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ORIGINAL CONTRIBUTIONS

Massive Outbreak of *Escherichia coli* O157:H7 Infection in Schoolchildren in Sakai City, Japan, Associated with Consumption of White Radish Sprouts

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In July 1996, an outbreak of *Escherichia coli* O157:H7 infection occurred among schoolchildren in Sakai City, Osaka, Japan. This outbreak developed in 13 North-East District and 34 Middle-South District elementary schools in the city. All children hospitalized on July 17–19 had presented on July 8 (North-East District) and July 9 (Middle-South District). School lunches served on July 1 and 8 in the North-East District and on July 1, 4, 8, and 9 in the Middle-South District were proposed by a food consumption study to be associated with infection. White radish sprouts from a single farm were the only uncooked food common to the most highly implicated meals on the involved days in two school districts (sweet and sour chicken with lettuce on July 8 in the North-East District and chilled Japanese noodles on July 9 in the Middle-South District). Two incidents of *E. coli* O157:H7 in neighboring areas were also related to white radish sprouts from the farm. The pulse-field gel electrophoresis patterns of isolates from patients in these two districts and the neighboring areas were identical. Thus, it was concluded that the cause of the outbreak was the white radish sprouts shipped on July 7–9 from one particular farm. *Am J Epidemiol* 1999;150:787–96.

disease outbreaks; enterocolitis; *Escherichia coli* infections; *Escherichia coli* O157; food handling; food inspection

Since a report from the United States in 1983 (1), many outbreaks of *E. coli* O157:H7 infection have been described, mainly in Canada, Great Britain, and the United States (2–12). Currently, there are about 20,000 infections and more than 100 deaths each year in the United States (13).

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Abbreviations: CI, confidence interval; OR, odds ratio; PFGE, pulse-field gel electrophoresis.

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In Japan between 1991 and 1995, incidences involving more than 10 patients were limited to four at child care centers and two at elementary schools (14).

Between May and December 1996, there were 9,451 cases and 12 deaths from *E. coli* O157:H7, representing 16 total outbreaks involving more than 10 patients each (7,900 patients), including the Sakai City incident. Sources of infection were school lunches served at elementary schools and child care centers (nine incidents, 7,470 patients), meals prepared at nursing homes (three incidents, 123 patients), a meal served at an industrial facility (one incident, 47 patients), a commercially prepared box lunch (one incident, 191 patients), and an unknown origin (two incidents, 69 patients).

Japanese elementary schools have a unique school lunch system: The same menu is prepared for all schools in an each area or each school, providing an opportunity for many children to be infected simultaneously. The teachers encourage children to eat every-

thing on the menu, so most of the food is consumed, making it very difficult to pinpoint the contaminated food item when an outbreak occurs.

The Ministry of Health and Welfare, along with the Bureau of Environment and Public Health of Osaka Prefecture and the Health Department of Sakai City, conducted a study of the mass infection in Sakai, which reportedly involved the largest number of individuals in history.

Late on the evening of July 12, 1996, the Health Department of Sakai City received a report from local medical facilities that a large number of elementary schoolchildren had developed conditions characterized by diarrhea and bloody diarrhea.

At the initial stage of the investigation, the school lunch appeared to be the most likely source of infection because 1) all patients identified by July 16 were schoolchildren or their teachers; 2) the area water supply was shared by neighboring cities; 3) the outbreaks in the schools were unrelated to the use of water tanks; 4) no major work on the water system had been done in early July; and 5) no irregularities were found in the residual chlorine concentrations in the water supply at any school. Sakai City is divided into three districts: the North-East, Middle-South, and Sakai-West. Each district prepares a common menu and purchases food for the district lunches, and the food is cooked in a kitchen at each school.

MATERIALS AND METHODS

Symptoms

On July 17–19, children hospitalized during that period (termed “hospitalized children”) were interviewed individually by food inspectors and public health nurses using a questionnaire to determine the day of onset, major symptoms, and the days on which the children were absent from July 1 to 10.

