JETIR.ORG ISSN: 2349-5162 | ESTD Year : 2014 | Monthly Issue
JOURNAL OF EMERGING TECHNOLOGIES AND INNOVATIVE RESEARCH (JETIR)
An International Scholarly Open Access, Peer-reviewed, Refereed Journal

Prevalence and antimicrobial sensitivity of Enterohemorrhagic E.coli O157:H7 isolated from beef and lambs meat in tree Marked (Gougi, Diguél and Démbe) in Ndjamena city.

Saleh Bakhit Sinio[1] , Nada Abbas Mohammed Elamin[2], Mahamat Faiz Abakar[3], Mahamat Ibet Chaib[1] Abdrahman Mahamat Saleh[5] Mahamat Koulbou Abdoullay[4] and Mouktar Abaya Adoum[4]

[1]department of biomedical and pharmaceutical science, National Higher institute of science and technology Abeche-chad

[2]University of Nyala faculty of Education department of biology

[3]School of Human science Ndjamena- Chad

[4]Laboratory service and university hospital of children and mother Ndjamena Chad

[5]Department of Biology, Faculty of science and technology Adam Barka University Abeche – Chad

Abstract:

This study aimed to investigate the occurrence of *enterohemorrhagic E. coli (EHEC) O157* and evaluate its antimicrobial susceptibility in beef and lamb meat sold at three markets in N'Djamena, Chad. 746 meats samples during tows season were examined using standard microbiological techniques to isolate and identify *E. coli* and determine the occurrence of *E. coli O157*. Antimicrobial susceptibility was assessed using the disc diffusion method on Muller-Hinton agar. The inhibition zones produced by the tested antibiotics were measured, and the results were subjected to statistical analysis. The study reported the presence of *E. coli* and *E. coli O157* in the examined meat samples. The findings also indicated that the tested treatments exhibited inhibitory activity against *E. coli O157*. These results highlight the importance of microbiological monitoring of meat products and antimicrobial susceptibility testing to support food safety measures and reduce the potential risk of foodborne infections in N'Djamena, Chad.

Meat , Antimicrobial. Enteroheamorrhagic E. Coli.

Introduction

Escherichia coli (E. coli) is a Gram-negative bacterium commonly found in the intestinal tract of humans and warm-blooded animals. Although most strains are harmless, certain pathogenic strains, particularly Shiga toxin-producing *E. coli (STEC)*, are associated with serious foodborne infections. Among these, *E. coli O157:H7* is one of the most clinically significant serotypes because of its ability to produce Shiga toxins, which can cause severe gastrointestinal illness, including hemorrhagic colitis and, in some cases, hemolytic uremic syndrome (HUS). Young children and older adults are particularly vulnerable to severe complications of STEC infections (Mesari *etal.*, 2024).

Meat and meat products are important potential vehicles for the transmission of *E. coli O157:H7* to humans. Cattle are recognized as major reservoirs of this pathogen, while sheep and other ruminants may also carry and shed the organism without showing clinical signs. Contamination of beef and lamb may occur during slaughter, carcass dressing, transportation, storage, and retail handling, particularly when hygienic practices are inadequate. In addition, cross-contamination from contaminated equipment, surfaces, water, or food handlers may contribute to the dissemination of the pathogen along the meat production and distribution chain. Consumption of raw or inadequately cooked contaminated meat is a well-recognized route of infection, highlighting the importance of effective microbiological surveillance and food safety measures (Moon *etal*., 2000).

The occurrence of *E. coli O157:H7* in meat represents a significant public health concern because infections may range from uncomplicated diarrhea to severe and potentially life-threatening complications. Evidence from African studies indicates that this pathogen has been detected in human, animal, environmental, and food samples, although reported prevalence varies considerably among countries and sample types. A systematic review and meta-analysis published in 2025 reported a pooled prevalence of 4.7% across the African studies included in the analysis, with animal samples showing a higher pooled prevalence than human and environmental samples. However, the geographical distribution of the available evidence was uneven, emphasizing the need for further investigations in countries and settings where published surveillance data remain limited (Maite *etal.*, 2006)

