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Molecular Epidemiology of Methicillin-Resistant *Staphylococcus aureus* in Israel: Dissemination of Global Clones and Unique Features

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From 2006 to 2009, 315 clinical methicillin-resistant *Staphylococcus aureus* (MRSA) isolates were collected from 5 hospitals across Israel. Most isolates (64%) were related to the global clones *spa* types t001-SCC*mec*-I (SCC*mec*-I stands for staphylococcal cassette chromosome *mec* type I) ($n = 99$; 31%), t002-SCC*mec*-II ($n = 82$; 26%), and t008-SCC*mec*-IV ($n = 21$; 7%), five of which were identified as MRSA strain USA-300. Seventeen strains unique to Israel were identified. SCC*mec* types IV and V were common among hospital-acquired isolates.

Methicillin-resistant *Staphylococcus aureus* (MRSA) is an increasing problem throughout the world, both in hospitals and in the community. Several epidemic health care-associated MRSA (HA-MRSA) clones have emerged since the 1970s. In the last decade, five major pandemic clones, designated the Iberian, Brazilian, Hungarian, New York/Japan, and pediatric clones, have been identified, while other new or preexisting clones have emerged in certain areas (12). The recent worldwide spread of several community-associated MRSA (CA-MRSA) clones, and their dissemination into hospitals, has made the understanding of this epidemiology even more complex (10). In Israel, the proportion of MRSA among all *S. aureus* isolates in 2008 was 35%, similar to the proportion in Southern Europe and the United Kingdom (15) and lower than that in other Middle Eastern countries (4). There is little data on the molecular epidemiology of MRSA in Israel. The epidemiology of CA-MRSA was studied in the pediatric population of southern Israel, where 5.7% of infants were found to be colonized, mostly by a unique MRSA staphylococcal cassette chromosome *mec* type IV (SCC*mec*-IV) clonal complex (CC), 913 (1). However, data regarding the molecular epidemiology of community-acquired MRSA among adults, and data on HA-MRSA infections in Israel, are limited to small series or case reports (6, 29, 30).

In 2008, the National Center for Infection Control initiated a national survey of MRSA in Israel. The aims of the study were (i) to describe the molecular epidemiology of hospital-associated and community-associated MRSA infections in Israel and (ii) to determine whether internationally known MRSA strains (e.g., the USA-300 strain) have spread into Israel. Five general hospitals throughout Israel participated in the study: (i) Tel-Aviv Sourasky Medical Center (TA) in the Tel-Aviv area, (ii) Rambam Medical Center (RA) in northern Israel, (iii) Barzilai Medical Center (AS) in southern Israel, (iv) Meir Medical Center (ME) in the Sharon region of central Israel, and (v) Bikur Cholim (BC) in the Jerusalem area. Each laboratory was asked to submit prospectively collected MRSA isolates from blood samples or wounds, isolated within 72 h of admission to the hospital (CA-MRSA), or from 72 h on (HA-MRSA). MRSA isolates collected from 2006 to 2010 were eligible to be included in the study. Isolates were shipped and analyzed at the central study laboratory in Tel-Aviv, Israel.

TABLE 1 Epidemiological characteristics of MRSA isolates in this study

Center	No. of MRSA isolates[a]				Total no. of MRSA isolates
	Blood		Wound		
	CA	HA	CA	HA	
TA	37	37	40	37	151
RA	21	34	27	16	98
ME	13	9	0	2	24
AS	11	4	13	12	40
BC	0	2	0	0	2
Total (all)	82	86	80	67	315

[a] The number of community-associated (CA) MRSA isolates is the number of isolates cultured within 3 days of admission to the hospital. The number of health care-associated (HA) MRSA isolates is the number of isolates cultured from ≥3 days of admission to the hospital.

Identification and susceptibility testing of MRSA isolates were done by using the VITEK2 (bioMérieux, Marcy l'Etoile, France) system; DNase testing and cefoxitin disk diffusion testing were performed according to Clinical and Laboratory Standards Institute guidelines (7). Genetic relatedness was determined by *spa* typing for all strains (18) and by pulsed-field gel electrophoresis (PFGE) for t008 isolates (27). *spa* types were determined with Ridom StaphType software version 2.2.1 (Ridom GmbH, Würzburg, Germany) and analyzed by the BURP algorithm, with the following parameters: *spa* types with fewer than five repeats were considered nongroupable, and *spa* types belonged to the same *spa* clonal complex if the cost was less than or equal to six (18). In addition, the corresponding multilocus sequence typing (MLST)

