Original Article

Outcomes and Treatment of Chronic Methicillin-Resistant *Staphylococcus aureus* Differs by Staphylococcal Cassette Chromosome *mec* (SCC*mec*) Type in Children With Cystic Fibrosis

Sonya L. Heltshe,[1,2] Lisa Saiman,[3,4] Elena B. Popowitch,[5,6] Melissa B. Miller,[5,6] Margaret Kloster,[2] Valeria Thompson,[2] Thomas W. Ferkol,[7,8] Wynton C. Hoover,[9] Michael S. Schechter,[10,a] and Marianne S. Muhlebach[11]

[1]Division of Pediatric Pulmonology, Department of Pediatrics, University of Washington School of Medicine, Seattle; [2]Cystic Fibrosis Foundation Therapeutics Development Network Coordinating Center, Seattle Children's Research Institute, Washington; [3]Department of Pediatrics, Columbia University Medical Center, New York; [4]Department of Infection Prevention & Control, New York-Presbyterian Hospital; Departments of [5]Medicine and, [6]Microbiology and Immunology, University of North Carolina at Chapel Hill; Departments of [7]Pediatrics, [8]Genetics, Washington University, St. Louis, Missouri; [9]Department of Pediatrics, University of Alabama at Birmingham; [10]Emory University School of Medicine, Atlanta, Georgia; and [11]Division of Pediatric Pulmonology, Department of Pediatrics, University of North Carolina at Chapel Hill

Corresponding Author: Marianne S. Muhlebach, MD, 450 MacNider, 333 S. Columbia Street, CB #7217, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599–7217. E-mail: Marianne_Muhlebach@med.unc.edu.

[a]**Present Affiliation:** Division of Pediatric Pulmonary Medicine, Children's Hospital of Richmond at Virginia Commonwealth University, Richmond, Virginia.

STAR-CF Study Team – listed alphabetically by CF center, * denotes authors who fully meet the criteria for authorship: Children's Hospital of Pittsburgh, Pennsylvania-Daniel J Weiner, MD; Columbia University, New York-Lynne Quittell, MD; Emory University, Georgia-Michael Schechter*, MD, MPH (MS relocated to VA); Riley Hospital for Children, Indiana-Michelle Howenstine, MD; St. Louis Children's Hospital, Missouri-Thomas Ferkol*, MD; University of Alabama at Birmingham, Alabama-Wynton Hoover*, MD; University of North Carolina at Chapel Hill, North Carolina-Marianne Muhlebach*, MD

Received February 20, 2014; accepted April 30, 2014; electronically published June 9, 2014.

Background. Methicillin-resistant *Staphylococcus aureus* (MRSA) infects ~25% of patients with cystic fibrosis (CF) in the United States. We hypothesized that health-related outcomes differed between healthcare-associated (staphylococcal cassette chromosome *mec* [SCC*mec*] II) vs community-associated (SCC*mec* IV) MRSA strains in patients chronically infected with CF.

Methods. At 7 CF centers, MRSA isolates were prospectively obtained from patients ≤18 years old with 2 or more positive MRSA cultures within 1 year. Isolates were classified by SCC*mec* type and Panton-Valentine-leukocidin (PVL) status at a core laboratory, and sites remained blinded to SCC*mec* type and PVL results. Prospective clinical data including antibiotic use, respiratory symptoms, and pulmonary exacerbations were obtained.

Results. Among the 295 cohort participants with typeable MRSA isolates, 69.5% had SCC*mec* II PVL(–), 13.2% had SCC*mec* IV PVL(–), and 17.3% had SCC*mec* IV PVL(+) strains. During follow-up of 287 patients with prospective data after enrollment, the risk for pulmonary exacerbations was significantly higher among participants with SCC*mec* II than SCC*mec* IV strains (risk ratio [RR] = 1.13; $P = .03$) and higher in those with SCC*mec* IV PVL(–) than SCC*mec* IV PVL(+) strains (RR = 1.62; $P < .0001$). Neither decline in lung function nor changes in nutritional outcomes differed by SCC*mec* type or PVL status during the study period.

Conclusions. Participants harboring chronic SCC*mec* II MRSA received more antibiotics and may have more lung disease than those with SCC*mec* IV; PVL(+) isolates were not associated with more advanced disease.

Key words. cystic fibrosis; methicillin-resistant *Staphylococcus aureus*; Panton-Valentine leukocidin; pulmonary exacerbation; PVL; SCC*mec* type.

