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Food Poisoning Caused by Mannitol Nonfermenting *Staphylococcus aureus*: Case Report

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On August 4, 2005, 20 people who participated in the closing ceremonies of a community festival experienced nausea, vomiting, diarrhea and abdominal pain. The community where the festival took place was under the jurisdiction of the Aizu Health Center of Fukushima Prefecture. The foods that all of these patients had consumed were rice balls prepared by caterer A. Another 8 people, who did not participate in the festival but who had ingested the rice balls, also became ill.

The Aizu Branch of the Fukushima Institute of Public Health examined 3 specimens of the remaining rice balls, 6 finger swab specimens from the cooks, 6 kitchen utensil swab specimens, 6 stool specimens from the patients with symptoms and 5 stool specimens from the cooks. Additionally, the Niigata City Institute of Public Health and the Niigata Prefectural Instiute of Public Health and Environmental Sciences examined one stool specimen from each of the other patients. The Aizu Branch isolated 7 *Staphylococcus aureus* strains and the Niigata City Institute of Public Health isolated 2 strains. The characteristics of the 9 isolates are summarized in Table 1. Bacterial identification was conducted using an ID test SP-18 (Nissui Pharmaceutical Co., Ltd., Tokyo, Japan), a coagulase test for Staphylocoagulase antisera (Denka Seiken, Co., Ltd., Tokyo, Japan) and the detection and typing of staphylococcal enterotoxin by reversed passive latex agglutination (RPLA) for types A - D (Denka

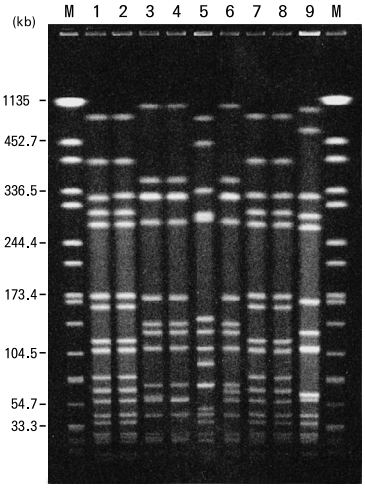

Fig. 1. Pulsed-field gel electrophoresis of *Sma*I digests of the chromosomal DNAs of *S. aureus* isolates. Lane 1, rice ball (pickled plum); 2, rice ball (seaweed); 3, fried food; 4, swab specimen from an employee (caterer A shop B); 5, swab specimen from an employee (caterer A shop B); 6, swab specimen from the handle of the refrigerator (caterer A shop B); 7, swab specimen from a cook (caterer A); 8-9, patient's stool specimen (Niigata City Institute of Public Health); the same patient. M, molecular size marker (*Salmonella* Braenderup strain H9812, *Xba*I digest).

Table 1. Characteristics of *S. aureus* isolated

		Enterotoxin	Coagulase	Colony characters on egg yolk plates	
				egg yolk reaction	mannitol fermentation
1	rice ball (pickled plum)	A	IV	+	−
2	rice ball (seaweed)	A	IV	+	−
3	fried food	−	III	+	+
4	swab specimen from an employee (caterer A shop B)	−	III	+	+
5	swab specimen from an employee (caterer A shop B)	−	UT	+	+
6	swab specimen from the handle of the refrigerator (caterer A shop B)	−	III	+	+
7	swab specimen from a cook (caterer A)	A	IV	+	−
8*	patient's stool specimen (Niigata City Institute of Public Health)	A	IV	+	−
9*	ditto	D	II	+	+

*: 8,9, the same patient. UT, untypable.

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Seiken) and by polymerase chain reaction (PCR) for types A - E (Takara Bio, Otsu, Japan).

Samples that were positive for *S. aureus* for type IV coagulase and enterotoxin A were isolated from pickled plum and seaweed rice balls, a finger swab specimen from a cook and a stool specimen examined at the Niigata City Institute of Public Health. The strain grew as a nearly pure culture on egg yolk isolation plates. The colonies were not typical for *S. aureus* in that, though positive for the egg yolk reaction, they were only faintly yellow and negative for mannitol fermentation.

The 9 isolates listed in Table 1 were submitted to pulsed-field gel electrophoresis (PFGE) of *Sma*I digests of chromosomal DNAs (Fig. 1). The conditions for electrophoresis were the following: pulse time, 5.3 - 34.9 sec; voltage, 6V/cm; and duration, 19 h. Two isolates from the rice balls (lanes 1 and 2), one isolate from a finger swab from the cook and one isolate from a patient's stool specimen examined at the Niigata City Institute of Public Health showed the same pattern. We therefore assume that the rice balls were responsible for the food poisoning.

Mannitol nonfermenting colonies are often discarded as non-staphylococcal colonies. In the present case, the nearly pure culture of the colonies led us to suspect them as the causative agent. It is important to keep in mind that biochemical markers should not be relied upon too heavily in the laboratory identification of *S. aureus*, especially where atypical phenotypes are involved. Massive growth as a pure culture such as that in the present case should be taken as an indication for further investigation.

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