Pulsed-Field Gel Electrophoresis Typing of Oxacillin-Resistant
Staphylococcus aureus Isolates from the United States:
Establishing a National Database

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Received 2 June 2003/Returned for modification 10 July 2003/Accepted 22 August 2003

Oxacillin-resistant Staphylococcus aureus (ORSA) is a virulent pathogen responsible for both health care-associated and community onset disease. We used Smal-digested genomic DNA separated by pulsed-field gel electrophoresis (PFGE) to characterize 957 S. aureus isolates and establish a database of PFGE patterns. In addition to PFGE patterns of U.S. strains, the database contains patterns of representative epidemic-type strains from the United Kingdom, Canada, and Australia; previously described ORSA clonal-type isolates; 13 vancomycin-intermediate S. aureus (VISA) isolates, and two high-level vancomycin-resistant, vanA-positive strains (VRSA). Among the isolates from the United States, we identified eight lineages, designated as pulsed-field types (PFTs) USA100 through USA800, seven of which included both ORSA and oxacillin-susceptible S. aureus isolates. With the exception of the PFT pairs USA100 and USA800, and USA300 and USA500, each of the PFTs had a unique multilocus sequence type and spa type motif. The USA100 PFT, previously designated as the New York/Tokyo clone, was the most common PFT in the database, representing 44% of the ORSA isolates. USA100 isolates were typically multiresistant and included all but one of the U.S. VISA strains and both VRSA isolates. Multiresistant ORSA isolates from the USA200, -500, and -600 PFTs have PFGE patterns similar to those of previously described epidemic strains from Europe and Australia. The USA300 and -400 PFTs contained community isolates resistant only to β-lactam drugs and erythromycin. Noticeably absent from the U.S. database were isolates with the previously described Brazilian and EMRSA15 PFGE patterns. These data suggest that there are a limited number of ORSA genotypes present in the United States.

Oxacillin-resistant Staphylococcus aureus (ORSA), more commonly referred to as methicillin-resistant S. aureus (MRSA) (even though methicillin is rarely tested in U.S. laboratories), is a frequent cause of infections both in health care and community settings and is endemic in many U.S. hospitals (12, 26, 27, 32, 48, 56). While a variety of strain typing methods have been used over the years to track the spread of ORSA (64), most outbreaks of ORSA have been characterized by bacteriophage typing or pulsed-field gel electrophoresis (PFGE). Although public health institutions in several countries, such as Australia (68), Denmark (41, 53, 67), The Netherlands (41, 66, 67), Canada (59), and the United Kingdom (41), have tracked ORSA strain types over the years, this has not been true in the United States for the most part.

Recent reports of vancomycin-intermediate S. aureus (VISA) (vancomycin MICs, 8 to 16 μg/ml) (5, 22, 28, 33, 50, 60, 65), of vanA-positive ORSA showing high-level vancomycin resistance (8, 9, 11), and of ORSA causing severe disease and death in children (6, 27, 42) all argued that better tracking of ORSA strains nationwide was needed to monitor the spread of such organisms. Thus, the Centers for Disease Control and Prevention (CDC), in collaboration with state health departments, has undertaken the goal of assembling a national database of S. aureus PFGE profiles similar to the PulseNet program for Escherichia coli O157:H7 (62).

The overall goal of this study was to assemble a database of ORSA PFGE profiles and to identify major lineages of ORSA present in the United States. PFGE was chosen over other typing methods, such as multilocus sequence typing (MLST), staphylococcal protein A gene (spa) typing, and restriction fragment length polymorphism-based methods, because the infrastructure and expertise for PFGE typing already exist within the state health departments in the United States. However, a national PFGE-based typing system for S. aureus would have to be validated with MLST and spa typing data to maintain continuity with the nomenclature already established in the literature. This would make the national database information useful for global tracking of ORSA, an organism with a limited number of lineages that exists endemically in health care and non-health-care settings. Thus, we typed a large number of ORSA isolates and developed a PFGE nomenclature scheme that was consistent with MLST and spa data yet was also useful for microbiologists and epidemiologists who were specifically studying ORSA infections in the United States. Herein, we present a framework for describing the lineages of ORSA present in the United States and correlating Smal PFGE profiles with the spa and MLST types already established in the literature (18, 19, 40, 46, 47, 58).