BACKGROUND

The incidence and prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) has been increasing in patients with cystic fibrosis (CF) in the United States. By 2010, 25.8% of patients reported to the Cystic Fibrosis Foundation National Patient Registry had a positive respiratory culture for MRSA [1, 2]. Epidemiologic studies performed in patients with CF have suggested that lung function may be lower with MRSA compared to methicillin-susceptible *S aureus*, and chronic MRSA infection is associated with increased mortality [3–5] and morbidity [6].

Methicillin resistance is encoded by the staphylococcal cassette chromosome *mec* (SCC*mec*) gene complex. In general, healthcare-associated strains carry SCC*mec* types I–III and are negative for Panton-Valentine leukocidin (PVL), whereas strains more likely to be community-associated carry the smaller SCC*mec* IV–V elements [7–9]. However, SCC*mec* types II and IV are increasingly found in community and hospital settings, respectively [10–12]. Studies evaluating the molecular epidemiology of MRSA in patients with CF in the United States have reported approximately two-thirds of MRSA strains were SCC*mec* II [13–15]. In non-CF MRSA disease, outcomes may be different based on SCC*mec* status [16, 17]. However, outcomes associated with different SCC*mec* types have not been addressed in CF patients with chronic lung infections, and SCC*mec* status is not currently used in clinical care. Nonetheless, because SCC*mec* II encodes for more antibiotic resistance genes [18, 19], there may be a poor response to oral antibiotics or choices may be limited; thus, the question arises whether knowledge of the SCC*mec* type would be relevant to care.

We hypothesized that, among children with CF who are chronically infected with MRSA, outpatient antibiotic use, exacerbations, and lung function and growth metrics would differ by SCC*mec* type and that PVL(+) isolates would be associated with worse outcomes. We present a multicenter observational study to evaluate the clinical outcomes and treatment associated with different SCC*mec* types and PVL status in children with CF who have persistently MRSA-positive respiratory tract cultures.

METHODS

Study Design, Participants, and Sites

An observational study was conducted at 7 CF centers from different regions of the United States between 2008 and 2012. Patients diagnosed with CF [20], ≤18 years of age, with chronic MRSA infection (≥2 positive cultures within at least 1 of the 2 years before enrollment) were eligible. They were included in the study if their culture was positive for MRSA at enrollment or at 1 of the 2 subsequent CF clinic visits. Patients who had positive cultures for *Burkholderia cepacia* complex or had undergone lung transplantation were ineligible to participate. Sites were selected based on MRSA prevalence (>20%) and size (>200 pediatric patients). The study was approved by each center's Institutional Review Board, written consent was obtained from parents, and assent was obtained from children able to read.

Typing of MRSA Isolates

Participants' respiratory cultures (deep pharyngeal swab, sputum, or bronchoalveolar lavage) were processed at each site's clinical microbiology laboratory using the Cystic Fibrosis Foundation's guidelines [21]. Three colonies of MRSA from the same culture were sent to the core laboratory at the University of North Carolina to determine the SCC*mec* type and presence of the PVL gene using polymerase chain reaction methods previously described [15, 22, 23]. Treating physicians were not informed about the typing results.

Data Collection

Site investigators prospectively recorded each participant's clinic visits from patient entry into the study until April 2012 or for a maximum of 3 years of follow-up. Weight, height, spirometry as forced expiratory volume in 1 second (FEV_1), and respiratory tract culture results were recorded at visits when new antibiotics were prescribed (oral, inhaled, or intravenous [IV]) for respiratory indications. Spirometry was recorded for those participants older than 6 years of age and expressed as percentage predicted per Wang's equation [24]. Body mass index (BMI) percentile (in participants 2 years of age or older) or weight for length percentile (in participants less than 2 years of age) according to US standards [25, 26] were recorded. Organisms other than *S aureus* present in the respiratory tract cultures were collected, but antibiotic susceptibility patterns were not recorded for any CF pathogens. Antibiotic use was collected for oral, inhaled, and IV antibiotics. Oral antimicrobial agents included trimethoprim-sulfamethoxazole (TMP-SMX), fluoroquinolone agents, linezolid, tetracycline, rifampin, clindamycin, and other agents without activity against MRSA (eg, amoxicillin).

