Detection of *Escherichia Coli* O157:H7 in Imported Meat Products from Saudi Arabian Ports in 2017

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Short report

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Abstract

Escherichia coli O157:H7 is a foodborne pathogen, which causes various health conditions in humans, including fatigue, nausea, and bloody diarrhoea, and in some cases, even death. In 2017, 15.71% of the total imported food products in Saudi Arabia were meat-based. India and Brazil are two of the top five countries from where Saudi Arabia imports meat. According to the Saudi Food and Drug Authority, in 2017, at least 562, 280, and 50 samples of imported beef, chicken, and sheep meat, respectively, were tested for the presence of *E. coli* O157:H7. Amongst these, *E. coli* O157:H7 was detected in respectively 6.5% and 2.2% of the tested beef meat samples imported from India and Brazil as well as in respectively 6.96% and 3.57% of the tested chicken samples imported from Brazil and Ukraine. Moreover, the pathogen was detected in 2.1% of the tested sheep meat samples imported from India. The present report provides evidence that imported meat can serve as the carrier of *E. coli* O157:H7, leading to epidemics, within the Kingdom of Saudi Arabia.

Introduction

Escherichia coli is gram-negative, facultative, anaerobic bacterium, considered a commensal organism within the human body (Santos et al. 2018). However, the *E. coli* strain O157:H7 is a human pathogen, posing threat to the human life by causing several diseases, such as haemolytic–uraemic syndrome, which may be fatal in some cases (Luna et al. 2018). The primary reservoir of *E. coli* O157:H7 is meat; however, it has also been isolated from vegetables and fruit (Haramain and Yagoub 2021; Riley 2020). *E. coli* O157:H7 was first detected in 1982. Since its discovery, the pathogen has been responsible for 73,000 illnesses every year in the United State alone, causing as many as 350 outbreaks within only two decades (1982–2002) (Rangel et al. 2005). Furthermore, illnesses caused by *E. coli* O157:H7 have been reported in over 30 countries across 6 continents (Rahal et al. 2012).

E. coli strains that produce Shiga toxins (Stx1 and Stx2) are called Shigatoxigenic E. c*oli* (STEC) (Mesele and Abunna 2019) [e.g. *E. coli* O157:NM (Rangel et al. 2005)], and those that produce Shiga-like toxins (verotoxin) are called verotoxigenic E. c*oli* (VTEC) (Shinde et al. 2020). STEC isolates are further divided into two groups, namely O157 and non-O157 (Santos et al. 2018). O157 isolates present the H7 serogroup, whereas and non-O157 isolates present the O26, O45, O103, O111, O121, and O145 serogroups (Santos et al. 2018). Collectively, *E. coli* O157:H7 strains that cause illnesses in humans are called enterohaemorrhagic E. c*oli* (EHEC).

To date, no *E. coli* O157:H7 outbreak has been reported in Saudi Arabia, and the prevalence of this pathogen remains in the country unknown. However, it has been isolated from several local cattle farms (Bosilevac et al. 2015). Reporting an outbreak in Saudi Arabia has many challenges due to an inefficient data collection system (Al-Mazrous 2004). Therefore, since 2003, the Saudi Food and Drug Authority (SFDA) has acquired the control of all food safety regulations, which has also helped avoid any overlap with other Saudi Arabian authorities (Alrobaish et al. 2021). Approximately 80% of the available food in

Saudi Arabian markets is imported and 15.71% of these imported food products are meat-based (Alrobaish et al. 2021).

To this end, the present study evaluated the possibility of detecting *E. coli* O157:H7 in meat products imported in Saudi Arabia using the monitoring system of SFDA to provide foundational data for creating a database of this fatal bacterium.

Methods

Sample Collection. All data were obtained from the database of the monitoring system of SFDA. Typically, upon arrival of shipments contain imported-meats-to-consume at the Saudi Customs Ports, SFDA inspectors collect samples from those shipments and send to SFDA labs for approval. Thereafter, depending on the type and request, the samples are referred for *E. coli* counting or *E. coli* O157:H7 detection. Data in this study was gathered from testing the raw whole parts only (not mince nor cooked meats).

