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Investigation into an outbreak of food poisoning

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SUMMARY

During an outbreak of food poisoning at a church camp, 16 of the 25 people attending were affected. Despite a thorough search for a bacterial pathogen none was identified. An examination of the *Escherichia coli* serotypes present suggest that *E. coli* O159.H9 may have been the organism causing the outbreak.

INTRODUCTION

Outbreaks of food poisoning appear to be increasing. Despite the increase in the variety of organisms shown to cause gastrointestinal symptoms, e.g. enterotoxigenic *Escherichia coli*, *Clostridium perfringens*, *Campylobacter jejuni*, *Bacillus cereus* and *Yersinia enterocolitica*, it is still a major problem to find the causes in many cases. Thus, even a small outbreak involving a limited number of people can require a great deal of work in order to try to ascertain the causes. An investigation into the causative organisms in a recent food-poisoning outbreak was carried out. Examination for the presence of all the above-named organisms as well as the more common enteropathogens is presented.

THE OUTBREAK

During August 1979, 16 people reported illnesses after eating several meals at a church camp that they attended over the weekend of 5 and 6 August.

The families attending had breakfast in their own homes, before going to the camp on the morning of Saturday 5 August. Meals eaten communally were morning tea, lunch and an evening meal. They slept at their own homes on Saturday night and returned to the camp after breakfast on Sunday morning. Sunday lunch was served at the camp but the participants returned home before dinner on Sunday night. Lunch on Saturday consisted of pumpkin soup, sandwiches, bread and butter, biscuits, tea and coffee. Sunday's lunch was the same but also included vegetable soup. Saturday-night dinner was prepared by a hired caterer and consisted of sweet and sour pork, beef stroganoff and rice, potatoes, beans, coleslaw, bread and butter, fruit salad in trifle, ice cream, spanish cream, tea and coffee.

By the time the food poisoning was reported, none of the suspect foods was available for analysis. Symptoms included vomiting, nausea, diarrhoea, prostration and dizziness. Faecal specimens were obtained from 25 persons who ate the communal meals during the weekend.

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BACTERIOLOGICAL METHODS

Faecal samples from all 25 persons were streaked directly onto Salmonella/Shigella, MacConkey, XLD and bismuth sulphite agars and inoculated into Rappaport's and Selenite broths, and after 24 h incubation at 37 °C the broths were streaked onto brilliant green Bile and MacConkey agars. All eight agar plates were examined for characteristic colonies of *Salmonella* and *Shigella* after 24 h incubation and the bismuth sulphite agar was re-examined after a further 24 h incubation. Suspect colonies were tested serologically and biochemically to determine whether they were salmonellas or shigellas.

Faecal samples were also streaked onto TCBS agar to test for the presence of *Vibrios*, and onto egg-yolk agar for the presence of *Bacillus cereus*.

Where enough material was available, a count for *Clostridium perfringens* was performed on egg-yolk agar containing neomycin sulphate. Thirteen specimens were inoculated into Robertson's cooked meat and spread onto blood agar after 24 h incubation. After 24 h anaerobic incubation at 37 °C the blood agar plates were examined for typical *Cl. perfringens* colonies. Three typical colonies from each sample were serotyped.

To examine for *Yersinia enterocolitica*, faecal samples were streaked directly onto bismuth sulphite agar and inoculated into phosphate buffered saline. The broths were incubated at 5 °C for 4 weeks and streaked at weekly intervals onto bismuth sulphite agar. All plates were incubated at 25 °C and examined for typical *Yersinia enterocolitica* colonies after 24 and 48 h.

The method of Skirrow (1977) was used for the detection of *Campylobacter* species, plating the faecal sample directly onto selective blood agar.

All faecal specimens were streaked directly onto MacConkey agar and, after 24 h incubation at 37 °C, six typical lactose-fermenting, *E. coli*-like colonies were picked. They were characterized (Cooke, Ewins & Shooter, 1969) and serotyped using 163 var. O antisera and 55 var. H antisera by the previously described methods (Chandler & Bettelheim, 1974; Meekin, Bettelheim & Bacon, 1979). Representative strains of each serotype obtained from each patient were tested for the production of heat-stable and heat-labile enterotoxins by the methods of Dean *et al.* (1972) and Sack & Sack (1975) and for the production of Vero cell cytotoxin (Konowalchuk *et al.* 1978).

