



## Antibiotic resistance and MAR index of *Escherichia coli* O157:H7 and *Salmonella* species isolated from smoked rat meat sold in Zaria, Nigeria

Terseer Iyene Addai

Department of Veterinary Public Health and Preventive Medicine, Faculty of Veterinary Medicine, Ahmadu Bello University, Nigeria

### Abstract

*Salmonella* spp. and *Escherichia coli* (*E. coli*) O157:H7 are important food-borne pathogens. This study determined the antibiotic resistance and multiple antibiotic resistance (MAR) index of *Salmonella* spp. and *E. coli* O157:H7 isolated from smoked rat meat sold in Zaria, Nigeria. A total of 384 samples were obtained for this study. The samples were analyzed in the laboratory by conventional biochemical techniques, Microgen GN-ID A+B kit for gram negative bacteria, and Rapid latex agglutination kit for *E. coli* O157:H7. Confirmed isolates were further subjected to antimicrobial susceptibility testing by the Agar disc diffusion technique. Results showed that both *Salmonella* and *E. coli* O157:H7 isolates exhibited multidrug resistance. Of the total (6) *Salmonella* isolates, 100% were resistant to tetracycline (30µg), doxycycline (30µg), amoxicillin/clavulanic acid (30µg), trimethoprim/sulfamethoxazole (25µg) and ampicillin (10 µg), 83.3% resistant to chloramphenicol (30µg) and cefixime (5µg), 66.6% resistant to gentamicin (30µg), 33.3% resistant to azithromycin (15µg), 16.6% resistant to kanamycin (30µg) and cefuroxime sodium (30µg), with no isolate (0%) resistant to ciprofloxacin (5 µg). Of the total (5) *E. coli* O157:H7 isolates, 100% were found to be resistant to ampicillin (10 µg) and penicillin (10 i.u), 80% resistant to streptomycin (10 µg), 60% resistant to trimethoprim/sulfamethoxazole (25 µg) and doxycycline (30µg), 40% resistant to chloramphenicol (30µg) and gentamicin (30µg), with no isolate (0%) resistant to imipenem (30µg). All isolates had MAR index of >2. The multiple antimicrobial resistance exhibited by the *Salmonella* and *E. coli* O157:H7 strains in this study is an indication of possible antibiotic abuse. Therefore, indiscriminate use of antibiotics should be discouraged.

**Keywords:** *Salmonella*, *E. coli* O157:H7, antibiotic resistance, multiple antibiotic resistance (mar) index

### 1. Introduction

Resistance of microbes to antibiotics is a global problem (Schroeder *et al.*, 2002) [51]. Antimicrobial resistance in *Enterobacteriaceae* is a threat to public health, especially in the developing countries (Karlowsky *et al.*, 2003; WHO, 2008) [23, 65]. Resistance could be caused by a variety of factors. The development of resistance to antimicrobial agents can occur through stable genetic change heritable from generation to generation through specific mechanisms including mutation, transduction, transformation and or conjugation (Goodman *et al.*, 1990) [16]. Also, the indiscriminate use of antibiotics in both veterinary and human medicines has led to the emergence of antibiotic resistant species of bacteria (Adetosoye and Rotilu 1984; Maurer, 2012) [2, 31]. However, van den Bogaard *et al.* (2001) explained that “usage” is the most significant factor responsible for antimicrobial resistance in bacteria.

*Salmonella* spp. and *E. coli* O157:H7 infections represent major threat to health worldwide (Schlundt 2001; Akinyemi *et al.*, 2007) [51, 4]. *Salmonella* spp. is identified as one of the most common causes of food borne infection worldwide, resulting in mortality and morbidity annually (Ellaine *et al.*, 2011). Enteric or typhoid fever and colitis/diarrhoeal disease are two major clinical syndromes caused by *Salmonella* infection (Fabrega and Villa, 2013) [15]. *Salmonella* spp. clinical manifestations include, abdominal pain, headache, fever, and transient diarrhoea or constipation and infection can cause fatal respiratory, hepatic, spleen, and/or neurological damage. If patients are not treated, mortality is

