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SHORT REPORT
Escherichia coli O157 outbreak associated with fresh unpasteurized goats' cheese

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SUMMARY

A family cluster of three cases of *Escherichia coli* O157 infection was identified in France. Two cases developed haemolytic–uraemic syndrome. The source was fresh unpasteurized goats' cheese, produced by an independent producer. Three *E. coli* O157 strains, isolated from one HUS case and faeces of one cow and one goat, were indistinguishable by toxin type and PFGE pattern.

Shiga toxin-producing *Escherichia coli* (STEC), especially *E. coli* O157:H7, is an important cause of foodborne disease in industrial countries [1]. The main natural reservoir of STEC is represented by ruminants, especially cattle. Human infections are mainly foodborne, but person-to-person and direct contact with contaminated cattle or goats are also identified as transmission routes [2].

In France, the majority of medical laboratories do not routinely examine stools for STEC, and STEC infections are not mandatory notifiable. Therefore, since 1996, the surveillance of STEC infections has been based on a nationwide surveillance system for haemolytic–uraemic syndrome (HUS) in children <15 years old. Thirty paediatric nephrology units in public hospitals notify HUS cases on a voluntary basis to the National Institute of Public Health (Institut de Veille Sanitaire). The aim of this surveillance is to assess the incidence of HUS in children <15 years old, identify STEC serotypes associated with HUS, and detect clusters of HUS or STEC infections.

Between 1996 and 2003, the annual HUS incidence has been stable in France (mean 0·74/100 000 children <15 years old) with most cases being sporadic and the majority caused by *E. coli* O157 infection. Outbreaks due to *E. coli* O157 infection are rare in France and over the last 15 years, only one outbreak could be linked to a foodborne source (sausages) [3].

On 4 June 2004, routine surveillance for HUS identified two cases in a family living in the district of Vosges. An investigation was prompted to determine whether a community-wide outbreak had occurred and, if so, to identify the source of infection.

The two HUS cases were a boy (aged 13 months) and his sister (aged 3 years), who presented with diarrhoea (bloody for the boy) and no fever on 30 May 2004. Both developed HUS and required

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hospitalization on 2 June 2004 for haemodialysis and red blood cell transfusions. The outcome was favourable for the girl after 15 days in hospital. The boy presented renal sequelae after 2 months in hospital and still requires follow-up.

The mother was interviewed by phone, using a hypothesis-generating questionnaire designed to enquire about possible exposure (consumption of foods and drinks, contact with animals and or with persons with diarrhoea, etc.) in the week before onset of diarrhoea of the children with HUS. The interview revealed that the father had suffered from diarrhoea and abdominal cramps on the same day as his sick children and that all three had consumed fresh unpasteurized goats' cheese, 6 days before onset of illness. The mother and the third child, who had not consumed the cheese, did not become ill. The suspected cheese was purchased on 23 May 2004 from a dairy farm located in the district of Bas-Rhin. No other exposures (contact with goats or cattle or with their environment, etc.) were identified.

On 7 June 2004, the district veterinary inspection services visited the farm to examine milking, cheese-production processes and operational hygiene practices, in order to assess potential sources of contamination of the goats' cheese. The farm comprised 14 cows, 14 goats and two pigs, and produced 100–150 l/day cows' milk (14 kg unpasteurized cheese) and 38 l/day goats' milk (5 kg fresh unpasteurized cheese). Inspection of the dairy farm revealed that manual milking took place in the cattle shed and hygiene conditions for cheese preparation and assembly were inadequate. Approximately 45 raw goats' cheeses were produced daily, stored for 2 days at the farm, and sold directly to customers (40 home-delivered customers and one village market) or distributed locally to three inns and three food stores. The distribution zone was limited to a radius of 20 km, including 13 villages of the Vosges and Bas-Rhin districts. All food produced locally (butter, cows' milk, goats' milk, fresh unpasteurized goats' cheese, unpasteurized cows' cheese and ham), all animals (cows, goats and pigs) and water from the drinking trough, the dairy and the river were sampled for *E. coli* O157 (in total, 42 samples).

Active case-finding was performed in the community by contacting general practitioners, hospital emergency and paediatric units, as well as the customers of the implicated dairy farm and through national HUS surveillance, in order to identify patients with bloody diarrhoea between 20 May and 20 June 2004, living in the districts of Vosges or Bas-Rhin. No additional cases were identified.

