Escherichia coli O157:H7 Outbreak Associated with the Consumption of Beef and Veal Tartares in the Province of Quebec, Canada, in 2013

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Abstract

Objectives: An Escherichia coli O157:H7 outbreak occurred in 2013 that was associated with the consumption of beef and veal tartares in the province of Quebec. This report describes the results of the ensuing investigation. *Materials and Methods:* As the outbreak was identified, all individuals in the province of Quebec affected with the same strain of *E. coli* O157:H7 as defined by pulsed-field gel electrophoresis were interviewed using a standardized questionnaire. Cases reported from other provinces in Canada were interviewed by their public health authorities and the results were reported to the Quebec public health authorities. Microbiological and environmental investigations were conducted by the Sous-ministériat à la santé animale et à l'inspection des aliments du Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec, by the Ville de Montréal's Food Inspection Branch, and by the Canadian Food Inspection Agency at the restaurants, suppliers, and slaughterhouses identified.

Results: In total, seven individuals in three different Canadian provinces became ill following infection with the same outbreak strain of *E. coli* O157:H7. Two cases were hospitalized and one had severe hemolytic uremic syndrome. No deaths were reported. Two restaurant locations serving different tartare meals including, beef, veal, salmon, tuna, and duck were identified as potential sources of the outbreak. No deficiencies at the restaurant locations were observed during inspections by food inspectors.

Conclusions: The risk of consuming tartare can be lowered when basic hygienic rules are followed, temperature is strictly controlled, and fresh meat is used. However, even if handling, chopping, and temperature control during storage of the meat are considered adequate, tartare is a raw product and the risk of contamination is present. Consumers should be advised that consuming this product can lead to serious illness.

Introduction

S HIGA TOXIN–PRODUCING *Escherichia coli* (STEC) is a bacterial pathogen that can cause a range of symptoms from uncomplicated diarrhea to bloody diarrhea (hemorrhagic colitis) and that may progress to the life-threatening condition known as hemolytic uremic syndrome (HUS). One third of STEC cases require hospitalization (Scallan *et al.*, 2011; Page *et al.*, 2013) and 5–15% of cases may progress to HUS (Karch *et al.*, 2005).

A large number of human illness outbreaks have been traced worldwide to consumption of undercooked ground

beef and other beef products contaminated with STEC. Other important vehicles for STEC infection include raw milk products and uncooked vegetables, but beef products remain the most common source of STEC (Rangel *et al.*, 2005; Hussein, 2007; Ravel *et al.*, 2009; Kozak *et al.*, 2013). Person-to-person transmission may also occur.

Reports of the involvement of steak tartare in STEC outbreaks have been documented (Doorduyn *et al.*, 2006; Greenland *et al.*, 2009). Other pathogens such as *Salmonella* Typhimurium and *Salmonella* Newport were associated with the consumption of beef tartare (CDC, 2002b; Ethelberg *et al.*, 2007; Kivi *et al.*, 2007). Steak tartare is a meat dish

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made from finely chopped or minced raw beef prepared most commonly from "filet mignon." The outer surface of the filet mignon is generally removed or trimmed and the remaining meat is chopped or minced. By removing the outer surface of the meat, the risk of exposure should be reduced. Crosscontamination remains possible.

Steak tartare is often served with onions, capers, and seasonings; however, raw eggs and mustards have also been documented ingredients in this type of dish. According to a report of the World Health Organization (WHO), the risk of bacterial infection is considered low when basic hygienic rules are followed and fresh meat is used (WHO, 2009). This report describes the results of an investigation of a STEC outbreak associated with beef tartare.