10 to 20%, decreasing to 1% among patients treated with the right antibiotics (Ohl and Miller, 2001; Parry *et al.*, 2002) [37, 44]. *E. coli* O157:H7 is a major cause of food borne illnesses such as acute diarrhea, hemolytic uremic syndrome and hemorrhagic colitis (Kinsinger *et al.*, 2017; Suardana *et al.*, 2017) [24, 55]. *E. coli* O157:H7 strains cause morbidity and mortality in animals and human, and they have become important zoonotic agents (Nataro and Kaper, 1998) [36]. *E. coli* O157:H7 infections can be acquired by coming in direct contact with animals or by person-to-person spread (Cho *et al.*, 2006) [12]

Even though the use of antibiotics has been proven to be an effective means for preventing and controlling bacterial infections, it can promote the selection and prevalence of drug resistant microbial populations when indiscriminately used (Braude, 1978; Threfall *et al.*, 1997) [8, 59]. A likely consequence of this is that life expectancy could fall due to people dying from easily treatable diseases (Sandle, 2014) [49]. Therefore, in providing treatment to the ever-changing resistance patterns of pathogenic bacteria, examining the drug resistance patterns of pathogens is important (Reda *et al.*, 2011) [46]. There have been several reports of *Salmonella* and *E. coli* O157:H7 isolated from different sources showing multidrug resistance (Reuben *et al.*, 2013; Tafida *et al.*, 2014; Agada *et al.*, 2014; Mshelbwa *et al.*, 2018) [47, 48, 56, 3, 35], but to the best of my knowledge nothing has been reported on the antibiotic susceptibility patterns of *Salmonella* and *E. coli* O157:H7 isolated from smoked rat meat sold, even though rat meat is widely traded and

consumed (Oyarekua and Ketiku, 2010; Tee *et al.*, 2012; Doyle, 2014; Addai, 2019) <sup>[43, 57, 13, 1]</sup>. In this light, this research was aimed bridging this knowledge gap. This is with the aim of providing evidence-based information to policy makers and public health workers which will aid in mapping out preventive strategies against *Salmonella* spp. and *E. coli* O157:H7 mortality and morbidity.

## 2. Materials and Methods

### 2.1 Study Area

The study was carried out in Zaria, Nigeria which is positioned between latitude 11° 07' N and 12° 00' N. and longitude 07° 44' E and 8° 00'. Zaria is located at central northern Nigeria and situated on a plateau at a height of 2200ft above sea level (Mortimore, 1970) <sup>[34]</sup>. The area is characterized by wet and dry seasons with fluctuation in temperature, with a monthly mean rising from January (21°C) and attaining a maximum in April (29°C) (Yakubu, 2009) <sup>[67]</sup>.

### 2.2 Sampling

Sample size was determined using the formula described by Thrusfield (1997) <sup>[61]</sup>. A total of 384 samples were obtained for the study. Samples were purchased from markets in the study area just the way they were sold to any customer but obtained in separate labeled sterile polythene bags and immediately transported to the Bacterial Zoonoses Laboratory, Department of Veterinary Public Health and Preventive Medicine, Ahmadu Bello University, Zaria, Nigeria for bacteriological analyses.

### 2.3 Laboratory procedures

#### 2.3.1 Non-selective pre-enrichment

Samples were cut and weighed aseptically to ten (10) grams, placed in 90ml (1.0%) buffered peptone water, homogenized using a laboratory stomacher and incubated at 37°C for 24 h.

#### 2.3.2 Isolation of *Salmonella*

Using sterile pipette, one (1) ml of the pre-enriched culture was transferred to 9ml of Rappaport Vassiliadis (RV) broth, which was then incubated at 37°C for 24 h. A loopful of the enriched Rappaport Vassiliadis (RV) broth was streaked onto Xylose Lysine Deoxycholate (XLD) agar plate and incubated at 37°C for 24 h. Two or three characteristic *Salmonella* colonies appearing pinkish-white, with or without black center on XLD agar were stored on nutrient agar slants, incubated at 37°C for 24 h and then refrigerated for biochemical confirmation.

