AN OUTBREAK OF *ESCHERICHIA COLI* O157 INFECTION ON THE GOLD COAST

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Abstract

An outbreak of bloody diarrhoea associated with *Escherichia coli* O157 infection in young children on the Gold Coast in Queensland was investigated. This outbreak was the first involving the O157 serotype in Australia. Fifty-seven people were screened and *E. coli* O157 was isolated from six people, all of whom had consumed different food items from a delicatessen. No single food item was identified as the source of the infection. One of the food handlers who was positive for *E. coli* O157 had minor symptoms of gastroenteritis preceding the onset of disease in the identified cases. This person had prior contact with an animal that showed clinical signs of infection, suggesting a possible method of entry for the organism into the delicatessen. However, it is also possible that a contaminated food product entered the delicatessen and contaminated other food products during handling. Therefore, cross contamination within the delicatessen was a likely associated factor in the transmission of this disease.

Introduction

Escherichia coli O157:H7 is one of a group of enterohaemorrhagic *E. Coli* (EHEC) capable of causing severe disease in humans. Since 1982, this pathogen has been increasingly recognised overseas as a major cause of morbidity and mortality, with an estimated 21,000 infections and as many as 250 deaths annually in the United States of America alone[1]. Overseas, the number of outbreaks associated with *E. coli* O157:H7 have been increasing in recent years, with the largest outbreak involving approximately 500 cases in the United States of America in 1993[2,3,4].

Infection with enterohaemorrhagic *E.coli* may result in a broad spectrum of clinical manifestations. These range from asymptomatic infection, diarrhoea or haemorrhagic colitis to haemolytic uraemic syndrome (HUS) in children or thrombotic thrombocytopenic purpura (TTP) in adults. The pathogenesis of these conditions in cases of EHEC infection is mediated by the bacteria producing verocytotoxin or shiga-like toxin (SLT)[5]. HUS is characterised by haemolytic anaemia, thrombocytopenia and acute renal failure (oliguria or anuria with elevated serum urea and creatinine). This infection is more frequently encountered in children under five years of age. Usually, HUS occurs in two to seven per cent of cases although in outbreaks, up to 30% of cases develop this complication[6, 7]. Mortality from HUS ranges from 3% to 17%, while long-term renal impairment may also occur[8].

Sporadic cases of EHEC infection have been recognised in Australia since 1987, although the proportion of *E. coli* O157:H7 isolates has been low[9]. A 1991 survey of children with diarrhoea at a Sydney hospital found that *E. coli* O157:H7 was an uncommon cause of acute gastroenteritis in that particular Australian context[10]. The isolates of EHEC from Australian cases of haemorrhagic colitis or HUS have been other serotypes such as O111:NM, which was implicated in the South Australian outbreak of HUS in 1995, and O157:NM[9, 11].

Most outbreaks of EHEC disease have been associated with the consumption of beef, primarily under-cooked ground beef. Other implicated means of infection include inadequately washed vegetables[12], yoghurt[7], unpasteurised milk[13] and apple cider[14]. Modes of transmission other than foodborne spread have also been identified, including person-to-person transmission[15], contact with infected farm animals[16], and from swimming in a faecally contaminated lake[17]. Cattle are considered to be the main reservoir of the infection, with EHEC organisms occurring as part of the normal intestinal flora of a small percentage of cattle[6]. Meat for human consumption may be contaminated during the slaughtering process. The infectious dose of EHEC organisms for humans appears to be extremely low[18].

This report describes an outbreak of *E.coli* O157 disease which occurred on the Gold Coast in south-east Queensland during March 1996. This is the first reported outbreak of *E.coli* O157-associated disease in Australia.

On 12 March 1996, the first two of three notifications of *E. coli* O157 infection were received by the Southern Zone Public Health Unit, Brisbane.

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Methods

Upon notification of the first two cases, parents of the affected children were interviewed. A standard foodborne illness questionnaire was used. Enquiries determined that a number of other close family members and school contacts had reported diarrhoea in the preceding or similar period. All close family contacts (symptomatic and asymptomatic) of the cases and school contacts who had diarrhoea provided faecal specimens and completed a modified foodborne illness questionnaire which specifically focussed on risk factors for EHEC infection. Initial responses from the parents of the two cases implicated a common food outlet (a delicatessen), but not a common food item. The delicatessen was inspected, food samples and environmental swabs were taken and two separate faecal specimens were requested from all staff (food handlers) working in the premises.