Background

On December 19, 2013, the Bureau de Surveillance et de Vigie (BSV) at the Ministère de la Santé et des Services sociaux (MSSS) in the Province of Quebec, Canada, which is responsible for the management of provincial outbreak investigations, was notified of four *E. coli* O157:H7 (STEC) cases from three different regions in the province. All four cases had consumed various forms of minced raw (tartare) products prepared from beef, salmon, veal, duck, or tuna, between December 3 and 6 at two restaurants located in the same city. Symptom onset dates were between December 7 and December 12, 2013. The age of patients varied from 17 to 43 years old. Two cases were hospitalized and one of them developed HUS. This prompted an epidemiological and microbiological investigation.

Materials and Methods

Epidemiological investigation

Case definition. A case was defined as a Quebec resident or someone who had visited the Province of Quebec during the incubation period (which was maximum 10 days) and experienced diarrhea and/or vomiting after the date of November 15, 2013, and:

- tested positive when cultured for *E. coli* O157:H7 with the same pulsed-field gel electrophoresis (PFGE) pro-file, or
- had a HUS without positive stool culture and an epidemiological link with a confirmed *E. coli* O157:H7 case involving the outbreak strain.

The detected outbreak PFGE profile was designated as EC PFGE 1005 according to the Quebec nomenclature and EC-XAI.0248/ECBNI.0215 according to the PulseNet Canada nomenclature.

STEC is a reportable disease in Quebec and Canada. Each case is reported by regional laboratories to provincial public health authorities and then to the Public Health Agency of Canada. During the last 5 years, an average of 120 cases of foodborne STEC were reported annually in Quebec, generally involving different PFGE profiles. An outbreak investigation is usually started when three or more STEC cases involving the same PFGE profile are reported to public health authorities during the same time frame. The public health network (including public health authorities and laboratories) is then alerted and investigation procedures are initiated. To

facilitate case-finding during this outbreak, a public health alert was posted on a secure Internet-based platform that reaches across Canada. All communications contained information about the investigation, the symptoms of illness, general risk factors, and the necessity to notify public health authorities if new cases were identified.

After the identification of the initial cluster of four cases. the three subsequent E. coli O157:H7 cases were interviewed before the PFGE profiles became available. Telephone interviews were performed by nurses or physicians from regional public health authorities. A standardized questionnaire was used to assess the demographic data, date and time of onset, duration of illness and complications, as well as to identify food and drink items consumed during the incubation period and any other exposures (restaurants, contact with animals, etc.) All the cases were classified after receiving the results of PFGE profile as either meeting or not meeting the case definition. Cases reported in other provinces were interviewed by their local public health units and the information was reported to Quebec's public health authority. Cases went to the restaurant with companions or family members, some of whom consumed tartare meals as well. No meal companions or family members were sick. They were interviewed about their meal consumption and can be seen as a convenience sample. However, comparisons must be looked upon with caution, since the number of cases was small and in some cases, some food was exchanged between the cases and the companions. For this reason, comparisons will be presented, but no statistical test will be done. Descriptive analysis was conducted using the software Microsoft Excel, version 2010 (Microsoft Corp., Redmond, WA).

Laboratoire de Santé Publique du Quebec (LSPQ)

The identity of bacterial isolates from patients as *E. coli* O157:H7 was determined as follows: Isolates were confirmed as *E. coli* by biochemical testing with the API 20 E, (bio-Mérieux), sorbitol negative on sorbitol-MacConkey agar plate (March *et al.*, 1986) and in Andrade sorbitol 0.5% broth (Farmer *et al.*, 1980; Clark *et al.*, 1984) and as O157:H7 by slide agglutination assay positive with the O157 and H7 using the latex agglutination Wellcolex kit as per the manufacturer's instructions (Remel Europe Ltd., UK). Molecular characterization was performed by PFGE (CDC, 2002a) using the enzymes *XbaI* and *BlnI*. The interpretation of the profiles was done according to Tenover's criteria (Tenover *et al.*, 1995). PFGE profiles were designated using BioNumerics and then submitted for comparison to the PulseNet Canada database.

Confirmed *E. coli* O157:H7 strains were sent to the National Microbiology Laboratory in Manitoba, which conducted a multiple-locus variable-number tandem repeat analysis (MLVA) (Hyytia-Trees *et al.*, 2006, 2010).