#### 2.3.3 Isolation of *E. coli*

Using sterile pipette, one (1) ml of the pre-enriched culture was transferred to 9ml of EC broth, which was then incubated at 37°C for 24 h. A loopful of the enriched EC broth was streaked onto Eosin Methylene Blue (EMB) agar using sterile wire loop and the plates were incubated at 37°C for 24 h. Characteristic *E. coli* colonies appearing as greenish metallic sheen and with dark centers were selected, stored on nutrient agar slants, and refrigerated for further biochemical and serological analyses.

#### 2.3.4 Conventional biochemical tests

The following conventional biochemical tests were carried out on the presumptive *Salmonella* and *E. coli* O157:H7

isolates: Carbohydrate fermentation test in Triple Sugar Iron (TSI) agar (Oxoid, UK); Sulfur reduction, indole production and motility test in Sulfide Indole Motility (SIM) agar (Oxoid, UK); glucose fermentation pathway in Methyl Red-Voges Proskauer (MRVP) broth; Citrate utilization in Simmons Citrate agar and hydrolysis of urea in Urea agar (Oxoid, UK), according to Manufacturer's instructions. The isolates were further tested for sugar fermentation: maltose, manitol, lactose, glucose, sucrose and manose, according to the Manufacturer's instructions.

#### 2.3.5 Selective Plating and Identification of *E. coli* O157:H7 Colonies

Biochemically confirmed *E. coli* were streaked on Cefixime-Tellurite Sorbitol MacConkey agar (CT-SMAC agar) supplemented with Cefixime 50µg/L and Potassium tellurite 2.5mg/L (March and Ratnam, 1986) <sup>[29]</sup>. The plates were then incubated at 37°C for 24 h. Suspected colonies of *E. coli* O157 appeared as non-sorbitol fermenters, which is characterized as having a slightly transparent, almost colourless with a weak pale brownish appearance. Three to four colonies from the CT-SMAC agar plates were selected and stored on nutrient agar slants, and incubated at 37°C for 24 h. These were stored in a refrigerator for further biochemical and serological analyses.

#### 2.3.6 Standardized micro-substrate (Microgen GN-ID A+B) detection kit for gram negative bacteria

A 24 h culture of presumptive *Salmonella* and *E. coli* O157:H7 colonies on selective media were obtained and the test was carried out according to the Manufacturer's instruction (Oxoid, UK). A 4-digit code was then obtained and inputted into the computer identification software; which gave the probable identity of the organism tested in percentage score. All tests that gave less than 75% were not accepted as *Salmonella* or *E. coli*

#### 2.3.7 Serological Test

*E. coli* O157:H7 positive colonies were further confirmed serologically by using *E. coli* O157:H7 latex agglutinations assay (ISO 2003) <sup>[18]</sup>, containing latex particles coated with antibodies specific for *E. coli* O157 and *E. coli* H7 antigen. Colonies that agglutinated were considered to be *E. coli* O157:H7 and the test was carried out according to the Manufacturer's instruction

#### 2.3.8 Antimicrobial Susceptibility Testing

*In vitro* susceptibility of the *Salmonella* isolates to 13 antimicrobial agents, and the *E. coli* O157:H7 isolates to 8 antimicrobial agents, was investigated according to the Kirby-Bauer disk diffusion susceptibility test protocol (Bauer *et al.*, 1996) <sup>[7]</sup>. The antibiotics tested were penicillin (10 i.u), tetracycline (30µg), doxycycline (30µg), chloramphenicol (30µg), amoxicillin/clavulanic acid (30µg), ciprofloxacin (5µg), trimethoprim-sulphamethoxazole (25µg), cefixime (5µg), kanamycin (30µg), gentamicin (30µg), Azithromycin (15µg), imipenem (30µg), cefuroxime sodium (30µg), Streptomycin (10 µg) and ampicillin (10µg). The isolates were uniformly streaked on Muller-Hinton agar plates and the antibiotic impregnated discs were applied onto the inoculated plates using a pair of sterile forceps. The plates were then incubated at 37°C for 24 h, after which clear zones of inhibition for each antibiotic were measured using a transparent ruler. The results were

interpreted using the Clinical and Laboratory Standards Institute (CLSI) criteria (CLSI, 2015).