E. coli O157:H7 Detection. Twenty-five gram samples were selected for enrichment and homogenised with 225 mL of modified Tryptone Soya Broth (mTSB) supplemented with novobiocin. The samples were incubated at 41.5°C for 12–18 h. For DNA extraction, the samples were prepared using the PrepMan™ Ultra Sample Preparation Reagent Kit (LOT number: 1809191) according to the manufacturer's protocol. Next, real-time polymerase chain reaction (PCR) was performed to amplify the *E. coli* O157:H7-specific target DNA sequences using the MicroSEQ™ *E. coli* O157:H7 Detection Kit (LOT number: 1804034) according to the manufacturer's protocol.

Results

E. coli O157:H7 strains were detected at varying frequencies in imported beef, sheep meat, and chicken products in Saudi Arabia in 2017. Table 1 summarises *E. coli* O157:H7 prevalence determined in the present study. *E. coli* O157:H7 was the most prevalent in chicken samples (6.07%), followed by beef (5.9%) and sheep meat (2%) samples.

Table 1
Prevalence of *Escherichia coli* O157:H7 (as inspected by the Saudi Food and Drug Authority) in imported beef, chicken, and sheep meat samples in Saudi Arabia in 2017

Product	Samples	Total	Contaminated	Prevalence (%)
Beef	All samples	562	33	5.90
	Australia	8	0	0.00
	Brazil	91	2	2.20
	Jordan	6	0	0.00
	New Zealand	2	0	0.00
	UAE	15	1	6.70
	Philippines	4	0	0.00
	Spain	1	0	0.00
	India	428	29	6.80
Chicken	All samples	280	17	6.07
	Brazil	230	16	6.96
	Jordan	20	0	0.00
	India	1	0	0.00
	Tunisia	1	0	0.00
	Ukraine	28	1	3.57
Sheep meat	All samples	50	1	2.00
	Australia	1	0	0.00
	New Zealand	2	0	0.00
	India	47	1	2.13

Regarding chicken, the highest number of samples contaminated with *E. coli* O157:H7 (6.96%) was imported from Brazil, followed by Ukraine (3.57%). Regarding beef, the highest number of contaminated samples were imported from India (6.8%), followed by Brazil (2.2%). Finally, only sheep meat samples imported from India were contaminated with *E. coli* O157:H7 (2.1%) (Table 1).

In the present study, the highest frequency of *E. coli* O157:H7 contamination was reported in products imported from Indian companies (30 of 476 samples: 8 from company A, 5 from company B, 4 from company C, 3 from company D, 2 from company E, 2 from company F, and 6 from other companies) (Table 2). More beef samples than sheep meat samples imported from India were screened given the high demand of the former in the Saudi Arabian markets in 2017 (Alrobaish et al. 2021); therefore, the

prevalence of *E. coli* O157:H7 in the beef samples was greater than that in the sheep meat samples (6.8 % and 2.1%, respectively). Products imported from Brazilian companies were also frequently contaminated (18 of 321 samples: 4 from company G, 2 from company H, 2 from company I, 2 from company K, and 8 from other companies) (Table 2). However, the prevalence of *E. coli* O157:H7 in the chicken samples was greater than that in the beef samples (6.96 % and 2.2%, respectively). To ensure anonymity, companies from where the contaminated products were imported are labelled with letters A to K.

Table 2
Sources of contaminated samples classified by countries and companies

India	Company codes	Number
	A	8
	B	5
	C	4
	D	3
	E	2
	F	2
	Other	6
	Total	30
Brazil	G	4
	H	2
	I	2
	K	2
	Other	8
	Total	18

Discussion

Contaminated raw meat is the source of 90% of foodborne infections (Hadi and Jabbar 2020). Thirty-one pathogens, including *E. coli* O157:H7, were responsible for 10 million annual episodes of foodborne illnesses in the United States (Riley 2020). In the present study, samples of imported raw meat were obtained from imported meats in the ports of Saudi Arabia, and the prevalence of *E. coli* O157:H7 in these samples was confirmed (Table 1). Meat products imported from India and Brazil were the most frequently contaminated (Table 1).

