

fections. Prevalence of MRSA and distribution of MRSA sequence types in livestock in Italy are not known. However, surveys of foods of animal (pig) origin have showed an MRSA prevalence of 3.7% (1,10). In view of the low prevalence of MRSA ST398 in patients with no exposure to animals, food products currently seem to play a negligible role. However, this clone is likely spreading because of the large animal reservoir of ST398 and the global market for meat and livestock. The changing epidemiology of MRSA indicates that collaborative surveillance plans integrating human and animal information should be increased.

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Panton-Valentine Leukocidin-Positive MRSA, Shanghai, China

To the Editor: The development of methicillin resistance in community strains of *Staphylococcus aureus* is a notable step in the evolution of this pathogen. Unlike their equivalents in the hospital environment, community-associated methicillin-resistant *S. aureus* (CA-MRSA) strains tend to cause infections in children and young adults who have few known healthcare risks (1). CA-MRSA strains usually possess the Panton-Valentine leukocidin (PVL) genes and staphylococcal cassette chromosome (SCC) *mec* type IV or V (1,2).

We studied 72 *S. aureus* isolates (49 MRSA and 23 methicillin-susceptible [MSSA]) by pulsed-field gel electrophoresis and by SCC*mec*, staphylococcal protein A (*spa*), and multilocus sequence typing (1,3). These isolates were recovered from clinical specimens (52 respiratory specimens, 9 wound, 4 urine, 2 blood, and 5 other body fluids) from 72 patients treated in 5 district hospitals in Shanghai, People's Republic of China, during October 2005 through January 2007. The isolates were randomly chosen. In the hospitals, ≈1,000 *S. aureus* isolates were recovered annually during the time period of our study. The 5 hospitals are estimated to serve a population of 3.4 million, equivalent to one fourth of the total population in Shanghai. Hospital D is a children's hospital. The other 4 hospitals (A, B, C, and E) have all the major clinical specialties, emergency departments, and outpatient clinics.

The isolates were identified as *S. aureus* by Gram stain, latex agglutination (Slide StaphPlus; bioMérieux, Marcy l'Etoile, France), and tube coagulase, mannitol, ornithine, and deoxyribonuclease reactions (1,4). Methicillin resistance in the isolates was

detected by cefoxitin disc screening and confirmed by *mecA* PCR (1,4). For patients with PVL-positive MRSA, the computerized discharge records in the hospitals were retrospectively reviewed to ascertain demographic and clinical information. A MRSA case was considered to be community associated if it was isolated from an out-patient or within 2 days of a patient's hospitalization. Exclusion criteria included a history of hospitalization for illness (except birth), surgery, or dialysis in the previous year or the presence of indwelling catheters or other medical devices (1). Conversely, healthcare-associated MRSA was defined by isolation >2 days after hospitalization or presence of any of the aforementioned healthcare risks.

PVL genes were detected in 9 (18.4%) of the 49 MRSA isolates (Table) and 4 (17.3%) of the 23 MSSA isolates. The 9 MRSA case-patients included 8 infants with pneumonia and 1 adult with prostatitis. Pulsed-field gel electrophoresis clustered 8 of the 9 PVL-positive MRSA isolates into 2 groups: 6 isolates as SH100 and 2 isolates as SH200. Strains of SH100 were *spa* type/MLST-SCC*mec* type t318/ST30-IV or t318/ST1114-V, and SH200

strains had t1376/ST88-V. Similar to the PVL-positive MRSA isolates, a limited number of *spa* types were found among the 40 PVL-negative MRSA isolates. These were t037/ST239-III (n = 19), t002/ST5-II (n = 14), t030/ST239-III (n = 5), t459/ST239-III (n = 1), and t1764/ST88-IV (n = 1).

In contrast, *spa* and sequence types (STs) among the 23 MSSA isolates were highly diverse. There were 20 *spa* types and 14 STs, giving a total of 20 distinct patterns. Three patterns (t091/ST7, t3388/ST630, t3389/ST15) had 2 isolates, and 17 patterns (t002/ST5, t1077/ST121, t127/ST1, t1376/ST88, t189/ST188, t2024/ST30, t2092/ST121, t2207/ST1206, t2471/ST25, t258/ST25, t3383/ST20, t3386/ST630, t377/ST630, t437/ST1205, t548/ST5, t701/ST6, t796/ST7) had 1 isolate only. The 4 PVL-positive MSSA isolates were t1376/ST88, t2471/ST25, t258/ST25, and t3383/ST20.

Mupirocin resistance rates among the PVL-positive and PVL-negative MRSA isolates were 33.3% (3/9) and 7.5% (3/40), respectively (p = 0.07). All MSSA isolates were susceptible to mupirocin.

It is notable that of the 9 PVL-positive MRSA isolates, 8 of them came

from hospital D and all were from children 1–4 months of age. Others have noted that the epidemiology of MRSA differs for children and adults (1,2,7,8). Molecular typing showed that the PVL-positive CA-MRSA isolates were attributed to 2 clones with genotypes t318/ST30-IV (or t318/ST1114-V) and t1376/ST88-V. Detection of t318/ST30 strains in 4 patients with healthcare-associated infections suggested hospital transmission of this CA-MRSA clone, corroborating reports elsewhere (9). Worldwide, ST30 is a common CA-MRSA genetic lineage (1,2). Besides t318, strains related to the ST30 clone have been reported to be *spa* types t019, t021, and t1273 (2). ST88 PVL-positive MRSA is relatively less common but has been found in Wenzhou (People's Republic of China), Bangladesh, Belgium, and Nigeria (2,7,8,10).