Upon notification of a third case, and the detection of *E. coli* O157 isolates in a family contact and two food handlers, a case control study was commenced. The case definition used was 'isolation of *E. coli* O157 from a faecal specimen, with or without the presence of an acute enteric illness characterised by fever, abdominal pain, diarrhoea or bloody diarrhoea'. Controls included *E. coli* O157 culture-negative family members, school contacts, food handlers and other children seen at the Gold Coast Hospital with bloody diarrhoea of different aetiology.

Faecal and serological specimens

Initial *E. coli* O157 isolates were detected by the Gold Coast Hospital Pathology Laboratory. This was made possible by the laboratory's protocol of routinely setting up sorbitol MacConkey agar plates when testing faecal specimens of patients with bloody diarrhoea. Non-sorbitol fermenting colonies on these plates were forwarded to Public Health Microbiology Laboratory (PHML) Queensland Health Scientific Services, where they were confirmed using a commercial antisera for *E. coli* O157. All faecal specimens collected in this investigation were examined at the PHML.

Isolated colonies of *E. coli* O157, and the faecal specimens from which they were derived, were tested for the presence of shiga-like toxin (SLT) using an ELISA (enzyme-linked immunosorbent assay) method. Separate tests were performed to determine the flagella antigen. Pulsed field gel electrophoresis using restriction enzymes was also carried out by PHML to examine the clonal characteristics of these isolates. Phage typing was performed at the Microbiological Diagnostic Unit (MDU) using a specific set of *E. coli* O157 phages.

Serological testing for *E. coli* O157 is a developmental, non-standardised ancillary test. Recognising these limitations, the investigators obtained blood samples for serology from *E. coli* O157 culture-negative family contacts with previous symptoms of diarrhoea and one symptomatic child who was culture-negative for *E. coli* O157.

Serological and faecal specimens were also obtained from an animal that showed clinical signs of infection, from one of the positive food handlers, and other animals associated with the person.

Food and environmental specimens

Quantities of food samples were obtained from patients' homes and the delicatessen suspected of being responsible for the infection. They were cultured specifically for the presence of *E. coli* O157 using methods supplied by the Institute of Medical and Veterinary Science (IMVS) in Adelaide. This involved a qualitative method, sampling 25 grams of food and plating directly onto sorbitol MacConkey agar plates after 24 hours enrichment in buffered peptone water. The quantitative method involved a standard five-tube Most Probable Number (MPN) method with all cultures being incubated at 37^0C, followed by plating onto Eosin Methylene Blue (EMB) agar and sorbitol MacConkey agar. Those suspect colonies which were confirmed as *E. coli* were tested for agglutination with specific *E. coli* O157 antisera. Environmental swabs taken from the delicatessen were examined qualitatively in a similar manner.

Results

In all, 57 individuals provided 84 faecal specimens for this investigation. This included 18 specimens from the six infected people, 17 family contacts, two school contacts, 24 delicatessen staff and eight others (two patients with symptoms and six of their family contacts). Thirty of these people completed a questionnaire and were eligible as controls

Six people were identified during this outbreak as having evidence of infection with *E.coli* O157. Three people were identified through screening procedures.

Case 1

Case 1 was a ten year old male who developed a sore throat and slight fever on 1 March 1996. This progressed to an illness with fever, abdominal cramps and associated bloody diarrhoea. He presented to the Gold Coast Hospital on 3 March 1996. Pathology tests showed a normal full blood count and biochemistry. Subsequent faecal samples showed *E. coli* O157. The illness lasted for six days and the patient made a full recovery without hospital treatment.

Case 2

Case 2 was a five year old male who developed abdominal cramps and malaise on 5 March 1996 with subsequent bloody diarrhoea. This patient also presented to the Gold Coast Hospital where his blood count and biochemistry were normal. A faecal specimen grew *E. coli* O157. This child had a brief illness and made a complete recovery.

Case 3

Case 3 was a 22 month old female who developed fever, nausea and vomiting on 7 March 1996, followed by watery diarrhoea ten days later. Examination of a stool

specimen obtained on 19 March 1996 detected a growth of *E.coli* O157. Other pathology results were normal. Despite the duration of her illness (17 days), this child also made a complete recovery.

Case 4

The fourth person was the asymptomatic 58 year old grandmother of case 1.

The last to be identified were two food handlers working in a delicatessen.

