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Occurrence of verocytotoxigenic *Escherichia coli* (VTEC) in processed chicken from retail chicken markets in FCT, Abuja, Nigeria

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Chicken meat is one of the predominantly consumed foods of animal origin in Nigeria with constant increase in demand normally met by local retail market. Processed chicken at the retail chicken meat markets in Abuja were screened for the presence of verocytotoxigenic *Escherichia coli* (VTEC) strains. A total of 273 faecal samples were collected using moistened sterile swabs and processed for *E. coli* isolation following standard cultural and biochemical procedures. Isolated *E. coli* samples were cultured on sorbitol McConkey (SMAC) and cefiximetellurite sorbitol McConkey (CT-SMAC) agar to assess their ability to ferment sorbitol. Samples were further characterized using commercially procured dry spot polyvalent serocheck and specific seroscreen agglutination test kits. Two (0.73%) of the samples tested positive to O157VTEC, while 5 (1.83%) tested positive to non-O157VTEC. There was no significant association (p>0.05) between VTEC infection and season. The study indicated that processed chicken meat sold at the retail chicken market may serve as a potential vehicle for the spread of VTEC infection and other food borne pathogens. Consumer food safety education is important in control programmes.

Key words: Occurrence, verocytotoxigenic *Escherichia coli* (VTEC), processed chicken, retail market, strain.

INTRODUCTION

Since the first isolation of *E. coli* O157:H7 from an outbreak of human bloody diarrhea in 1982, it has been reported from hundreds of sporadic cases and outbreaks in more than thirty countries of the world (Carter et al., 1987). In addition to *E. coli* O157, many other serogroups of VTEC cause disease and are called non O157 VTEC [Centre for Disease Control (CDC), 2005]. Most attempts to isolate VTEC have been done in dairy and beef cattle because they are most often associated with human disease episode (Dorn, 1995). Cattle are believed to be the principal reservoir for the organisms (Borczyk et al., 1987). Other animal species including birds sometimes pick up VTEC from the environment and may spread it.

VTEC comprise a diverse group that elaborate one or

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both shiga toxins (stx1 and stx2) and can cause diarrhea, haemorrhagic colitis and haemolyticuraemic syndrome (HUS) in human beings (Grant et al., 2011; Gyles, 2007). Meat obtained from farmed and wild game animals contaminated with VTEC has the potential to cause infections in humans, while most of these *E. coli* infections are caused by *E. coli* O157:H7, 20 – 70% of VTEC infections throughout the world are attributed to non-O157 VTEC (Brooks et al., 2005). Of the 81 serotypes identified worldwide, 71% of the isolates recovered from human beings belonged to the "top 6" 'O'-groups (O26, O45, O111, O103, O121 and O145) (Brooks et al., 2005). In 2012, these 6 non – O157 VTEC O-groups in addition to O157 earlier considered were included as adulterants in beef produced in the United States [United States Department of Agriculture-Food Safety Inspection Services (USDA-FSIS), 2012].

Production of safe poultry meat requires systematic and continuous control of the carcasses during all the production steps including slaughterhouses and retail shops (Daci et al., 2016). Microbiological risk from poultry meat is due to contamination during rearing, slaughtering process, and the marketing conditions at the retail shops. Contamination with specific pathogens is common and one of the main concerns of the public Health authorities worldwide (Mead et al., 1994). The importance of this study is linked to the role of poultry meat as one of the main sources of food borne diseases (Fitzgerald et al., 2000).

There has been an increased participation in poultry production in the country in recent time. Live chicks and chicken including their products have been imported and distributed among poultry farmers in all parts of the country. This may lead to introduction of the organism into the environment and then to the food chain hence the need to investigate the organism in processed chicken.

The study is aimed at investigating the occurrence of VTEC O157 and non O157 in processed chicken from retail chicken market in Abuja, FCT, Nigeria in order to evaluate their zoonotic potentials.

MATERIALS AND METHODS

The study was carried out in the Federal Capital Territory (FCT), Abuja located between 8° and 9°25' North of the equator and longitude 6°45' and 7°45' East of the Greenwich meridian (Dawan, 2000). Selected processed chickens through cluster sampling method from retail markets and outlets were studied in 3 out of the 6 Area Councils, selected by simple random sampling. The study design was cross sectional.

Faecal samples from 273 processed chickens were collected using moistened sterile swab immediately after de-feathering and before the chicken was opened in the retail market. Precautionary measures were taken to prevent cross-contamination of samples in transit and at the laboratory. An enriched medium of buffered peptone water (BPW) supplemented with 8 mg/l vancomycin, 10 mg/l cef sulodin and 0.05 mg/l cefixime (BPW-VCC) was prepared to suppress the growth of Gram positive organisms, *Aeromonas* and *Proteus* spp. (Pritchard, 2000). Approximately, 0.5 g of the faecal sample was inoculated into 5 ml of the prepared BPW-VCC and incubated at 37°C for 6-8 h (Pritchard, 2000). Samples were first cultured on McConkey agar (MCA) then those lactose fermenting organisms (pinkish colonieson MCA) were streaked on eosine methylene blue (EMB) agar and incubated at 37°C for 24 h. The cultured isolates exhibited the typical greenish sheen colouration characteristic of *E. coli* on EMB agar. Biochemical tests were carried out to confirm the isolates as typical *E. coli* and they exhibited similar IMViC pattern of + + - - and showed negative to both urease and hydrogen sulphide production.

E. coli isolates ex-EMB were sub-cultured into plates of Sorbitol McConkey agar (SMAC) and Cefixime-Tellurite Sorbitol McConkey agar (CT-SMAC) and incubated at 37°C for 18 to 24 h (March and Rotnam, 1986). Non sorbitol fermenting (NSF) isolates that appear as colourless or neutral gray with smokey center and 1-2 mm in diameter on the two plates were presumptive of *Escherichia coli* O157, while sorbitol fermenting (SF) isolates that appear pinkish in colour were presumptive of *E. coli* non-O157 (Zadik et al., 1993).