From July 22 to 27, nonhospitalized schoolchildren from the North-East, Middle-South, and Sakai-West Districts were interviewed individually by schoolteachers using the same form to determine the presence (or absence) of symptoms suggestive of *E. coli* O157:H7 infection (abdominal pain, diarrhea, fever, vomiting, or nausea). Children who reported one or more symptoms were defined as “symptomatic children,” and the day of onset and the symptoms were recorded. School records were also examined to determine the days on which the children were absent between July 1 and 10.

Food consumption

All schoolchildren were interviewed to analyze consumption of the individual items in the school lunches

between July 1 and 10. For each item served, the odds ratio and its 95 percent confidence interval were calculated.

Food distribution and preparation; water supply

The Sakai City Education Committee and the Sakai City School Lunch Association were asked to submit data on the distribution of foods used between July 1 and 10. Simultaneously, those employees who prepared food at some of the elementary schools in the North-East and Middle-South Districts were interviewed to document the cooking procedures followed during this period.

Information on the water supply system in Sakai City was collected from the Water Supply Office in the city government, and residual chlorine concentrations in the water supplies of individual schools were tested.

Laboratory investigations

E. coli O157:H7 strains isolated from the children (13 from the North-East and 18 from the Middle-South Districts) were analyzed by pulse-field gel electrophoresis (PFGE) at the National Institute of Health (15).

At the Sakai City Health Institute, samples preserved at each school between July 8 and 12 and food samples and smears collected from the facilities that supplied products used from July 1 to 10 were cultured for *E. coli* O157:H7 by an immunomagnetic separation method (16).

Some *E. coli* O157:H7 strains isolated in the outbreaks and sporadic cases in Osaka Prefecture between July 10 and 20 were also analyzed by PFGE at the National Institute of Health (15).

Environmental investigation

Epidemiologic studies confirmed white radish sprouts as the probable source of this infection. In late July and August, samples were collected from the well water, drainage from sprout production facilities, seed culture fluid, waterways surrounding the facilities, and drainage from animal quarters. These samples were cultured at the Institute of Public Health of Osaka Prefecture with an immunomagnetic separation method to isolate *E. coli* O157:H7 (16).

Laboratory investigation of *E. coli* O157:H7 was also performed at public health laboratories of the local governments on seeds shipped from the identified seed producer and from other seed producers at sprouting facilities in other prefectures.

Related cases

Information was obtained from health departments of Osaka Prefecture, Osaka City, and Kyoto City on

incidents and sporadic cases of an *E. coli* O157:H7 infection that occurred during the same period as the Sakai incident.

RESULTS

Symptoms

Symptoms and dates of onset. Hospitalized children included 86 (52 boys and 34 girls) from the North-East District and 312 (151 boys and 161 girls) from the Middle-South District. No children from the Sakai-West District were hospitalized.

All nonhospitalized elementary schoolchildren were also studied: 12,850 from the North-East District, 19,648 from the Middle-South District, and 15,145 from the Sakai-West District. There were 1,849 children (14.4 percent) in the North-East District, 6,376 (32.5 percent) in the Middle-South District, and 130 (0.9 percent) in the Sakai-West District with one or more symptoms between July 1 and the day of the interview (table 1).

Major symptoms of hospitalized and nonhospitalized children in the North-East and the Middle-South Districts were abdominal pain, diarrhea, and fever. Bloody diarrhea among those hospitalized occurred in 49 children (57.0 percent) in the North-East District and 159 children (51.0 percent) in the Middle-South District, while it occurred in 288 (15.6 percent) and 1,039 (16.3 percent) of symptomatic children, respectively. In Sakai-

West District, only four (3.1 percent) developed bloody diarrhea. Major symptoms were frequent among hospitalized children, and rates were similar in the North-East and Middle-East Districts (table 2).

Study of the hospitalized children showed that the number of patients began to increase on July 10. In the North-East District, 15 children (17.4 percent) had a July 10 onset, compared with only 12 children (3.8 percent) in the Middle-South District. The peak increase in patients occurred in the North-East District on July 11 and in the Middle-South District on July 12, a time difference of 1 day. In nonhospitalized schoolchildren, the number of children with symptoms also began to increase on July 10. Those with symptoms and onset on July 10 represented 8.6 percent (154 children) of those with symptoms in the North-East District and 2.7 percent (158 children) in the Middle-South District. Symptomatic children with onset by July 12 involved 60.7 percent (1,084 children) in the North-East District and 46.6 percent (2,762 children) in the Middle-South District. As with the hospitalized children, a certain time difference was noted, but peak occurrence in symptomatic children was recorded on July 12 in both districts (figure 1).

