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EFFECT OF BEEF CHAIN ON THE PREVALENCE OF ENTEROHAEMORRHAGIC *ESCHERICHIA COLI* O157:H7 AND ITS PUBLIC HEALTH IMPLICATIONS

*** [1]Akinnibosun, F.I. and [2]Imade, O.S.**

[1]Department of Microbiology, Faculty of Life Sciences, University of Benin, PMB 1154, Benin City, Nigeria
[2] Department of Biological Sciences, College of Natural and Applied Sciences, Igbinedion University, Okada, Edo State, Nigeria
*faith.akinnibosun@uniben.edu; imade.stanley@gmail.com

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ABSTRACT

This study was carried out to analyze the prevalence of Escherichia coli O157:H7 in beef, and it implication on public health of consumers in Edo State, Nigeria. Prevalence was determined by direct plating on sorbitol MacConkey agar supplemented with cefexime (50 µg/l) and potassium tellurite (25 mg/l), after an initial pre-enrichment of samples on tryptic soy broth (TSB). Results of this study showed that overall prevalence of E. coli O157:H7 in the faeces and split carcasses at the abattoir were 53.00 % and 75.00 % respectively. Findings of this study indicated that the retail environment did not significantly contribute to change in the microbial load of E. coli O157:H7 when microbial load at the abattoir and at the retail market were compared. Logarithmic reductions of 1.5 - 0.5 log_{10} cfu/100 g rare done (undercooked) beef and ≥ 5 log_{10} cfu/100 g well cooked beef were recorded when their microbial load were compared with those at the retail market. There is therefore, an urgent need for the relevant regulatory agencies to intensify the monitoring of abattoirs, which is the major source of E. coli O157:H7 contamination, so as to ensure compliance to standard best practices in the entire beef chain.

1. INTRODUCTION

The beef chain is essentially categorized into the pre-harvest (pre-processing) stage, harvest (processing) stage, and post-harvest (post-processing) stage (Dahl et al., 2004; Minihan et al., 2004; Koohmaraie et al. 2005). The pre-harvest stage refers to the live animal in the farm or at the feedlot (Dahl et al., 2004). Stress and long periods of transport, whether to farms or

feedlots, increase faecal shedding of *E. coli* O157:H7, and as a consequence, the contamination of beef carcasses (Arthur et al., 2010). Therefore, efforts to limit stress of cattle prior to transport should be intensified to reduce shedding of *E. coli* O157:H7 upon arrival at the destination (Bach et al., 2004). Contamination routes and the population dynamics of *E. coli* O157:H7 on farms are currently unclear, limiting interventions and control options to those recommended within general guidelines for hygienic practices, quality assurance programs and/or application of HACCP principles to the extent possible (Arthur et al., 2010).

The harvest stage is widely considered to include transport of cattle from the farm to the lairage in the abattoir including slaughtering and dressing of the animal in the abattoir (Minihan et al., 2004). While some cross contamination may occur from carcass-to-carcass through contact with common equipment and workers hands in the abattoir, there is no published evidence, mainly in the developed countries, that *E. coli* O157:H7 has ever become established and multiplied in slaughtering and dressing operations and contaminated subsequent lots of beef (Swanson, 2011). This is because abattoir operations in developed countries employ modern technology to decontaminate hides prior to removal, minimize carcass contamination during the slaughtering process, reduce the likelihood of microbial attachment to exposed tissues, and decontaminate carcasses using steam, hot water or organic acid sprays prior to and after chilling (Bacon et al., 2000; Koohmaraie et al., 2005). Abattoir operations in most developing countries are a sharp contrast to what obtains in the developed countries and this can cause *E. coli* O157:H7 to become established and multiplied during abattoir operations. Abattoir operations in most developing countries are characterized by unhygienic practices that are associated with potential health risk to consumers due to the presence of pathogens in meat, and environmental contamination (Abdullahi et al., 2006). The abattoir attracts wild and domestic carnivores, rodents, flies and other insects that serve as vectors of diseases transmission to humans. In Nigeria, most butchers are poor and have not received occupational training. There is no compensation if meat is condemned and butchers may strongly and even violently resist condemnation of diseased and unwholesome meat (Okoli et al., 2005).