Antimicrobial resistance is another important dimension of foodborne bacterial infections. The presence of antimicrobial-resistant *E. coli* in food-producing animals and meat products raises concerns about the dissemination of resistant bacteria through the food chain and the environment. A systematic review of African studies published in 2023 documented antimicrobial resistance and multidrug resistance among E. coli isolates from food, animal, human, and environmental sources. These findings support the importance of monitoring antimicrobial susceptibility patterns to better understand the distribution of resistance and inform appropriate public health interventions. It is important, however, to distinguish the detection of antimicrobial resistance in bacterial isolates from decisions about clinical treatment, particularly because antibiotics are generally not recommended for STEC infections and may increase the risk of complications in some patients.(Laurent *etal.*,2018)

In Chad, the microbiological safety of meat sold in traditional and retail markets warrants attention because beef and lamb are widely consumed foods. Nevertheless, the availability of published, market-specific data on the prevalence of *enterohemorrhagic E. coli O157:H7* and its antimicrobial susceptibility in meat products sold in N'Djamena needs to be established through a focused review of the literature. Data obtained from different markets can help characterize the local occurrence of this pathogen and identify potential differences in contamination between meat types and points of sale. Such evidence may contribute to strengthening food safety surveillance, improving meat-handling practices, and supporting public health strategies in Chad.(Doutoum *etal.*,2019)

2. Material and method

2.1 study area

The study was conducted in N'djamena, the capital city of Chad located in central Africa. Meat samples were collected from three major Markets: Démbe, Diguél and Gougi the study area is situated at approximately 12°o6 N latitude and a15° 03 E longitude with an elevation ranging from 294 to 298 m above sea level**.**

2.2 Study population

The study population consisted of raw beef and lambs meat samples offered for sale in the selected markets during the study period. Samples were collected during two season winter (December 2025 – January 2026) and

summer(May-June 2026) . approximately 50 meats sample were collected per sampling day. From selected markets

2.3 Study design and sampling method

A cross sectional study design was employed to investigate the occurrence of *Escherichia coli O157:H7* in row beef and lambs meat samples collected during the tow study period. The sample were collected using and a random sampling approach from tree markets (Démbe, Diguél and Gougi) in N'djamena.

2.4 Sample collection

A total of 746 raw meat samples were collected, comprising 472 lamb meat samples and 274 beef samples. Samples were obtained from selected markets in N'djamena. Approximately 25g of each sample was collected aseptically using sterile gloves and placed in sterile container the samples were transported to the laboratories university hospital Mather and children.(Cowan and Steels, 2002)

2.5 Sample preparation

In the laboratory, 25g of each meat sample was mixed with 225ml of sterile buffered peptone water to obtain an initial homogenized suspension. The mixture was homogenized using a sterile instrument for 2-3 minute. The resulting suspension was used for subsequent microbiological.(Cheesbreugh,2006)

2.7 Isolation and identification of *E. coli O157:H7*

The prepared meat suspension were processed for isolation of presumptive *E.coli* aliquots were streaked onto eosin methylene blue (EMB) agar (Difco Laboratories, USA). followed by aerobic incubation at 37°C for 24 hours. Colonies exhibiting characteristic morphological feature including, a green metallic sheen were selected for further examination.

Presumptive *E.coli* colonies were subjected to biochemical identification. The confirmed isolates were subsequently subcultured onto MacConkey sorbitol agar (SMAC., Oxoid)) and incubated at 37°C for 24 hours. Following incubation, the plates were examined for the colonies not ferment sorbitol. These colonies appeared relatively transparent or nearly colorless and were approximately 1mm in diameter. The colonies exhibiting the expected characteristic on SMAC agar were considered presumptive *E.coli O157:h7* isolates and were subjected to further identification, were applicable. (Cheesbreugh,2006)

2. 8 Antimicrobial Susceptibility Test

Antimicrobial susceptibility testing was performed on 215 *E. coli O157:H7* isolates using the agar disk diffusion method, based on the procedure described by Bauer *etal*., 2009). Pure cultures were suspended in sterile normal saline, and the resulting suspensions were inoculated onto Mueller–Hinton agar (Oxoid, Basingstoke, UK) using a sterile cotton swab. Antibiotic disks were then placed on the inoculated agar surfaces.