Case 5

Food handler one, a 20 year old female, reported mild abdominal cramps on 26 February 1996, suggesting that she might have been the index case of this outbreak. This person had contact with a pet dog that had had bloody diarrhoea in the week before the onset of her own symptoms. Food handler one and the pet had frequently visited a rural property prior to the onset of symptoms. However, there were no cattle resident on that property or in the vicinity. The faecal specimens from all sampled animals, including the pet dog of the food handler, were negative for *E. coli* O157.

Case 6

Food handler two, a 17 year old female, reported no gastroenteric symptoms.

The case-control study did not reveal any statistically significant associations between disease and particular food items. A salient finding was that all six infected people had consumed different food items from the same delicatessen. Inspection of the food handling procedures in the delicatessen revealed a generally satisfactory standard of food hygiene in the workplace. Protocols, including periodic training, were in place to emphasise the importance of food hygiene to employees. However, the attention of management was directed to several areas of importance, including the adequate maintenance of hand washing facilities and the appropriate storage of products in display cabinets. Upon the identification of the infected food handlers and their potential as asymptomatic carriers to contaminate food items, the management cooperated in the disposal of any items which the food handlers may have contaminated and in the thorough sanitisation of the workplace. The infected staff members were excluded from food handling duties until they had demonstrated microbiological clearance of the organism (defined as two negative faecal specimens taken at intervals of not less than 48 hours)[19]. All manufacturers of smallgoods retailed at the outlet were subjected to review of quality assurance procedures.

Faecal and serological specimens

All isolates of *E. coli* O157, and some of the faecal specimens from which they were derived, tested positive for SLT. Pulsed field gel electrophoresis of all six isolates demonstrated greater than 95% homology, suggesting that the isolates were clonally identical. All isolates were phage typed as phage type 14.

Tests for the flagella antigen have demonstrated motility, but have not yet identified the H7 antigen. However, these tests are being repeated at the Fairfield Hospital, Melbourne. The final results of serological testing are currently unavailable.

Food and environmental specimens

Although a large number of food samples were provided from the cases' residences and the delicatessen thought responsible for the spread of the infection, *E. coli* O157 was not detected. Similarly, environmental swabs of surfaces, cutting machinery, utensils and other areas in the delicatessen did not detect *E. coli* O157.

Discussion

No particular food item could be identified bacteriologically or epidemiologically as the source of this outbreak of *E. coli* O157 infection. However, a common epidemiological link was established with the consumption of different food items from a particular delicatessen. It is likely that cross contamination of multiple food items within the delicatessen was a factor in the spread of this infection. The dates of onset of illness suggest that transmission of this pathogen commenced during late February. The onset of symptoms in food handler one, prior to onset in the documented cases, suggests that she (and hence the animal (dog) with the bloody diarrhoea) may have been the source of the infection. This investigation cannot exclude the possibility that a contaminated food product entered the delicatessen and contaminated other food products during its handling. The possibility exists that lapses in personal hygiene amongst the staff may also have contributed to the transmission of this organism.

An important factor in the identification of this outbreak was the use of sorbitol MacConkey agar plates by the Gold Coast Hospital Pathology Laboratory in the routine investigation of patients with bloody diarrhoea. The Australian experience with EHEC has suggested that *E. coli* O157 was not a prevalent pathogen. However, this experience suggests that the use of sorbitol MacConkey agar plates in the investigation of bloody diarrhoea should be reconsidered as an aid to detecting further outbreaks. It is important to remember that not all EHEC serotypes fail to ferment sorbitol. Laboratories also need to be aware of the necessity to serotype and test for SLT production in any heavy growth of *E. Coli* associated with bloody diarrhoea in the absence of other pathogens[9].

Another important feature of this investigation was the role of enhanced surveillance. All pathology laboratories and medical practitioners on the Gold Coast were notified about the occurrence of the first two cases of *E. coli* O157 infection. Surveillance was also carried out in the schools that the first two cases attended. This enhanced surveillance facilitated the detection of the third case and assisted in further defining the nature of the outbreak.

The occurrence of this outbreak emphasises the importance of correct food handling procedures and strict hygiene and sanitation precautions in the food industry, at both the manufacturing and retail levels. Inspection, and reinforcement of education about food hygiene should be a standard practice in food outlets, particularly for those dealing with smallgoods.