In the Sakai-West District, the total number of children who developed symptoms between July 1 and 27 was 130 (0.85 percent). By July 16, no *E. coli* O157:H7 was isolated from any of them.

The Sakai-West District differed from the North-East or Middle-South Districts in the following epidemiologic features: the number of children with symptoms was markedly lower, symptom patterns (such as frequency of diarrhea and the condition of the stool) differed, and the number of symptomatic children was within the normal range. According to the Sakai City Educational Committee, the average number of children who exhibit some symptoms is between 1 and 2 percent daily throughout the year. There has been no mass incidence of diarrhea in this district.

TABLE 1. Number of symptomatic and hospitalized children in each district of Sakai City, Japan, July 1996

	North-East		Middle-South		Sakai-West	
	No.	%	No.	%	No.	%
Total	12,850	100.0	19,648	100.0	15,145	100.0
Hospitalized	86	0.7	312	0.4	0	0.0
Symptomatic	1,849	14.4	6,376	32.5	130	0.9

TABLE 2. Major symptoms reported by hospitalized and symptomatic children in each district, Sakai City, Japan, July 1996

	North-East				Middle-South				Sakai-West (symptomatic)	
	Hospitalized		Symptomatic		Hospitalized		Symptomatic		No.	%
	No.	%	No.	%	No.	%	No.	%		
Total	86	100.0	1,849	100.0	312	100.0	6,376	100.0	130	100.0
Abdominal pain	85	98.8	1,640	88.7	304	97.4	5,407	84.8	91	70.0
Diarrhea	83	96.5	1,631	88.2	307	98.4	5,388	84.5	96	73.8
>12/day	34	39.5	264	14.3	139	44.6	805	12.6	4	3.1
Bloody	49	57.0	288	15.6	159	51.0	1,039	16.3	4	3.1
Fever (>37°C)	69	80.2	802	43.4	251	80.4	2,737	42.9	63	48.5
Nausea	36	41.9	356	19.3	151	48.4	1,166	18.3	25	19.2
Vomiting	31	36.0	246	13.3	146	46.8	813	12.8	19	14.6

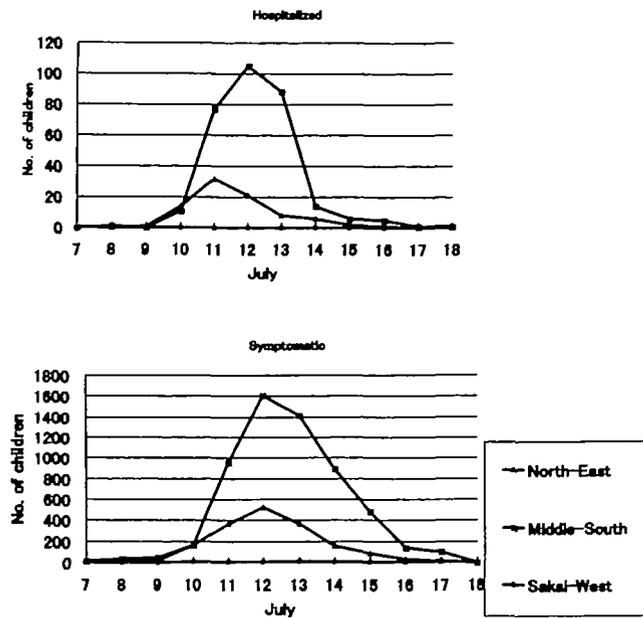


FIGURE 1. Date of onset of illness in children from the North-East, Middle-South, and Sakai-West districts, Sakai City, Japan, July 7-17, 1996.

Hospitalized children were from approximately one half of the schools (13 of 25 schools, 52.0 percent) in the North-East District and almost all of the schools (34 of 35 schools, 97.1 percent) in the Middle-South District.

Absenteeism of hospitalized and symptomatic children. None of the hospitalized children were absent from school on July 8 in the North-East District and on July 9 in the Middle-South District.