MATERIALS AND METHODS

Bacterial isolates. A total of 957 S. aureus isolates were examined in the study; 722 were oxacillin resistant and 235 were oxacillin susceptible according to NCCLS criteria (43). Of the 957 isolates, 300 S. aureus isolates were chosen at
random from the CDC *S. aureus* strain collection. These strains were from outbreaks in hospitals, food-borne disease, and community-acquired infections and were submitted to CDC for typing between 1995 and 2000. They were selected mainly for geographical and source diversity. An additional 381 ORSA isolates were obtained from health care and community infections. PFGE was performed by CDC and state health departments from 2001 to 2003. These included two *unc*-positive VRSA strains from Michigan (8) and Pennsylvania (9), eight U.S. VISA isolates for which the vancomycin MICs were 8.0 μg/ml and 22 isolates for which the vancomycin MICs were 4 μg/ml (22, 60). A further 221 isolates were from various CDC-sponsored surveillance studies. Also included in the database were 34* S. aureus* isolates from Japan (28), one from France (50), one from Hong Kong, one from Korea (33), and two from Scotland; 15 donal-type isolates (graciously provided from the collection of H. de Lencastre, Rockefeller University, New York, N.Y.) (13); 15 epidemic MRSA isolates (EMRSA; isolates obtained from H. Aucken, Public Health Laboratory Service, Colindale, United Kingdom); four representative epidemic Canadian MRSA isolates (CMRSA) (59) (courtesy of B. Willey, Mt. Sinai Hospital, Toronto, Ontario, Canada); 12 isolates from community outbreaks in Australia (39, 44) (courtesy of J. Bell, Women and Children’s Hospital, Adelaide, Australia), and three staphylococcal SCC mec-type control isolates (31) (courtesy of K. Hiramatsu, Juntendo University, Tokyo, Japan).

**Antimicrobial susceptibility testing.** The antimicrobial susceptibility profiles of the isolates were determined by broth microdilution with cation-adjusted Mueller-Hinton broth (BD Biosciences, Sparks, Md.) as described in the NCCLS publication M7-A5 (43). The antimicrobial agents tested were chloramphenicol, clindamycin, erythromycin, gentamicin, levofloxacin, linezolid, oxacilin, penicillin, quinupristin-dalfopristin, rifampin, tetracycline, trimethoprim-sulfamethoxazole, and vancomycin. Quality control strains included *S. aureus* ATCC 29213, *E. coli* ATCC 29222, and *Enterococcus faecalis* ATCC 29212. Multiresistance was defined as resistance to three or more classes of antimicrobial agents. Inducible clindamycin resistance was determined with a disk diffusion test in which an erythromycin disk was placed 12 mm from the edge of a clindamycin disk on a blood agar plate after inoculating the plate with a suspension of the test organism that was adjusted to the turbidity of a 0.5 McFarland standard. The presence of a D-shaped zone of inhibition indicated the induction of methylethyl production by erythromycin (69). Inducible clindamycin resistance was presumed to be mediated by a *erm* gene. If no clindamycin induction occurred, the resistance mechanism was assumed to be efflux of the macrolide via *mef* (35). High-level resistance to spectinomycin (300 μg/ml), which is associated with the majority of the ORSA strains that carry the transposon Tn554 (49, 55), was determined by disk diffusion with use of a 300-μg disk (made in-house). The absence of a measurable zone of inhibition after 24 h of incubation at 35°C was indicative of resistance. *S. aureus* strains HDE1 and HP107, which are known to be susceptible and resistant to spectinomycin, respectively (55), were used as controls.