Food safety inspections

Food safety inspections were conducted by Ministère de l'Agriculture, des Pêcheries et de l'Alimentation du Québec (MAPAQ) in collaboration with Ville de Montréal, Division de l'inspection des aliments's inspectors and Canadian Food Inspection Agency (CFIA) inspectors.

Two restaurant locations were identified as a potential source of the outbreak. Ville de Montréal's food inspectors started their activities on December 19, 2013, and continued intermittently between December 21, 2013 and January 31, 2014. They visited the restaurant and did a complete review of food preparation, handling, chopping, and cooling practices.

Food samples were collected using official MAPAQ procedures (MAPAQa, 2013), and were sent to either the Direction du Laboratoire d'Expertise d'Analyses Alimentaires, which is MAPAQ's food analysis laboratory or the City of Montreal's food inspection service laboratory, depending on the specimen's origin and the availability of the respective laboratories. Standardized procedures were used to detect and identify STEC in both laboratories. MAPAQ's laboratory used their published methods (MAPAQ, 2007; MAPAQb, 2013) and the City of Montreal's food laboratory used MFLP-82 (Health Canada, 2014) to detect *E coli* O157:H7 followed by MFLP-80 for identification (Health Canada, 2008).

If *E. coli* O157:H7 was identified by culture from food samples, isolates were sent to the LSPQ for characterization and for comparison to human isolates.

MAPAQ and CFIA's inspectors conducted a traceback investigation of suspected products.

Results

Epidemiological investigation

A total of seven cases from which *E. coli* O157:H7 (EC1055) was isolated were identified as part of this outbreak: five cases in Quebec and two cases (from two other Canadian provinces) that had visited Quebec during the incubation period. Symptom onset dates ranged from November 26, 2013 to December 15, 2013. The median age was 25 years old (range: 17–43 years). Two cases were hospitalized and one case developed HUS requiring dialysis. The average incubation period was 5 days (range 1–10 days). There were no deaths.

All cases reported eating a variety of tartares at either site A1 or A2 between November 16, 2013 and December 10, 2013 (Fig. 1). None of the individuals reported eating tartare elsewhere. No other *E. coli* O157:H7 cases were reported in Quebec during this period of time. Five cases consumed tartares at site A1 and two at site A2. Overall, there were 24 days between the consumption of the first and the last case. The common item to each menu consumed by the *E. coli* O157:H7 cases was beef, except for one case who only ate veal tartare (Table 1).

As represented in Table 1, meal companions ate different tartares. The proportion of cases who ate veal tartare is higher for cases versus meal companions but only one *E. coli* O157:H7 case consumed only veal tartare. In two situations, cases shared the same plate with their meal companions. One case shared a tartare tasting plate with one meal companion and one case shared a combination of tartare including salmon, beef, and duck with three companions. Information about the quantity of tartare consumed for each case or meal companions was not available. None of the meal companions were sick. No statistical tests were performed because the number of cases and meal companions were limited and cross-contamination is possible because mixed tartares are served in the same plate, which may confound results.

Tartares could be served with different seasonings as per customer choice, that is, once the meat or fish was chosen from the menu, the customer could select from seasoning options. The same seasonings may be added to each of the tartares; however, green onions were the only probable common ingredient among possible seasonings. Green onions were excluded from further investigation beyond the initial epidemiological analysis because if green onions were contaminated, we would have had significantly more cases. Furthermore, cases occurred over a 1-month period, which

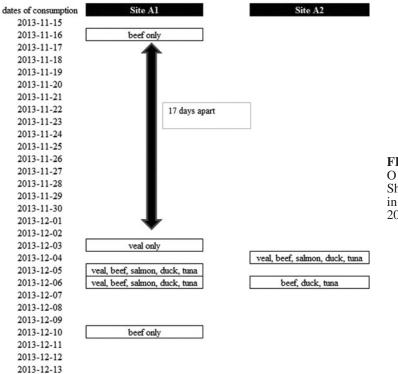


FIG. 1. Tartares consumed by *Escherichia coli* O157:H7 cases and dates of consumption, in the Shiga toxin–producing *Escherichia coli* outbreak in the Province of Quebec, November–December 2013, N=7.