This outbreak is the first documented outbreak of *E. coli* 0157 infection and only the second recognised outbreak of EHEC infection in Australia. It may be that the apparent increased number of outbreaks overseas is about to be experienced in Australia. The lessons from this outbreak include the value of routine microbiological surveillance for EHEC organisms (including the use of sorbitol MacConkey agar plates), the role of extensive screening and enhanced surveillance in detecting further cases and the importance of obtaining industry cooperation in the prevention and management of outbreaks of this disease.

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References

1. Boyce TG, Swerdlow DL, Griffin MD. *Escherichia coli* O157:H7 and the Hemolytic-Uremic Syndrome. *N Engl J Med* 1995; 333:364-368.
2. Bell BP, Goldoft M, Griffin PM, *et al.* A multistate outbreak of *Escherichia coli* O157:H7 - Associated bloody diarrhoea and hemolytic uremic syndrome from hamburgers, *JAMA* 1994; 272: 1349-1353.
3. Jay JM. Foods with low numbers of microorganisms may not be the safest foods OR Why did human listeriosis and hemorrhagic colitis become foodborne diseases? *Dairy, Food Environ Sanitation* 1995; 15:674-677.
4. Neill MA. Pathogenesis of *Escherichia coli O157:H7 infection. Current Opinion Infect Dis* 1994; 7:295-303.
5. Moake L. Haemolytic-uraemic syndrome: basic science. *Lancet* 1994; 343:393-397.
6. Griffin PM, Tauxe RV. The epidemiology of infections caused by *Escherichia coli* O157:H7, other enterohemorrhagic *E. coli,* and the associated hemolytic uremic syndrome. *Epidemiol Rev* 1991; 13:60-98.
7. Morgan D, Newman CP, Hutchinson DN, *et al.* Verotoxin producing *Escherichia coli* O157 infections associated with the consumption of yoghurt. *Epidemiol Infect* 1993; 111:181-187.
8. Subcommittee of the PHLS Working Group on Vero cytotoxin producing *Escherichia coli* (VTEC). Interim guidelines for the control of infections with Vero cytotoxin producing *Escherichia coli* (VTEC). *CDR Rev* 1995; 5:R77-R81.
9. Goldwater PN, Bettelheim KA. The role of enterohaemorrhagic *Escherichia coli* serotypes other than O157:H7 as causes of disease in Australia. *Comm Dis Intell* 1995; 19:2-4.
10. Ong J, Zhe L, Robins-Browne R, *et al.* Prevalence of verocytoxigenic *Escherichia coli* serotype O157:H7 in children with diarrhoea attending a Sydney hospital, *J Paediatr Child Health* 1993;29:185-187.
11. Cameron AS, Beers MY, Walker CC, *et al.* Community outbreak of hemolytic uremic syndrome attributable to *Escherichia coli* O111:NM - South Australia, 1995, *MMWR Morb Mort Wkly Rep* 1995;44:550-558.
12. Cieslak PR, Barrett TJ, Griffen PM, *et al. Escherichia coli* O157:H7 infection from a manured garden [letter]. *Lancet* 1993; 342:367.
13. Chapman PA, Wright DJ, Higgins R. Untreated milk as a source of verotoxigenic *Escherichia coli* O157. *Vet Rec* 1993; 133:171-172.
14. Besser RE, Lett SM, Weber JT, *et al.* An outbreak of diarrhoea and hemolytic uremic syndrome from *Escherichia coli* O157:H7 in fresh-pressed apple cider. *JAMA* 1993; 269:2217-2220.
15. Spika JS, Parsons JE, Nordenberg D, *et al*. Hemolytic uremic syndrome and diarrhoea associated with *Escherichia coli* O157:H7 in a day care centre. *J. Pediatr* 1986; 109:287-291.
16. Sharp JCM, Ritchie LD, Curnow J, Reid TMS. High incidence of haemorrhagic colitis due to *Escherichia coli* O157 in one Scottish town: clinical and epidemiological features. *J Infect Dis* 1994; 29:343-350.
17. Keene WE, McAnulty JM, Hoesly FC, *et al*. A swimming-associated outbreak of hemorrhagic colitis caused by *Escherichia coli* O157:H7 and *Shigella sonnei. N Engl J Med* 1994; 331: 579-584.
18. Benenson A (ed.). *Control of Communicable Diseases Manua*l. American Public Health Association. Washington, 1995: 140-144.
19. PHLS Salmonella Committee. The prevention of human transmission of gastrointestinal infections, infestations, and bacterial intoxications. *CDR Rev* 1995; 11:R164.