Symptomatic children absent during the same period were fewest on July 8 in the North-East District, and in the Middle-South District, the fewest number of symptomatic children were absent on July 9 (table 3). Those who were absent on those days were questioned individually, but it was not possible to determine from the symptoms and other information that any in these districts had been infected by *E. coli* O157:H7.

Food consumption

In the North-East District, the foods thought to be associated with children who were later hospitalized are listed in descending order: milk and sweet and sour chicken with lettuce served on July 8 (odds ratio (OR) = ∞); milk served on July 1 (OR = 4.7, 95 percent confidence interval (CI): 1.1, 19.0); and potatoes and beef served on July 1 (OR = 4.6, 95 percent CI: 1.1, 18.9) (table 4).

In the Middle-South District, foods thought to be associated with children who were later hospitalized are listed in descending order: milk served on July 9 (OR = 6.7, 95 percent CI: 1.7, 27.2); chilled Japanese noodles served on July 9 (OR = 3.3, 95 percent CI: 1.2, 8.8); chilled Chinese noodles served on July 4 (OR = 2.5, 95 percent CI: 1.2, 5.3); milk served on July 8 (OR = 2.3, 95 percent CI: 1.0, 5.2); curry rice served on July 1 (OR = 2.0, 95 percent CI: 1.1, 3.7); and milk served on July 1 (OR = 1.8, 95 percent CI: 1.0, 3.1) (table 5).

Food distribution and preparation; water supply

In Sakai City, foods purchased for each district are brought to a distribution center, transported to each elementary school by eight trucks, and cooked in a kitchen at the school. The menus in the three districts sometimes differed, were sometimes shared by two of the three districts, or might be uniform throughout the three districts. Milk, bread, eggs, and rice were delivered by registered dealers directly to each school kitchen.

The distribution center to which the foods were delivered from wholesalers and the trucks used for delivery to each school had no freezing or refrigeration equipment. Other than a refrigerator to store milk and food samples, the cooking area at the school had no refrigerators or freezers.

No inspections or spot-checks were made of hygienic conditions when foods were delivered to either the distribution center or the school kitchens.

TABLE 3. Absenteeism in hospitalized and symptomatic children between July 1 and July 10, 1996, in Sakai City, Japan

	Date of absenteeism							
	July 1	July 2	July 3	July 4	July 5	July 8	July 9	July 10
Hospitalized								
North-East	2	1	6	2	4	0	4	4
Middle-South	9	7	5	5	11	2	0	2
Symptomatic								
North-East	36	21	65	19	41	11	43	18
Middle-South	216	164	147	90	147	61	18	35

TABLE 4. Correlation of recorded consumption among schoolchildren in the North-East District, Sakai City, Japan, July 1996