TABLE 1. CONSUMPTION OF TARTARES BY <i>Escherich</i>	ĪΑ
COLI O157:H7 CASES AND THEIR MEAL COMPANIONS	5,
December 2013, Province of Quebec	

Tartares	Cases $(n=7)$	Meal companions $(n=13)$			
Beef	6 (85.7%)	6 (46.1%)			
Veal	4 (57.1%)	1 (7.6%)			
Duck	4 (57.1%)	4 (30.7%)			
Salmon	4 (57.1%)	3 (23.1%)			
Tuna	3 (42.9%)	4 (30.7%)			
Scallops	0	1 (7.6%)			

means that different lots of green onions would have been involved or different brands. Finally, if green onions were involved, there would be an extremely low probability of a delay of 17 days between the first 2 cases.

LSPQ

From December 16, 2013 to December 27, 2013, 4 *E. coli* O157 isolates were sent to LSPQ. These isolates had been collected between December 9, 2013 and December 14, 2013. The isolates all had the same PFGE profile, EC1005 (ECXAI.0248/ECBNI.0215). This profile had not been previously identified in either Quebec or Canada. The isolates also had the same MLVA profile (15-7-11-2-4-7-8-7), confirming the specificity of the strain. This cluster was posted on Pulse-Net Canada on December 23, 2013. One case associated with this cluster had HUS, but *E. coli* O157:H7 could not be isolated from the case's stool culture. Analysis by LSPQ of the HUS stool specimen confirmed the presence of Shiga toxin genes by polymerase chain reaction, but no STEC were isolated.

Food safety investigation

Food inspectors focused their activities at a restaurant that had two sites (A1 and A2) because no other *E. coli* O157:H7

cases with the same PFGE profile were reported in the province during this period. Both sites have the same owner and share the same tartares menu. Because beef and veal were identified as the common item among cases, food inspectors focused their inspection on those food items.

The owner ordered beef and veal meat every 1–2 weeks or when needed. According to food inspectors, beef and veal are delivered to site A1 in cryovac packaging (i.e., vacuumpackaged) and stored in a refrigerator until the cryovac package is opened. There was no electronic thermometer or alarm in the refrigerator to monitor temperature. When opened, the outer surface of the meat is removed and the remaining meat is chopped or minced. The chopped meat is divided into 100-g portions that are shaped and stored in a refrigerator for a maximum of 3 days. The date of chopping is clearly identified on each 100-g container. There is no other handling until the tartare is ordered. When ordered, seasonings are added to the meat and served to the customer. When needed, prepackaged 100-g portions of meat can be delivered to site A2 where only seasonings are added prior to serving. There is no meat chopping at site A2.

Beef and veal turnover from November 3 to December 2 only are summarized in Table 2. Cryovacs delivered to the restaurant are not necessarily opened the same week they were delivered. Thus, cryovacs may accumulate week after week. If there is enough product in cryovac accumulated to support an usual volume of tartare served during a week, the owner will not order more. The shelf life of these products in a cryovac is about 90 days, following Canadian regulations (Government of Canada, 1990).

When ordered, each crate of beef contained three cryovac packages of beef filet and each crate of veal contained one cryovac package of veal filet. Each single package with a filet of beef can provide up to 50 portions of 100 g and each single package with a filet of veal can provide 40 portions of 100 g.

Different portions of tartare are available on the restaurant menus. Clients may order 50-, 100-, 150-, or 200-g portions.