The presence of specific signs and symptoms were collected at the time of prescribing a new antibiotic to evaluate whether the event qualified as a study-defined pulmonary exacerbation [27, 28]. In brief, an exacerbation was defined as receiving new antibiotic treatment plus 1 or 2 of the 2 major criteria (≥10% decrease in FEV_1 from 6-month baseline, or oxygen saturation <90%, or 5% decline) or

2 or more of the 8 minor criteria (new or increased: cough, fatigue, chest congestion or sputum, respiratory symptoms or adventitial sounds; decreased: activity, appetite, or weight). Clinical study data were linked with the following data from the US Cystic Fibrosis Patient Registry: demographic characteristics, CF transmembrane regulator (CFTR) genotype, MRSA culture dates, and other microbiology results before enrollment, and spirometry and BMI collected for the duration of the study.

Statistical Analyses

Demographic characteristics and clinical data were described using summary statistics and compared among participants with different SCC*mec* types and PVL status using 2-sided, 0.05 alpha level *t* tests for continuous variables or Fisher's exact test for categorical variables. Cystic fibrosis clinic visits, antibiotic use, symptoms, and pulmonary exacerbations (defined above) were analyzed following enrollment by SCC*mec* type and PVL status. Oral antibiotic agents were summarized and compared as percentage of oral prescriptions, overall use, and use at the center level. Repeated measures analysis of covariance was used to compare FEV_1% predicted, BMI (or weight for length) percentile, and symptoms corresponding to antibiotic treatment adjusted for sex, age at first study visit, time since MRSA onset, study site, and presence of *Pseudomonas aeruginosa* at time of study enrollment. All available FEV_1% predicted and anthropometric percentile data from the Patient Registry were modeled linearly with a compound symmetric correlation structure. Pulmonary exacerbations were modeled as rate per year using Poisson regression adjusted for aforementioned covariates to estimate risk ratio (RR) estimates and 95% confidence intervals (CIs). SAS version 9.2 (SAS Institute, Cary, NC) was used for all analyses.

RESULTS

From October 2008 to April 2010, 314 MRSA-positive patients with a history of chronic MRSA infection were recruited from 7 CF centers. Of these, 295 participants had typeable MRSA isolates: 205 (69.5%) SCC*mec* II all PVL (–), 39 (13.2%) SCC*mec* IV PVL(–), and 51 (17.3)% SCC*mec* IV PVL(+). The final study population consisted of 287 participants for whom the date of first detection of MRSA was available in the Cystic Fibrosis Patient Registry, and prospective study data were collected.

Participant Characteristics by MRSA Type

Mean age, sex, CFTR genotypes, lung function (FEV_1% predicted), and nutritional status (BMI or weight for length percentile) were similar in participants with SCC*mec* II vs SCC*mec* IV MRSA type and among those with SCC*mec* IV PVL(+) and SCC*mec* IV PVL(–) isolates (Table 1). However, there were more participants ≥12 years of age infected with SCC*mec* II isolates. A greater percentage of participants with SCC*mec* II isolates had *P aeruginosa*-positive cultures at study enrollment (30.2% compared to 19.3% in SCC*mec* type IV), but the difference was not statistically significant ($P = .056$).

Most (71%) of the participants had 2 or more years of prospective follow-up; 91% (n = 261) had 1 year or more of follow-up, and 3.8% (n = 11) had only data collected during the enrollment visit. Duration of follow-up was comparable for participants with SCC*mec* II and SCC*mec* IV (mean [standard deviation; SD] = 2.2 [0.71] vs 2.1 [0.75] years, respectively; $P = .15$). At the time of enrollment, participants with SCC*mec* II had significantly longer duration of MRSA infection than those with SCC*mec* IV (mean = 4.7 [3.1] and 3.1 [2.5] years,

Table 1. Participant Characteristics by MRSA Type at Time of Study Enrollment

Characteristic		SCC*mec* II N = 199	SCC*mec* IV N = 88	*P* Value	SCC*mec* IV PVL(+)N = 50	SCC*mec* IV PVL(–)N = 38	*P* Value
Female	n (%)	102 (51.3%)	37 (42.1%)	0.16	21 (42.0%)	16 (42.1%)	1.0
Age in years	Mean (SD)	11.5 (4.6)	10.3 (5.0)	0.40	10.0 (5.2)	10.7 (4.6)	0.41
Age group	n (%)						
	<6 years	31 (15.6%)	16 (18.2%)	0.03*	11 (22.0%)	5 (13.2%)	0.63[a]
	6–11 years	65 (32.7%)	41 (46.6%)		22 (44.0%)	19 (50.0%)	
	≥12 years	103 (51.8%)	31 (35.2%)		17 (34.0%)	14 (36.8%)	
CFTR genotype	n (%)						
	F508 del homozygous	113 (56.8%)	43 (48.9%)	0.52*	27 (54.0%)	16 (42.1%)	0.39[a]
	F508 del heterozygous	71 (35.7%)	36 (40.9%)		18 (36.0%)	18 (47.4%)	
	Other	10 (5.0%)	7 (8.0%)		3 (6.0%)	4 (10.5%)	
	Not done/missing	5 (2.5%)	2 (2.3%)		2 (4.0%)	0 (0%)	
Pseudomonas aeruginosa	n (%)	60 (30.2%)	17 (19.3%)	0.056	12 (24.0%)	5 (13.2%)	0.20
BMI or Wt/length Percentile[†]	n: mean (SD)	176: 44.3 (27.3)	79: 46.4 (29.8)	0.34	44: 41.2 (30.1)	35: 53.0 (28.5)	0.75
FEV_1% predicted[‡]	n: mean (SD)	141: 81.9 (22.0)	64: 81.7 (22.5)	0.82	34: 77.9 (22.7)	30: 85.9 (21.9)	0.16