**PFGE.** A single colony of the test isolate was inoculated into 5 ml of brain heart infusion broth and incubated with vigorous shaking at 35 to 37°C. The cell suspensions were adjusted with saline either by the turbidity method and incubated with vigorous shaking at 35 to 37°C (57). The concentrations of the cell suspensions were adjusted with saline either by the turbidity method or the ORSA strains that carry the transposon Tn554 (49, 55), were used as controls. The concentrations of the cell suspensions were adjusted with saline either by the turbidity method or the ORSA strains that carry the transposon Tn554 (49, 55), were used as controls. Of isolates from health care-associated infections, USA100 and USA300 were determined by PFGE. A dendrogram of percent similarity, calculated with Dice coefficients from the PFGE data using a cutoff of 80%, revealed eight major clusters of isolates, designated as PFTs USA100 through USA800 (Fig. 1). Of the 667 U.S. ORSA isolates, 622 (93%) clustered within these eight PFTs. In addition, 134 of the 235 Ossa isolates (57%) had PFGE patterns that fell within the same eight PFTs. The results of MLST, spa typing, SCC mec typing, antimicrobial resistance profiles, and other relevant properties of ORSA strains are summarized in Table 1. All but four PFTs (USA300 and -500, and -100 and -800) had a unique MLST sequence type and spa type motif. Five of the eight PFTs (USA100, -200, -500, -600, and -800) contained isolates that were predominantly obtained from health care-associated infections, while the isolates from two PFTs (USA300 and -400) were from community infections. USA700 isolates were obtained from patients in both community and health care settings.

Of isolates from health care-associated infections, USA100 was the largest and most diverse of the PFTs, containing 292 ORSA isolates from throughout the United States. For the seven housekeeping genes, these isolates shared a common MLST allelic profile (1- 4- 1- 4- 12- 1- 10), which is designated as ST 5, and a common *spa* motif (MDMGMK). USA100 isolates were usually spectinomycin resistant (consistent with SCC mec II) and multiresistant to commonly used therapeutic agents. This group included seven of eight U.S. VISA isolates and VISA isolates from Japan (Mu50) and Korea. USA100 also included the two U.S. VRSA isolates from Michigan and Pennsylvania. All isolates were resistant to erythromycin: 72%...
were constitutively clindamycin resistant, and 28% showed inducible clindamycin resistance. *S. aureus* isolates BK2464, PA237, and JA48, from the New York/Japan clone (1, 51, 52), belonged to this PFT.

USA800 isolates shared the same MLST sequence type and *spa* motif as the USA100 isolates. These isolates were primarily from community surveillance studies, were spectinomycin susceptible (suggesting the lack of Tn554 [consistent with SCC*mec* type I or IV]) and were generally resistant only to β-lactam drugs. Twenty-three percent were erythromycin resistant. When we tested the isolates, SCC*mec* typing showed that most of them were SCC*mec* type IV. *S. aureus* isolates HDE1, HDE288, and COB94 from the Pediatric clone (24, 55) belonged to this group. Other isolates, such as EMRSA3, which carried SCC*mec* type I and fell just outside the USA100 and -800 clusters, shared the same MLST and *spa* motif.

USA200, the second most common health care-associated PFT among U.S. isolates, contained ORSA isolates that were spectinomycin resistant (consistent with SCC*mec* II) and were multiresistant to therapeutic agents. All ORSA isolates were erythromycin resistant, with 98% showing constitutive resistance to clindamycin. The isolates had the same MLST profile, i.e., ST 36 (2-2-2-2-3-3), and *spa* type motif (WGKAK AOMQQQ) as isolates from the EMRSA16 epidemic clone.

