with those of known taxa. Sorbitol fermenting *Escherichia coli* was further identified using the latex agglutination kit (Oxoid, England). The antibiotics susceptibility pattern of isolates was determined using the Kirby-Bauer disc diffusion method and read according to the Clinical and Laboratory Standards Institute (CLSI) recommendation.

Results: Out of the 428 stool specimens examined, 336 were positive for gastroenteritis. The prevalence of *Escherichia coli* O157:H7 in respect to age group reveals that patients between the age group of 0-9 years had the highest frequency of occurrence 19(39.58%) when compared to other age group in the study. Although there were more males 21(10.82%) than females 27(14.84%), *Escherichia coli* O157:H7 was not significantly related to gender ($p>0.05$). The antibiotic profile reveals that *Escherichia coli* O157:H7 isolates were susceptible to Ciprofloxacin (72.92%), Chloramphenicol (60.42%) and Tetracycline (43.75%). However, it was completely not susceptible to ampicillin (0.00%). Chi square test was used for statistical analysis.

Conclusion: It was observed from this study that there is a need to enlighten the public on the use/misuse of antibiotics, this is due to the fact that *Escherichia coli* O157:H7 isolates were not susceptible to most of the drugs used in this study. This data also provide valuable information for health personnel's in determining the appropriate antibiotic to be administered. There is also need to improve personal hygiene, environmental sanitation, provision of safe drinking water and health education in order to reduce infection with gastroenteritis.

Keywords: Gastroenteritis; prevalence; antibiotic susceptibility; *E. coli* O157:H7.

1. INTRODUCTION

Gastroenteritis is said to be the inflammation of the lining of the stomach, the small and large intestines. Infection due to gastroenteritis has been known to be caused by microorganisms and also by ingestion of some food items, chemical toxins or drugs [1]. Gastroenteritis in healthy adults causes discomfort and inconvenience. However, it can cause life-threatening dehydration and electrolyte imbalance in the very ill weak, young and old individuals. Children and workers in daycare centers and nurseries, students living in dormitories, military employees, and travelers could also be susceptible to gastroenteritis [1].

Escherichia coli O157:H7 is an emerging public health concern in most countries of the world [2]. Verotoxigenic producing *Escherichia coli* (VTEC) or Shiga toxin-producing strains of *Escherichia coli* (STEC) are recognized as an important human pathogen of public health concern [3]. STEC isolates belong to many different serotypes. *E. coli* O157:H7 is the most common serotype associated with human illness. Isolates of this pathogen are a major cause of haemorrhagic colitis (HC) and mild diarrhoeal illness and haemolytic uraemic syndrome (HUS) [4]. Haemolytic uraemic syndrome is characterized by a prodrome of gastroenteritis, frequently including bloody diarrhoea, followed by acute haemolytic anaemia, thrombocytopenia and renal failure. Infections are usually linked to

the consumption of VTEC-contaminated vegetables, raw milk and improperly cooked beef. VTEC is also transmitted through contaminated water and direct transmission of VTEC from animals to man [5].

The potent cytotoxins produced by STEC are divided into two major classes (Shiga-like-toxin 1 (stx1) and Shiga-like toxin 2 (stx2). These strains produce verotoxins 1 and 2 (Shiga-like-toxin 1 (stx1) and Shiga-like toxin 2 (stx2) and their variants [6]. In addition to toxin production, another virulence-associated factor expressed by VTEC is a protein called intimin, which is responsible for intimate attachment of VTEC to the intestinal epithelial cells, causing attaching and effacing (AE) lesions in the intestinal mucosa [7].

E. coli O157:H7 has been isolated from a wide variety of hosts, especially cattle, sheep, goat, pig, poultry, dogs, horses, deer, wild birds [8].

Microbial resistance to antibiotics is a worldwide problem [9]. Resistance to antimicrobial agents has resulted in morbidity and mortality from treatment failures and increased health care costs [10]. Antimicrobial resistance in Enterobacteriaceae poses a critical public health threat, especially in the developing countries [11,10]. The development of resistance to antimicrobials is known to occur through stable genetic change heritable from generation to generation through specific mechanisms

including mutation, transduction, transformation and or conjugation [12].

2. METHODOLOGY

2.1 Study Area

The study was conducted in Minna, Niger State. Niger state is located in the Northern part of Nigeria. Niger state lies on latitude 8° to 11°30' North and longitude 3° 30' to 7° 40' East. The State is bordered to the North by Zamfara State, West by Kebbi State, South by Kogi State, South West by Kwara State, North-East by Kaduna State and South East by FCT. Niger State also has an International boundary with the Republic of Benin along Agwara and Borgu LGAs to the North West [13].