Abbreviations: BMI, body mass index; CFTR, cystic fibrosis transmembrane regulator; FEV_1, forced expiratory flow in 1 second; MRSA, methicillin-resistant *Staphylococcus aureus*; PVL, Panton-Valentine-leukocidin; SCC*mec*, staphylococcal cassette chromosome *mec*; SD, standard deviation; Wt/length, weight for length.

*P value for Fisher's test of distributional differences between groups.

[†]For children <2 years of age, weight for length is used in place of BMI.

[‡]FEV_1% predicted is recorded only in participants >6 years of age (see Methods).

respectively; $P < .001$). Among those with SCC*mec* IV isolates, the duration of time since initial detection of MRSA was longer in those with PVL(–) isolates (mean = 4.0 [3.0] years) than PVL(+) isolates (mean = 2.4 [1.9] years) ($P < .01$).

Clinic Visits and Antibiotic Treatment

The number of CF clinic visits and the rate of antibiotic prescriptions by SCC*mec* type and PVL status are described in Table 2. For all participants with MRSA, the mean number of clinic visits was 6.0 (SD = 2.9) visits per year and ranged from 1 to 62 clinic visits per participant during the study period. Participants with SCC*mec* IV PVL(–) isolates had more CF clinic visits per participant and per year of follow-up than those with SCC*mec* IV PVL(+) isolates.

Antibiotics (oral, additional inhaled or IV) were prescribed at 58.1% of all CF clinic visits in association with increased signs and symptoms. Oral agents were most common and were prescribed at 45.8% of visits (46.9% of visits of participants with SCC*mec* II isolates compared to 42.9% of visits of participants with SCC*mec* IV isolates; $P = .025$). Overall, the oral antibiotics TMP-SMX, fluoroquinolone agents, and linezolid were most frequently prescribed. Linezolid was used more often in those with SCC*mec* II (23.2%) compared to SCC*mec* IV (18.2%) isolates ($P = .01$) (range per center, 5.6%–34.8%). A single oral antibiotic was prescribed for 72.8% of treatment events, dual therapy was prescribed for 24.0%, and 3 or more agents were prescribed for 3.2% of events. Rifampin and clindamycin were rarely prescribed. Agents without MRSA activity were prescribed in approximately 7% of treatment events.

Symptoms were recorded at 85.6% of visits in which new antibiotics were prescribed. The number of symptoms was similar for participants with SCC*mec* II vs SCC*mec* IV isolates (mean [SD] 3.02 [1.99] vs 2.89 [2.01], respectively, adjusted; $P = .68$). The number of symptoms among SCC*mec* IV PVL(–) vs PVL(+) isolates was also similar (mean [SD] = 3.01 [2.05] vs 2.76 [1.95], respectively, adjusted; $P = .67$).

Pulmonary Exacerbations

Study criteria for a pulmonary exacerbation were met in 77.0% of events for which treating clinicians prescribed antibiotics. The annual rate of study-defined exacerbations was higher in those with SCC*mec* II isolates than those with SCC*mec* IV isolates (2.9 [95% CI, 2.7–3.0] vs 2.4 [95% CI, 2.2–2.6], respectively; $P = .0003$). After adjusting for sex, age at first MRSA detection, time since initial MRSA detection, study site, and presence of *P aeruginosa*, the risk for pulmonary exacerbations remained significantly higher among those with SCC*mec* II than among those with SCC*mec* IV isolates (adjusted RR = 1.13 [95% CI, 1.01–1.27]; $P = .03$). Among participants infected with SCC*mec* IV isolates, the rate of exacerbations was higher in those with PVL(–) isolates than those with PVL(+) isolates (annual rate 2.8 [95% CI, 2.5–3.2] vs 2.0 [95% CI, 1.7–2.3], respectively; $P = .0002$). The adjusted risk for exacerbations remained significantly higher among those with PVL(–) isolates (RR = 1.62 [95% CI, 1.28–2.04]; $P < .0001$).