Date and menu	Hospitalized		Healthy		Odds ratio	95% confidence interval
	Exposed	Unexposed	Exposed	Unexposed		
July 1						
French roll	75	7	9,941	1,189	1.3	0.6, 2.8
Milk	81	2	9,520	1,097	4.7	1.1, 19.0
Potatoes and beef	79	2	9,493	1,113	4.6	1.1, 18.9
Vinegared dish	75	6	9,392	1,196	1.6	0.7, 3.7
Soybean butter	71	9	9,149	1,368	1.2	0.6, 2.4
July 2						
French roll	82	2	10,078	531	2.2	0.5, 8.8
Milk	82	2	10,156	461	1.9	0.5, 7.6
Cold Chinese noodles	82	1	10,137	466	3.8	0.5, 27.1
Watermelon	81	3	10,010	589	1.6	0.5, 5.0
Mixed nuts	75	8	9,653	852	0.8	0.4, 1.7
July 3						
Curry rice	78	6	10,063	678	0.9	0.4, 2.0
Milk	77	7	10,021	715	0.8	0.4, 1.7
Salad	75	8	9,913	805	0.8	0.4, 1.6
Pickled vegetables	73	10	9,924	793	0.6	0.3, 1.1
July 4						
Bread	80	3	10,013	594	1.6	0.5, 5.0
Milk	81	2	10,091	525	2.1	0.5, 8.6
French fries	80	2	10,117	498	2.0	0.5, 8.0
Fried squid	80	3	10,039	567	1.5	0.5, 4.8
Minestrone	74	6	9,901	653	0.8	0.4, 1.9
July 5						
Sushi	79	4	10,014	590	1.2	0.4, 3.2
Milk	77	6	10,063	567	0.7	0.3, 1.7
Soup	78	4	10,044	553	1.1	0.4, 2.9
Fruit gelatin	79	4	10,042	567	1.1	0.4, 3.1
July 8						
Bread	80	2	10,151	459	1.8	0.4, 7.4
Milk	84	0	10,212	404	∞	∞
Chicken with lettuce	79	0	10,096	460	∞	∞
Noodle soup	78	1	10,180	403	3.1	0.4, 22.3
July 9						
Bread	79	4	9,969	647	1.3	0.5, 3.5
Milk	80	4	10,040	583	1.2	0.4, 3.2
Curry stew	79	5	10,049	568	0.9	0.4, 2.2
Watermelon	78	6	9,974	643	0.8	0.4, 1.9
July 10						
French roll	77	6	10,096	515	0.7	0.3, 1.5
Milk	80	4	10,175	443	0.9	0.3, 2.4
Cooked pumpkin	75	8	10,065	529	0.5	0.2, 1.0
Soup	78	4	10,159	431	0.8	0.3, 2.3
Boiled young beans	78	4	10,122	482	0.9	0.3, 2.5

The Sakai City School Lunch Association had not required registered dealers to submit reports on periodic voluntary inspections for hygiene control.

The kitchen personnel were questioned about the cooking procedures on July 1–10. These had generally

been done in accordance with the specifications given in their manual. Meat, pre-cut into small pieces, was placed in plastic containers and delivered to each kitchen. It was defrosted in the preprocessing room that is set aside in the cooking area.

TABLE 5. Correlation of recorded consumption among schoolchildren in the Middle-South District, Sakai City, Japan, July 1996

Date and menu	Hospitalized		Healthy		Odds ratio	95% confidence interval
	Exposed	Unexposed	Exposed	Unexposed		
July 1						
Curry rice	294	11	11,197	844	2.0	1.1, 3.7
Milk	273	13	11,109	928	1.8	1.0, 3.1
Salad	272	31	10,827	1,178	1.0	0.7, 1.4
Pickled vegetable	275	29	10,905	1,115	1.0	0.7, 1.4
July 2						
French roll	283	19	11,167	854	1.1	0.7, 1.8
Milk	292	11	11,303	727	1.7	0.9, 3.1
Cooked pumpkin	273	27	10,965	1,028	0.9	0.6, 1.4
Soup	288	12	11,242	750	1.6	0.9, 2.9
Cooked young beans	279	22	11,106	918	1.0	0.7, 1.6
July 3						
French roll	291	13	11,252	772	1.5	0.9, 2.7
Milk	291	10	11,461	573	1.5	0.8, 2.7
French fries	293	9	11,531	511	1.4	0.7, 2.8
Fried squid	279	23	11,347	680	0.7	0.5, 1.1
Minestrone	262	36	10,986	927	0.6	0.4, 0.9
July 4						
French roll	287	16	11,314	721	1.1	0.7, 1.9
Milk	296	9	11,414	626	1.8	0.9, 3.5
Cold Chinese noodles	297	7	11,351	673	2.5	1.2, 5.3
Watermelon	267	18	11,211	827	1.1	0.7, 1.8
Mixed nuts	267	36	10,849	1,153	0.8	0.6, 1.1
July 5						
Sushi	274	20	11,122	862	1.1	0.7, 1.7
Milk	284	15	11,241	780	1.3	0.8, 2.2
Soup	280	16	11,208	780	1.2	0.7, 2.0
Fruit gelatin	276	20	11,215	801	1.0	0.6, 1.6
July 8						
French roll	289	14	11,376	644	1.2	0.7, 2.0
Milk	298	6	11,487	538	2.3	1.0, 5.2
Japanese stew	292	9	11,428	556	1.6	0.8, 3.1
Pickled cucumbers	278	22	11,187	786	0.9	0.6, 1.4
Minitomato	271	31	11,075	947	0.7	0.5, 1.1
July 9						
French roll	291	12	11,421	598	1.3	0.7, 2.3
Milk	303	2	11,520	513	6.7	1.7, 27.2
Cold Japanese noodles	300	4	11,509	501	3.3	1.2, 8.8
Saute with sausage	294	8	11,441	574	1.8	0.9, 3.7
July 10						
Bread	287	16	11,475	538	0.8	0.5, 1.4
Milk	297	7	11,583	438	1.6	0.8, 3.4
Chicken with lettuce	280	20	11,286	639	0.8	0.5, 1.3
Noodle soup	281	19	11,479	485	0.6	0.4, 1.0

