

Illness outbreak associated with *Escherichia coli* O157:H7 in Genoa salami

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Abstract

Background: An outbreak of *Escherichia coli* O157:H7 infection was identified in the spring of 1998, with a 7-fold increase in the number of laboratory-confirmed *E. coli* O157:H7 cases in southern Ontario. This prompted an intensive investigation by local, provincial and federal public health officials.

Methods: Case interviews of 25 people from southern Ontario were conducted using a broad food history and environmental exposure survey. Laboratory investigations involved both case and food sampling. Specimens of foods sold locally and reportedly consumed by those affected were tested. Common suppliers of suspected foods were identified by cross-referencing suppliers' lists with stores frequented by those who fell ill. A case-control study involving 25 cases and 49 age-matched controls was conducted. This was followed by a comprehensive environmental investigation of the meat processing plant identified as the source of the *E. coli*.

Results: Thirty-nine outbreak-related cases occurred between April 3 and June 2, 1998. Of the 36 case specimens tested all were positive for *E. coli* O157:H7. The case-control study identified Genoa salami as the most probable (odds ratio 8 [confidence interval 2–35]) source of the outbreak. Samples of Genoa salami produced by the most commonly identified supplier later tested positive for *E. coli* O157:H7, and the pathogen matched the same pulsed-field gel electrophoresis pattern and phage type of the case specimens.

Interpretation: Our investigation, which led to a national recall of the brand of dry fermented Genoa salami identified as the source of the outbreak, supports an adherence to stringent manufacturing requirements for fermented meat products. A review of the Canadian standards for fermented meat processing and the effectiveness of their implementation is warranted.

Escherichia coli O157:H7 is the most common of the pathogenic *E. coli* serovars identified in humans.¹ Between 1990 and 1996, the number of cases of verocytotoxigenic *E. coli* reported annually in Canada ranged between 1200 and 1700, with 1233 cases reported in 1996 (4.1 cases per 100 000 Canadians).² *Escherichia coli* O157:H7 became a reportable disease in Ontario in 1989. Since then rates of notified cases for Ontario have been similar to those for the rest of Canada; the average annual number of cases reported in Ontario between 1994 and 1998 was 462 (380 cases in 1998) (Dr. Chuck Le Ber, Ontario Ministry of Health, Toronto: personal communication, 1999). Although the number of laboratory-confirmed cases of *E. coli* O157:H7 infection in Canada has dropped from a peak of close to 2500 cases in 1989,¹ the isolate continues to be a public health concern because of its pathogenicity. In Canada in 1995 the hospitalization rate was reported to be 365 per 1000 cases, with a fatality rate 39 per 1000.¹ In addition, *E. coli* O157:H7 is the most common cause of hemolytic uremic syndrome in children.³

Research

Recherche

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Since the first reports of outbreaks of *E. coli* O157:H7 infection in 1982,⁴ public health investigations of outbreaks and individual cases have led to a long list of confirmed food sources that includes undercooked beef, unpasteurized milk, unpasteurized apple juice, yogurt, cheese, water and salad products.⁵ More recently, dry fermented salami has been linked to outbreaks of *E. coli* O157:H7 infection.⁶ These products are made from raw ground meat, usually beef and pork, and are preserved through fermentation and drying, with the addition of salt and spices. It was a common belief among manufacturers of raw meat products that pathogens like *E. coli* were unable to grow in foods that were processed under low pH, low water activity and high salinity conditions and that fermented sausages, therefore, did not require cooking and were ready to eat with no further preparation.

In early May of 1998 a 7-fold increase in the number of illnesses related to *E. coli* O157:H7 was noted through laboratory surveillance in 2 adjoining public health department areas in southern Ontario (population close to 1 million). An epidemiological investigation was initiated to identify a possible common source of the *E. coli* pathogen.