**2.4 Determination of multiple antibiotic resistance (MAR) index**

Multiple antibiotic resistance (MAR) index was determined for each isolate by using the formula  $MAR = a/b$ , where *a* represents the number of antibiotics to which the test isolate depicted resistance and *b* represents the total number of antibiotics to which the test isolate has been evaluated for susceptibility (Krumperman, 1983) [27].

**3. Results**

Table 1, shows the antimicrobial resistance of the 6 *Salmonella* isolates recovered from smoked rat meat, using 13 antibiotics. Of the total (6) *Salmonella* isolates, 100% were found to be resistant to tetracycline, doxycycline, amoxicillin/clavulanic acid, trimethoprim/ sulfamethoxazole and amoxicillin, 83.3% resistant to chloramphenicol and cefixime, 66.6% resistant to gentamicin, 33.3% resistant to azithromycin, 16.6% resistant to kanamycin and cefuroxime sodium, with no isolate (0%) resistant to ciprofloxacin. Figure 1, shows the zones of growth inhibition of *Salmonella* isolates to antibiotic panel.

Table 2, shows the MAR index of 6 *Salmonella* isolates. Two isolates each showed MAR indices of 0.5,0.6 and 0.7.

Table 3, Frequency of resistance of *Salmonella* isolates to panels of antibiotics. A resistant pattern: tetracycline-amoxicillin-ampicillin-doxycycline-sulphamethoxazole-chloramphenicol-kanamycin-azithromycin-cefixime-cefuroxime sodium was shown by 1 isolate. The most predominant pattern as shown by 6 (100%) of the *Salmonella* isolates was tetracycline-amoxicillin-ampicillin-doxycycline-sulphamethoxazole.

Table 4, shows the antimicrobial resistance of the 5 *E. coli* O157:H7 isolates, from smoked rat meat, using 8 antibiotics. Of the total (5) *E. coli* O157:H7 isolates, 100% were found to be resistant to ampicillin and penicillin, 80% resistant to streptomycin, 60% resistant to trimethoprim/sulfamethoxazole and doxycycline, 40% resistant to chloramphenicol and gentamicin, with no isolate (0%) resistant to imipenem. Figure 2, shows the zones of growth inhibition of *E. coli* O157:H7 isolates to antibiotic panel.

Table 5, shows the MAR index of 5 *E. coli* O157:H7 isolates. One isolate gave a MAR index of 0.3. a MAR index of 0.5 and 0.9 was observed by 2 isolates each.

Table 6, shows the frequency of resistance of *E. coli* O147:H7 isolates to panels of antibiotics. The most predominant resistant was ampicillin-penicillin and it was shown by 5 (100%) of the isolates.

**Table 1:** *In vitro* resistance of 6 *Salmonella* isolates to 13 antimicrobial agents.

Drug class	Antibiotic/ Disc potency	susceptibility pattern (n=6)		
		S (%)	I (%)	R (%)
Carbapenem	Imipenem (30µg)	66.7	33.3	0
Tetracyclines	Tetracycline (30µg)	0	0	100
	Doxycycline (30µg)	0	0	100
Beta-lactam	Amoxicillin/Clavulanic acid (30µg)	0	0	100
Aminoglycosides	Gentamicin (30µg)	0	33.3	66.7
	Kanamycin (30µg)	50	33.3	16.7
Cephems	Cefixime (5µg)	16.7	0	83.3
	Cefuroxime Sodium (30µg)	33.3	50	16.7
Macrolides	Azithromycin (15µg)	66.7	0	33.3
Phenicols	Chloramphenicol (30µg)	16.7	0	83.3
Penicillin	Ampicillin (10 µg)	0	0	100
Folate Pathway inhibitors	Trimethoprim/Sulfamethoxazole (25µg)	0	0	100
Fluoroquinolones	Ciprofloxacin (5 µg)	100	0	0