Lung Function and Growth

Using all spirometry recorded in the National Patient Registry for the duration of the study (independent of antibiotic usage), the estimated annual rate of decline in

Table 2. Clinic Visits and Antibiotic Treatment by SCC*mec* Type and PVL Status

Characteristic		SCC*mec* II (N = 199)	SCC*mec* IV (N = 88)	*P* Value	SCC*mec* IV PVL(+) (N = 50)	SCC*mec* IV PVL(–) (N = 38)	*P* Value
Clinic visits/participant	Mean (SD)	13.3 (7.6)	11.8 (7.6)	.15	9.9 (4.5)	14.3 (9.9)	**.014**
Clinic visits/year follow-up	Mean (SD)	6.1 (3.0)	5.8 (2.6)	.40	5.3 (2.0)	6.4 (3.1)	**.05**
Visits with inhaled, oral, or IV antibiotics prescribed	n (%)	1642 (60.6%)	549 (51.8%)	**<.001**	271 (50.1%)	278 (53.4%)	.28
Prescriptions of TMP-SMX*	%	33.0	35.0	.37	31.0	39.2	**.049**
Prescriptions of fluoroquinolone*	%	23.4	25.9	.25	22.9	28.8	.12
Prescriptions of linezolid*	%	23.2	18.2	**.01**	20.7	15.8	.15
Prescriptions of tetracycline*	%	8.6	7.3	.37	11.4	3.2	**<.001**
Prescriptions of rifampin*	%	4.7	3.8	.47	5.6	2.2	**.046**
Prescriptions of clindamycin*	%	0.2	1.6	**<.001**	0.7	2.5	.18
Prescriptions of non-MRSA active oral antibiotic*,†	%	6.9	8.0	.39	7.8	8.3	.88

Abbreviations: IV, intravenous; MRSA, methicillin-resistant *Staphylococcus aureus*; PVL, Panton-Valentine-leukocidin; SCC*mec*, staphylococcal cassette chromosome *mec*; SD, standard deviation; TMP-SMX, trimethoprim-sulfamethoxazole. Bold text indicates *P* Value <0.05.

*Expressed as a percentage of oral antibiotics prescribed.

†These were antibiotics classically not considered active for MRSA (eg, amoxicillin).

FEV_1% predicted was not significantly different between SCC*mec* types ($P = .73$): −1.8% (95% CI: −3.5, −0.1) for SCC*mec* II compared to −1.2% for SCC*mec* IV (95% CI: −4.1, 1.7) adjusted for sex, age at first study visit, time since MRSA onset, study site, and presence of *P aeruginosa*. Among those with SCC*mec* IV, PVL was not associated with differing rates of decline in FEV_1% predicted ($P = .88$).

Likewise, changes in BMI/weight for length percentile did not differ between the SCC*mec* types ($P = .74$): −0.6% per year (95% CI: −2.9, 1.9) for SCC*mec* II compared to −1.3% per year for SCC*mec* IV (95% CI, −5.1, 2.5). Body mass index/weight for length percentile per year did not differ by PVL status ($P = .97$).

DISCUSSION

This is the first multicenter study to analyze the association between chronic infection with different MRSA SCC*mec* types with clinical outcomes in patients with CF. At the time of enrollment into this study, those participants with SCC*mec* II isolates were older, had a longer duration of MRSA infection, and were more likely to be coinfected with *P aeruginosa*. We demonstrated that, when adjusting for these and other covariates known to impact CF lung disease, the rate of exacerbations was higher in participants with SCC*mec* II isolates compared to those with SCC*mec* IV isolates. Furthermore, among those with SCC*mec* IV isolates, the rate of exacerbations was higher in those with PVL(−) isolates than those with PVL(+) isolates. Likewise, the number of CF clinic visits per participant and per year of follow-up was greater in those infected with PVL(−) isolates. Notably, treating clinicians did not know the SCC*mec* type infecting their patients; thus, treatment decisions were made without consideration of MRSA type. Forty-four percent of our SCC*mec* IV isolates were PVL negative, which allowed comparison within this group by PVL status. Previous reports in CF showed worse presentation at MRSA onset in PVL(+) MRSA isolates in a case series [29]. Furthermore, PVL has been postulated to be the cause of necrotizing pneumonia in previously healthy people [30]. However, these earlier observations have been questioned in other studies [18]. Our findings do not lend support to PVL(+) isolates causing worse disease in patients with CF.

