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DETECTION OF *ESCHERICHIA COLI O157:H7* FROM IMPORTED AND LOCALLY PRODUCED BURGER IN BAGHDAD

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ABSTRACT
During the period April till June (2016), fifty burger samples (twenty five locally produced and twenty five imported) were collected randomly at weekly intervals from different markets and butcher shops in Baghdad province in which they processed and analyzed by different food microbiological procedures to identify the *E. coli* and *E. coli O157:H7*. The Results Showed that the prevalence rate of *E. coli* contamination in locally produced and imported produced burger samples was 100% with the mean value of *E. coli* contamination in all the positive locally produced burger samples was 4.4441 cfu\g^{-1} whereas the mean value of *E. coli* contamination in all the positive imported burger samples was 4.4089 cfu\g^{-1} in which the *E. coli* O157:H7 was detected in 6 (24%) isolates from locally produced burger samples with the mean value 2.2958 and 7 (28%) isolates from imported burger samples with the mean value 3.0903 which indicate there is a significant difference for mean $\log_{10}$ count of *E. coli O157:H7* at level (P≤0.05) between the locally produced and imported burger samples.

KEYWORDS: Detection, *E. coli O157:H7*, burger.

INTRODUCTION

Red Meat is one of the most important sources of verocytotoxin-producing *E. coli* (VTEC) infection and some of those implicated were burger meat, bruised lettuce, unchlorinated water and mayonnaise (McDowell and Sheridan, 2001). Cattle are considered to be one of the principal sources of *E.coli O157:H7*, that is expand through faecal contamination of food. Other ruminants were also an important reservoir as shedding occurs intermittently. Thus, human beings may be infected at any time and all measures should be taken to reduce the risk to public health. Verocytotoxin-producing *E.coli* (VTEC) O157:H7 is a globally prominent illness frequently associated with hemolytic uremic syndrome (HUS) (Hajian et *al*., 2011). HUS occurs in all age groups but is more frequent in infants and young children (Gianviti *et al*., 1994). It is characterized by the sudden onset of haemolytic anemia with fragmentation of red blood cells, thrombocytopenia and acute renal failure after acute gastroenteritis (Temelli *et al*., 2012). *E. coli* O157:*H7* belongs to the categorize of enterohaemorrhagic *E. coli* (EHEC) that expresses its pathogenicity with the yielding of one or more Shiga-like toxins, also called verocytotoxins (VT1, VT2), and with the shade of several accessory virulence genes acquired by horizontal gene transfer. VTEC that generate disease in humans belong to a restricted number of serogroups: *O157:H7* is the most dominant group followed by a numerous more serogroups, such as O26, O111, O103, O145 and O121 (Karch *et al*., 2005). In general, the infected patients are usually vulnerable members of the community such as the very young or very old and immunocompromised individuals. The infectious dose of *E. coli O157:H7* which may be as low as 10 organisms (Gillespie *et al.*, 2003). In an outbreak survey recorded by Willshaw *et al*. (1994) contamination levels in an implicated product were reportedly as low as 2 cells per 25 g. and the incubation period to the onset of diarrhea can vary from 1 to 8 days. Surveillance data demonstrate that the VTEC *O157*:*H7* infection is evolving .Since the early description of this illness in the USA in 1982 (CDC, 2014), the geographic range of the organism is growing and the pattern of disease transmission is changeable.

The satisfactory microbiological quality for *E. coli O157* was <20 *CFU \g* with the acceptable range being 20 to <100 *CFU \ g* (De Giusti *et al*., 2007: Gilbert *et al*., 2000). However, it is the opinion of the Advisory Committee for Food and Dairy Products (ACFDP) of the UK that ready to-eat foods should be free from *E. coli O157*:*H7* and other VTEC organisms (Gilbert *et al*., 2000). The Centers for Disease Control in the USA has estimated that E. *coli O157:H7* generates 73 000 illnesses and 61 deaths per year in the USA (CDC, 2003). In the UK, the Health Protection Agency (formerly, the Public Health Laboratory Service) has indicated that in 2001, there were 85 468 food poisoning notifications, which represent a six fold increase from 1982. Of these, 768 were because of *E. coli O157:H7* (Health Protection Agency., 2003). The aim of this survey was to determine the existence and prevalence of *E.Coli* and its serotype *O157:H7* from locally produced and imported burger samples.

MATERIALS & METHODS

Collection and processing of samples

Fifty burger samples (25 locally produced and 25 imported) were collected randomly weekly during June till April (2016) from different markets and butcher shops in Baghdad province in which they prepared and processed by different food microbiological procedures. Samples were collected aseptically in sterile plastic bags and containers in which conserved cooled in an ice box during the transfer to the research laboratory as soon as possible. A 25-g portion of each burger sample was blend in a stomacher with 225 ml of peptone water for 2 min then incubated at 37°C for 24 h. The culture was diluted in peptone water (1%), inoculated onto MacConkey agar, and incubated overnight at 37°C. Twenty to fifty colonies per sample were chosen and screened for lactose fermentation (blue-black colony with a greenish metallic sheen) on eosin methylene blue agar and for sorbitol non fermentation (colorless, smooth, circular colonies) on sorbitol MacConkey agar. For further identification procedures All sorbitol negative colonies were tested for the *O157:H7* antigen by latex agglutination (Oxoid) (BAM., **2015;** Dontorou *et al*., 2003). All sorbitol non-fermenting, colonies were examined by latex agglutination these beads are coated with antibodies which bind to any O157 or H7 antigens on the test organisms, forming a visible antigen antibody precipitate (De Boer and Heuvelink, 2000). Colonies giving a precipitation reaction were confirmed as *E. coli O157:H7* positive.