The post-harvest stage refers to handling of the beef carcasses during retail at the market through to consumption by the consumers. This is probably the most diverse stage of the process, as elements of it will vary depending on the final product that is marketed by the meat processor; the conditions of retail and of distribution; whether it is supplied in bulk to catering establishments or is purchased in small amounts for consumption in the home; and finally how it is prepared before final consumption (Koohmaraie et al., 2005). In Nigeria, the conditions of retail are a sharp contrast to what obtains in the developed countries, for example, in the United States of America and Europe. Most beef are sold in outdoor markets. Meat retailers are often inadequately and poorly dressed, thereby negatively impacting meat quality due to a probable cross-contamination of beef with pathogens of human origin (Okoli et al., 2005). The humid tropical environment encourages the breeding of flies which swarm and perch on meat displayed for sale. This constitutes a major nuisance at these markets and

aid in the spread of agents of diseases of significant public health importance (Okoli et al., 2005).

United States Center for Disease Control and Prevention (CDC) has indicated that the risk of *E. coli* O157:H7 from beef continues to be associated with processors and consumers who have not changed their handling/cooking habits to control the risk of the pathogen. Because beef handling habits have consistently remained unchanged in Nigeria, this study was carried out on beef processed and sold in Edo State, Nigeria to evaluate the prevalence of *E. coli* O157:H7, a major causative agent of diseases, such as, haemorrhagic colitis (Ina et al., 2003), haemolytic uraemic syndrome (Olsen et al., 2002; Thorpe, 2004), and thrombotic thrombocytopenia purpura (Tsai, 2003; Thorpe, 2004), that are of grave consequence to public health.

2. MATERIALS AND METHODS

2.1. Study Site

Edo State government abattoir situated at Ikpoba slope in Benin City, Edo State, Nigeria was the municipal abattoir selected for evaluating the processing stage of the beef chain in this study. Five municipal markets (New Benin, Oba, Ogida, Oliha, and Uselu markets), where beef processed at the abattoir are sold were selected for assessing the post-processing stage.

2.2. Study Design

The abattoir was visited twice weekly between July and December, 2013 covering the wet and harmattan seasons. On each visit 8 cattle were randomly selected from the lot of cattle to be slaughtered, which often ranged between 70 to 80 heads of cattle per day. A sample size of 400 cattle was employed in this study. To determine prevalence and microbial load of *E. coli* O157:H7 in the abattoir, faecal samples were collected from the randomly selected cattle before processing (pre-processing). Samples were also taken from the brisket and flank of the carcasses at two critical processing locations (skinning or pre-evisceration operation and splitting or post-evisceration operation), as well as from retailers at the respective markets where beef are sold (post-processing). The beef collected from the retailers were also subjected to two cooking preferences (rare cooking and well cooked preferences).

2.3. Prevalence of *E. coli* O157:H7 in Cattle Faeces Prior to Processing

Faecal samples were collected by inserting a hand covered with sterile latex gloves into the rectum of the randomly selected cattle after they had been stunned, and collecting about 100 grams of faeces into sterile containers which were stored on ice while being transported to the laboratory. Prevalence was done according to methods specified in ISO 16654:2001 (ISO, 2001). Ten grams of the faecal samples were each homogenized in 90 ml 2 % (w/v) sterile peptone water. One ml of each of the homogenates was inoculated into 9 ml sterile tryptic soy broth (TSB) and then incubated at 37°C for 18 hours for pre-enrichment of the homogenate. One ml of the TSB culture was then mixed with 9 ml sterile peptone water and

serial dilutions of up to 10^{-7} were made. One ml of each of the dilutions was placed on duplicate petri-plates and mixed with 19 ml of sterile molten sorbitol-MacConkey (SMAC) agar supplemented with cefixime ($5x10^{-5}$ g/ml) and potassium tellurite (0.0025 g/ml). The agar plates were allowed to solidify and then incubated at 37ºC for 18 to 24 hours. After incubation, straw coloured colonies on the plate presumptively identified as those of *E. coli* O157 were counted to obtain the microbial load of *E. coli* O157. These colonies were subsequently isolated and pure cultures of the isolates were then subjected to morphological and biochemical examinations as well as serological test to confirm *E. coli* O157:H7. The actual microbial load of *E. coli* O157 in the faecal samples was subsequently deduced and the prevalence of *E coli* O157:H7 in the faeces of cattle was estimated as a function of the detection rate of *E coli* O157:H7 in the faeces of all the cattle examined.

