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Full Length Research Paper

The incidence and antibiotics susceptibility of *Escherichia coli* O157:H7 from beef in Ibadan Municipal, Nigeria

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The incidence of *Escherichia coli* 0157: H7 was assessed in meat samples from slaughtered cattle in Ibadan metropolis by culturing on sorbitol MacConkey agar and confirmed using serological agglutination kits. The isolates were tested for susceptibility to seven commonly used antimicrobial agents. Out of the total of the 116 non-sorbitol fermenting (NSF) *E. coli* isolated from the samples, 71 (comprising of 18.4, 2.0, 3.2 and 4.8% from Bodija abattoir, Bashorun, Apata and Iwo Road slaughter slabs, respectively) were confirmed as *E. coli* O157:H7 serotype. Antibiotics susceptibility profile showed that all the isolates were resistant to one or multiple antibiotics, resulting in eight different resistance patterns. Tetracycline resistant isolates were the highest with 91.4% incidence. The results are of public health significance confirming cattle as major reservoir of EHEC and antimicrobial-resistant organisms. The high level of carcass contamination with microorganisms may be due to unhygienic slaughtering and meat processing engaged in these abattoir and slabs. Indiscriminate and misuse of antimicrobials on livestock in Nigeria could also be responsible for the multiple resistance pattern of the organism. Application of food hygiene practices such as HACCP is recommended for high quality farm to fork wholesome and safe meat for public consumption in Nigeria.

Key words: EHEC, *Escherichia coli*, beef, food safety Ibadan, Nigeria.

INTRODUCTION

Escherichia coli is a widespread intestinal commensal organism found in human and animal resulting from fecal contamination or contamination during food animal slaughter it is often found in soil, water and foods. Shiga toxin-producing *E. coli* (STEC) O157 has emerged as a public health threat following its initial identification as a pathogen in a 1982 outbreak of illness associated with the consumption of undercooked ground beef (Riley et al., 1983). There are many pathogenic strains causing a variety of illness in man and animals with associated clinical features and virulence factors depending on the serogroups from a food safety perspective, the EHEC groups are most important. Specifically, *E. coli* O157:H7 and O157: NM (non-motile) is recognized as major etiologic agent in hemorrhagic colitis (HC) and hemolytic-uremic syndrome (HUS) in humans (Thielman and Guerrant, 1999).

In the recent years, there are growing concern of bacterial adaptation and evolution resulting in the emergence of a number of zoonotic microorganisms in the food and water. Food-borne disease is a global public health concern. Mead et al. (1999) reported an estimated food-borne 76 million illnesses, 325,000 hospitalizations and 5,000 deaths annually in United States and in the United Kingdom, an estimated 2.37 million cases of food-borne gastroenteritis occurred in 1995 (Adak et al., 2002). Available data from United States Department of Agriculture Food Safety and Inspection Service indicated that 13 million Kg of ground beef were contaminated with *E. coli* O157:H7 on August 12, 1997 and 9.5 million Kg of beef trimmings and ground beef potentially contaminated with *E. coli* O157:H7 on July 19, 2002 (Sofos, 2008).

Abbreviations: NSF, Non-sorbitol fermenting; **HC,** hemorrhagic colitis; **HUS,** hemolytic-uremic syndrome; **LGAs,** local government authorities.

Table 1. Result of screening of slaughtered cattle from ibadan abattoir and slaughter slabs.

Abattoir/slaughter slabs	Number of samples	Number of NSF *E. coli* isolated	Number of *E. coli* O:157 isolated
Bodija	100	65 (26.0%)	46 (18.40%)
Basorun	50	13 (5.2%)	05 (2.00%)
Apata	50	16 (6.4%)	08 (3.20%)
Iwo road	50	22 (8.8%)	12 (4.80%)
Total	250	116 (46.4%)	70 (28.00%)

Transmission of pathogens to humans may occur from contaminated foods or water, or from infected persons, environments or animals. Food animals, in particular mature cattle are usually asymptomatic carriers of *E. coli* O157, including STEC (Meng et al., 1998) can also serve as reservoirs of antimicrobial-resistant bacteria. Resistance to antibiotics is highly prevalent in bacterial isolates worldwide, particularly in developing countries including Nigeria (Hart and Kariuki, 1998; Aibinu et al., 2007; Okeke et al., 2005; Ojo et al., 2009).