Although an observational study cannot determine causality, there are several possible explanations for the different outcomes related to PVL status and SCC*mec* type. Contrary to our hypothesis that PVL positivity would be associated with worse outcomes, we found that SCC*mec* IV PVL(−) and SCC*mec* II [all PVL(−)] isolates were associated with more overall CF clinic visits and with more visits at which antibiotics were prescribed, respectively. These outcomes may be affected by bacterial and host aspects in a multifactorial manner. Bacterial factors related to *S aureus* type with potential impact include antibiotic susceptibilities, virulence factors, or bacterial fitness. Although some non-CF studies show that a higher vancomycin minimal inhibitory concentration has been associated with worse outcomes [31], we did not see vancomycin-resistant or vancomycin-intermediate isolates in this study [22]. Other bacterial factors were not assessed in this study and would only be speculative. In addition, outcomes observed here may be related to host characteristics rather than bacterial virulence. Observational studies of invasive MRSA infections in the United States have reported that the underlying disease severity of the infected individual, rather than the SCC*mec* type, was the main risk factor associated with mortality [17, 32].

We had hypothesized that the greater number of antibiotic resistance genes in SCC*mec* II MRSA would impact the choice of antibiotic therapy used to treat pulmonary exacerbations. We have previously shown that the SCC*mec* II strains were more often resistant to clindamycin and ciprofloxacin compared with the SCC*mec* IV strains [22]. We found that linezolid was used more often in those with SCC*mec* type II compared to SCC*mec* IV, which may be reflective of these differences in antimicrobial susceptibilities. However, this antibiotic should be used with caution because resistance can emerge [33–35], and it may be associated with potentially irreversible side effects including peripheral neuropathy. Our study definition of exacerbations included the use of oral agents as well as IV antibiotics; this definition allowed assessment of all exacerbations and reduced the potential bias toward more severe events and only very resistant isolates, which were expected to be more frequent in SCC*mec* II strains.

We acknowledge some limitations of this study. First, only 1 culture was used for classification of MRSA type; however, previous studies, including our own, have shown persistence of the same strain over time [36–38]. In addition, although this was a multicenter study, inclusion was limited to children, and it is unknown whether these findings are generalizable to adults, or to CF centers that have a prevalence of MRSA less than 20%, or to children with CF outside the United States, where different strains may be prevalent. Duration of follow-up was variable for the participants; however, there were not significant differences between groups, and our analysis accounted for follow-up duration; therefore, results are unlikely biased. Longitudinal spirometry and weight for length/BMI were compared, and no differences by

SCC*mec* type or PVL were observed, despite differences in pulmonary exacerbations. We speculated that these observations may reflect several factors related to this study design and population. The FEV_1% predicted and BMI percentiles in the Patient Registry had high a within-patient variation. In addition, 16% of the population was too young to reliably perform spirometry at study entry, whereas they were included in the exacerbation analysis. Furthermore, published studies of the association between lung function decline and exacerbations typically define exacerbations as events when IV antibiotics were used. In this study, however, exacerbations were also defined when oral antibiotics were used, which may have prevented lung function decline. The estimated rate of FEV_1 decline in this cohort was consistent with previously reported declines in lung function among CF patients persistently infected with MRSA [5]. Another possible source of bias is the fact that patient signs and symptoms were only collected when new antibiotics were prescribed, both to limit reporting burden to the sites and because we considered symptoms not associated with treatment to be relatively minor in scope. However, in this pediatric population, there may be an overall lower threshold for treating as seen by use of antibiotics in 23% of cases that did not meet exacerbation criteria. Lastly, we chose not to collect antibiotic susceptibilities for isolates tested at each visit and each study site to avoid variation in techniques and results; thus, study outcomes were not analyzed by success or failure of antibiotic course.

CONCLUSIONS

In conclusion, we show that children with CF chronically infected with SCC*mec* II MRSA isolates have higher exacerbation rates and use of oral antibiotics compared with those with SCC*mec* IV and that PVL(+) status was not associated with worse outcome. Although an epidemiologic study cannot address causality of higher exacerbation rates, our results do raise the following questions: (1) would routine clinical typing be advantageous to clinicians treating patients chronically infected with MRSA? and (2) are further, prospective studies that include subtyping warranted? The answers to these questions could allow us to more precisely direct care and to determine whether an eradication strategy should be favored based on *S aureus* type. Currently, data showing the benefit of early eradication for any MRSA type in CF are not available, but studies examining the clinical effect of eradication are in progress.