2.4. Prevalence of *E. coli* O157:H7 in the Carcasses of the Cattle during Processing

Samples were collected from two critical sites (briskets and flanks) on the carcasses during three critical processing operations. Sampling of the skinning operation was done by swabbing a 100 cm^2 area of each sampling site with 10 sterile cotton tipped sticks which had been pre-moistened with 2 % w/v peptone water according to the technique specified by ISO 17604:2003 (ISO, 2003). Area of sampling was delimited by sterile templates to ensure precise measurement of the 100 cm^2 sampling area. After swabbing, the swab sticks were put into a sterile container containing 100 ml of 2 % w/v peptone water, and was stored on ice while being transported to the laboratory. Sampling of the splitting operation involved the collection of about 100 g of the freshly split carcasses into sterile container and onward transportation to the laboratory in ice-pack. Microbiological analysis to determine the prevalence of *E. coli* O157:H7 in the carcasses was subsequently conducted within 6 hours of sampling according to the methods specified in ISO 16654:2001 (ISO, 2001).

2.5. Prevalence of *E. coli* O157:H7 in the Carcasses of the cattle after Processing

From eight retailers at the market, 300 g of beef cuts were collected in sterile containers for laboratory analysis. Prevalence during retail display was determined according to the methods specified in ISO 16654:2001 (ISO, 2001). To predict the prevalence of *E. coli* O157:H7 in beef cuts consumed by consumers using the methods of Juneja et al. (1997), 100 g portions of beef cuts each with equivalent surface area of 100 cm^2 were initially cooked to temperatures of 56ºC for 20 minutes (rare cooking preference). Similar portions of beef cuts were also cooked to temperatures of 100ºC for 20 minutes (well cooked preference which are often performed by consumers in their homes for mitigation of microbial load in the beef). Prevalence of *E. coli* O157:H7 in the two cooking preferences was subsequently determined by the methods specified in ISO 16654:2001 (ISO, 2001).

2.6. Statistical Analysis

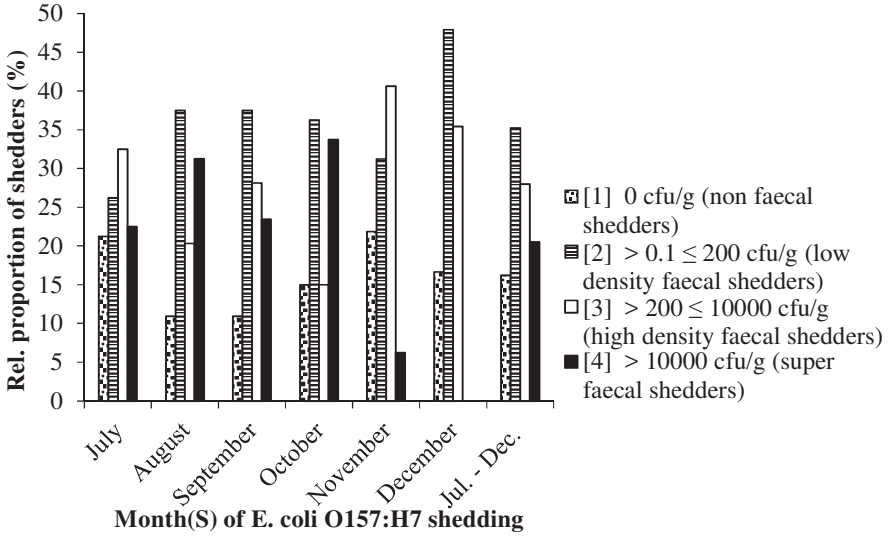
Statistical analysis was done using the statistical package for social sciences (SPSS) software, version 19. Microbial load of *E. coli* O157:H7 on carcasses and beef cuts were presented as colony forming units (cfu)/g. Sample size was determined using the SPSS sample power.

Chi-square test was used to assess how seasonal variations influenced the prevalence of *E. coli* O157:H7, while Pearson correlation test was used to evaluate the effect of thermal intervention on beef cuts displayed in the retail market prior to consumption.