Isolates from community onset infections belonging to PFT USA400 had the MLST ST 1 profile (1-1-1-1-1-1-1) and *spa* type motif (UJJJFE). These ORSA isolates were spectinomycin susceptible, carried SCC*mec* IV, and were not multiresistant. *S. aureus* MW2, an isolate from a rapid fatal infection in a child from Minnesota (6), was included in this group.

Although representative isolates from USA300 and USA500 had the same MLST allelic profile (ST 8, 3-3-1-1-4-4-3) and *spa* type motif (MBOBLO), these isolates clustered into separate, but contiguous, groups by *Sma*I PFGE. USA300 isolates carried SCC*mec* IV, were resistant to β-lactam drugs, were frequently resistant to erythromycin, and were predominantly from community onset skin infections. Eighty-five percent of the erythromycin-resistant isolates were susceptible to clindamycin and were not inducible with erythromycin (probably due to *msrA*). On the other hand, USA500 isolates were generally from health care-related infections. The majority of USA500 isolates were spectinomycin susceptible, indicating the absence of Tn554 (consistent with SCC*mec* type I or IV). Most of the isolates were resistant to clindamycin, erythromycin, gentamicin, levofloxacin, tetracycline, trimethoprim-sulfamethoxazole, and the β-lactams. The remaining U.S. VISA isolate and the three isolates for which the vancomycin MICs were 4 μg/ml had PFGE profiles belonging to this PFT. PFGE patterns of *S. aureus* isolates PER34, E2125, and HPV107 from the Archaic/Iberian clones (13, 14, 57); EMRSA isolates 2, 5, 10, 12, 13, and 14; VISA isolates from Hong Kong and France; and the Iberian clonal-type isolate from Scotland clustered near the USA500 isolates. EMRSA isolates 2, 6, 12, 13, and 14 shared the same MLST profile (3-3-1-1-4-4-3) as the USA300 and USA500 isolates, which differed at a single locus from ST 250 (3-3-1-1-4-4-16) of isolate Per34. *S. aureus* E2125 and HPV107 (Archaic/Iberian clonal-type isolates) and the *S. aureus* EMRSA5 isolate have the MLST profile ST 247 (3-3-1-1-12-4-4-16), which differed at one locus from ST 250 and two

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**FIG. 1.** Dendrogram of PFTs with type strain (most frequent pattern) and a variant strain. Also shown is the corresponding MLST for each PFT (18, 19, 20).
<table>
<thead>
<tr>
<th>Name of PFT</th>
<th>Total no. of U.S. ORSA strains in PFT (%)</th>
<th>MLST and allelic profile&lt;sup&gt;b&lt;/sup&gt;</th>
<th>spa type&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Clone name&lt;sup&gt;d&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>USA100</td>
<td>292 (44)</td>
<td>ST 5, 1- 4- 1- 4- 12- 1- 10</td>
<td>TJMBDMG</td>
<td>USA100 ORSA II</td>
</tr>
<tr>
<td>USA800</td>
<td>45 (6.7)</td>
<td>ST 5, 1- 4- 1- 4- 12- 1- 10</td>
<td>TJMBDMG</td>
<td>USA800 ORSA IV</td>
</tr>
<tr>
<td>USA200</td>
<td>57 (8.5)</td>
<td>ST 36, 2- 2- 2- 3- 3- 2</td>
<td>WGKAKAOMQ</td>
<td>USA200 ORSA II</td>
</tr>
<tr>
<td>USA400</td>
<td>42 (6.3)</td>
<td>ST 1, 1- 1- 1- 1- 1- 1- 1</td>
<td>UJJFE</td>
<td>USA400 ORSA IV</td>
</tr>
<tr>
<td>USA300</td>
<td>91 (13.6)</td>
<td>ST 8, 3- 3- 1- 1- 4- 4- 3</td>
<td>YHGF MBQBLO</td>
<td>USA300 ORSA IV</td>
</tr>
<tr>
<td>USA500</td>
<td>34 (5.1)</td>
<td>ST 8, 3- 3- 1- 1- 4- 4- 3</td>
<td>YHGC MBQBLO</td>
<td>USA500 ORSA IV</td>
</tr>
<tr>
<td>USA600</td>
<td>23 (3.4)</td>
<td>ST 45, 10- 14- 8- 6- 10- 3- 2</td>
<td>A2AKEEMBKB</td>
<td>USA600 ORSA II</td>
</tr>
<tr>
<td>USA700</td>
<td>38 (5.7)</td>
<td>ST 72, 1- 4- 1- 8- 4- 4- 3</td>
<td>UJGFMGGG</td>
<td>USA700 ORSA IV</td>
</tr>
<tr>
<td>Unnamed</td>
<td>14 (2)</td>
<td>Not tested</td>
<td>ZDMDMNKB</td>
<td></td>
</tr>
<tr>
<td>Unnamed</td>
<td>3</td>
<td>ST 239, 2- 3- 1- 1- 4- 4- 3</td>
<td>WGKAOMQ</td>
<td>ST239 ORSA III</td>
</tr>
<tr>
<td>Unnamed</td>
<td>0</td>
<td>ST 22, 7- 6- 1- 5- 8- 8- 6</td>
<td>TJEN12MN12MOM</td>
<td>ST 22 ORSA IV</td>
</tr>
</tbody>
</table>

