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Outbreaks of Enterohemorrhagic *Escherichia coli* O157 Infections among Children with Animal Contact at a Dairy Farm in Yokohama City, Japan

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On October 2, 2006, a clinic notified health and welfare center A of a case of enterohemorrhagic *Escherichia coli* (EHEC) O157:H7 (VT1 & 2) infection. The patient was a 1-year-old girl who developed diarrhea, fever, and hemorrhagic stool on September 26. The Yokohama City Institute of Health examined stool specimens from the four family members.

Stool specimens were inoculated on CT-SMAC agar (Oxoid, Hampshire, UK) for bacterial isolation. Polymerase chain reaction (PCR) (1) and RPLA (Denka Seiken, Tokyo, Japan) were carried out to examine the samples for the presence of Vero toxin.

EHEC O157:H7 (VT1 & 2) was also isolated from the patient's 9-year-old brother, but not from her mother, who developed diarrhea on September 27. This family had visited a dairy farm in Yokohama City on September 24 for an event that included activities such as milking a cow and handling animals (Fig. 1). The children participated in these events (Fig. 2). The stool samples from five cattle that were kept at the dairy farm were investigated. EHEC O157:H7 (VT1 & 2) was isolated from two cattle, one of which was used in the public cow-milking event on September 23.

The pattern of pulsed-field gel electrophoresis (PFGE) after digestion with *Xba*I was the same among two patient isolates and two bovine isolates (lanes 1-2 and 3-4, Fig. 3) (2,3). The results suggest that the outbreak was caused by a common EHEC O157 strain. The cow-milking event was discontinued as a result of the outbreak.

On October 18, health and welfare center B received a report that EHEC O157:H7 (VT1 & 2) was isolated from a 3-year-old girl. She developed abdominal pain and watery and hemorrhagic stool on October 13. The Yokohama City Institute of Health examined stool specimens from the three family members and identified O157:H7 (VT 1 & 2) in the stool of the patient's 11-month-old brother. Her latent period was calculated to be 20 days, which was longer than usual. The family of this 3-year-old girl had also visited the dairy farm on September 23 and participated in the event (Fig. 2). The pattern of PFGE after digestion with *Xba*I was same as that of the previous two patient isolates and two bovine isolates (lanes 5-6 and 3-4, Fig. 3).

A total of 123 stool specimens from children and staff members at a nursery school attended by two of the patients were examined, and all were negative for EHEC O157.

On October 16, 47 cattle at the dairy farm were examined



Fig. 1. Location of the dairy farm in Yokohama City.

Family No.	Age (y)	Sep.					Oct.																		
		23	24	25	26	27	28	29	30	1	2	3	4	5	6	13	14	15	16	17	18	19	20	21	
A	1			●	○																				
	2			●			△				×														
	3			●								△		×											
	4						○					△													
	5												△												
B	6			●													○	△						×	
	7			●																				△	×
	8			●																				△	
	9			●																				△	

Fig. 2. Summary of an outbreak of enterohemorrhagic *Escherichia coli* O157:H7 infections of two families. ○, Date of onset; △, Date of sampling; ×, Date of isolation from stool; ●, Date of participation in cow-milking event.

at the Kanagawa Prefectural Eastern Region Livestock Hygiene Station. O157:H7 (VT 1 & 2) strains were isolated from 11 cows. These 11 isolates were compared with those isolated from the 4 patients and 2 cattle mentioned above. The PFGE pattern of 10 of the 11 strains was the same as those of the 6 previous isolates. The results suggest that the outbreak was caused by an EHEC O157 strain from a cow used in the aforementioned public cow-milking event.

Akiba et al. reported a previous outbreak of EHEC derived from infected cattle on a dairy farm (4). The current study likewise emphasizes the importance of molecular epidemiological analysis by PFGE in epidemiological surveys.

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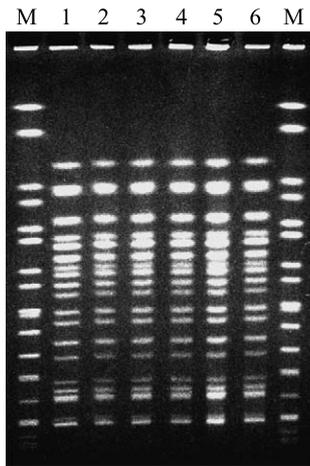


Fig. 3. PFGE profiles of EHEC O157:H7 digested with *Xba*I.
Lanes 1 to 2, nos. 1 and 3 in Fig. 2; lanes 3 to 4, strains isolated stools of cows; lanes 5 to 6, nos. 6 and 7 in Fig. 2; M, *Salmonella* Braenderup digested with *Xba*I.

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