3. RESULTS AND DISCUSSION

Carcasses of cattle during processing in the abattoir are often contaminated from the hides which have been previously contaminated by faeces from *E. coli* O157:H7-colonized cattle (Arthur et al., 2010). Therefore, there is a need to understand the dynamics of transmission of *E. coli* O157:H7 between animals in cattle-associated environments. These cattle-associated environments include production (that is, pasture or feedlots) and harvest (transportation and lairage prior to entering into the processing at the abattoir). The probability of faecal contamination of the hides of cattle is largely dependent on the shedding intensity of the *E. coli* O157:H7-colonized cattle, with the super shedders essentially responsible for the transmission of *E. coli* O157:H7 among cattle (Robinson et al., 2004; Matthews et al., 2006). The results of the data presented by Robinson et al. (2004) and Matthews et al. (2006) led to the conclusion that if 5 % of cattle in the lairage of an abattoir were super shedders, then the spread of *E. coli* O157:H7 among cattle could be controlled.

Figure 1 represents the relative proportions of cattle that shed *E. coli* O157: H7 in their faeces during the period of sampling. It shows that the shedding cattle were categorized into four main groups (non faecal shedders, low density faecal shedders, high density faecal shedders, and super faecal shedders) based on colonization by *E. coli* O157: H7. The proportion of the non faecal shedders (cattle that did not shed faecal *E. coli* O157: H7 (0 cfu/g)) ranged from 21.25 % in July to 10.94 % in August and September. In October, November, and December the values recorded were 15.00 %, 21.88 %, and 16.67 % respectively. The overall mean proportion of the non faecal shedders for the entire period of sampling was estimated at 16.25 %. For low density faecal shedders (cattle shedding below 200 cfu/g), the cattle sampled in December had the highest proportion (47.92 %), while the lowest proportion was recorded in July (26.25 %). The mean proportions recorded in August, September, October, and November were 37.50 %, 37.50 %, 36.25 %, and 31.25 % respectively. Overall mean proportion for the entire period of sampling was 47.92 %. The mean proportions of high density faecal shedders (those cattle shedding above 200 cfu/g but below 10^4 cfu/g) were 32.50 %, 20.31 %, 28.13 %, 15.00 %, 40.63 %, and 35.42 % in July, August, September, October, November, and December respectively; while for the super faecal shedders (cattle shedding above 10^4 cfu/g) they were 22.50 %, 31.25 %, 23.44 %, 33.75 %, 6.25 %, and 0.00 % in July, August, September, October, November, and December respectively. The overall mean proportions of the faecal shedders for the entire period of sampling were 35.25 %, 28.00 %, and 20.50 % for low density, high density, and super faecal shedders respectively, while it was 16.25 % for the non faecal shedders.



Rel: Relative; 1: non faecal shedders; 2: low density faecal shedders; 3: high density faecal shedders; 4: super faecal shedders

Figure 1: Relative proportions of *E. coli* O157:H7-faecal shedders

The data presented in Figure 1 showed that it was only in December that super shedders (0.00 %) were less than 5 % in the lot of cattle that were taken from the lairage for processing. Unlike the months of July to October, the proportion of super shedders obtained in the month of November (6.25 %) was not significantly different from the threshold value of 5 %. Thus, in agreement with the studies of Robinson et al. (2004) and Matthews et al. (2006), findings in the present study do suggest that in November and December *E. coli* O157:H7 faecal contamination of cattle hides may be significantly controlled when compared with the other months of July to October. Chi-square test on the effect of seasonal variations on *E. coli* O157:H7-faecal shedding, as shown in Table 1, also revealed that super shedders were significantly ($P < 0.05$) most likely to be dependent on the season of sampling.