Key: µg = Microgramm, S = susceptible, I = intermediate, R = resistant

**Table 2:** MAR indices of *Salmonella* species (n=6)

MAR Index	Number
0.5	2 (33%)
0.6	2 (33%)
0.7	2 (33%)

**Table 3:** Frequency of resistance of *Salmonella* isolates to panels of antibiotics

Resistance pattern	Number of isolates resistant to panel of antibiotics
TE, AMC, AMP, DO, RL, C, CN, AZM, CFM, CXM	1
TE, AMC, AMP, DO, RL, C, CN, AZM	2
TE, AMC, AMP, DO, RL, C, CN,	3
TE, AMC, AMP, DO, RL, C	5
TE, AMC, AMP, DO, RL	6

Key: TE= Tetracycline; AMC= Amoxicillin/Clavulanic acid; AMP= Ampicillin; DO= Doxycycline; CXM= Cefuroxime Sodium; RL= Sulfamethoxazole; C= Chloramphenicol CN= Kanamycin, AZM= Azithromycin, CFM= Cefixime

**Table 4:** *In vitro* resistance of 5 *E. coli* O157:H7 isolates to 8 antimicrobial agents.

Drug class	Antibiotic/ Disc potency	Susceptibility patterns (n=5)		
		S (%)	I (%)	R (%)
Carbapenem	Imipenem (30µg)	100	0	0
Tetracyclines	Doxycycline (30µg)	40	0	60
Penicillins	Penicillin (10 i.u)	0	0	100
	Ampicillin (10 µg)	0	0	100
Aminoglycosides	Gentamicin (30 µg)	60	0	40
	Streptomycin (10 µg)	0	20	80
Phenicols	Chloramphenicol (30 µg)	60	0	40
Folate Pathway inhibitors	Trimethoprim/ Sulfamethoxazole (25 µg)	40	0	60

Key: µg = Microgramm, S = susceptible, I = intermediate, R = resistant

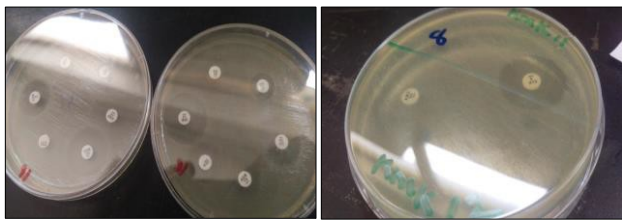
**Table 5:** MAR indices of *E. coli* O157:H7 (n=5)

MAR index	Number
0.3	1 (20%)
0.5	2 (20%)
0.9	2 (20%)

**Table 6:** Frequency of resistance of *E. coli* O147:H7 isolates to panels of antibiotics

Resistance pattern	Number of isolates resistant to panel of antibiotics
AMP, P	5
AMP, P, S, DO	3
AMP, P, S, DO, CN, C, RL	2

Key: AMP = Ampicillin; DO = Doxycycline; RL= Trimethoprim; C = Chloramphenicol; CN= Kanamycin; S = Streptomycin; P = Penicillin



**Fig 1:** Resistant patterns shown by *Salmonella* isolates to a panel of antibiotics.



**Fig 2:** Resistant patterns shown by *E. coli* O157:H7 isolates to a panel of antibiotics

**4. Discussion**

It was observed in this study that multiple antibiotic resistance was common among *E. coli* O157:H7 and *Salmonella* spp. These results are in agreement with previous reports in Nigeria (Raji *et al.*, 2007; Olonitola *et al.*, 2015) [45, 41].

This study revealed multi-drug resistance of the *Salmonella* isolates to commonly used antibiotics, with each isolate resistant to at least five (5) of the antibiotics tested. Nigeria has recorded increasing resistance to antimicrobials over the past 25 years (Okeke *et al.*, 2000; Iwalokun *et al.*, 2001) [38, 19] and increasing resistance to antimicrobials has also been recorded in other parts of the world such as Kenya (Kariuki *et al.*, 2006) [22], Singapore (Ling *et al.*, 2002) [28] and the Netherlands (Kivi *et al.*, 2005). The 100% susceptibility of