<sup>a</sup> PFT is based on Smal PFGE with ≥80% similarity identified on an UPGMA-derived dendrogram using Dice coefficients.
<sup>b</sup> Allelic profile is based on the DNA sequences of seven housekeeping genes: *arcC*, *aroE*, *gplF*, *gmk*, *pta*, *tpi*, and *yqiL*.
<sup>c</sup> DNA sequence of polymorphic region of protein A gene. The spa type motif is shown in boldface.
<sup>d</sup> The clone name consists of the following three parts: PFT or MLST sequence type, the oxacillin phenotype (all are ORSA in this table), and the SCC*mec* type (when available).
<sup>e</sup> Isolates in this PFT subgroup were susceptible to spectinomycin.
<sup>f</sup> β-Lactams (oxacillin and penicillin); chl, chloramphenicol; cli, clindamycin; ery, erythromycin; gen, gentamicin; lvx, levofloxacin; tet, tetracycline; sxt, trimethoprim-sulfamethoxazole.
<sup>g</sup> CMRSA strains are epidemic type strains from Canada (59). Four strains are included in this study.
<sup>h</sup> EMRSA strains are epidemic type strains from the United Kingdom (16, 54). Fifteen strains are included in this study.
loci from ST 8. All of these isolates shared a similar spa type motif (ST 8).

USA600 contained 23 isolates. Representative strains were ST 45 (10-14-8-6-10-3-2) and spa type A2AKEEMBKB. All but four of the ORSA isolates in this PFT were spectinomycin resistant (SCCmec II) and multiresistant; the remaining four were spectinomycin susceptible. Isolates from USA700 shared a unique MLST type, i.e., ST 72 (1-4-1-8-4-3), and spa motif (UJGFMGGM) from community surveillance and a hospital-acquired outbreak.

There were two clusters within the remaining 45 U.S. ORSA isolates. The first cluster (14 isolates) included surveillance isolates, mainly from Alaska, and isolates from an outbreak of soft-skin infections among Vermont wrestlers (37). The second cluster included three isolates that clustered with S. aureus isolates HU25, HSJ216, and HUSA304 (Brazilian and Hung-
garian type strains [13, 63]); EMRSA isolates 1, 4, and 11; and MRSA and VISA isolates from Scotland. These isolates had the same MLST profile (ST 239) and spa type motif (WGKAOMQ). Twenty-eight isolates had either miscellaneous PFGE patterns or patterns that could not be identified within an 80% PFT cutoff.