Table 1: Chi-Square test of association between *E. coli* O157:H7-shedding cattle and season of sampling

				Season						Total
				Aug.	Dec.	Jul.	Nov.	Oct.	Sept.	
Shedding cattle	High density faecal shedders	Count	119	0	0	0	0	0	0	119
		Expected Count	62.4	9.3	10.7	8.7	10.3	8.9	8.7	119.0
		% within Season	37.8%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	19.8%
	Low density faecal shedders	Count	100	0	0	0	0	0	0	100
		Expected Count	52.4	7.8	9.0	7.3	8.7	7.5	7.3	100.0
		% within Season	31.7%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	16.6%
	Non faecal shedders	Count	0	47	54	44	52	45	44	286
		Expected Count	149.9	22.4	25.7	20.9	24.7	21.4	20.9	286.0
		% within Season	0.0%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%	47.6%
	Super shedders	Count	96	0	0	0	0	0	0	96
		Expected Count	50.3	7.5	8.6	7.0	8.3	7.2	7.0	96.0
		% within Season	30.5%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	16.0%
	Total	Count	315	47	54	44	52	45	44	601
		Expected Count	315.0	47.0	54.0	44.0	52.0	45.0	44.0	601.0
		% within Season	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%	100.0%

Chi-square tests

	Value	DOF	P - value
Pearson Chi-Square	601.000	18	0.000
Likelihood Ratio	831.763	18	0.000
Number of Valid Cases	601		

Table 2 represents the prevalence of *E. coli* O157: H7 in the faeces of shedding cattle. The lowest prevalence of *E. coli* O157: H7 shed in the faeces of cattle was recorded in December (45.83 %), while it was highest (56.25 % and 56.25 %) in July and September respectively. The overall prevalence for the entire period of sampling was estimated at 53.00 %.

Table 2: Prevalence of *E. coli* O157: H7 in faeces of shedding cattle

Month(s) of visitations	Number of cattle sampled	Mean prevalence of *E. coli* O157: H7 in faeces	
		Frequency of occurrence (F)	Percentage F (P)
July	80	45/80	56.25
August	64	34/64	53.13
September	64	36/64	56.25
October	80	44/80	55.00
November	64	31/64	48.44
December	48	22/48	45.83
July to December	400	212/400	53.00

$$F = \frac{Number\ of\ \ cattle\ positive\ for\ E.\ coli\ O157:H7}{Total\ number\ of\ \ cattle\ tested} \quad (1)$$

$$P = \frac{Number\ of\ \ cattle\ positive\ for\ E.\ coli\ O157:H7}{Total\ number\ of\ \ cattle\ tested} \times 100 \quad (2)$$

However, the findings of the present study did not satisfy the recommendation proposed in the study of Authur et al. (2009), which states that the *E. coli* O157:H7 faecal prevalence of cattle in the lairage should be maintained below 20% and levels of shedding need to be kept below 200 cfu/g to minimize the contamination of cattle hides. All the *E. coli* O157:H7 faecal prevalence of cattle in the lairage of the abattoir under study recorded from the entire period of sampling (Table 2) significantly exceeded the recommended value of 20%, thus, suggesting that cattle hides may be grossly contaminated irrespective of the month of sampling. Such variations in the *E. coli* O157:H7 faecal prevalence in the lairage of abattoirs is not uncommon, since data of faecal shedding from the work of several authors (Robinson et al., 2004; Matthews et al., 2006) have also found large variability in the prevalence of *E. coli* O157:H7 among different cattle populations. These variations in the faecal *E. coli* O157:H7 prevalence may be due to differences in environment suitability for *E. coli* O157:H7 growth, cattle movement onto and away from farms, as well as carriage level and persistence in some cattle. Therefore, to comply with the recommendation of Arthur et al. (2009), cattle from the farm feedlots must first be screened for *E. coli* O157:H7 faecal prevalence and microbial load before they are transferred into the lairage prior to processing. Even though cattle were not screened prior to entering the lairage in this study, the significant increase in the proportion of cattle shedding *E. coli* O157:H7 at levels below 200 cfu/g during the months of November and December corroborate the previous suggestion that faecal contamination of cattle hides may be significantly controlled during this period.