RESULTS

From all the faecal specimens no organisms of the following bacterial groups could be isolated: *Salmonella*, *Shigella*, *Campylobacter*, *Yersinia enterocolitica*, *Bacillus cerus* and *Vibrio parahaemolyticus*. Strains of *Clostridium perfringens* were isolated from four persons. The numbers generally were low and serotyping did not reveal a single serotype.

In Table 1 are listed, in family groups A–G, the people who took part in the church camp as well as their symptoms and the serotypes of *E. coli* that were isolated from their faeces.

Of the 69 strains representing all serotypes from all patients only one strain of *E. coli* was positive for enterotoxin production. It was Vero cell toxic, was isolated from patient B4 and had the serological structure O39.H8.

Table 1. *Serotypes of* Escherichia coli *isolated from patients*

Patients in family groups	Symptoms: Vomiting	Nausea	Diarrhoea	Prostration	Dizziness	Serotypes of *Escherichia coli* found
A 1	−	−	−	−	−	O2.H−; O121.H−; Ont. H−; R.Hnt; R.H−
A 2	−	+	−	−	−	Ont. H10; Ont. HR−; OR. Hnt; OR. HR
A 3	+	+	+	+	+	O2. H1; Ont. H1
A 4	+	+	+	+	+	O2. H1; O15. Hnt
A 5	−	+	−	−	−	O16. H38; O159. H9
A 6	+	+	−	−	+	O23. H15
B 1	−	+	−	+	−	O7.H−
B 2	−	−	−	−	−	O75.H−
B 3	−	−	−	−	−	O75.H−; O102.H−; Ont. H2; OR.H2
B 4	−	−	−	−	−	O18ac.H1; O39.H8; O159.H8
C 1	+	+	−	+	−	O91.H28−; O159.H9
C 2	−	−	−	−	−	O75.H−; Ont. H1; Ont. H−; OR.H−
C 3	−	−	−	−	−	O73.H18; O73.H−; O153.Hnt; O159.H9
D 1	−	−	−	−	−	O69.H39; O91.H28; O148.H28; O148.H53
D 2	−	−	−	−	−	O6.H1; Ont. H19; Ont. Hnt; Ont. H−
D 3	+	+	+	+	−	O132.Hnt; O132.HR
D 4	+	+	−	+	−	O6.H1; O159.H9; O159.H−
D 5	−	−	−	−	−	O73.H18; O73.H−
E 1	+	−	+	−	−	O2.H6; O6.H1; O159.H9
E 2	+	−	+	−	−	O159.H9; O159.H−
E 3	+	−	+	−	−	O75.H−
E 4	+	−	+	−	−	O1.H−
E 5	+	−	+	−	−	O75.H−
F 1	−	+	+	+	−	Ont. H45
G 1	+	+	−	+	−	O159.H9

DISCUSSION

This study shows that, despite an extensive search for a possible enteropathogenic bacterium, none was specifically found. Although strains of *Clostridium perfringens* could be isolated, their numbers and distribution did not suggest that they might have been involved in this outbreak. It was because of this lack of isolation of a specific established enteropathogenic organism that the *E. coli* were further investigated.

A study of the *E. coli* isolated from these people provides an insight into the type of problems which such an investigation can produce. Two people yielded strains which at first sight could be considered likely causes of the outbreak. These are types O39.H8 which was Vero cell toxic (VT+) isolated from B4, and type O148.H28 isolated from D1. This latter serotype has been associated with diarrhoea in many parts of the world since it was first described in Aden (Rowe, Taylor & Bettelheim, 1970). However, neither B4 nor D1 had significant symptoms. These serotypes could have been present in the other people but were possibly not isolated owing to the known complexity of the human faecal flora (Bettelheim, Faiers & Shooter, 1972).

An examination of the serotypes found shows that only *E. coli* O159.H9 occurred with any frequency among all the people taking part in this camp, being isolated from seven people; A5, C1, C3, D4, E1, E2 and G1 (Table 1). Of these people only C3 did not have symptoms, also a number of people with symptoms did not yield *E. coli* O159.H9. Enterotoxigenic *E. coli* belonging to this 'O' group but with different 'H' antigens have recently been associated with intestinal symptoms (Gross *et al.* 1978) and thus it can be suggested that this may be the causative organism. It must nevertheless be stressed that extreme caution should always be taken when making such an assessment.

It also seems noteworthy that those people with no symptoms had a greater variety of serotypes (Table 1). This might suggest that this could be an advantage in preventing colonization of the bowel by a potential pathogen. Only further studies of other similar outbreaks can help to clarify this aspect.

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