Methods

Case series and case-control study

A hypothesis-generating case questionnaire was designed to collect demographic information on cases and included a broad food and environmental exposure history, taking into account foods and environmental sources previously associated with *E. coli* O157:H7 outbreaks. The questionnaire was administered to all cases in the Niagara and Hamilton regions with symptom onset after Apr. 3, 1998, and to anyone with laboratory-confirmed *E. coli* O157:H7 infection, clinical symptoms of bloody diarrhea and abdominal cramping with an epidemiological link to a laboratory-confirmed case and anyone with hemolytic uremic syndrome.

For each case in the case-control study up to 4 age-matched neighborhood controls were identified either by the families or through neighborhood schools. Children were matched on the basis of preschool, elementary or high school age, and adults were matched on age to within 5 years. Case interviews were conducted in person by a public health nurse or public health inspector, or both, and controls were interviewed in person or by telephone.

The case-control questionnaire included questions about foods frequently reported in the case series investigation and focused on usual eating patterns to reduce recall bias and assess probable exposure to these foods. Respondents were asked whether they normally ate any of: 17 different deli meats, 6 deli-sliced cheeses, common sandwich condiments, other ready-to-eat foods and fresh or precooked ground beef. To test for an association between illness and the potential for exposure to each food during the 8-day incubation period, respondents were also asked how often (i.e., daily, weekly, every 2 weeks, once a month, or less than once a month) they ate each of the foods in question. Matched odds ratios (ORs) were calculated for each food item in relation to whether they were normally consumed. Variables with ORs found to be statistically significant (1-tailed *p* value) were selected for conditional logistic regression modelling. Matched ORs

were also calculated for the 17 deli meats and ground beef on the question of consumption frequency after dichotomizing frequency to "frequently" (i.e., daily, weekly or every 2 weeks) and "infrequently" (i.e., once a month or less).

Laboratory investigation

Laboratory tests were conducted on stool specimens from those who were ill and food samples from their homes, on deli foods selectively sampled by public health inspectors from the stores frequented by the cases and on closed samples of Genoa salami from suppliers who distributed to the region. Stool and food samples were emulsified and incubated in enrichment broths and the supernatants were tested for verocytotoxin using a commercial enzyme immunoassay.⁷ Verocytotoxin-positive samples were subjected to cultural confirmation using standard methods.⁸ A verotoxin-immunoblot method⁹ was also used to increase the sensitivity for isolating the pathogen from the broth cultures of deli-meat specimens; once isolated, these cultures were further verified using official methods. All cultures identified as *E. coli* O157:H7 were genotyped using pulsed-field gel electrophoresis¹⁰ and phage typed.¹¹

Food investigation

Detailed cross-referencing of deli-product suppliers of stores frequented by cases was conducted to identify common suppliers in the outbreak region. The plant of the meat-product producer implicated as the probable source of the *E. coli* was then inspected by the Canadian Food Inspection Agency.

Results

Case series and case-control study

Between April 3 and June 2, 1998, 39 primary cases of *E. coli* O157:H7 infection were identified (Fig. 1): 22 from the Niagara region, 9 from the Hamilton region, 3 from

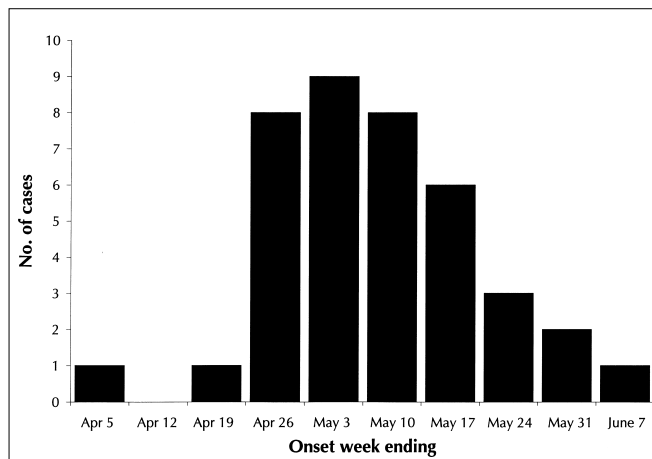


Fig. 1: Epidemic histogram of confirmed outbreak-related cases of *Escherichia coli* O157:H7 by date of onset in Ontario — April 3 to June 2, 1998.