It was understood that kitchen personnel washed their hands and used antiseptic before the start of the shift. Sinks are located in the toilets, the preprocessing room, and the cooking area and are equipped with soap, nail brushes, and antiseptic. Different cutting

boards are used for food to be heated and food served without heating.

Pasteurization records kept at the two milk processing facilities confirmed that the milk had been pasteurized. Milk deliveries confirmed were unrelated to the

incidence of the infection. Investigation confirmed that milk from the same processing facilities was also consumed at the Sakai-West District, where no infection was found.

Examination of food consumption listed on menus uncooked food items (other than milk) whose lower 95 percent confidence interval was more than 1.0. In the North-East District, there was no uncooked food in the potato and beef served on July 1, but lettuce and white radish sprouts were in the sweet and sour chicken with lettuce served on July 8. In the Middle-South District, the chilled Japanese noodles served on July 9 contained fish paste, cucumber, and white radish sprouts; the chilled Chinese noodles served on July 4 contained cucumber; and the curry rice served on July 1 contained no uncooked food.

The suspect white radish sprouts were produced by a single farm and were used in the North-East District on July 8 and in the Middle-South District on July 9. They were also included in the chicken with lettuce served on July 10 in the Middle-South District. It was confirmed that three of the four hospitalized patients who did not consume the chilled Japanese noodles on the July 9 had eaten the chicken with lettuce on July 10.

The white radish sprouts were delivered on July 5 and 7 to be served for lunch on July 8 at the North-East District, those for the lunch served at the Middle-South District on July 9 were shipped on July 8 and 9, and those for the lunch served on July 10 at the Middle-South District were shipped on July 9 and 10.

For the school lunches served in Sakai City between July 1 and 11, white radish sprouts were used seven times. In addition to July 8–10, sprouts were served on July 3 in the Sakai-West District and on July 11 in the Middle-South, North-East, and Sakai-West Districts. The white radish sprouts used in these districts on these dates had been shipped by farmers other than the one responsible for the shipments on the July 8–10.

The area water supply in Sakai City is shared by neighboring cities, and the hospitalized children were both from schools with water tanks and those with water supplied directly from the city system. No major work on the water system had been done in early July, and no irregularities were found in the residual chlorine concentrations in the water supply at any school.

Laboratory tests

The number of hospitalized children from whom *E. coli* O157:H7 was isolated was 129 (32.4 percent). Thirty-one isolates from children in the North-East and the Middle-South Districts were analyzed by PFGE, and all patterns were identical (15).

Tests on 1,626 food and other samples (mainly meat and vegetables served in school lunches at Sakai City between July 1 and 10) found no *E. coli* O157:H7. These included 950 food samples related to school lunches served between July 1 and 10 and 676 samples collected from cooking utensils, drinking water, and drainage water from the school kitchens and meat processing plants. A total of 226 other samples that had been preserved at each school from lunches served between July 8 and 12 also found no *E. coli* O157:H7.

Environmental investigation

Samples were collected from the farm that shipped the white radish sprouts consumed in the North-East District on July 8 and in the Middle-South District on July 9 and 10. Tests were conducted on 78 samples; 14 collected on July 24 included samples of well water, sewer water, seeds, culture fluid of seeds, and white radish sprouts. On August 8, 64 additional samples were tested (including fecal samples from employees). No *E. coli* O157:H7 was isolated from these samples.

Tests were extended to include water from rivers, streams, and waterways around the farm, fecal matter of cattle reared in the areas along the rivers, and sewage from the animal quarters. A total of 214 samples were examined, but no *E. coli* O157:H7 was isolated.