Data were statistically analyzed by t-test in accordance with SPSS (SPSS, 2014).

RESULTS & DISCUSSION

Human infections by food borne *E. coli O157:H7* have principally been recognized to be originated from animal source foods (Jo MY *et al*., 2004). Domestic ruminants, chiefly cattle, sheep, and goats, have been settled as dominant natural reservoirs for STEC and play a vital role in the epidemiology of human infections (Griffin *et al*., 1991). In the present survey, the imported and locally produced burger samples were analyzed for *E. coli* and *E. coli O157:H7* contamination. The prevalence rate of *E. coli* contamination in locally produced and imported produced burger samples was 100% with the mean value of *E. coli* contamination in all locally produced burger samples was 4.4441 cfu\g^{-1} whereas the mean value of *E. coli* contamination in all imported burger samples was 4.4089 cfu\g^{-1} in which the *E. coli O157:H7* was detected in 6 (24%) isolates from locally produced burger samples and 7 (28%) isolates from imported burger samples. This indicates that the Freezing didn't prevent the survival of *E. coli O157:H7* in imported burger samples (Ansay *et al.,* 1999).

TABLE 1: The mean log^{10} count of *E.coli* from burger in Baghdad

Type of sample	No. of samples	Isolation %	Mean log^{10} count cfu\g^{-1}
Locally produced burger	25	25 (100%)	4.4441 a
Imported burger	25	25 (100%)	4.4089 a

(a): indicate there is no significant difference for mean $\log_{10}$ count vertically at level (P≤0.05).

TABLE 2: The mean log^{10} count of *E.Coli O157:H7* from burger in Baghdad

Type of sample	No. of samples	Isolation %	Mean log^{10} count cfu\g^{-1}
Locally produced burger	25	6 (24%)	2.2958a
Imported burger	25	7 (28%)	3.0903b

(a, b):indicate there is significant difference for mean log^{10} count vertically at level (P≤0.05).

Unrestricted hygienic monitoring systems and food policies like absence of bio-safety and hazard analysis critical control points during manufacturing and handling of healthy meat, lack of risk assessments during importation of meat and meat products, all the above mentioned and others result in contamination of meat and meat products in Baghdad markets with different invaders. The initially principal occurrences were in the USA in 1982 which involved burgers from fast food chains in Oregon and Michigan (CDC, 2014). In 1988, 30 students at a secondary school in Minnesota take sick back of consuming partially-cooked beef patties, and in late 1992 and soon 1993, four deaths in four states (Washington, Idaho, California and Nevada) were documented. These were once again attributed to burgers in 1996. Also in that year, an outbreak in Lanark shire, Scotland claimed 20 lives at an old people's home, this was due to the victims consuming beef contaminated with the organism (Bell and Kyriakides, 1998). In August 1997, Hudson Foods recollected 25 million pounds of ground beef after an *E. coli* outbreak was traced to its plant in Nebraska (Snyder, 1998). In 1999, a dangerous outbreak affected New York state where 1000 people were affected with two deaths registered (Charatan, 1999). outbreaks have been reported in Croatia (Matica *et al*.,1999), Turkey (Kucuker *et al*.,1999), Italy (Payne *et al*., 2009: Caprioli *et al*.,1997) Scotland (SCIEH,1997), Japan (Mermin and Griffin.,1999),one other in Scotland (Currie *et al*., 2007) and one other in Japan (McCartney *et al*., 2010) which the Outbreaks have been associated mainly with the consumption of ground beef (Swerdlow *et al*., 1992). In this study the isolation % of *E. coli O157:H7* is higher In comparison to diverse countries, conducted in the UK (1.1%) (Chapman *et al*., 2001), Swiss study (2.3%) (Fantelli and Stephan, 2001) and study in Argentina (3.8%) (Chinen *et al*., 2001). Such high standards represent point source contamination at the primary meat manufacturing and processing, and/or subsequent temperature abuse of the meat and meat products within the production/retail chain. Observed variation in prevalence in

connection with other studies probably attributed to controversy in sampling and isolation procedures, fluctuation in sampled populations, different geographical origins of cattle, study design, season, abattoir conditions and treatment with antimicrobial substances during the process (CDC,1996 : Chapman *et al*., 2001:Varela *et al*., 2007).The higher contamination levels of the above mentioned locally produced and imported burger samples could be hazardous considering the very low virulent dose of this pathogen, its detection in such concentrations in retail beef products poses significant public health risks. While it is to be hoped that most retail beef products will be adequately cooked before consumption, leading to the destruction of the pathogen, the existence of contaminated meat at retail and consumer levels places, consumers at risk. *E. coli O157:H7* may persist in undercooked beef burgers (Bell *et al*., 1994) or be transferred from such raw meats to cooked products, or products that do not receive heat treatments prior to domestic consumption, e.g. salad items, by cross contamination of hands, utensils or surfaces (Little and de Louvois, 1998) during domestic food preparation. Our study calls for developing preventive approach to control *E. coli O157:H7* contamination in meat production chain by imposing strict hygienic meat processing practices. This can be done by ensuring Good healthful Practice, Good Manufacturing Practice and if possible Hazard Analysis of Critical Control Points (HACCP) at whole stage of the beef deliver chain, from the farm, through the abattoir, to the butcher houses, and the above mentioned involved with the handling and processing Further investigations employing molecular typing should be conducted.

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