The antibiotic disc listed in the study include Cefotaxime(30 µg) ,Gentamicin(120 µg) ,Ofloxacin(5 µg) ,Amoxicillin(30 µg), Clavulanic acid(5 µg),Ciprofloxacin (CIP) (30 µg),Ceftriaxone(30 µg) ,Cefepime(30 µg) ,Tetracycline(75 µg), Amoxicillin(25 µg) ,Ampicillin(10 µg). These antimicrobial were selected on the basis of their availability and reported use in Laboratory service and at university hospital of mothers and children in Ndjamena Chad .

Following incubation at 37°C for 24 hours, the diameters of the inhibition zones were measured. The susceptibility results were interpreted according to the procedures used by the national committee for clinical standard(NCCLS) (Cheesbreugh,2006). Isolates exhibiting resistance to at least one antimicrobial agent in three or more antimicrobial classes were classified as multidrug-resistant (MDR).

The multiple antibiotic resistance (MAR) index was also assessed using the method adopted by study laboratories.

2.9 Data Analysis

The collected data were entered, coded, and organized using Microsoft Excel 2016. Descriptive statistics, including frequencies and percentages, were used to summarize the findings. Overall prevalence was calculated by dividing the number of positive samples by the total number of samples examined and multiplying the result by 100. Prevalence was also calculated separately for beef and lamb samples. The resulting data were organized to facilitate the comparison of findings by meat type and sampling period.

Result

the Isolation of *E.coli* and *Enterohemorrhagic E.coli O157:H7* during the tow session from tree markets during tow season.

A total of 306 sample were examined during winter comprising Lambs and beef meets samples collected from Gougi, Diguél and Démbe showed that 167(54.57%) positive of E.coli and 100 (59.88%) positive of *Enterohemorrhagic E.coli* while 139 (45.42%)sample were Negative

But a total of 440 sample were examined during summer comprising Lambs and beef meets samples collected from Gougi, Diguél and Démbe showed that 238(54.09%) positive of *E.coli* and 115 (48.31%) positive of *Enterohemorrhagic E.coli O157:H7* while 202 (45.42%) sample were Negative

Table (1) showed the distribution of *E.coli* and *Enterohemorrhagic E.coli O157:H7* isolation from meat samples collected from Démbe, Gougi and Diguél Market during tow season (summer and winter).

No	Season	Number of sample	Type of meat	Number of *E.coli*	Negative	*Enterohemorrhagic E.coli O157:H7*	Negative
1	Winter	218	Lamb meat	135(61.92%)	83(27.12%)	87(64.44%)	48(35.55%)
		88	Beef meat	32(36.36%)	56(63.63%)	13(40.62%)	19(59.37%)
	Total	306		167(54.57%)	139(45.42)	100 (59.88%)	67(40.11%)
2	Summer	254	Lamb meat	135(53.14%)	119(46.85%)	71(52.59%)	48(40.33)
		186	Beef meat	103(55.37%)	83(44.66%)	44(42.71%)	39(46.98%)
	Total	440		238(54.09%)	202(45.90%)	115(42.71%)	87(43.06)
3	Winter	306		167(54.57%)	139(45.42%)	100(59.88%)	67(40.11%)
4	Summer	440		238(54.09%)	202(45.90%)	115(48.31%)	87(43.06)
	Total	746		404(54.28)	342(45.84%)	215(53.21%)	127(37.13)

3.2 Antimicrobial sensitivity

3.2. **the Antimicrobial sensitivity of *Enterohemorrhagic O157:H7 E.* Isolated from lambs and beef meat from three Markets (Gougi , Démbe and Diguél) during the winter season**

The antimicrobial susceptibility profiles of 100 *Enterohemorrhagic E.coli O157:H7* isolates recovered from lambs and beef meat samples collected from three Markets(Gougi, Démbe and Diguél) during the winter season were determined against ten commonly used antimicrobial agent. table (2) and figure (1).

