

## Laboratory and Epidemiology Communications

### Molecular Epidemiology of Methicillin-Resistant *Staphylococcus aureus* in a Sendai Hospital in 2003

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Methicillin-resistant *Staphylococcus aureus* (MRSA) is a major hospital-acquired pathogen. Molecular epidemiological is essential for assessing hospital infection control measures (1-7).

Forty-five MRSA isolates were obtained from 24 inpatients during October 2003 in a hospital with 15 wards and 650 beds in Sendai city. Of these, 22 isolates, each derived from a single patient, were analyzed in terms of the following: chromosomal DNA typing with a contour-clamped homogeneous electric field system (CHEF Mapper™: Bio-Rad Laboratories, Hercules, Calif., USA), antibiotic resistance (disk diffusion tests by the National Committee for Clinical Laboratory standards method), enterotoxin serotyping (SET-RPLA™: Denka Seiken Co., Tokyo), toxic shock syndrome toxin-1 (TSST-1) production (TST-RPLA™: Denka Seiken), and coagulase serotyping (Denka Seiken).

Thirteen different pulsed-field gel electrophoresis (PFGE) patterns of *Sma*I DNA digests were detected (Fig. 1). A band-based cluster analysis (Molecular Analyst™: Bio-Rad), in which PFGE-band similarity exceeding 70% was used as the criterion of cluster formation, revealed the three following clusters, A, AV/AU, and BH (Fig. 2A). The frequency distribution of the PFGE patterns is shown in Fig. 2B. Pattern AV5 was shared by six isolates, pattern A3 by four, and pattern A4 by two. The other 10 PFGE patterns appeared in only a single isolate.

As shown in Table 1, pattern AV5 was spread over five wards (e6, w2, w3, w4, and w6). Three isolates from the neonatal intensive care unit (NICU) were of pattern A3, and two from wards e6 and s6 were of pattern A4. Other patterns were unique to each ward.

The isolates showed a wide spectrum of drug resistance, with a total of 12 different patterns (Table 2). The isolates were resistant to 7-12 of 15 tested drugs. All of the isolates

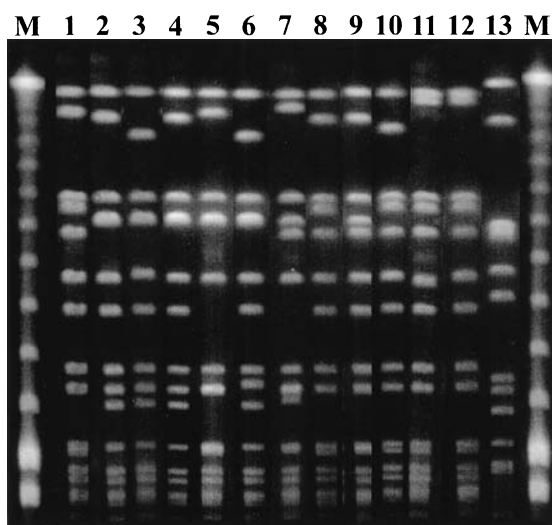


Fig. 1. Pulsed-field gel electrophoresis of *Sma*I-digested genomic DNA from MRSA isolates. M: low range PFG Marker. Lanes 1 to 13: MRSA isolates with different PFGE patterns A3 to BH shown in Fig. 2.

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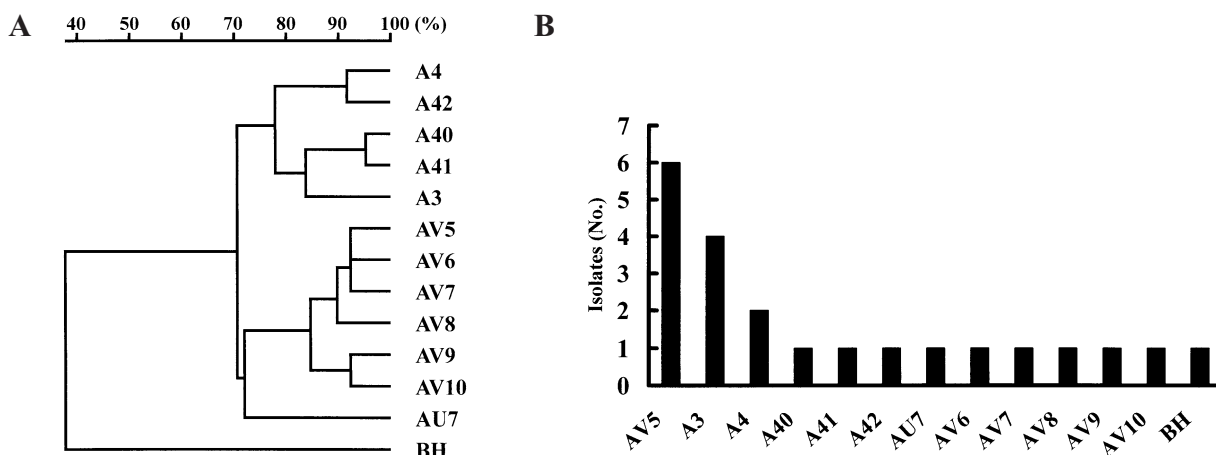


Fig. 2. Cluster analysis of MRSA isolates based on PFGE patterns of *Sma*I-digested genomic DNA.