Weeks starting:	Cryovac of veal and beef delivered at site A1 and portions			Veal and beef tartares ordered by site (portions of 100g)				
				Site A1		Site A2		
	Veal and beef	No. of cryovac bags	Portions of 100 g	Beef	Veal	Beef	Veal	Total
November 3	3 crates of beef 2 crates of veal	9 2	450 80	na	na	na	na	
November 10	4 crates of beef 3 crates of veal	12 3	600 120	217	53	241	29	458 82
November 17	0			270	58	226	41	496 99
November 24	3 crates of beef 4 crates of veal	9 4	450 160	209	23	264	34	473 57
December 2	4 crates of beef 4 crates of veal	12 4	600 160	216	51	237	39	453 90

TABLE 2. TURNOVER IN THE MANAGEMENT OF BEEF AND VEAL AND CONSUMPTION OF TARTARES AT BOTH SITES(A1 AND A2), DURING THE TIMEFRAME OF THE SHIGA TOXIN–PRODUCING ESCHERICHIA COLI OUTBREAKIN THE PROVINCE OF QUEBEC, NOVEMBER–DECEMBER 2013

One crate of beef contains 3 cryovac bags; 1 crate of veal contains 1 cryovac bag; 1 cryovac bag of beef filet contains approximately 50 portions of 100 g; 1 cryovac bag of veal filet contains approximately 40 portions of 100 g.

na, not available.

In a tartare tasting plate, 50 g of each meat or fish product is served. Only one food handler is responsible for chopping or mincing the meat and this is done only at restaurant A1. There were no employees at either site who had reported symptoms of diarrheal disease and thus, no stool samples were requested.

Preparations of beef and veal tartare (chopped meat with spices) were requested by food inspectors, and samples made for specifically for testing were collected. Beef and veal samples directly from the cryovac format were also collected and tested for *E. coli* O157:H7. In total, 11 food samples were submitted for analysis. No samples from lots of beef or veal consumed by the cases were available for testing. A total of nine environmental samples were collected (knives, work tables, freezer, etc.) at site A1. All food and environmental specimens tested negative for *E. coli* O157:H7. The kitchen is cleaned every day.

Traceback activities done by MAPAQ and Ville de Montréal food inspectors identified only one supplier/distributor that delivered beef and veal under cryovac format to site A1 during this period. As the meat is already in a cryovac format, there is no meat manipulation at this level. Chopped beef and veal meat are delivered to site A2 from site A1 when needed. CFIA's food inspectors identified two slaughterhouses that provided beef and veal in a cryovac format to this distributor. Beef meat came from slaughterhouse A, which slaughters >4500 animals a day, and veal meat came from slaughterhouse B, which slaughters <400 animals a day (80,000 annually).

Records of both slaughterhouses were reviewed by CFIA's food inspectors, and no deficiencies were observed during the period of time around the outbreak. No trim samples tested positive for *E. coli* O157:H7 during this timeframe. Most of the time, only trims are tested for *E. coli* O157:H7 because trims are intended to make ground beef. Beef filet is rarely tested unless needed.

CFIA's inspectors checked the distribution of beef and veal filets during the identified timeframe. Slaughterhouse A distributed their product to >80 restaurants or markets in 8 regions in Quebec. Slaughterhouse B distributed their product to <10 clients in 4 regions in the province of Quebec. Both slaughterhouses distributed these products across Canada, and no other cases of *E. coli* O157:H7 EC1055 were reported and associated with other restaurants or institutions according to the provincial surveillance program results.

Discussion

Investigation of this outbreak of *E. coli* O157:H7 focused on contaminated beef or veal from site A1 as the most likely cause of the outbreak. All of the cases had consumed beef or veal tartare prepared at site A1. No other common ingredients were identified as a potential source of *E. coli* O157:H7.