Thunder Bay and 1 from Newmarket; the other 4 had travelled to the outbreak area.

Twenty-nine (74%) of the 39 cases presented with bloody diarrhea, and 27 (69%) presented with abdominal cramping; 14 (36%) were admitted to hospital, and 2 children (5%) were diagnosed with hemolytic uremic syndrome. An equal proportion of males and females were affected, and the median age of the group was 16 years (range 18 months to 69 years).

The most commonly consumed ready-to-eat food was deli-cut meat (purchased from different deli counters); 37 (95%) people reported eating a deli meat or cheese product cut at a deli counter in the week before their illness, and 26 (67%) reported eating Genoa salami in the week before they fell ill.

Data from the 25 cases that had been identified when the case-control study was initiated and from 49 matched controls were included in the case-control statistical analysis. Univariate analysis of individual foods revealed a matched OR of 3 or greater and $p < 0.05$ for 7 foods (Table 1). Genoa salami was most strongly associated with illness (matched OR 8, 95% confidence interval [CI] 2–35). Foods with significant ORs were introduced individually into a conditional logistic regression model with Genoa salami. Only fresh ground beef in combination with Genoa salami provided an improved regression model over the model with Genoa salami alone (likelihood ratio 20.6 v. 14.4, $p = 0.013$). The adjusted OR for Genoa salami was 19 (95% CI 3–816) and for fresh ground beef was 20 (95% CI 1–1625).

In the univariate analysis of consumption frequency (i.e., probability of exposure) only Genoa salami was significantly associated with illness (matched OR 5, 95% CI 1–22, $p = 0.01$). The matched OR for fresh ground beef was not significant.

Laboratory and food-source investigation

Stool samples of 36 of the 39 cases we investigated were confirmed by laboratory tests to be *E. coli* O157:H7 posi-

tive; the other 3 cases were symptomatic and epidemiologically linked (i.e., had contact with a confirmed case with same exposure, symptoms and onset). All isolates displayed the same pulsed-field gel electrophoresis pattern, and 34 (95%) of the 36 isolates were phage type 14, 1 was phage type 4 and the other was phage type 8.

Escherichia coli O157:H7 was not detected in any of the original food samples taken from the retailers and the homes of cases. However, evidence from the case-control study led to a more selective screening of Genoa salami, and 39 samples of 13 different lots from different manufacturers were subsequently tested; 3 of 7 samples tested from 1 lot were positive for *E. coli* O157:H7. Phage typing and pulsed-field gel electrophoresis patterns were indistinguishable from those of the clinical isolates. This lot of Genoa salami was manufactured in mid-February and, after processing and drying, was available at retail outlets by the middle of April, 1998; *E. coli* O157:H7 was subsequently isolated from other lots manufactured at the same plant during the same period.

Over 150 suppliers of deli products were identified by the 16 retailers that were frequented by cases. The most common supplier, who provided Genoa salami to 13 of the 16 stores, supplied the salami from which the culture-positive samples were obtained. An investigation of the implicated plant by Canadian Food Inspection Agency in May and June of 1998 revealed significant problems with the manufacturing process — the use of natural fermentation in manufacturing 4 of its products including Genoa salami, poor record keeping, no microbiological tests of incoming raw ingredients or final products, no records of lot-specific pH or degree-hours measurements, faulty pH measurement methods and no written procedures for how to manage products with abnormal results.

The Genoa salami from this plant was recalled nationally and another health hazard evaluation¹² was carried out to assess the risks posed by other products produced at this plant. The recall included the removal of products that had been distributed in Ontario, British Columbia and the United States.

Table 1: Matched odds ratios (univariate) for selected food preferences*

Food consumed	No. (and %) of cases†	No. (and %) of controls‡	Odds ratio (and 95% CI)‡	<i>p</i> value
Genoa salami	14/23 (61)	6/46 (13)	8 (2–35)	< 0.001
Fresh ground beef	22/23 (96)	38/48 (79)	7 (1–159)	0.04
Margarine with deli meats	11/24 (46)	11/48 (23)	6 (1–42)	0.02
Capicola ham	10/24 (42)	8/47 (17)	4 (1–20)	0.02
Cheese with deli meats	18/23 (78)	21/49 (43)	5 (2–17)	0.004
Submarine sandwich	12/24 (50)	15/47 (32)	4 (1–18)	0.04
Mozzarella cheese	14/24 (58)	16/47 (34)	3 (1–12)	0.04

Note: CI = confidence interval.