The isolated exhibited variable susceptibility patterns to the antimicrobial agents tested. The proportion of susceptibility isolated range from 0% to 99%, with intermediate susceptibility ranged 0% to 34% the proportion of resistant isolate range from 0% to100 % the highest susceptibility rate observed for Cefepime (FEP) with

99(99%) isolate being followed by amoxicillin/clavulanic acid(AUG)with 81(81%) isolate, gentamycin (GM) with 74(74%), ceftriaxone (CR) 75(75%) and ciprofloxacin with 70(70%).

The moderate susceptibility rate were recorded for tetracycline (TC), Ofloxacin (OFX) and Cefotaxime (CTX) with 56(56%), 51(51%), and 53(53%) isolate respectively.

The highest proportion of intermediate susceptibility was observed for amoxicillin (AML), with 34(34%) isolate, followed by tetracycline with 22(22%), Ofloxacin with 20(20%) and gentamycin with 18(18%). All isolated (100%) were resistant to ampicillin(AMP). Resistant was also observed for Cefotaxime (34%), amoxicillin (33%, Ofloxacin(29%), ciprofloxacin(21%) and tetracycline(22%).

Lower resistant rates were recorded for ceftriaxone (11%), gentamycin (8%) and amoxicillin/clavulanic acid (8%). No resistance was detected against Cefepime, as all 100% isolates were susceptible.

Table (2) **showed the Antimicrobial sensitivity of *Enterohemorrhagic O157:H7 E.coli* Isolated from lambs and beef meat in three Markets (Gougi, Démbe and Diguél) during the winter season**

	Type of sample	Dis conc µg	Sensitive	Intermediate	Resistance
			NO%	NO%	No%
1	Cefotaxime (CTX)	30 µg	53(53.00%)	13(13.00%)	34(34.00%)
2	Gentamicin (GM)	120 µg	74(74.00%)	18(18.00%)	8(8.00%)
3	Ofloxacin (OFX)	5 µg	51(51.00%)	20(20.00%)	29(29.00%)
4	Amoxicillin/Clavulaniacid (AUG)	30 µg	81(81.00%)	11(11.00%)	8(8.00%)
5	Ciprofloxacin (CIP)	5 µg	70(70.00%)	17(17.00%)	21(21.00%)
6	Ceftriaxone (CR)	30 µg	75(75.00%)	14(14.00%)	11(11.00%)
7	Cefepime (FEP)	30 µg	99(99.00%)	1(1.00%)	00(00.00%)
8	Tetracycline (TC)	75 µg	56(56.00%)	22(22.00%)	22(22.00%)
9	Amoxicillin (AML)	25 µg	33(33.00%)	34(34.00%)	33(33.00%)
10	Ampicillin (AMP)	10 µg	00(00.00%)	00(00.00%)	100(100.00%)