2.2 Study Population

The study population comprised of outpatients (children and adults of both gender) with signs and symptoms suggestive of gastroenteritis visiting the hospitals / clinics.

2.3 Ethical Consideration

Approval for the study was obtained from the Research, Ethics and Publication Committee of Niger State Hospital Management Board, General Hospital, Minna. Consent of all participants were also sought and obtained.

2.4 Collection of Samples

Fresh faecal specimens were collected into sterile universal containers. For children whose faecal specimens could not be collected, rectal swabs were collected using sterile swab sticks [14].

2.5 Sample Processing

2.5.1 Macroscopic examination

Gross examination of the faecal specimen was performed to note characteristics such as color, consistency and the presence of blood/mucus [14].

2.5.2 Culturing of specimens

Macroscopic examination of faecal specimens was carried out [15]. The fecal specimens were inoculated onto sorbitol MacConkey agar (Oxoid

medium code CM0813) and incubated for 24 hours at 37°C [16].

2.5.3 Serological assay and antibiotics susceptibility

Non-sorbitol fermenters on Sorbitol MacConkey agar (SMA) plates were tested with latex agglutination test kits. Isolates that did not ferment sorbitol within 24 hours, and were positive to latex agglutination test were confirmed as *E. coli* 0157:H7 [17].

The antibiotics susceptibility pattern of isolates was determined using the Kirby-Bauer disc diffusion method and read according to Clinical and Laboratory Standard Institute [18].

Escherichia coli isolates were tested for susceptibility to Ciprofloxacin (5 µg), Sulphamethoxazole / Trimethoprim (25 µg), Chloramphenicol (30 µg), Tetracycline (30 µg), Nalidixic acid (30 µg), Ampicillin (10 µg) and Cefuroxime (30 µg). The choice of antibiotics/discs used was based on those commonly available in most pharmacy within the locality. Also they are groups of antibiotics recommended in previously published susceptibility profiles for their potential efficacy as antimicrobial agents on gastrointestinal infection [19].

3. RESULTS

Table 1 shows the distribution of bacterial isolates from cases of gastroenteritis of subjects screened in this study. Out of the total number of 428 faecal specimens examined, 336 (78.50%) specimens were positive for gastroenteritis, in which *Escherichia coli* had the highest frequency of occurrence 117(31.12%) while the bacteria with the least frequency of occurrence was *Shigella* species 5(1.33%).

Table 1. Distribution of bacterial isolates in the studied population

Bacteria	Frequency (%)
<i>Escherichia coli</i>	117(31.12)
<i>Escherichia coli</i> 0157:H7	48(12.77)
<i>Salmonella</i> species	82(21.81)
<i>Shigella</i> species	5 (1.33)
<i>Vibrio cholera</i>	51(13.56)
<i>Vibrio parahaemolyticus</i>	33(8.78)
Total	336

In Table 2, the frequency of occurrence of *E. coli* 0157:H7 associated with gastroenteritis on the basis of age was observed. The age group 0-9

years showed the highest number of gastroenteritis with *E. coli* 0157:H7 19(39.58) while two age brackets 60-69 and ≥70 years had the least frequency (2.08%) of infection with *Escherichia coli* 0157:H7.

Table 2. Prevalence of *Escherichia coli* 0157:H7 associated with gastroenteritis based on age group

Age groups (years)	Number of isolates	Number positive (%)
0-9	146	19(39.58)
10-19	50	8(16.67)
20-29	55	9(18.75)
30-39	44	4(8.33)
40-49	29	3(6.25)
50-59	26	3(6.25)
60-69	14	1(2.08)
≥70	12	1(2.08)
Total	376	48(12.77)

Table 3 represents the frequency of *E. coli* 0157:H7 in relation to gender, it shows that although the males had the highest number of isolates positive for gastroenteritis (10.82%), the females showed the highest frequency of infection with *E. coli* 0157:H7 (14.84%).

The antibiotics susceptibility of *E. coli* 0157:H7 from cases of gastroenteritis by Kirby-Bauer disc diffusion method is shown on Table 4. *E. coli* 0157:H7 was susceptible to the following antibiotics, Ciprofloxacin (72.92%), chloramphenicol (60.42%) and tetracycline (43.75%). The organism was completely resistant to ampicillin (0.00%).