Carcass contamination from hides, skin and gut contents of animals can occur during bleeding, handling and processing of meat which include slaughtering, scalding, eviscerating and washing (Ikeme, 1990). Unhygienic floor dressing of carcasses is a common practice in this part of the world resulting in carcass contamination and isolation of pathogenic microorganisms from meat and slaughtering facilities in Nigeria (Umolu et al., 2006, Ojo et al., 2009).

Ibadan in Oyo State, Nigeria is located on geographic grid reference longitude 3° 5E, latitude 7° 20N with a population of over 2 million (Filani, 1994) and having Federal, State and Local Government participation in meat processing hygiene and inspection. The local government authorities (LGAs) are allowed legally to own slaughter slabs and abattoirs within their boundaries, subject to the approval of the supervising State Veterinary Division. This study investigated the incidence of STEC and characterize antimicrobial susceptibility of the isolates of *E. coli* O157 obtained from the meat samples from the main municipal abattoir and 3 other slaughter slabs at different LGAs to assess the wholesomeness and safety of this product resulting from contamination with STEC, a zoonotic food-borne pathogen.

MATERIALS AND METHODS

Two hundred and fifty replicates meat samples were randomly obtained from 800 carcasses of cattle during slaughter for public consumption at Bodija abattoir, Bashorun, Iwo Road and Apata slaughter slabs within Ibadan metropolis for a period of four weeks. The samples were aseptically collected using the method of carcass scraping adopted by Adams et al. (1980) in sterile sample bags and immediately transported to the laboratory for processing and culture. One gram of meat samples were suspended in 9 ml of peptone water and vortexed. After which, 0.1 ml of the same buffer was spread onto the surface of MacConkey agar and incubated for 24 h at 37°C and sub-cultured onto sorbitol-MacConkey agar plates. The non-sorbitol fermenting colonies were picked and characterized using standard biochemical tests. The sorbitol negative colonies were serologically typed using latex agglutination kit for *E. coli* O157:H7 (Oxoid DRO 120 M, UK). Hemolytic activity of the isolates was tested by culturing the isolates in 7% sheep blood agar (Oxoid Columbia blood agar) and incubating at 37°C for 24 h. Colonies with morphologic characteristics of *E. coli* were confirmed by conventional biochemical tests. The standard disk diffusion method was used for susceptibility testing (according to NCCLS, 1990). The antibiotic disks used include Nitrofurantoin 200 µg, Cefuroxime 25 µg, Norfloxacin 30 µg, Cotrimoxazole 25 µg, Gentamycin 10µg, Tetracycline 30 µg, Ciprofloxacin 25µg, Nalidixic acid 30 µg, Chloramphenicol 30 µg and Ampicillin 25 µg (Abtek Biological Ltd, England). *E. coli* NCTC 10418 and K-12 C600 were used as controls.

RESULTS AND DISCUSSION

E. coli O157:H7 serotypes were isolated from a 28.4% of the samples from the four meat processing center comprising of 18.4, 2.0, 3.2 and 4.8% from Bodija abattoir, Bashorun, Apata and Iwo Road slaughter slabs, respecttively (Table 1). Antibiotic susceptibility profile showed that all the isolates were resistant to one or multiple antibiotics. Tetracycline resistance was the highest in 91.4% of the isolates, while 72.9% resistance to nitrofurantoin and Chloramphenicol, 65.7% to cefuroxime, 44.3% resistance to cotrimozole, 35.7% resistance to nalidixic acid, 11.4% resistance to gentamicin (Figure 1). Eight different resistance patterns were observed (Table 2).