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TABLE 2 Molecular and microbiological features of MRSA strains (other than *spa*-t008) in Israel[a]

SCC*mec* type and *spa* type	*spa* CC[b]	MLST CC[c]	No. of isolates	SCC*mec* type	No. of isolates that were:			Comment(s)[g]
					Clin S[d]	CA[e]	W[f]	
SCC*mec* types I and III ($n \geq 2$)								
001	002	5	99	I[h]	0	49	51	Presumed Southern Germany clone (8); previously reported from Israel (6)
002	002	5	82	II[i]	8	44	33	Presumed New York/Japan clone (8); previously reported from Israel (6)
004	065/004	045	8	II	0	5	3	Novel report*
010	002	5	3	II (2)[h]/I (1)[i]	0	1	2	Novel report*
051	008	8	2	I	0	2	1	Presumed Iberian clone (12)
052	008	8	6	I	0	2	2	Presumed Iberian clone (12)
535	E	5	5	II	1	1	0	Novel report*
5212	008		2	I	0	0	1	Novel report*
SCC*mec* types IV/V								
002	002	5	28	V	15	14	12	Previously reported from hospitals in Israel (6)
024	008	8	2	IV	2	0	1	Presumed EMRSA-2/-6 (8); previously reported from Denmark (21)
032	S	22	2	IV	1	1	0	Presumed EMRSA-15; reported from both animals and humans in Germany (34) and the United Kingdom (25, 31)
064	008	8	1	IV	1	1	1	Previously isolated from domestic animals in Europe (20, 25, 32)
065	065/004	45	8	IV	6	4	4	Previously reported from Israel (1, 28, 29), presumed Berlin clone (12)
105	002	5	1	V	1	0	1	Novel report*
127	S	1	1	IV	0	1	1	Associated with pigs in Europe (16)
159	S	121	1	IV	1	1	1	Novel report*
214	002	5	1	V	1	0	1	Novel report*
223	223/8610	22	1	IV	1	0	1	Presumed EMRSA-15 (24)
242	002	5	1	V	0	1	0	Novel report*
318	037	30	1	IV	0	0	1	A *pvl*-positive strain, previously reported from Israel (1) and Shanghai (19)
437	S	59	1	V	0	1	1	A *pvl*-positive strain, commonly reported from Taiwan (5).
509	002	5	1	IV	0	0	0	Presumed "pediatric clone," reported from France (9)
570	002	5	1	V	1	0	1	Novel report*
723	008	8	1	V	0	0	0	Novel report*
796	S	7	1	IV	1	0	0	Previously reported from China (22)
991	E	913	4	IV	3	4	4	A common colonizing strain in Bedouin children in Israel (1)
1774	008	8	1	IV	1	0	1	Previously reported from South Africa (26)
1911	008	8	1	IV	1	1	B	Novel report*
2849	008		2	IV/V	2	1	2	Novel report*
5160	008		1	IV	0	1	1	Novel report*
6274	065/004		1	IV	0	1	0	Novel report*
7544	002		1	V	0	0	1	Novel report*

[a] Data are presented for all strains excluding t008 strains (Fig. 1) and single-isolate, SCC*mec* type I to III *spa* types ($n = 23$). Additional *spa*-CC-002 strains presented in Fig. S1 in the supplemental material are t088, t105, t2358, and t5712.

[b] *spa* CC, *spa* clonal complex. E, excluded from *spa* BURP analysis due to low (5>) repeat number; S, singleton on *spa* BURP analysis.

[c] MLST CC, multilocus sequence typing clonal complex (presumed allocation was based on the Ridom *spa* database and Monecke et al. (24), unless specified otherwise in the comment).

[d] Clin S, isolates tested that were susceptible to clindamycin.