4. DISCUSSION

Our study reveals the prevalence and antibiotic susceptibility pattern of *Escherichia coli* 0157:H7

associated with gastroenteritis in Minna, Niger state. Similar studies from other parts of Nigeria conducted on humans and documented are scanty, thereby making the comparison of data on prevalence of this organism with data from other parts of Nigeria limited. *Escherichia coli* had the highest frequency (31.12%) of bacteria isolates from cases of gastroenteritis. This is similar to the study conducted by Okeke, Lamikanra, Steinrück and Kaper [20], who reported an incidence of 40.4% of diarrhoeagenic *Escherichia coli* in children and another study in Jordan showed 30% incidence Shehabi, Bulos and Hajjaj [21]. The prevalence of *E. coli* 0157:H7 in this study was 12.77%. This prevalence is higher than the incidence reported by [22-24] in similar studies conducted in Lagos, Zaria and Jos, Nigeria who reported a prevalence of 6%, 5.4% and 3.1% respectively. Poor sanitary living conditions, inadequate portable drinking water could be responsible for the high incidence of *Escherichia coli*.

Table 3. Prevalence of *Escherichia coli* 0157:H7 in relation to gender

Gender	Number of isolates	Number positive (%)
Male	194	21(10.82)
Female	182	27(14.84)
Total	376	48

Bacterial isolates other than *Escherichia coli* reported from this study include *Salmonella* species (21.81%), *Shigella* species (1.33%), *Vibrio cholera* (13.56%) and *Vibrio parahaemolyticus* (8.78%). This corroborates with the report of [25], which states that, the most common bacteria that cause foodborne illnesses are: *Salmonella* species, *Shigella* species, *E. coli* 0157:H7, *Listeria monocytogenes* and *Vibrio* species.

Table 4. Antibiotics susceptibility of *Escherichia coli* 0157: H7 from patients with gastroenteritis

Bacteria	Antibiotics (µg)						
	CIP (%)	SXT (%)	C (%)	TE (%)	NA (%)	AMP (%)	CXM (%)
<i>E. coli</i> n=117	93(79.49)	72(61.54)	73(62.39)	31(26.50)	73(62.39)	2(1.71)	7(5.98)
<i>E. coli</i> 0157:H7 n=48	35(72.92)	15(31.25)	29(60.42)	21(43.75)	11(22.92)	0(0.00)	1(2.08)

Keys: CIP=Ciprofloxacin (5 µg), SXT=Sulphamethoxazole / Trimethoprim (25 µg), C=Chloramphenicol (30 µg), TE=Tetracycline (30 µg), NA=Nalidixic acid (30 µg), AMP=Ampicillin (10 µg), CXM=Cefuroxime sodium (30 µg)

The prevalence of *E. coli* O157:H7 in relation to age group shows that the highest prevalence of (39.58%) of *E. coli* O157:H7 was isolated from patients belonging to the age group of 0-9 years. In a similar study conducted in Lagos, Nigeria by Ogunsanya, Rotimi and Adenuga [26], children under the age of 5 years recorded a prevalence of (3%). In this study, there was no significant relationship ($p>0.05$) between the infection and age group as shown in Table 2. This agrees with the findings of Isibor, Ekundayo and Ohenhen [27] who observed that *E. coli* O157: H7 infection was independent of a person's age in a study conducted in Edo State.

Out of 376 isolates from cases of gastroenteritis, the females had the highest frequency (14.84%) of isolates positive for *E. coli* O157:H7 which is similar to separate studies conducted by Ademokoya, Adebolu and Ogundare; Nweze [28,29] who recorded a high frequency of 48.20% and 45.45% among females. The occurrence of *E. coli* O157:H7 had no significant relationship between gender and infection ($p>0.05$), this is in relation to a report by Ogunsanya, Rotimi and Adenuga [26] which showed that there was no significant association between gender and the infection. Thus, it can be deduced that a person's gender does not constitute a risk factor for *E. coli* O157:H7 infection.

E. coli O157:H7 isolates recorded (0.00%) susceptibility to ampicillin. This agrees with the findings of Musa, Shikieri, Ahmed and Kafi [30] who reported that *E. coli* O157:H7 was (0.00%) not susceptible to ampicillin. This could be due to the misuse or overuse of drugs. *E. coli* O157:H7 was susceptible to Ciprofloxacin (12.41%), Chloramphenicol (12.50%) and Tetracycline (18.10%).

5. CONCLUSION

This study reveals that *E. coli* O157:H7 is associated with gastroenteritis and is responsible for infectious diarrhoea among children. It also suggests that this pathogen is likely to be an important aetiology of acute gastroenteritis in the country. The high rates of antimicrobial resistance to ampicillin and cefuroxime was observed. Thus, the indiscriminate use of antibiotics for the treatment of diarrhoea needs to be avoided. It is equally important to improve sewage and refuse disposal system, provide safe potable water, sanitation, personal hygiene and health education in order to reduce infection with

this and other enteric pathogens. Since the infection primarily occurs via fecal-oral route, food hygiene measures like proper cooking of meat, consumption of pasteurized milk, washing fruits and vegetables especially those to be eaten raw and drinking chlorine- treated water could be very useful.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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