Jpn. J. Infect. Dis., 65, 551-552, 2012

Short Communication

Presence of *Staphylococcus aureus* ST398 and ST9 in Swine in Japan

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(Received July 30, 2012. Accepted October 1, 2012)

SUMMARY: Livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA) is mainly associated with swine and is capable of causing zoonotic infections. The methicillin-resistant *S. aureus* (MRSA) multilocus sequence type (ST) 398 of swine origin is predominant in Europe and North America, whereas ST9 is predominant in Asia. To evaluate the possible emergence of MRSA in swine, we examined the ST and *spa* type of 15 methicillin-susceptible *S. aureus* (MSSA) isolates obtained from swine in 8 different prefectures from north to south Japan between 2003 and 2009. Sequence analyses revealed that 6 porcine MSSA isolates belonged to ST398; 6 to ST9; and 1 each to ST5, ST97, and ST705. Of the 6 MSSA ST398 strains, 4 were classified as *spa* type t034. This study illustrated that there is a reservoir in Japanese swine of livestock-associated MSSA types.

The emergence of livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA), belonging to the multilocus sequence type (ST) 398, has recently been recognized as a possible threat to human health (1,2). LA-MRSA ST398 isolates are found in domestic animals (mainly pigs); humans that are in close contact with affected animals: and retail meats in Europe (1), North America (3,4), and Asia (5,6). In Asian countries, ST9, a distinct type of methicillin-resistant *S. aureus* (MRSA), has been identified in swine in China (7,8), Malaysia (9), and Thailand (10). In Japan, a previous study found no MRSA ST398 or ST9, but MRSA ST221 was isolated from a pig. (11).

MRSA strains evolve from methicillin-susceptible *S. aureus* (MSSA) strains by acquisition of the *mecA* gene, which is carried in a mobile genetic element called the staphylococcal cassette chromosome *mec* (SCC*mec*). This gene confers resistance to virtually all β-lactam antimicrobials by encoding an alternative penicillin-binding protein (PBP2a), which has a lowered affinity for β-lactam antibiotics (12). Epidemiological information on *S. aureus* can be obtained through various methods such as multilocus sequence typing (MLST) and *spa* typing, which is based on the sequencing of the polymorphic X region of the *S. aureus* protein A gene (*spa* gene). European surveillance studies (1) show that 90% of LA-MRSA ST398 isolates typically belong to 4 major (t011, t108, t034, and t899) and several minor *spa* types. The diversity of *spa* types and SCC*mec* types (IVa and V) indicates that MRSA ST398 strains may evolve from a variety of MSSA strains. Recent studies have reported the prevalence of MSSA ST398 in swine (8,13). In the present study, we examined the ST and *spa* types of MSSA isolates obtained from swine in order to evaluate the possibility of the emergence of MRSA in pigs in Japan.

In total, 15 MSSA isolates from diseased pigs were kindly provided by various Livestock Hygiene Service Centers in Japan. These isolates were collected from 8 different prefectures from north (Hokkaido) to south (Kyushu) Japan between 2003 and 2009, and were subjected to MLST and *spa* typing. Of these isolates, 7 were obtained from dermatitis lesions and 1 was obtained from arthritis lesions; the source of the remaining isolates was not known.

MLST and *spa* typing of MSSA isolates were performed according to previously published methods (14,15); STs and *spa* types were determined using the MLST website (www.mlst.net) and Ridom database website (http://spaserver2.ridom.defjndex.shtml), respectively.

MLST analysis of the MSSA isolates revealed that 6 belonged to ST398; 6 to ST9; and 1 each to ST5, ST97, and ST705 (Table 1). Porcine MSSA isolates belonging to ST398 and ST9 have been detected in Denmark (13) and China (8), but have now been isolated in Japan for the first time. A few European studies have provided information on the *spa* types associated with MRSA ST398 (1,13). In the present study, the *spa* types t034, t1298, and t3934 were identified among the MSSA ST398 isolates, whereas the *spa* types t337, t526, t1430, and t6158 were identified in porcine isolates belonging to ST9 (Table 1). The *spa* type t034 was the third most predominant type of MRSA ST398 isolated from swine in the European Unit (1) and the most dominant type in Denmark (13). However, MRSA ST9 isolates of the *spa* type t1430 have also been identified in swine in European countries (1,13). The current study showed the existence of an MSSA reservoir containing *spa* types resembling those of well-known LA-MRSA strains.

Our previous study showed that MRSA ST398 strains were not present among Japanese pigs despite the fact that many animals were imported from countries where

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Table 1. Molecular typing of MSSA from swine in Japan

MLST (allelic profile)	*spa* type (repeat succession)	District (isolation year)	No. of isolates
ST398 (3-35-19-2-20-26-39)	t034 (08-16-02-25-02-25-34-24-25)	Tokai (2003)	1
		Chugoku (2005)	1
		Hokkaido-Tohoku (2007)	2
	t1298 (15-16-02-16-02-25-17-24)	Hokkaido-Tohoku (2007)	1
	t3934 (07-02-25-34-24-25)	Kanto (2009)	1
ST9 (3-3-1-1-1-1-10)	t337 (07-16-23-23-02-12-23-02-34)	Kanto (2003, 2008, 2009)	3
	t526 (07-16-23-23-02-12-23-34)	Hokkaido-Tohoku (2005)	1
	t1430 (07-16-23-02-12-23-02-34)	Koshinetu (2008)	1
	t6158 (07-16-23-02-12-23-02-02-34)	Koshinetu (2008)	1
ST5 (1-4-1-4-12-1-10)	t179 (26-23-17-34-17-20-17-12-12-16)	Hokkaido-Tohoku (2009)	1
ST97 (3-1-1-1-1-5-3)	t2112 (26-23-12-21-17-34-34-34-34-33-34)	Kanto (2009)	1
ST705 (6-72-50-43-49-67-59)	t529 (04-34)	Kyushu (2006)	1

MRSA strains are prevalent (10). Yet, the present study warns of a possibility of the emergence of LA-MRSA in Japan; therefore, it is necessary to continue MRSA surveillance in livestock animals and retail meats.

Acknowledgments We thank the members of the Livestock Hygiene Service Centers for providing *S. aureus* isolates.

This work was supported in part by a grant-in aid from the Japanese Ministry of Health, Labour and Welfare (H21-Shokuhin-Ippan-013).

Conflict of interest None to declare.

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