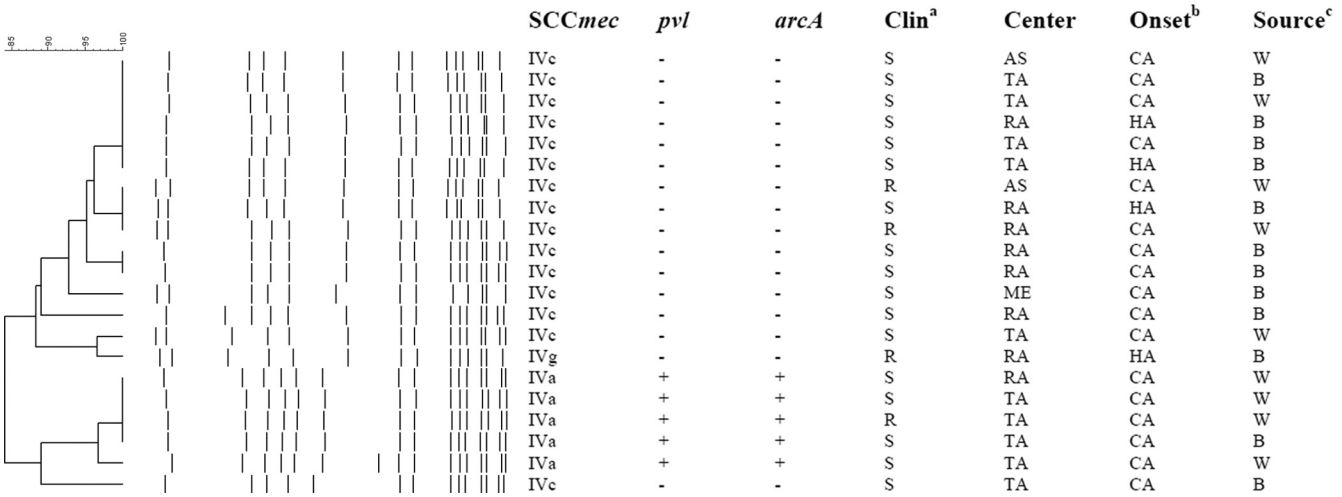
[e] CA, community associated (cultured within 3 days of admission to the hospital).

[f] W, wound (cultured from wound or other nonblood sites).

[g] An asterisk designates either a newly reported *spa* type or a new *spa*-SCC*mec* type combination.

[h] Two isolates harbored SCC*mec*-II.

[i] One isolate harbored SCC*mec*-I.



SCC*mec*	*pvl*	*arcA*	Clin[a]	Center	Onset[b]	Source[c]
IVc	-	-	S	AS	CA	W
IVc	-	-	S	TA	CA	B
IVc	-	-	S	TA	CA	W
IVc	-	-	S	RA	HA	B
IVc	-	-	S	TA	CA	B
IVc	-	-	S	TA	HA	B
IVc	-	-	R	AS	CA	W
IVc	-	-	S	RA	HA	B
IVc	-	-	R	RA	CA	W
IVc	-	-	S	RA	CA	B
IVc	-	-	S	RA	CA	B
IVc	-	-	S	ME	CA	B
IVc	-	-	S	RA	CA	B
IVc	-	-	S	TA	CA	W
IVg	-	-	R	RA	HA	B
IVa	+	+	S	RA	CA	W
IVa	+	+	S	TA	CA	W
IVa	+	+	R	TA	CA	W
IVa	+	+	S	TA	CA	B
IVa	+	+	S	TA	CA	W
IVc	-	-	S	TA	CA	B

FIG 1 Molecular features of *spa*-t008 strains in Israel. Molecular, microbiological, and epidemiological features of 21 *spa*-t008 isolates in Israel. The SCC*mec* type, presence (+) or absence (−) of the *pvl* and *arcA* genes, clindamycin drug resistance, center, onset, and source are shown in the columns to the right of the dendrogram and PFGE pattern. The Clin[a] column shows the resistance (R) or susceptibility (S) of the strain to clindamycin. The Onset[b] column shows whether the isolate was community associated (CA) (cultured within 3 days of admission to the hospital) or health care associated (HA) (cultured from ≥3 days of admission to the hospital). The Source[c] column shows whether the isolates were derived from blood cultures (B) or wound cultures (W).

clonal complex was assigned for each *spa* type based on the *spa* types in the Ridom StaphType database (http://spa.ridom.de/spatypes.shtml) and the study by Monecke et al. (24). The Panton-Valentine leukocidin (*pvl*) and *arcA* (t008 isolates only) genes were tested for by PCR (13, 27). SCC*mec* typing was assigned according to the *mec* and *ccr* complexes (36); subtyping of SCC*mec*-IV was done for t008 isolates (23).

A total of 315 MRSA isolates were collected from 2006 to 2009. A summary of the epidemiological features of the MRSA isolates in this study is presented in Table 1. Almost half of the isolates were collected at the Tel-Aviv Sourasky Medical Center (TASMC), Tel-Aviv, Israel. Eighty-six (51%) and 66 (45%) of the blood and wound cultures, respectively, were collected at least 72 hours after admission to the hospital.