Acknowledgments

We acknowledge Emily Knapp and the Cystic Fibrosis Foundation for providing patient-specific data from the US Cystic Fibrosis Patient Registry. We also gratefully acknowledge the anonymous referees whose thoughtful reviews improved this manuscript.

Author contributions. The listed authors provided direct contribution to this manuscript. Investigators from the other study STAR-CF sites were involved in sample collection, study-related conference calls, and read the manuscript.

Financial support. This work was supported by Cystic Fibrosis Foundation Therapeutics (Grant MUHLEB08A0; to M. S. M., S. L. H., M. K., V. T., and study sites) and the National Institutes of Health National Institute of Diabetes and Digestive and Kidney Diseases (Grant P30DK089507; to S. L. H.).

***Potential conflicts of interest*:** M. S. M. has received research funding from Cystic Fibrosis Foundation for this project as stated in the financial support statement. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Razvi S, Quittell L, Sewall A, et al. Respiratory microbiology of patients with cystic fibrosis in the United States, 1995–2005. Chest **2009**; 8:8.
2. CFF-Registry. Available at: http://www.cff.org/UploadedFiles/research/ClinicalResearch/2011-Patient-Registry.pdf. Accessed 2 June 2014.
3. Ren CL, Morgan WJ, Konstan MW, et al. Presence of methicillin resistant *Staphylococcus aureus* in respiratory cultures from cystic fibrosis patients is associated with lower lung function. Pediatr Pulmonol **2007**; 42:513–8.
4. Dasenbrook EC, Checkley W, Merlo CA, et al. Association between respiratory tract methicillin-resistant *Staphylococcus aureus* and survival in cystic fibrosis. JAMA **2010**; 303:2386–92.
5. Dasenbrook EC, Merlo CA, Diener-West M, et al. Persistent methicillin-resistant *Staphylococcus aureus* and rate of FEV_1 decline in cystic fibrosis. Am J Respir Crit Care Med **2008**; 178: 814–21.
6. Sanders DB, Bittner RC, Rosenfeld M, et al. Failure to recover to baseline pulmonary function after cystic fibrosis pulmonary exacerbation. Am J Respir Crit Care Med **2010**; 182:627–32.
7. Ito T, Katayama Y, Asada K, et al. Structural comparison of three types of staphylococcal cassette chromosome mec integrated in the chromosome in methicillin-resistant *Staphylococcus aureus*. Antimicrob Agents Chemother **2001**; 45:1323–36.
8. Okuma K, Iwakawa K, Turnidge JD, et al. Dissemination of new methicillin-resistant *Staphylococcus aureus* clones in the community. J Clin Microbiol **2002**; 40:4289–94.
9. Tenover FC, McAllister S, Fosheim G, et al. Characterization of *Staphylococcus aureus* isolates from nasal cultures collected from individuals in the United States in 2001 to 2004. J Clin Microbiol **2008**; 46:2837–41.
10. Maree CL, Daum RS, Boyle-Vavra S, et al. Community-associated methicillin-resistant *Staphylococcus aureus* isolates causing healthcare-associated infections. Emerg Infect Dis **2007**; 13:236–42.
11. Miller LG, Perdreau-Remington F, Bayer AS, et al. Clinical and epidemiologic characteristics cannot distinguish community-associated methicillin-resistant *Staphylococcus aureus* infection from methicillin-susceptible *S. aureus* infection: a prospective investigation. Clin Infect Dis **2007**; 44:471–82.
12. Knight GM, Budd EL, Whitney L, et al. Shift in dominant hospital-associated methicillin-resistant *Staphylococcus aureus*