Besides the contamination of cattle hides by super shedders, various studies have showed that another important factor in cattle hide contamination and subsequent carcass contamination is the lairage environment at the abattoir (Small et al., 2002; Avery et al., 2002; Arthur et al., 2007; Arthur et al., 2008; Mather et al., 2008). With the high cattle density and confined spaces associated with lairage environments in abattoirs, it is presumed that if cattle from respective farm feedlots were not preliminarily screened for *E. coli* O157:H7 faecal prevalence and microbial load, super shedding cattle would have a large impact on the overall contamination of cattle currently in these environments and those that will enter these areas at subsequent times (Arthur et al., 2010). In these areas hundreds to thousands of cattle transverse the same approximate path each day. Cattle shedding over 10^4 *E. coli* O157:H7 cfu/g of faeces could readily deposit enough pathogen-laden material to contaminate a significant portion of cattle lots passing through the spaces for the remainder of the processing cycle. Since the hides are a major source of *E. coli* O157:H7 entering the processing unit of abattoirs, one of the most critical steps in preventing beef carcass contamination is the hygienic removal of the cattle hides. Therefore, several hide-directed decontamination interventions have been embraced by abattoirs (Byrne et al., 2000; Bosilevac et al., 2005; Arthur et al., 2007; Brichta-Harhay et al., 2008). Hide decontamination interventions ranged from spraying of solutions of acetic acid (10 %), sodium hydroxide (3 %), sodium metasilicate (4 %), followed by high pressure washing to deluging with solutions of potassium cyanate (2.4 %) or sodium sulphide. The abattoir in this study, however, utilized a deluging procedure in which cattle hides were cremated, thereby eliminating or significantly reducing contamination of carcasses by the hides. However, upon

the removal of the hides, prevalence of *E. coli* O157:H7 on carcasses is assumed to be proportional to the prevalence of cattle shedding the pathogen (Cassin et al., 1998).

The prevalence of *E. coli* O157:H7 recovered from carcasses during processing is presented in Table 3. Pre-evisceration-*E. coli* O157:H7 prevalence was highest in October (46.88 %) and lowest in December (7.29 %). In July, August, September, and November, pre-evisceration prevalence were 41.25 %, 44.53 %, 31.25 %, and 14.38 % respectively. Overall mean pre-evisceration prevalence for the period of July to December was estimated at 33.50 %. The lowest post-evisceration *E. coli* O157:H7-prevalence was also recorded in December (47.29 %), while it was highest in August (87.50 %). Post-evisceration *E. coli* O157:H7 prevalence for July, September, October, and November were 81.25 %, 80.47 %, 82.50 %, and 60.16 % respectively. Overall mean post-evisceration prevalence for the period of July to December was estimated at 75.00 %. Despite cremation (the most proficient means of anti-microbial decontamination) of the cattle hides, *E. coli* O157:H7 was recovered from the carcasses (Table 3) sampled in this study during processing (slaughtering); particularly during July to October (the peak cattle *E. coli* O157:H7 shedding season), where it was most prevalent. This finding agreed with the work of Bosilevac et al. (2005). However, in contrast to the finding of Bosilevac et al. (2005), the number of positive carcass samples recorded during pre-evisceration (skinning) was significantly increased after the carcasses were eviscerated and split (post-evisceration). The overall prevalence of carcass contamination with *E. coli* O157:H7 during processing was significantly greater than that of fecal prevalence prior to processing (Tables 2 and 3). Furthermore, in cattle which was initially *E. coli* O157:H7-faecal negative prior to processing, carcass samples from the same cattle were found to be *E. coli* O157:H7 positive during processing. This finding suggests that cross-contamination of carcasses may be occurring during processing in the abattoir. Finding *E. coli* O157:H7 at these levels during processing is not completely unexpected, given the potential for bacterial contamination to occur when large numbers of animals infected with *E. coli* O157:H7 are being processed. Several mechanisms may be responsible for dissemination of *E. coli* O157:H7 during processing: Carcasses may come directly in contact with each other during processing with the potential for transfer of microorganisms. Air and water-borne contaminations are also possible. Besides, processing interventions to mitigate cross contamination of carcasses may be grossly inefficient.