Of the 235 U.S. OSSA isolates, 134 (57%) clustered within the eight PFTs. All of the PFTs, with the exception of USA500, contained OSSA. Twenty OSSA isolates were USA800, 1 isolate was USA100, 78 isolates were USA200, 9 isolates were USA300, 1 isolate was USA400, 22 isolates were USA600, and 3 isolates were USA700. Twenty-eight OSSA isolates clustered in a unique PFT. There were 10 OSSA clusters with 3 to 13 isolates, and 23 isolates with unique SmaI PFGE profiles.

**DISCUSSION**

Because of its high discriminatory power, PFGE is a valuable tool for investigating outbreaks of S. aureus infections, particularly in hospital settings (3, 4, 16, 27, 61, 64, 66). PFGE has also been used to discriminate among community- and health care-acquired ORSA strains (42). In previous work, de Lencastre and colleagues used PFGE to delineate lineages of ORSA that circulate in Europe, South America, and the United States, and they have given the lineages names, such as the Archaic clone, the Iberian clone, the Pediatric clone, and the New York/Tokyo clone (13). Similarly, Simor et al. have used PFGE to designate four lineages of ORSA in Canada, designated as CMRSA 1 through 4 (59). These and other studies have shown that PFGE can identify stable lineages of ORSA and can be used to track the spread of these lineages from continent to continent over extended periods of time.

In the past, the sharing of PFGE data among laboratories was difficult (15), and typing results for the same strains performed in different laboratories often lacked concordance (66). However, recent advances in gel analysis software programs allow the creation and storage of large databases of normalized fragment patterns in which similarity calculations and cluster analyses can be performed with relative ease. Normalization of the fragment patterns using established standards (such as S. aureus NCTC 8325) and the advent of new database sharing tools both serve to facilitate the exchange of PFGE strain typing data and epidemiologic information among reference laboratories, even in different countries. Thus, the traditional barriers to the sharing of PFGE patterns, even those run using different switching parameters, have to a large extent been overcome with powerful new software programs (62).

However, the appropriateness of using PFGE to study the long-term evolution of S. aureus lineages in general, and ORSA isolates in particular, remains a concern (19, 40). While MLST, which examines changes in the sequences of seven neutral loci on the S. aureus genome, may be more amenable to long-term population studies of ORSA, the higher discriminatory ability of PFGE profiles over that of MLST for S. aureus is advantageous. This holds true for epidemiologic studies of particular clusters of isolates, such as those isolates causing community onset disease (12, 27, 44) and for assessing the effectiveness of targeted-prevention programs. For example, USA300 and -500 isolates were assigned different PFTs because they separated into two distinct pulsed-field clusters and had different antimicrobial susceptibility patterns and epidemiology, even though they both shared ST 8 (Table 1). USA300 isolates are predominantly from community onset infections and are resistant only to β-lactam drugs and macrolides, while USA500 isolates differ from representatives of the Archaic and Iberian clones by only one or two locus variants (14, 20), tend to be from health care-associated infections, and are multidrug resistant. USA300 isolates have been identified in multiple community onset ORSA outbreaks associated with soft-skin infections in correctional facilities (7, 10), athletic teams (10), and nurseries (10). Isolates from prisons in Mississippi, Texas, Tennessee, and Georgia all shared this PFT, as did isolates from an outbreak of ORSA infections in football players. Thus, the epidemiology of USA300 isolates is quite distinct from the health care-associated USA500 isolates, even though they are indistinguishable by MLST typing. Similarly, although USA100 and -800 isolates shared a common MLST sequence type (ST 5) and spa motif (MDMGMK), the isolates carried different SCCmec structures and had different susceptibility profiles. USA100 isolates cluster with representatives of the multiresistant New York/Japan clone containing SCCmec II, while isolates from USA800 cluster with representatives of the Pediatric clone containing SCCmec IV. These critical epidemiologic differences are obscured by MLST. Thus, the PFGE typing system described here will likely be more useful than MLST, particularly for public health agencies, for targeted epidemiologic studies aimed at understanding the epidemiology of specific clonal groups, and for developing interventional studies to halt the transmission of the disease. Given the large data sets of PFGE profiles already established in the United States and around the world (15, 16, 41), the development of a surveillance system that can integrate these databases and make the information available to other reference centers is critical.