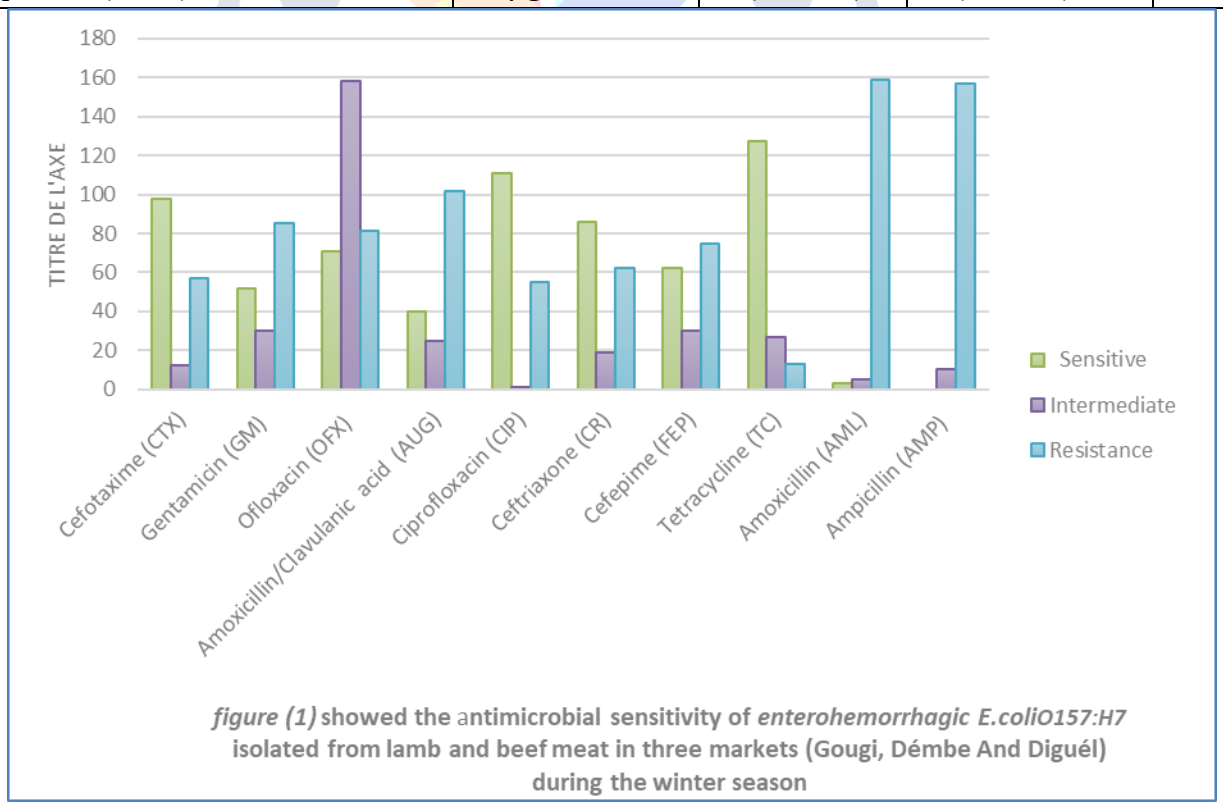

figure (1) showed the antimicrobial sensitivity of *enterohemorrhagic E.coliO157:H7* isolated from lamb and beef meat in three markets (Gougi, Démbe And Diguél) during the winter season

3.2.1. A**ntimicrobial sensitivity of *Enterohemorrhagic O157:H7 E.coli* Isolated from lamb and beef meat from three Market (Gougi, Démbe and Diguél) during the summer season**

The antimicrobial susceptibility profiles of 115 *Enterohemorrhagic E.coli O157:H7* isolates recovered from lambs and beef meat samples collected from three Markets(Gougi, Démbe and Diguél) during the summer season were determined against ten commonly used antimicrobial agent. table (3) and figure (2).

The isolated exhibited variable susceptibility patterns to the antimicrobial agents tested. The proportion of susceptibility range from o% to 75.65, with intermediate susceptibility ranged 2.60 to 26.08%, the proportion of resistant isolate range from 0.86% to 95% %. The highest susceptibility rate observed for Tetracycline (TC) with 87(75.65%) isolate being followed by ciprofloxacin (CIP) with 76(66.08%) isolate, Cefotaxime (CTX) with 70(60.86%), ceftriaxone (CR) 70(60.86%)and Ofloxacin (OFX) with 49(42.60).

The moderate susceptibility rate were recorded for Cefotaxime (CTX) and Ceftriaxone (CR) with 70(60.86%) and 59(51.30%) isolate respectively.

The highest proportion of intermediate susceptibility was observed for Gentamycin (GM), with 30(26.08%) isolate, followed by tetracycline with 27(23.47%) Cefepime with 21(18.26%) and Amoxicillin/Clavulanic acid (AUG) with 18(15.65%). Most isolated (93.91%) were resistant to Amoxicillin (AML) (95.65%). Resistant was also observed for ampicillin (AMP) (93.91%) Amoxicillin/Clavulanic acid (60%), Ofloxacin (47.82%) , Cefepime (FEP) (45.21%), Ceftriaxone (CR) (35.65) and ciprofloxacin(32.17%).

Lower resistant rates were recorded for tetracycline (0.86%), gentamycin (28.69%) and Cefotaxime (CTX) (28.69%). No Sensitive was detected against Ampicillin (AMP), as most (93.91%) isolates were resistant.