Both the NSF and the SF stored in nutrient agar slants were further characterized using latex agglutination test kits called dry spot *E. coli* seroscreen and polyvalent serocheck which were commercially procured from Oxoid ™ England. The kit contained seroscreen for O157 and serocheck for O26, O103, O111, O91 and O145.

Table 1. Prevalence of O157 and non-O157 VTEC in processed chicken.

Serogroup	No. tested	No. positive	Positive (%)
O157	273	2	0.73
Non O157	273	5	1.83

Table 2. Distribution of the 5 non-O157 detected in processed chicken.

Serotype	No. positive	Positive (%)
O26	1	0.37
O103	-	-
O111	1	0.37
O145	1	0.37
O91	1	0.37
Untyped	1	0.37

RESULTS

The prevalence of VTEC O157 in processed chicken was 0.73% (2) while the prevalence for non-O157 VTEC was 1.83% (5) (Table 1). The distribution of the 5 non-O157 detected were also determined. VTEC O26, O111, O145 and O91 and the untyped were 1 each, while VTEC O103 was 0 (Table 2).

The seasonal distribution of VTEC O157 and non-O157 VTEC was studied. Of the 273 samples collected, 145 were during the dry season, while 128 were during the wet season. The number of isolates for VTEC O157 was

Table 3. Seasonal distribution of VTEC O157 and non-O157 in processed chicken.

Season	No collected	No positive for0157	No positive for Non-0157
Dry	145	2	4
Wet	128	-	1
Total	273	2	5

p>0.05.

2 in the dry season and none in the wet season, while for non-O157 VTEC, 4 was positive in the dry season and 1 positive in the wet season. There was no significant difference between infection and season (p>0.05) (Table 3).

DISCUSSION

This work detected the presence of O157, O26, O111, O145 and O91 in processed chicken. The results carried out elsewhere on processed chicken agreed with the result of this study. Doyle and Schoeni (1997) assayed 263 fresh, uncooked chicken samples and isolated the organisms from 1.5% (4) of the samples. In Thailand, Suthienkul et al. (1990) found 1% (9) out of 107 chicken carcasses contaminated with *E. coli* that did not produce shiga like toxin. In another study performed in UK, 1 to 2% of chicken lamb and pork meat samples were found to be contaminated by *E. coli* O157:H7 (Adams and Moss, 1995). Vemozy-Rozand et al. (1997) analyzed 250 samples of meat and meat products in France and found 4 chicken meat samples (1.6%) to be positive to non verotoxin producing *E. coli*. El-Jakere et al. (2012) collected 12 VTEC isolates from chicken to study their diversity and O2, O6, O8, O26, O27, O78, O86, O111, O128, O136 and O157 were typed from the chicken. Although, genetically diverse, avian *E .coli* isolates share several genes or traits associated with virulence in chicken (Ngeleka, 1996). *E. coli* isolates were recovered by Cook et al. (2012) from the skin-off chicken breasts, 33% (33) of 99, than from the skin-on chicken breasts, 41% (77) of 187 (p=0.204) and VTEC was detected on a single skin-off chicken breast. Verocytotoxigenic *E. coli* are a specialized group of *E. coli* that can cause severe colonic disease and renal failure.

Although, Heuvalink et al. (1998) could not find any VTEC O157 in chicken faeces, 1.3% of 459 pooled samples from turkeys were positive and one isolate contained genes for type 2 verotoxin, attaching-and-effacing capability and the relevant haemolysin. Akkaya et al. (2006) isolated *E. coli* O157:H7 from 2 (1.0%) of the 190 samples of poultry meat examined with all the strains producing both VT1 and VT2 verotoxin indicating that poultry meat can also be a source of VTEC infection to humans.

In Egypt, Abdul-Raouf et al. (1996) examined 50 boneless chicken meat samples and were able to detect *E. coli* O157:H7 in 2 samples. Zhao et al. (2001) reported 38.7% prevalence of *E. coli* in chicken meat in a study in Washington D.C, USA. Chapman et al. (1997) recorded no positive result from 1000 chicken tested for O157 VTEC.

The isolation of VTEC strains from processed chicken may be due to cross-contamination during slaughter, processing or transportation. The contamination might occur in the chicken slaughter house at various stages such as during evisceration, scalding, plucking and or cutting processes. The presence of *E. coli* O157:H7 on chicken carcasses suggests that chickens may be natural carriers of the microorganisms (Akkaya et al., 2006).

The result of this research showed low prevalence in processed chicken suggesting that the organism is rare in these products in the study area. Published research findings show that VTEC O157 is rare in poultry whether in live birds or on processed products and when it was found, tests for the necessary virulence factors was not carried out (Schmidt et al., 1990). Read et al. (1990) recorded zero isolation in chicken in South Western Ontario indicating that the organism is actually rare in chicken and chicken products. VTEC O157:H7 has been documented to colonize the gastrointestinal tract of chickens under experimental conditions (Barry et al., 1985; Stavric et al., (1993).

There was no significant difference between season and infection with VTEC in processed chicken. No available records showed that research has been done on seasonal distribution of VTEC in processed chicken.

Conclusion

The isolation of VTEC organisms in processed chicken meat as determined in this study is indicative that processed chicken meat may serve as a source of infection to humans. Control strategies aimed at protecting public health should be adopted to eliminate contamination of any sort with the pathogen. Consumer food safety education is important as prevention and control measures.

Conflict of Interests

The authors have not declared any conflict of interests.

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