Because the number of isolates tested in this study is relatively small, no firm conclusion could be drawn on the prevalence of PVL-positive CA-MRSA among *S. aureus* isolates. Nonetheless, our findings agree with previous reports that the genotypes of MSSA isolates are more diverse than are those for PVL-positive and -nega-

Table. Epidemiologic and microbiologic characteristics for Panton-Valentine leukocidin–positive MRSA infections in 9 case-patients, Shanghai, People's Republic of China, 2006*

Strain	Admission		Onset	Diagnosis†	Resistance pattern‡	Resistance determinants§	ST§	SCC <i>mec</i>	<i>spa</i> type	PFGE
	Age/sex	month								
D8	1 mo/F	Feb	HA	Pneumonia	Chl, Ery, Gen, Mup	<i>aacA-aphD</i> , <i>ermC</i> , <i>ileS-2</i>	ST30	IV	t318	SH100
D9	1 mo/F	Feb	CA	Pneumonia	Gen, Mup	<i>aacA-aphD</i> , <i>ileS-2</i>	–	IV	t318	SH100
D13	1 mo/M	Mar	HA	Pneumonia	Gen	<i>aacA-aphD</i>	–	IV	t318	SH100
D14	3 mo/M	Mar	CA	Pneumonia	Gen	<i>aacA-aphD</i>	–	IV	t318	SH100
D16	1 mo/M	Mar	CA	Pneumonia	Gen, Mup	<i>aacA-aphD</i> , <i>ileS-2</i>	–	IV	t318	SH100
D12	3 mo/F	Mar	HA	Pneumonia	Ery	<i>ermC</i>	ST1114	V	t318	SH100
A18	29 y/M	Mar	HA	Prostatitis	None	–	ST30	IV	t3384	Singleton
D7	1 mo/F	Jan	CA	Pneumonia	Ery, Gen	<i>aacA-aphD</i> , <i>ermC</i>	ST88	V	t1376	SH200
D18	4 mo/M	Apr	CA	Pneumonia	Ery, Gen	<i>aacA-aphD</i> , <i>ermC</i>	–	V	t1376	SH200

*Strains are designated by hospital code and strain number. MRSA, methicillin-resistant *Staphylococcus aureus*; ST, sequence type; SCC, staphylococcal cassette chromosome; *spa*, staphylococcal protein A; PFGE, pulsed-field gel electrophoresis; HA, healthcare-associated; Chl, chloramphenicol; Ery, erythromycin; Gen, gentamicin; Mup, mupirocin; *aacA-aphD*, aminoglycoside resistance gene encoding the bifunctional enzyme, 6'-aminoglycoside N-acetyltransferase/2"-aminoglycoside phosphotransferase; *ermC*, gene encoding macrolide-lincosamide-streptogramin B resistance; *ileS-2*, gene encoding high-level mupirocin resistance mediated by isoleucyl tRNA-synthetase; CA, community-associated.

†According to the diagnosis given in the computerized record. In the 8 children, MRSA was isolated from sputum specimens. One child also had MRSA isolated from pleural fluid. Because limited information was provided in the computerized records, some of the isolations may represent respiratory colonization. The outcomes of the 8 children were unknown.

‡According to disk diffusion test (1,5).

§Determined by PCR as described (4,6). ST30 (allelic profile 2–2–2–2–6–3–2) and ST114 (139–2–2–2–6–139–2) belonged to clonal complex 30.

tive MRSA isolates and that genotypes for some CA-MRSA strains are shared by a few of the MSSA strains (1).

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Clostridium difficile in Ground Meat, France

To the Editor: *Clostridium difficile* is a toxigenic enteropathogen responsible for 15%–20% of antimicrobial drug-associated diarrhea and for almost all cases of pseudomembranous colitis. Two protein toxins (TcdA and TcdB) play a major role in the pathogenesis of infections. *C. difficile* is also recognized as a cause of disease in several animal species, which could be potential reservoirs (1). In the past few years, the presence of *C. difficile* in raw diets for dogs and cats and in retail meat sold for human consumption has been reported in the United States and Canada at rates from 6% to 42% (2–5). To determine *C. difficile* contamination of meat in France, we evaluated 105 packages of ground beef (vacuum packed or not), 59 pork sausages, and 12 packages of feline raw diet meat purchased from 20 urban and suburban Paris retail stores and supermarkets during September 2007–July 2008.

C. difficile spores or vegetative forms in samples were found as described by Rodriguez-Palacios et al. (4). Briefly, 5 g of each sample was cultured in 100 mL of prerduced brain–heart infusion (BHI) broth supplemented with cefoxitin (10 µg/mL), cycloserine (250 µg/mL), and taurocholate (0.1%). After the samples were incubated under anaerobic conditions at 37°C for 72 h, subculturing with and without alcohol shock for spore selection was performed. The BHI broth culture was treated with 2 mL of absolute ethanol (1:1 vol/vol) for 30 min and centrifuged at 3,800 × g for 10 min, and the pellet was resuspended in 200 µL of prerduced BHI broth. Serial dilutions of the BHI broth and the pellet were injected onto Columbia cysteine agar supplemented with cefoxitin-cycloserine, taurocholate, and 5% horse blood and incubated anaerobically for 48 h at 37°C.