***Table (3)* showed the A**ntimicrobial sensitivity of *Enterohemorrhagic E.coli O157:H7* **Isolated from lambs and beef meat from three markets (Gougi, Démbe and Diguél) in Ndjamena city during the summer season.**

No	Type of sample	Dis conc µg	Sensitive	**Intermediate**	Resistance
			NO%	NO%	No%
1	Cefotaxime (CTX)	30 µg	70(60.86%)	12(10.43%)	33(28.69%)
2	Gentamicin (GM)	120 µg	52(45.21%)	30(26.08%)	33(28.69%)
3	Ofloxacin (OFX)	5 µg	49(42.60)	11(9.56%)	55(47.82%)
4	Amoxicillin/Clavulanic acid (AUG)	30 µg	28(24.34)	18(15.65%)	69(60%)
5	Ciprofloxacin (CIP)	5 µg	76(66.08%)	1(0.86%)	38(32.17%)
6	Ceftriaxone (CR)	30 µg	59(51.30%)	15(13.04%)	41(35.65%)
7	Cefepime (FEP)	30 µg	42(36.52%)	21(18.26%)	**52(45.21%)**
8	Tetracycline (TC)	75 µg	87(75.65%)	27(23.47%)	1(0.86%)
9	Amoxicillin (AML)	25 µg	2(1.73%)	3(2.60)	110(95.65%)
10	Ampicillin (AMP)	10 µg	0(00.00%)	7(6.08%)	108(93.91%)

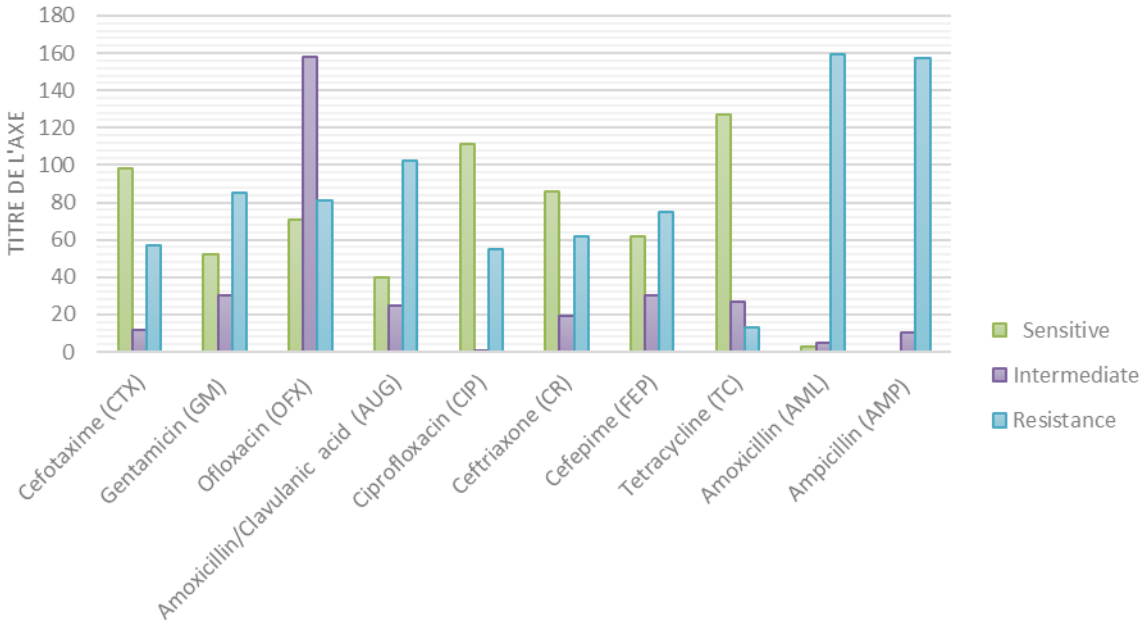

figure (2) showed the antimicrobial sensitivity of enterohemorrhagic *E.coli O157:H7* isolated from lambs and beef meat from three markets in Ndjamena during the summer season