The seeds of white radish sprouts were then examined. Those used in early July had been imported from North America in January 1996 and delivered to the facility on June 8. Tests were conducted on the two seed samples used in early July at the suspected farm. An additional 11 seed samples imported at the same time but collected from other seed producers were tested. No *E. coli* O157:H7 was detected, but another serotype of *E. coli* was found in one.

Related cases

It was confirmed that the identified farm shipped a total of 24.6 tons of white radish sprouts to seven prefectures between July 1 and 15.

Outbreak at a nursing home in Habikino City, in Osaka prefecture. Almost simultaneously with the outbreak of diarrhea in schoolchildren in Sakai, an incident of *E. coli* O157:H7 infection was reported at a nursing home in Habikino City, near Sakai. The number of patients with symptoms began to increase on July 13 and peaked on July 15.

Ninety-eight individuals with symptoms were reported between July 6 and 24. The only meal shared by the 33 individuals who were *E. coli* O157:H7 positive, including 12 without symptoms, was a lunch served on July 9 (beef curry, a white radish sprout salad, and pickled scallions). The white radish sprouts

used in this salad had been shipped on July 7 from the same farm that was associated with the Sakai outbreak (the same sprouts were consumed in the North-East District on July 7). The PFGE patterns of the six strains of *E. coli* O157:H7 isolated from patients in this nursing home were analyzed at the National Institute of Health; all matched those isolated from the schoolchildren of Sakai City (15).

Incident at a business office in Kyoto. *E. coli* O157:H7 infection occurred at about the same time at a large business office in Kyoto. The number of symptomatic patients began to increase on July 15 and peaked on July 17. *E. coli* O157:H7 was isolated from five of the 47 individuals who developed symptoms between July 15 and 22.

The lunch on July 11 or 12 was the most likely cause of the infection according to an investigation of food consumption. The white radish sprouts used in the salad served at lunch on July 11, which had been shipped to Sakai's Middle-South District, had been shipped on July 9 by the farm in question. The PFGE pattern of the *E. coli* O157:H7 isolated from symptomatic individuals in Kyoto matched that of the isolates from the schoolchildren in Sakai (15).

Sporadic incidences in Osaka prefecture on July 10–20. In addition to those at the nursing home, 157 symptomatic individuals with positive identification of *E. coli* O157:H7 were found in Osaka Prefecture between July 10 and 20. The incidence reached a peak on July 15. It was an unusual increase compared with number of patients in June.

The PFGE patterns of the 63 strains of *E. coli* O157:H7 isolated from these 157 symptomatic individuals were analyzed at the National Institute of Health. The PFGE patterns of 56 strains (88.9 percent) matched those isolated from the schoolchildren in Sakai (15).

DISCUSSION

In this outbreak of *E. coli* O157:H7 infection, only schoolchildren in Sakai and their teachers developed symptoms within 1 week of the initial discovery of the infection. Contamination of the water supply was ruled out.

We concluded that the white radish sprouts shipped by a specific farm on July 7–9 were associated with the Sakai school outbreak. The following evidence supports this conclusion.

First, white radish sprouts from a single farm were the only uncooked food in common in the most highly implicated meals in two different school districts (sweet and sour chicken with lettuce served on July 8 (OR = ∞) in the North-East District and chilled Japanese noodles served on July 9 (OR = 3.3, 95 per-

cent CI: 1.2, 8.8) in the Middle-South District. Second, outbreaks that occurred in neighboring areas at about the same time were also linked to white radish sprouts shipped by the same farm on July 7 and 9. The PFGE patterns of the *E. coli* O157:H7 isolated from the symptomatic individuals matched those isolated from the schoolchildren (15). Third, the 1-day lag in consumption of the presumed causative food item in the North-East and Middle-South Districts coincides with the 1-day difference in the peak onset of illness in these two districts.

There were fewer schools with children requiring hospitalization in the North-East District than in the Middle-South District. White radish sprouts served on July 8 in the North-East District were shipped on July 5 and 7. Although it could not be confirmed, it is possible that the white radish sprouts shipped on July 5 from the farm were not contaminated by *E. coli* O157:H7.