The molecular characteristics of MRSA strains according to *spa* and SCC*mec* types are presented in Table 2 and Fig. 1 (t008). The majority of isolates (229; 63%) belonged to 13 *spa* types which are related to *spa* CC-002 or MLST CC 5 (Table 2; see Fig. S1 in the supplemental material). The second largest was a group of 40 isolates (11%) and 11 *spa* types related to *spa* CC 008 or MLST CC 8 (Table 2 and Fig. S1). Except for five t008 isolates, all *spa* CC-008 isolates were negative by *pvl* testing.

The 21 t008 strains were divided into 2 major types (Fig. 1): (i) SCC*mec*-IVa, a *pvl*- or *arcA*-positive strain, identified as MRSA strain USA-300 by PFGE; (ii) SCC*mec*-IVc (1 isolate was SCC*mec*-IVg), a *pvl*- or *arcA*-negative strain, that possessed a PFGE pattern similar to that of a previously described Israeli t008 strain (30) presumably similar to the EMRSA-2/-6 clone (8). All 5 USA-300 isolates and 12 of 16 *pvl*-negative t008 isolates were categorized as CA based on the time of isolation.

Six novel MRSA *spa* types were found and nine isolates were typed with a unique combination of *spa* and SCC*mec* types (Table 2).

Almost all MRSA SCC*mec*-I to -III strains (Table 2) were resistant to clindamycin, whereas strains harboring SCC*mec*-IV/V were mostly susceptible (55/85). In contrast, most isolates were susceptible to rifampin (95%) and trimethoprim-sulfamethoxazole (98%); all strains were susceptible to vancomycin.

The present study is the first national survey of MRSA strains in Israel. The most common MRSA strains in our study were found to be related to common epidemic clones, such as the Southern Germany (t001/SCC*mec*-I), New York/Japan (t002/SCC*mec*-II), Berlin (t065/SCC*mec*-IV), EMRSA-2/-6 (t008/024/SCC*mec*-IV or *pvl* negative) and Iberian (t051/052/SCC*mec*-I) (8, 12) clones; these clones have been previously identified in Israel (1, 6). Interestingly, these clones were absent or rarely reported in a recent study from Lebanon (35).

Three *pvl*-producing strains were identified. The t318-SCC*mec*-IV strain ($n = 1$) has been previously reported from Israel (1) and other Middle Eastern countries (3, 14). t437-SCC*mec*-V, a common strain in Taiwan (5), has never been reported from our region, and USA-300 MRSA infections have never been reported from other countries in our region (3, 14, 35). Although we are not able to determine whether these USA-300 MRSA infections were acquired in Israel, these cases together with the recent report of a USA-300 MRSA infection in an Israeli child (17) suggest that this strain is likely to be present at a low level in our population.

The proportions of SCC*mec* types IV and V were 16% and 11%, respectively. The proportion of SCC*mec*-V is relatively high compared with other countries, as was also reported in a recent study from Israel (2). Many of the SCC*mec*-IV/V strains identified in our study are uniquely reported from Israel (Table 2). Four t991 SCC*mec*-IV isolates were cultured from wounds within 3 days of admission to the hospital, from AS, TA, and RA, indicating the presence of this clone in communities outside the Bedouin population in the Negev desert in Israel (1). t002-SCC*mec*-V, the most common SCC*mec*-IV/V-harboring strain in our study, is reported uniquely from Israel (6) and was isolated after at least 72 h of admission to the hospital in 14 out of 28 cases. Altogether, SCC*mec*-IV/V-harboring strains were isolated after at least 72 h of

admission to the hospital in 35 of 85 cases, indicating that the SCC*mec*-IV/V types, cannot serve as a marker for community acquisition in Israel (11).

Our study has several limitations concerning the extent of representation of the MRSA population in Israel. First, our collection lacked a significant number of isolates from certain parts of the country, such as Jerusalem and the Galilee region. Second, despite the collection instructions, the proportion of wound and blood culture isolates was not even in all centers. Third, we are not able to determine how many of the isolates labeled as CA represented actual community-acquired infection, as many of the patients from whose specimens these isolates grew may have been recently hospitalized.

Our study delineates molecular epidemiological features that our MRSA isolates have in common with those in other parts of the world, while highlighting unique features of the Israeli isolates. With global transmission of MRSA (33), our local epidemiology may be changing due to the introduction of new strains.

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