Discussion

Enterohemorrhagic E.coli strain belonging to serogroup *EHEC O157:H7* are most frequently associated with human disease. However, *EHEC* strain of serotypes from human, *E.coli O157:H7* was recovered at higher prevalence in meat samples obtained from beef (42.64%) and lambs (58.51%) this study finding is in close agreement with reported prevalence of (28.04%) Wogayehu *etal.*, 2026 in the Bahir Ethiopia., 37% Uzeh *etal*., 2019 in Logos Nigeria, (46.26%)% Getachew *etal.*, 2022 in Tigray-Ethiopia , (50%) honore *etal.*, 2014 in the Benin, and 75% Aschalew *etal.*, 2023 Awi Ethiopia. This might be due to the high contamination of around environment, no respect of hygiene by meat seller. The overall variation in the prevalence of *EHEC O157:H7* may be result of different sampling techniques employed and laboratories methodologies used and may be due to the season that the studies were conducted in different countries at the different times. Since virulence gene detection was not performed. However, the high prevalence of *EHEC O157:H7* strain identified with the limited available methodology gives cause for concern.

The *E.coli EHEC O157:H7* strain isolated in this study were some found susceptible to antibiotics tested. Similar finding have been reported by other researcher Adem *etal.*,2007 and Christina *etal* ., 2022 However, resistant strains do exist mainly to ampicillin and amoxicillin and were detected in the present study the multi-drug resistance was found in 1.19%of the strain similar finding have been observed in other studies Maryse *etal*., 2018 and Shawlu *etal.*, 2006.

Although other enterohemorrhagic *E.coli* serotypes and the virulence factors of *E.coli O157:H7* were not investigated because the research is exclusively focused on Isolation of *E.coli O157:H7*. This study revealed that in two different row meat (beef and lamb) contaminated with *E.coli O157:H7*, suggesting possible risk of infection to people through consumption of row/ undercooked meat or cross contamination of other foods.

The available evidence also shows that meats from foods of animal origin serve as potential vehicles for transmitting *E.coli O157:H7*. Interruption of transmission to human should be given a high priority. The importance of proper handling and cooking of meat origin are probably as important in preventing *E.coli O157:H7*.

Coordinated action are needed to reduce or eliminate the risk posed by this organism at various stages in the food chain. These include good meat handling, good manufacturing practice and Hazard analysis of critical control points at every stage of the meat supply chain, from the abattoir, to the retailer, and to those involved with the handling and processing of such raw meat products in home environment. Antibiotic resistance in strains in this study is low. MDR may become a public health risk should they enter the food chain.

Conclusion

The present study demonstrated a considerable prevalence of Escherichia coli and enterohemorrhagic Escherichia *coli O157:H7* in beef and lamb meat samples collected from Gougi, Diguél, and Démbe markets in N'Djamena, Chad. The findings revealed substantial bacterial contamination during both winter and summer seasons, with a higher prevalence of *EHEC O157:H7* recorded during winter (59.88%) than summer (48.31%). Antimicrobial susceptibility testing revealed alarming resistance levels, particularly against ampicillin and amoxicillin, whereas Cefepime showed the highest susceptibility (99%). These findings indicate that retail meat may serve as an important vehicle for the transmission of foodborne pathogenic and antimicrobial-resistant bacteria, representing a potential public health risk. Therefore, strengthening meat hygiene practices, improving storage and handling conditions, implementing regular microbiological surveillance, and promoting the rational use of antibiotics in livestock production are essential to minimize bacterial contamination and the spread of antimicrobial resistance.

Recommendations

Based on the findings of this study, the following recommendations are proposed:

Regular microbiological surveillance of beef and lamb meat should be conducted in local markets to monitor the prevalence of *E. coli O157:H7*.

Government laboratories ought to implement continuous surveillance programs to monitor antimicrobial resistance trends and safeguard the effectiveness of key therapeutic drugs like Cefepime and Ciprofloxacin and Further molecular investigations are recommended to characterize the virulence and antimicrobial resistance genes of the isolated strains.

Improve hygiene and sanitation in slaughterhouses and meat markets.

Implement safe meat handling and storage practices.

Prevent cross-contamination between raw meat and ready-to-eat foods.

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