The prevalence of *E. coli* O157:H7 was the highest in raw meat products imported from India, posing a threat to public health in the Kingdom of Saudi Arabia (Table 2). According to Shinde et al. (2020), *E. coli* O157:H7 was frequently isolated from healthy Indian cattle on both organised and non-organised farms in and around the Pune District in India during 2015. This can be explained by the fact that new generations of cattle may carry the pathogen but may not present any symptoms, thus appearing as heathy livestock; however, the consumption of meat from such asymptomatic carriers of *E. coli* O157:H7 may affect humans, representing a severe public health concern. Furthermore, subsequent studies in the same region revealed the presence of *E. coli* O157:H7 isolates resistant to a number of common antibiotics used for livestock animals against this pathogen, including cefotaxime, streptomycin, penicillin G, kanamycin, ampicillin, tetracycline, gentamycin, and piperacillin. These findings, in addition to our results, emphasise the need for the further of assessment of imported meat, specifically from India, to ensure public health safety. In another recent study in China, clinical isolates of *E. coli* exhibited high resistance to conventional antibiotics for livestock, including sulfamethoxazole, trimethoprim/sulfamethoxazole, tetracycline, nalidixic acid, and ampicillin (Yassin et al. 2017).

Amongst samples of meat imported from Brazil, *E. coli* O157:H7 was detected at different frequencies in products from several companies (Table 2). The prevalence of *E. coli* O157:H7 in samples from only specific companies (G, H, I, J, K, and others), but not others, indicates internal contamination through air during rearing at the livestock farms (Chmielowiec-Korzeniowska et al. 2021), slaughter (Irshaid et al. 2018), or processing (Rangel et al. 2005) (Figure 1). According to Santos et al. (2018), the prevalence of STEC in Brazilian food products was approximately 9.5%, which was primarily attributed to the development of multi-resistance to antibiotics in these strains (Santos et al. 2018). Notably, Brazil is the second largest exporter and the third major producer of beef worldwide (Santos et al. 2018).

The detection of *E. coli* O157:H7 in samples of meat imported from one company each in Ukraine and UAE also indicates unhygienic handling that led to contamination (Table 2), highlighting the need for the revision of processing and packaging steps in these regions (Rangel et al. 2005).

Of note, the present report only includes results from products that have been undergone *E. coli* O157:H7 testing from the port of Saudi Arabia. Many shipments may have been excluded from the examination for approval and owners may have only been asked to produce a list of essential documents (Alrobaish et al. 2021). In addition, to import food products into Saudi Arabia, the SFDA mandates a registration certificate authorised by the Saudi health ministry, an industry certificate authorised by the commerce ministry, and a quality certificate (e.g. International Organization for Standardization 9001 or 22000, Good Manufacturing Practice, and Hazard Analysis Critical Control Point) (Alrobaish et al. 2021). Therefore, to ensure public safety, the SFDA has announced a list of countries from where the import of food into Saudi Arabia is prohibited (available at bit.ly/2TOoNMm) (Alrobaish et al. 2021).

Summery

The presence of *E. coli* O157:H7 in samples of imported raw meat highlights the need for more regular surveillance at the borders of Saudi Arabia before the products are made available on the market for consumption by the public. Our results underscore the necessity of more stringent control protocols for the approval of imported food products, particularly from India and Brazil, which are the major suppliers of meat to Saudi Arabia. Moreover, the detected *E. coli* O157:H7 isolates should be tested against antibiotics that are commonly used to treat livestock.

Abbreviations

STEC shigatoxigenic *E. coli*

VTEC Verotoxigenic *E. coli*

EHEC enterohaemorrhagic *E. coli*

SFDA Saudi Food and Drug Authority

mTSB modified Tryptone Soya Broth

Declarations

- Ethical Approval and Consent to participate: Not applicable.

- Consent for publication: Not applicable.

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Disclaimer

The views expressed in this paper are those of the author(s) and not do not necessarily reflect those of the SFDA or its stakeholders. Guaranteeing the accuracy and the validity of the data is a sole responsibility of the research team.