The other lineage associated with community onset disease was USA400. This PFT included ORSA isolates that caused severe, and in some cases fatal, disease in children from Minnesota and North Dakota, and was associated with skin disease in Native Americans from eastern Washington (state) (6, 25, 27, 42). Also in this group were isolates from Australian Aborigines (39, 44). Although not present in the U.S. database, isolates of this same MLST type (ST 1) were obtained in a community study in England from healthy carriers of S. aureus and those with health care-associated infections (21). This may be another instance in which MLST obscures epidemiologically relevant information regarding clusters of strains with different virulence characteristics. Many S. aureus strains responsible for primary skin infections and necrotizing pneumonia harbor the Panton-Valentine leukocidin determinant (17, 23, 36), and preliminary studies suggest that many USA300 and -400 isolates harbor this virulence determinant (CDC unpublished observations) in addition to the more recent SCCmec IV.

Among the five PFTs associated predominantly with health care-related infections (i.e., USA100, -200, -500, -600, and -800), USA100 was by far the most common (44% of all U.S. ORSA isolates examined). U.S. isolates of this PFT carried SCCmec II. This PFT was previously designated as the New York/Japan clone (1). Interestingly, several other previously described global lineages that are common as causes of health care-associated infections (e.g., the Brazilian clone and EMRSA15) were not found among U.S. isolates.
Most of the ORSA PFTs also contained OSSA isolates, suggesting that each of these lineages had independently acquired mecA. Five types of SCCmec have been identified. Type I (34 kb) was detected in the first ORSA strain isolated in 1961 in the United Kingdom (strain NCTC 10442) (31); type II (52 kb) was identified in a MRSA strain isolated in 1982 in Japan (strain N315) (30); type III (66 kb) was identified in an MRSA strain isolated in 1985 in New Zealand (strain 82/2082) (31); type IV (20 to 24 kb) was identified in two community-acquired ORSA strains (38) and in isolates of the Pediatric clone from Poland and Portugal (45); and a new type 2 was identified in three community onset ORSA strains from Adelaide, Australia (45). SCCmec II and III contain transposon Tn554, which encodes erythromycin and spectinomycin resistance (31). SCCmec I and IV carry no other resistance genes (31, 38).

In conclusion, results of Smal PFGE typing, corroborated with those of limited MLST and spa typing, allowed us to identify genetic backgrounds and delineate the major U.S. ORSA lineages within a national database generated from PFGE fingerprinting and epidemiologic data from the CDC and U.S. state health laboratory investigations. PFGE has proven to be more discriminating than MLST for monitoring the spread of ORSA isolates in the United States, although periodic typing of selected isolates using MLST will be critical for assessing major changes in the lineages over time.

ACKNOWLEDGMENTS

We thank Mark Enright for providing the MLST data on several of the isolates cited in this paper; we also thank Molly Kellum, Bette Jensen, Glennis Westbrook, and Loretta Carson for pulsed-field gel electrophoresis as a replacement for bacteriophage typing of ORSA 5119. We thank Mark Enright for providing the MLST data on several of the isolates cited in this paper; we also thank Molly Kellum, Bette Jensen, Glennis Westbrook, and Loretta Carson for pulsed-field gel electrophoresis as a replacement for bacteriophage typing of ORSA 5119. We thank Mark Enright for providing the MLST data on several of the isolates cited in this paper; we also thank Molly Kellum, Bette Jensen, Glennis Westbrook, and Loretta Carson for pulsed-field gel electrophoresis as a replacement for bacteriophage typing of ORSA 5119.


coccal cassette chromosome mec integrated in the chromosome in methicil-


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