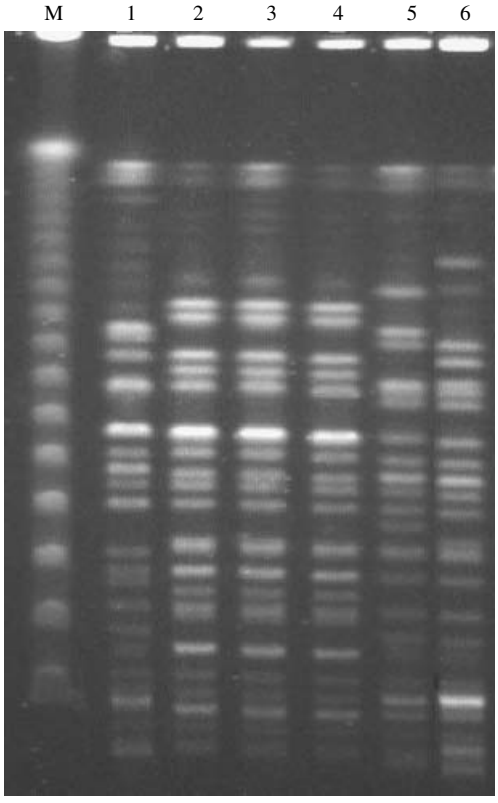

Fig. Pulsed field gel electrophoresis of genomic DNA of *E. coli* O157 strains digested with restriction enzyme *Xba*I, June 2004, France. M, lambda ladder; lane 1, strain CIP 105242; lane 2, cow's strain; lane 3, goat's strain; lane 4, HUS strain; lanes 5, 6, non-related strains.

Stool specimens of all family members, food, animal and environmental samples were cultured for the presence of *E. coli* O157:H7. All the isolates were examined for the presence of stx_1, stx_2 and *eae* by PCR [4, 5]. Pulsed-field gel electrophoresis (PFGE) of *Xba*I-restricted genomic DNA was used to establish clonal relation and diversity among the human and non-human *E. coli* O157 isolates. Patterns differing in up to four bands were considered related [6].

Human sera of all family members were tested for antibodies to the lipopolysaccharide (LPS) of seven major serogroups of STEC (O26, O91, O103, O111, O128, O145, O157) by line-blot immunoassay [7].

Among the five stool specimens from the family members, one *E. coli* O157 *stx2*+,*eae*+ strain was isolated from the 13-month-old child with HUS. *E. coli* O157 antibodies were detected only in the serum of the two HUS cases.

Two *E. coli* O157 *stx2*+,*eae*+ strains were isolated from faeces of one of the 14 cows (7%) and one of the

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14 goats (7%). All other 40 samples, including three samples from the goats' milk and two goats' cheeses, were O157:H7 negative.

All three *E. coli* O157 isolates obtained during the outbreak were subtyped with PFGE and were found to have patterns indistinguishable from one another (Fig.).

The unpasteurized cheese and raw milk production and sales were interrupted on 11 June 2004 and products were recalled from the three food stores and the village market. The three inns and the 40 home-delivered customers were informed.

All cheese produced in the weeks before the recall and stored on the farm were destroyed. Pasteurization of raw milk was recommended. In order to reduce the bacterial contamination of the farm environment, and to facilitate the renewal of farm activities, the owner was required to implement appropriate corrective measures: strict hygiene during milking, cheese production and cattle husbandry, use of tap water in the animals' water troughs, and separation of animals of different species.

The results of the present investigation indicate that this outbreak of *E. coli* O157 infection was caused by fresh unpasteurized goats' cheese from a single farm. Evidence was provided by the results of epidemiological, environmental and laboratory investigations.

This is the first outbreak of *E. coli* O157 infection linked to fresh unpasteurized goats' cheese in France. Before this outbreak, raw milk products have been suspected, but have never been confirmed as a source of *E. coli* O157 infection. In this investigation, no *E. coli* O157 was isolated from unpasteurized goats' cheese. Nevertheless, the link between consumption of raw milk products and disease has been well established for *E. coli* O157 in other countries [8–11] and transmission of bacterial pathogens, other than *E. coli* O157, by raw cheese has already been documented in France [12–14].

In France, faecal carriage of *E. coli* O157 in cattle is rare. *E. coli* O157 has been identified only once among 210 STEC isolates from 471 samples of cattle faeces [15] and to date, there are no published reports of *E. coli* O157 isolated from faeces of sheep and goats. The isolation of *E. coli* O157 with an indistinguishable PFGE pattern from two different species (cow and goat) in a single farm is unusual. This perhaps occurred because animals of different species were kept together in the same area where they shared water and food troughs, enhancing faecal–oral contact.

A contaminated environment, manual milking and insufficient basic hygiene practices suggested that cross-contamination could have occurred, either between unpasteurized milk and faeces at milking time, or possibly at some point during cheese preparation and/or assembly.

Although the farm produced 5 kg of fresh unpasteurized goats' cheese per day, no further cases were identified (in spite of active case-finding). The lack of further cases suggested that the contamination of cheese was temporally limited and that perhaps only a few unpasteurized goats' cheeses were contaminated.

French law regulates hygiene and microbiological requirements concerning the production and processing of raw milk products at farm level, but this does not address *E. coli* O157 (although inclusion of this is being considered in a new regulation currently being prepared).

This first outbreak of O157 infection linked to goats' cheese and the finding of faecal carrying among the farm animals should lead to increased awareness on the part of consumers of the risk of consuming raw milk products, especially if produced on small farms.

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DECLARATION OF INTEREST

None.

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