Table 4 represents the prevalence of *E. coli* O157:H7 recovered from beef cuts during retail display. Prevalence was highest in August (100.00 %) and lowest in December (66.67 %). In July, September, October, and November, prevalence were 90.00 %, 89.06 %, 87.50 %, and 79.69 % respectively. Overall prevalence was estimated at 86.50 %. The data obtained in the present study indicate that there was no significant difference when the prevalence of *E. coli* O157:H7 in processed beef cuts displayed in retail markets (Table 4) were compared to those obtained during processing at the abattoir (Table 3). This indicates that contamination of the beef cuts may largely be attributed to the efficiency of the intervention strategies that are employed during the pre-harvest and processing stages at the abattoir (Adam and Brülisauer, 2010; Loretz et al., 2011; Smith et al., 2012). While pre-harvest intervention (cremation of cattle carcasses prior to entering the processing stage at the abattoir in this study) was

relatively effective in mitigating *E. coli* O157:H7 contamination of cattle at the pre-harvest stage; interventions during the processing stage (washing of carcasses with water, and trimming of visible defects on carcasses) at the abattoir in this study were grossly inefficient and inadequate, and may have significantly contributed to the high prevalence of *E. coli* O157:H7-contaminated carcasses and beef cuts at the abattoir (overall prevalence estimated at 75.00 %) and in the retail market (overall prevalence estimated at 86.50 %) respectively.

Table 3: Prevalence of *E. coli* O157: H7 in cattle carcasses during processing

Processing operations	Month(s) of visitations	No. of sampling sites	Prevalence of *E. coli* O157:H7 in carcasses	
			F	P
Skinning (Pre-evisceration)	July	160	66/160	41.25
	August	128	57/128	44.53
	September	128	40/128	31.25
	October	160	75/160	46.88
	November	128	23/160	14.38
	December	96	7/96	7.29
	July to December	800	268/800	33.50
Splitting (Post-evisceration)	July	160	130/160	81.25
	August	128	112/128	87.50
	September	128	103/128	80.47
	October	160	132/160	82.50
	November	128	77/128	60.16
	December	96	46/96	47.92
	July to December	800	600/800	75.00

Table 4: Prevalence of *E. coli* O157: H7 on beef cuts during display at the retail markets

Month(s) of visitations	No. of beef cuts sampled	Prevalence of *E. coli* O157:H7 in	
		F	P
July	80	72/80	90.00
August	64	64/64	100.00
September	64	57/64	89.06
October	80	70/80	87.50
November	64	51/64	79.69
December	48	32/48	66.67
July to December	400	346/400	86.50

The prevalence of *E. coli* O157:H7 on the surface of beef cuts processed at the abattoir in this study which was cooked at a temperature of 56^{o}C for 20 minutes (rare cooking preference) are shown in Table 5. The lowest *E. coli* O157:H7-prevalence was recorded in December (8.33 %), while it was highest in July (61.25 %). *E. coli* O157:H7-prevalence on the rare beef cuts for August, September, October, and November were 57.81 %, 45.31 %, 51.25 % and 12.50 % respectively. Overall prevalence for the entire period of study was estimated at 42.00

%. Upon cooking of the beef cuts to a temperature of 100ºC for 20 minutes (well cooked preference), *E. coli* O157:H7-prevalence was recorded only in July (1.25 %). Overall prevalence was estimated at 0.25 %.

Table 6 represents the overall microbial load of *E. coli* O157:H7 in beef cuts during post-processing. For beef cuts displayed at retail market, microbial load of ≤ 50 cfu/100 cm^2 was obtained. Overall proportions of *E. coli* O157:H7-positive samples containing ≤ 10, 20, 30, 40 and 50 cfu/100 cm^2 were 53.00 %, 18.50 %, 10.75 %, 2.75 %, and 1.50 % respectively, while in 13.50 % of beef cuts sampled, no *E. coli* O157:H7 was recovered. For rare done beef cuts at consumption, microbial load of ≤ 40 cfu/100 cm^2 was recorded. Overall proportions of *E. coli* O157:H7-positive samples containing ≤ 10, 20, 30, 40 and 50 cfu/100 cm^2 were 23.50 %, 13.75 %, 3.00 %, 1.25 %, and 0.00 % respectively, while in 58.50 % of beef cuts sampled, no *E. coli* O157:H7 was recovered. The maximum microbial load obtained from the well cooked beef cuts at consumption was 5 cfu/100 cm^2, with an overall proportion of *E. coli* O157:H7-positive samples estimated at 0.25 %. No *E. coli* O157:H7 was recovered in 99.75 % of well cooked beef cuts.