Table 1. Distribution of MRSA in a hospital

PFGE pattern	Ward						pediatrics	NICU	Critical care center
	e		w		s				
	2	3	4	5	6	5			
A2							1		
A3			1#					3	
A4				1			1		
A40					1				
A41						1			
A42			1						
AV5			1	1	1	2	1		
AV6			1						
AV7					1				
AV8					1				
AV9					1				
BH					1				

#: Number of patients with MRSA.

Table 2. Antibiotic pattern classified by antibiotic pattern of 15 antibiotics against MRSA

Antibiotic pattern	Antibiotics								
	LVFX	GM	TC	MINO	AMK	ABK	ST	VCM	
a	R	R	R	R	R	S	S	S	
b	R	R	R	I	R	S	S	S	
c	R	R	S	R	R	S	S	S	
d	R	I	R	R	R	S	S	S	
e	R	R	R	R	I	S	S	S	
f	R	R	R	I	I	S	S	S	
g	R	I	R	I	R	S	S	S	
h	R	I	R	R	I	S	S	S	
i	R	I	R	I	I	S	S	S	
j	R	I	I	S	R	S	S	S	
k	R	I	S	S	R	S	S	S	
l	S	R	S	S	R	I	S	S	

All the isolates were resistant to MIPIC, ABPC, CEZ, FMOX, IPM, EM, CLDM.  
 MIPIC: oxacillin, ABPC: ampicillin, CEZ: cefazolin, FMOX: flomoxef,  
 IPM: imipenem/cilastatin, EM: erythromycin, CLDM: clindamycin,  
 LVFX: levofloxacin, GM: gentamicin, TC: tetracycline, MINO: minocyclin,  
 AMK: amikacin, ABK: arbekacin, ST: sulfamethoxazole/trimethoprim,  
 VCM: vancomycin, R: resistant, S: susceptible, I: intermediate.

were sensitive to sulfamethoxazole/trimethoprim and vancomycin. No antibiotic resistance pattern was found to be specific to a particular ward. No correlation was found between antibiotic patterns and PFGE patterns (data not

shown).

Among 22 MRSA isolates, 21 produced coagulase type II, and the remaining isolate produced coagulase type I. Eleven isolates produced enterotoxin type C, nine isolates entero-

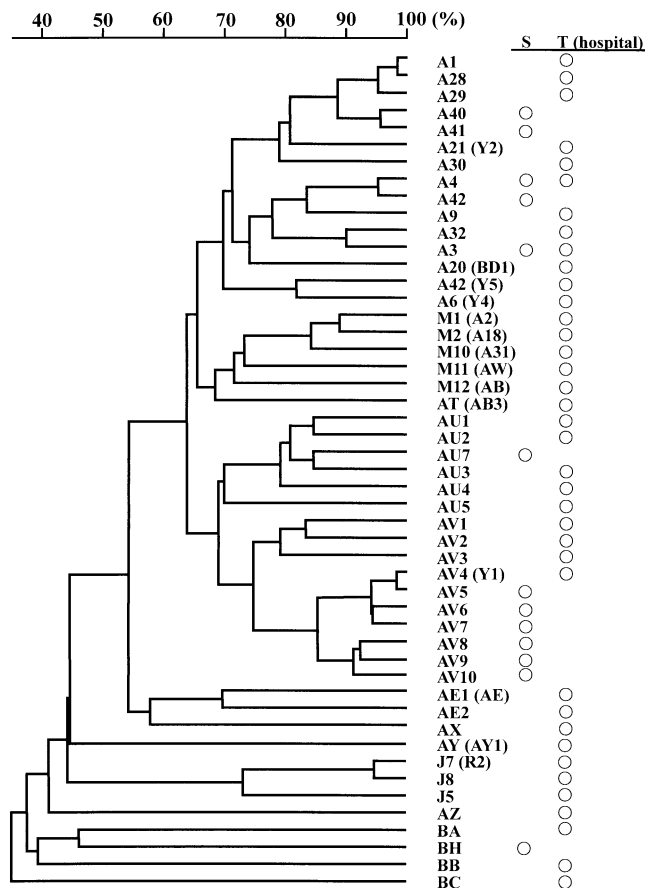


Fig. 3. Cluster analysis of MRSA isolates based on PFGE patterns. S: Sendai hospital, T: Tokyo hospital.

toxin type B, one isolate enterotoxin type D, and one isolate produced no enterotoxins. Twelve isolates produced TSST-1. Eleven isolates produced coagulase type II, enterotoxin type C, and TSST-1; their PFGE patterns were A3, A4, A40, A42, AU7, AV6, or AV7. Nine isolates produced coagulase type II, enterotoxin type B, but not TSST-1; their PFGE patterns were AV5, AV8, AV9, or AV10.

The PFGE patterns obtained in this study were compared with those identified in Tokyo in October 2003 (Fig. 3) (4). Patterns A3 and A4 were detected in both instances. Eleven patterns were unique to Sendai, and 36 were unique to Tokyo. Previous studies conducted in 2000-2003 in Tokyo (1-4) and in 2001-2003 in Kumamoto (5-7) indicated a clone propagation of the pattern A1 MRSA in both hospitals. In a Sendai hospital, a different clone with a PFGE pattern of AV5

was found to have spread (Table 1). Thus, it was determined that nosocomial infections in a hospital in Tokyo shared common clones with those in Sendai and those in Kumamoto. However, infections in hospitals in Sendai and in Kumamoto, which are geographically separated from each other, did not share clones. Considering the fact that patients, as well as doctors, nurses, and other health care workers are frequently sent to or visit large medical facilities in Tokyo, this data may help account for how MRSA spreads nationwide in Japan.

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