Contamination of meat and other animal products with entero-pathogenic bacteria and their contribution to the spread of antibiotic resistant bacteria to humans via food-chain is a global food safety concern. The results of this investigation showed high incidence microbial contamination of meat in the city as indicated by high incidence of *E. coli* (46.4%) which is usually an indicator organism. The high level of carcass contamination was envisaged due to the unhygienic slaughtering and meat processing engaged in these abattoir and slabs, where butchering of meat are done on concrete floor with inadequate slaughtering basic facilities including lack of potable water. The study also confirms cattle as a major reservoir of EHEC and antimicrobial-resistant organisms for meat consumers which have also been isolated from live cattle, meat and milk from other parts of the country by different researchers (Ojo et al., 2009; Aibinu et al., 2007; Lugal et

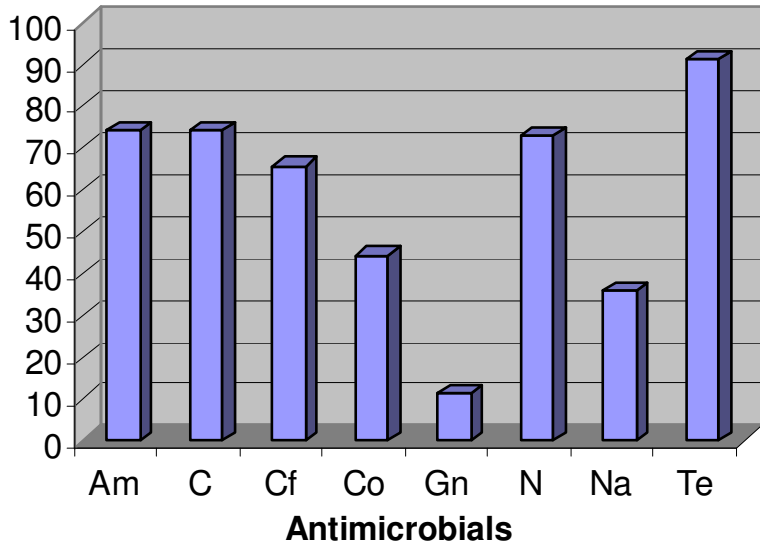

Figure 1. Percentage resistance of STEC isolates to common antimicrobials.

Table 2. Antibiotic resistance patterns of *E. coli* isolated from beef.

Pattern of resistance	Bodija	Bashorun	Apata	Iwo Road	Numbers of resistance isolates
Am,C,Cf,Co, Na,Nf,Te	8	0	2	3	13
Am,C,Cf,Co,Gn,N,Te	5	1	0	2	8
Am,C,Cf,N,Na,Te	10	1	0	1	12
Am C,Cf,N,Te	3	1	1	2	7
Am C,Co,N,Te	8	0	2	0	10
Am,C,N,Te	5	0	0	1	6
Am,C,Cf,N	4	0	1	1	6
Te	3	2	1	2	8
Total	46	5	7	12	70

Key: Am = Ampicillin, C = Chloramphenicol, Cf = Cefuroxime, Co = Cotrimoxazole, Gn = Gentamycin, N = Nitrofurantoin, Na = Nalidixic acid, Nf = Norfloxacin, Te = Tetracycline.

al., 2007; Umolu et al., 2006; Amosun et al., 2005). This study also confirms the widespread resistance to most commonly used antimicrobial agents in both human and animal health practice in Nigeria. The public health significance of these findings is that antimicrobial-resistant bacteria from food animals may colonize the human population via the food chain, contact through occupational exposure, or waste runoff from meat production facilities to the neighborhood. Indiscriminate and misuse of antimicrobials among livestock producers and marketers in Nigeria could also be responsible for the resistance pattern obtained in this study. Tetracycline resistance was the highest in 91.4% of the isolates; it is the most commonly available for use as growth promoter and routine chemoprophylaxis among livestock in Nigeria. The high prevalence of antibiotic resistance in bacteria in Nigeria and other developing countries has been associated with several factors including indiscriminate use due to unregulated access of non-professional to different classes of antimicrobial over-the-counter (Hart and Kariuki, 1998; Okeke et al., 1999). Application of food hygiene practices such as, Good Veterinary Practice and HACCP are therefore recommended for high quality farm to fork wholesome and safe meat processing for public consumption in Nigeria. There is also the need for further surveillance of resistant food borne pathogens and their genetic materials in animal and man ecosystem in order to achieve the global one health aspiration.

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