*Salmonella* spp. observed against the fluoroquinolone (ciprofloxacin) is similar to a report by Campos *et al.* (1990) [9], Sonstein and Burnham. (1993) [54] and Mathew *et al.* (2015) [30]. However, in Lagos, Nigeria, Akinyemi *et al.* (2007) [4] reported 18% reduced susceptibility of *Salmonella* spp. to ciprofloxacin. The high susceptibility of the *Salmonella* spp. to fluoroquinolones recorded in this study may be due to relatively [4] high cost of ciprofloxacin (Akinyemi *et al.*, 2007), thereby discouraging its indiscriminate use. The high (66.7%) susceptibility of the *Salmonella* isolates to imipenem (30µg) observed in this study agree with the findings of Mathew *et al.* (2015) [30] who reported 100% susceptibility of *Salmonella* enterica Serovar Typhi to imipenem (30µg), in Kaduna Metropolis, Nigeria. The high resistance of the *Salmonella* isolates, 100% to ampicillin and 83.3% to chloramphenicol could be due to the fact that these drugs most commonly used in human and poultry (Threlfall *et al.*, 2001) [60], therefore the development of resistance over time. The high percentage resistance of the *Salmonella* isolates to ampicillin and amoxicillin/clavulanic acid, which belongs to same class of antibiotics are similar with the findings of Mathew *et al.* (2015) [30] and Mshelbwala *et al.* (2018) [35].

Resistances to sulfamethoxazole-trimethoprim among *Salmonella* isolates have been documented from other parts of the world, Senegal (Bada-Alambedi *et al.*, 2006) [6], Mexico (Zaidi *et al.*, 2006) [68] and USA (Zhao. *et al.*, 2006) [69]. The *Salmonella* isolates in this study showed 100% resistance to the tetracyclines (tetracycline and doxycycline), which is similar to the findings of Kalu *et al.* (2008) [21] and Mathew *et al.* (2015) [30]. This finding was not surprising as both antibiotics belong to the same class. Eighty-three (83) percent resistance of the *Salmonella* isolates to cefixime, a cephalosporin was observed in this study. This trend is of particular concern because the extended spectrum cephalosporins are the antibiotics of choice for children (Weill *et al.*, 2004) [66].

Also, this study showed multidrug resistance of the *E. coli* O157:H7 isolates with each isolate resistant to at least to antibiotics tested. This result is in agreement with the findings by other studies: Kim *et al.* (1994) [25], Schroeder *et al.* (2002) [51] and Rueben *et al.* (2013) who reported multidrug resistance among *E. coli* O157:H7 isolates. The high resistance (100%) observed of the *E. coli* O157:H7 isolates to ampicillin and penicillin (Beta-Lactams) are similar with the findings reported by Shintandi and Sternesjo (2001) [53], Olatoye (2010) [40], Reuben *et al.* (2013) [47, 48] and Tafida *et al.* (2014) [56]. These are the most commonly available antibiotics used as growth promoters and routine chemoprophylaxis among livestock in Nigeria (Olatoye, 2010) [40] which may be a probable reason for the