*Food not associated with illness included: a variety of hams, a variety of salamis, roast beef, turkey, chicken, bologna, ground beef, a variety of cheeses, common sandwich condiments, various fruits, vegetables and dairy products and eating patterns (e.g., eating deli meats, eating at deli shops, eating prepackaged deli meats, buying food at farmer's markets).

†Although 25 cases and 49 controls were included in the analyses, all of those questioned did not respond to each question; thus, denominators differ and are indicated for each food.

‡Maximum likelihood estimate of odds ratio and 95% mid-P confidence limits.

Interpretation

This is the first Canadian publication documenting dry fermented sausage as the source of an outbreak of *E. coli* O157:H7 related illness. Through Central Public Health Laboratory surveillance public health officials recognized an increase in the number of *E. coli* cases in a limited geographic area. The epidemiological investigations identified a link between those infected and the consumption of dried fermented Genoa salami, and a more focused laboratory investigation using molecular subtyping then confirmed the link. There were, however, some inconsistencies in the results that require further discussion.

Although only 27 (69%) of the 39 people recalled eating Genoa salami during the week before they became ill, the majority (37 [95%] of 39) reported consuming deli-sliced products (i.e., cheese or deli meat) during the incubation period. It is common practice in delis to slice food products on the same slicer without cleaning between slicing jobs because it is assumed that all products are free of contaminants and proper food-handling practices are followed. However, when an unsafe product is sliced there is a potential for cross contamination. Thus, some of the cases in this investigation may not have been associated with the consumption of Genoa salami but with eating other deli foods contaminated by the Genoa salami.

Our case-control study implicated Genoa salami and fresh ground beef as possible food sources. However, fresh ground beef was not likely a second food source of the outbreak because there was no common point of exposure. Given that raw hamburger is the most publicized food source for *E. coli* O157:H7 infection, a reporting bias may have led to the elevated OR for hamburger.

Dry fermented sausages have been identified as a source of *E. coli* O157:H7 in other countries as well. In the 1994 outbreak in Washington and California, 20 laboratory-confirmed cases of *E. coli* O157:H7 were linked to the consumption of dry cured sausage.⁶ Studies have shown that *E. coli* O157:H7 can survive many of the typical dry fermentation processing conditions;¹³⁻¹⁵ *E. coli*'s tolerance of acidic conditions has also been reported in the processing of other foods such as apple cider¹⁶ and mayonnaise.¹⁷ These findings led to significant changes in the industry and in the manufacturing of dry fermented sausage in the US.¹³ Because the infective dose of *E. coli* O157:H7 is very low¹³ any survival in a ready-to-eat product has the potential to cause illness.

Escherichia coli O157:H7 may be present in raw ground beef that is used in the manufacturing of other meat products.⁶ Manufacturers must take this into account and be required to follow the appropriate procedures to control for *E. coli* O157:H7 in the final product. In 1996 the United States Department of Agriculture Food Safety Inspection Service recommended that deli meat manufacturers adopt 1 of 5 options for the control of *E. coli* when they make fermented sausages.¹⁸ Although these options have not been

adopted in Canada as standards they have been circulated by the Canadian Food Inspection Agency¹⁹ to all federal meat plants manufacturing fermented meats (Dr. Tom Gleeson, Health Canada, Ottawa: personal communication, 1999).

The results of this investigation indicate a breakdown in Canadian consumer protection. Although this outbreak was traced to 1 processing plant and a specific meat product, the potential for future outbreaks calls for an immediate collaborative review of ready-to-eat deli-meat processing methods, standards and regulations and for action by the Canadian government and the industry to improve consumer safety and minimize public health risk in the future.

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Competing interests: None declared.

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