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Figures

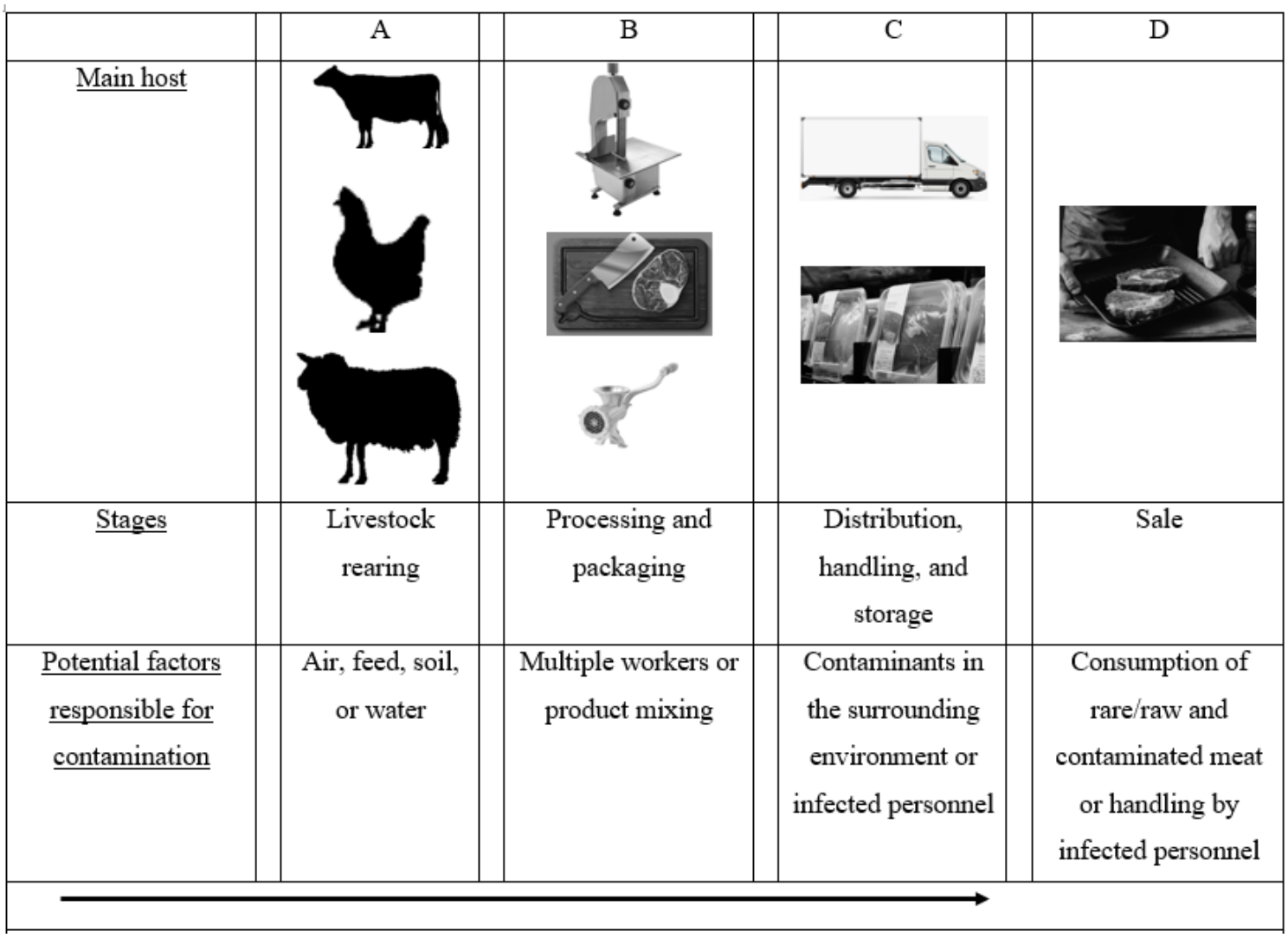

Figure 1

Schematic showing the meat production steps from livestock rearing on farms until sale (A, B, C, and D). Potential factors responsible for the contamination of meat are illustrated. The arrow indicates the direction of steps from the start to end.