According to the study of hospitalized children in this outbreak, the mean incubation period was 3.6 days, and the median was 3 days (range, 2–9 days) in the North-East District. In the Middle-South District, the mean incubation period was 3.2 days, with a median of 3 days (range, 2–9 days). These are within the ranges cited in the United States (1, 2, 4–6), but appear to be slightly shorter, perhaps because the patients were schoolchildren.

Examination of the school lunches showed more than 90 percent consumption of most meals by both the healthy and the hospitalized children. This is because the school lunch program is considered to be part of the educational system in Japan, and teachers strongly urge that children eat everything.

In the investigation of the production, processing, and distribution of food products, no *E. coli* O157:H7 was isolated from meat, raw vegetables, related facilities, or the trucks used to transport the food materials from the distribution center to the schools. Because the Sakai City School Lunch Association has not required that the dealers submit the results of voluntarily conducted tests concerning hygienic control of the products and the foods were not inspected when delivered, it was not possible to confirm the hygienic conditions at the time of the infection.

Except in the case of milk, the distribution center, trucks for transport, and distribution routes to the school kitchens are not equipped with refrigeration or freezing equipment, so it must be a major contributing factor in this outbreak.

At the National Institute of Hygienic Sciences and the National Institute of Health, tests were conducted on white radish sprouts that were inoculated with *E. coli* O157:H7 and left standing at 30°C for 5 hours.

These tests confirmed that the organism can proliferate under these conditions (Konuma et al., unpublished data). It was also confirmed that *E. coli* O157:H7 can proliferate on shredded lettuce (17).

No serious problem was noted in handling of the food material in the kitchen facilities at any of the schools investigated. Meat was processed separately from other food; pre-cut meat was placed in plastic containers so it would be unlikely to contaminate other food. At all of the schools investigated, adequate heat was used in cooking, as specified in the manual; there was little possibility that inadequate heat was used during the cooking process.

It is also highly improbable that human errors (such as insufficient heating or inadequate handling of meat) would occur simultaneously in all 47 schools where outbreaks occurred; therefore, unheated food materials were the most probable cause of the infection.

In addition, we found that no children later hospitalized had been absent on July 8 in the North-East District or on July 9 in the Middle-South District. The study of all children showed that absenteeism by symptomatic children was also lowest on these days. Therefore, the lettuce and white radish sprouts served on July 8 in the North-East District and the fish paste, cucumber, and white radish sprouts served on July 9 at the Middle-South District were identified as the food sources through which the causative organism most probably was transmitted.

Next, the possibility of contamination of the white radish sprouts during the production process was investigated. According to an experiment conducted at the National Institute of Hygienic Sciences with white radish sprouts grown hydroponically in *E. coli* O157:H7-contaminated water, contamination spreads to the upper part of the plant (18). A number of studies conducted in Japan indicated that in comparison with other vegetables, white radish sprouts are contaminated with greater numbers of bacterial species and coliform bacteria.

This investigation pointed to white radish sprouts as the probable causative food. In the United States as well, foods with a plant origin, such as salad and apple cider, were implicated in 19 percent of the 75 outbreaks that developed between 1982 and 1995. In particular, lettuce was considered responsible for the three outbreaks in 1995 (13).

Salmonella infections in the United States and Finland in 1995 were caused by contamination of alfalfa seeds (19). During investigation of the present outbreak, *E. coli* (not O157:H7) was isolated from the seeds of white radish sprouts. This indicates the need for further studies of the ability of the *E. coli* O157:H7 strain to survive on white radish seeds.

Vegetables other than white radish sprouts and fruits have also been implicated. *E. coli* O157:H7 infections due to contaminated apple cider (4, 7) and lettuce (13, 20) have been reported in the United States. In Japan, salad has been involved in outbreaks in an elementary school at Gifu City in June, in an elementary school in Morioka City in September, and in a kindergarten at Obihiro City in November, all in 1996.

E. coli O157:H7 has also been isolated in other vegetables and fruits (21). It is essential to establish adequate policies for control of the production, distribution, and cooking of foods other than meat and milk, i.e., foods with a plant origin.

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