No food handlers at site A1 reported being sick. No deficiencies while handling, chopping, and cooling the meat at site A1 were identified. Cases were exposed over a 24-day period, making it unlikely for an asymptomatic foodhandler to be responsible for this outbreak. Since no major deficiencies were observed in food handling at this restaurant, it seemed probable that the initial cause of the outbreak was contaminated meat purchased by the restaurant. STEC is a common part of the microflora of the gastrointestinal tract of cattle and calves, and is shed in the feces (Hancock *et al.*, 1994; Van Donkersgoed *et al.*, 1999; Gannon *et al.*, 2002; Murinda *et al.*, 2002). At the slaughterhouse, the contamination of beef or veal carcasses with STEC can occur during removal of the hide and evisceration (Elder *et al.*, 2000; McEvoy *et al.*, 2003; Hussein, 2007; Stanford *et al.*, 2013). STEC on carcass surfaces can then be transferred to primal cuts during subsequent processing of the carcass.

However, beef and veal products from slaughterhouses A and B, which supplied site A1, were broadly distributed across the province of Quebec and Canada and no other STEC infections involving the same PFGE profiles were reported during this period. Even in the context of a low level of the organism being present on the meat, handling and storage conditions may have provided an opportunity for E. coli to replicate in this specific environment. If beef product is of normal pH (<5.9) and remains under vacuum, there is no potential for growth of E. coli O157. Once vacuum packaging is opened, the environment becomes aerobic and growth of E. coli is limited only by temperature (USDA, 2003; Gill and Gill, 2010). Growth of *E. coli* has not been reported at 4°C, the stated temperature of storage, but the potential for growth cannot be excluded if the temperature fluctuates to 6°C or higher (Jones et al., 2004, 2013). This scenario is possible if the storage temperature had not been accurately recorded or the temperature in the refrigeration unit fluctuated throughout the day in response to the compressor cycle and workers periodically opening the unit. Pieces of meat chopped for tartare were kept in the refrigerator between several hours and up to a maximum of 3 days without electronic or alarm monitoring. In this outbreak, it was impossible to know the length of time between preparation and serving of the meat that any of individual cases consumed. As seen in Table 2, the turnover of beef and veal products was high. Because there is a possibility of bacterial growth once the cryovac is opened (if bacteria are already present) and tartares are consumed raw, this meal should be served as fresh as possible to limit or avoid any bacterial growth.

An alternative possibility is the transfer of *E. coli* O157:H7 from some initially contaminated product (such as beef from cryovac) to an unidentified environmental niche (cutting board, knives, etc.) where the bacterium could replicate and be a source for ongoing contamination (for example, contamination of veal). This scenario could explain the case that consumed only veal and the presence of the same PFGE profile in all cases. Another possibility that might explain the case that consumed only veal would be that beef was served instead veal. This might have been possible since both meats look similar when prepared as they were at site A1.

In summary, the first 2 cases consumed tartare 17 days apart and the period between the second and the seventh cases was 7 days. The delay between the consumption of tartare by cases may be consistent with the delivery of more than one cryovac of meat from the same shipment or different shipments contaminated with *E. coli* O157:H7 of the same PFGE pattern. Contamination of meat from a persistent source or unidentified niche within the restaurant is also possible. Bacterial growth in the refrigerator represents another possible hypothesis, since meat is chopped and may be kept in the refrigerator for a period as long as 3 days.

Even though we can only formulate hypotheses about product contamination, given that none of the tested samples were *E. coli* O157:H7 positive, this outbreak raised many concerns regarding the preparation and consumption of raw food products, particularly that of meat.

Recommendations

Consumers should be advised that tartares made from raw meat are products associated with a potential risk of serious infectious disease, and even death. Even if handling, chopping, and cooling of the meat are done properly, they cannot reduce the contamination of meat if already present, and may provide an opportunity for cross-contamination and pathogen replication. Additionally, because temperature fluctuations create a potential for the replication of *E. coli* under refrigeration, meat for raw meat products should be chopped and eaten as soon as possible after preparation, most preferably within the same day.

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Disclosure Statement

No competing financial interests exist.

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