Table 5: Prevalence of *E. coli* O157: H7 in beef cuts cooked using rare done preference

Month(s) of visitations	Number of rare beef cuts sampled	Prevalence of *E. coli* O157:H7 in rare beef cuts	
		F	P
July	80	49/80	61.25
August	64	37/64	57.81
September	64	29/64	45.31
October	80	41/80	51.25
November	64	8/64	12.50
December	48	4/48	8.33
July to December	400	168/400	42.00

Table 6: Microbial load of *E. coli* O157: H7 on beef cuts during post-processing

Samples	N	Overall microbial load of *E. coli* O157: H7 in beef cuts (cfu/100 cm^2)											
		>0.1 ≤ 10		> 10 ≤ 20		> 20 ≤ 30		> 30 ≤ 40		> 40 ≤ 50		> 50	
		A	P (%)	A	P (%)	A	P (%)	A	P (%)	A	P (%)	A	P (%)
Beef at retail market	400	212/400	53.0	74/400	18.50	43/400	10.75	11/400	2.75	6/400	1.50	0/400	0.0
Rare done beef	400	94/400	23.5	55/400	13.75	12/400	3.00	5/400	1.25	0/400	0.00	0/400	0.0
Well cooked beef	400	1/400	0.25	0/400	0.00	0/400	0.00	0/400	0.00	0/400	0.00	0/400	0.0

N: Number of beef cuts sampled; A: Relative proportion of the varying concentrations of *E. coli* O157:H7 in the sampled beef cuts; P: Percentage relative proportions, cfu: colony forming unit

The findings of the present study reveal that post-processing interventions are mandatory prior to consumption of beef cuts processed at the abattoir in this study, so as to mitigate or eliminate the exposure of humans to *E. coli* O157:H7-contaminated beef cuts. Thermal

inactivation of *E. coli* O157:H7 on the beef cuts by cooking to different internal temperatures (Table 6) were employed in this study as an intervention strategy.

The microbial load of *E. coli* O157:H7 present during retail in the market and those present on the rare beef cuts after cooking at 56^{o}C were correlated. The microbial load obtained during retail and after the rare cooking had a statistically significant ($P < 0.05$) linear relationship which was positively correlated ($r = 0.168$), indicating that the rare cooking preference employed did not significantly reduce the microbial load of *E. coli* O157:H7 present on the beef cuts during retail display. The microbial load of *E. coli* O157:H7 recovered from beef cuts during retail in the market and those present on the well cooked beef cuts after cooking at 100^{o}C were also correlated. The microbial load recovered during retail and after cooking at 100^{o}C did not have a statistically significant ($P > 0.05$) linear relationship and was weakly positively correlated ($r = 0.237$), indicating that the well cooking preference employed significantly reduced the microbial load of *E. coli* O157:H7 present on the carcasses during retail display. These findings agreed with the work of Cassin et al. (1998) and Smith et al. (2012), which showed that efficient thermal inactivation is likely the most effective barrier against exposure to *E. coli* O157:H7.

Unlike the well-done cooking preference which largely recorded a logarithmic reduction of ≥ 5 $\log_{10}$ cfu/100 g beef cut, thus, resulting in the significant elimination of *E. coli* O157:H7 on the beef cuts prior to consumption and absence of risk of illness to consumers; thermal inactivation by the rare done cooking preference which recorded a logarithmic reduction of 1.5 to 0.5 $\log_{10}$ cfu/100 g beef cut, was not significantly efficient in eliminating the risk of illness upon exposure of humans to *E. coli* O157:H7 when they consume *E. coli* O157:H7-contaminated beef cuts processed at the abattoir in this study.

4. CONCLUSION

Data of the present study revealed that consumers of rare done beef cuts processed at the abattoir in this study are likely exposed to significant risk of illness due to exposure to *E. coli* O157:H7 and possibly other pathogenic organisms; and this may result in human uraemic syndrome (HUS) complications and ultimately death. Therefore, there is an urgent need for the relevant regulatory agencies to intensify the monitoring of abattoirs (which are responsible for a significant proportion of *E. coli* O157:H7-contaminated beef prevalence) and the beef distribution chain; and to also enforce appropriate regulations, so as to ensure compliance to standard best practices in the entire beef chain.

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6. CONFLICT OF INTEREST

There is no conflict of interest associated with this work.

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