high resistance to these antibiotics observed in this study. Another probable reason for the high resistance of the isolates to doxycycline (a tetracycline) and penicillin could be due to the fact that Tetracycline and penicillin (ampicillin) are first-line drugs which are routinely prescribed or readily purchased over the counter for self-medication (Ayukekbong *et al.*, 2017) <sup>[5]</sup>. The development of antimicrobial resistance might limit their use leading to treatment failure and onset of complications (Reuben and Owuna, 2013) <sup>[47, 48]</sup>. The 60% resistance observed by the *E. coli* O157:H7 isolates to sulfamethoxazole / trimethoprim is similar to findings of Rueben *et al.* (2013) <sup>[47, 48]</sup> who observed 84.2% resistance of *E. coli* O157:H7 isolates from fermented milk samples to this antibiotic in Nasarawa State, Nigeria. This antimicrobial agent is commonly used to treat respiratory infections, diarrhoea, mastitis, and other infections in beef and dairy cattle (Rueben *et al.*, 2013) <sup>[48]</sup>, which could be a reason for the high resistance observed in this study. Eighty percent (80%) resistance of the *E. coli* O157:H7 was observed against streptomycin. This finding was rather surprising as Cheesbrough (2000) <sup>[10]</sup> reported that the antibiotic is administered intravenously thereby restricting indiscriminate use. Also, the *E. coli* O157:H7 isolates were observed to have with 60% susceptibility to gentamicin and chloramphenicol. This finding is in agreement with reports by Walsh *et al.* (2006) <sup>[64]</sup> and Okolocha (2006) <sup>[39]</sup> who observed that all *E. coli* isolates tested were highly susceptible to gentamicin and trimethoprim. The quinolones (ciprofloxacin and ofloxacin), the aminoglycoside (gentamicin) and the phenicol (chloramphenicol) all showed great activity, agreeing with the findings of Iwu *et al.* (2017) <sup>[20]</sup> that they are the drug of choice for *E. coli* O157 infections. Susceptibility of the *E. coli* O157:H7 isolates to imipenem (30µg) was observed to be the highest (100%). This makes this antibiotic a considerable drug for the treatment of *E. coli* O157:H7 infections. This high susceptibility to imipenem (30µg) was also observed by Goncuoglu *et al.* (2010) <sup>[17]</sup> who documented a 100% susceptibility of *E. coli* O157:H7 isolates to the carbapenem. Among the several hundreds of known beta lactams the carbapenems, possess the broadest spectrum of activity and greatest potency against both Gram-positive and Gram-negative bacteria. They are administered when infected patients are gravely ill or harbour resistant bacteria, hence, they are referred to as “antibiotics of last resort” (Torres *et al.*, 2007) <sup>[62]</sup>.

The multidrug resistance observed in the study is of serious public health concern and requires urgent attention. The finding that all *Salmonella* and *E. coli* O157:H7 isolates were susceptible to ciprofloxacin and imipenem respectively, gives assurance of that these drugs are effective in the treatment of *Salmonella* and *E. coli* O157:H7 infections. Also, the 100% resistance observed among the *Salmonella* isolates to tetracycline amoxicillin/clavulanic, ampicillin, doxycycline and trimethoprim and 100% resistance of *E. coli* O157:H7 to ampicillin and penicillin, calls for urgent attention to find alternatives to these drugs.

Monitoring of both antibiotic consumption and multiple antibiotic resistances (MAR) is required in effective containment programs and audit of such programs (Osundiya *et al.*, 2013) <sup>[42]</sup>. As compared to other bacteria source tracking methods such as genotypic characterization, the MAR indexing method is cost effective, quick and

without the necessity of specialized training (Sandhu *et al.*, 2016) <sup>[50]</sup>. Thenmozhi *et al.* (2014) <sup>[58]</sup> stated that MAR index values > 0.2 indicate existence of isolate from high risk contaminated source with frequent use of antibiotics while values ≤ 0.2 show bacteria from source with less antibiotic's usage. Also, Mishra *et al.* (2013) <sup>[32]</sup> reported that MAR index of 0.2 or higher suggests high risk contamination sources and MAR index of 0.4 or higher is associated with human faecal sources of contamination. All the *Salmonella* and *E. coli* O157:H7 isolates in the study showed MAR index greater than 0.2. This shows that the isolates are likely to have originated from an environment with high human faecal contamination and where several antibiotics are used.

## 5. Conclusion

Multidrug resistance of the *Salmonella* and *E. coli* O157:H7 isolates was observed in this study. The development of resistance by the *E. coli* O157:H7 and *Salmonella* isolates to these drugs poses a major threat to public health. Indiscriminate use of antimicrobials contributes to the selection of antimicrobial resistance and could be a probable reason for the antibiotic resistance observed in this study. Therefore, there is need for more public health programs to be set up, to sensitize the general public on the dangers of the indiscriminate use of antibiotics.

## 6. Limitation

As a limitation, the number of isolates recovered from the samples and used for antibiotic resistance testing was low. Notwithstanding, the research emphasized the problems affecting the human population in connection with spread of antimicrobial resistant bacteria associated with processed meat, leading to increased morbidity and mortality.

## 7. Acknowledgment

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