(HA-MRSA) clones over time. J Antimicrob Chemother **2012**; 67:2514–22.
13. Stone A, Quittell L, Zhou J, et al. *Staphylococcus aureus* nasal colonization among pediatric CF patients and their household contacts. Pediatr Infect Dis J **2009**; 28:895–9.
14. Glikman D, Siegel JD, David MZ, et al. Complex molecular epidemiology of methicillin-resistant *Staphylococcus aureus* isolates from children with cystic fibrosis in the era of epidemic community-associated methicillin-resistant *S aureus*. Chest **2008**; 133:1381–7.
15. Goodrich JS, Sutton-Shields TN, Kerr A, et al. Prevalence of community-associated methicillin-resistant *Staphylococcus aureus* in patients with cystic fibrosis. J Clin Microbiol **2009**; 47:1231–3.
16. Chen SY, Liao CH, Wang JL, et al. Methicillin-resistant *Staphylococcus aureus* (MRSA) staphylococcal cassette chromosome mec genotype affects outcomes of patients with healthcare-associated MRSA bacteremia independently of vancomycin minimum inhibitory concentration. Clin Infect Dis **2012**; 55:1329–37.
17. Haque NZ, Arshad S, Peyrani P, et al. Analysis of pathogen and host factors related to clinical outcomes in patients with hospital-acquired pneumonia due to methicillin-resistant *Staphylococcus aureus*. J Clin Microbiol **2012**; 50:1640–4.
18. Voyich JM, Otto M, Mathema B, et al. Is Panton-Valentine leukocidin the major virulence determinant in community-associated methicillin-resistant *Staphylococcus aureus* disease? J Infect Dis **2006**; 194:1761–70.
19. Wehrhahn MC, Robinson JO, Pascoe EM, et al. Illness severity in community-onset invasive *Staphylococcus aureus* infection and the presence of virulence genes. J Infect Dis **2012**; 205:1840–8.
20. Rosenstein BJ. What is a cystic fibrosis diagnosis? Clin Chest Med **1998**; 19:423–41.
21. Miller M, Gilligan PH. Laboratory aspects of management of chronic pulmonary infections in patients with cystic fibrosis. J Clin Microbiol **2003**; 41:4009–15.
22. Champion EA, Miller MB, Popowitch EB, et al. Antimicrobial susceptibility and molecular typing of MRSA in cystic fibrosis. Pediatr Pulmonol **2014**; 49:230–7.
23. Oliveira DC, de Lencastre H. Multiplex PCR strategy for rapid identification of structural types and variants of the mec element in methicillin-resistant *Staphylococcus aureus*. Antimicrob Agents Chemother **2002**; 46:2155–61.
24. Wang X, Dockery DW, Wypij D, et al. Pulmonary function between 6 and 18 years of age. Pediatr Pulmonol **1993**; 15:75–88.
25. Kuczmarski RJ, Ogden CL, Grummer-Strawn LM, et al. CDC growth charts: United States. Adv Data **2000**; (314):1–27.
26. Flegal KM, Wei R, Ogden C. Weight-for-stature compared with body mass index-for-age growth charts for the United States from the Centers for Disease Control and Prevention. Am J Clin Nutr **2002**; 75:761–6.
27. Rosenfeld M, Emerson J, Williams-Warren J, et al. Defining a pulmonary exacerbation in cystic fibrosis. J Pediatr **2001**; 139: 359–65.
28. Saiman L, Anstead M, Mayer-Hamblett N, et al. Effect of azithromycin on pulmonary function in patients with cystic fibrosis uninfected with *Pseudomonas aeruginosa*: a randomized controlled trial. JAMA **2010**; 303:1707–15.
29. Elizur A, Orscheln RC, Ferkol TW, et al. Panton-Valentine leukocidin-positive methicillin-resistant *Staphylococcus aureus* lung infection in patients with cystic fibrosis. Chest **2007**; 131: 1718–25.
30. Tseng MH, Wei BH, Lin WJ, et al. Fatal sepsis and necrotizing pneumonia in a child due to community-acquired methicillin-resistant *Staphylococcus aureus*: case report and literature review. Scand J Infect Dis **2005**; 37:504–7.
31. Haque NZ, Zuniga LC, Peyrani P, et al. Relationship of vancomycin minimum inhibitory concentration to mortality in patients with methicillin-resistant *Staphylococcus aureus* hospital-acquired, ventilator-associated, or health-care-associated pneumonia. Chest **2010**; 138:1356–62.
32. Lessa FC, Mu Y, Ray SM, et al. Impact of USA300 methicillin-resistant *Staphylococcus aureus* on clinical outcomes of patients with pneumonia or central line-associated bloodstream infections. Clin Infect Dis **2012**; 55:232–41.
33. Hill RL, Kearns AM, Nash J, et al. Linezolid-resistant ST36 methicillin-resistant *Staphylococcus aureus* associated with prolonged linezolid treatment in two paediatric cystic fibrosis patients. J Antimicrob Chemother **2010**; 65:442–5.
34. Endimiani A, Blackford M, Dasenbrook EC, et al. Emergence of linezolid-resistant *Staphylococcus aureus* after prolonged treatment of cystic fibrosis patients in Cleveland, Ohio. Antimicrob Agents Chemother **2011**; 55:1684–92.
35. Gu B, Kelesidis T, Tsiodras S, et al. The emerging problem of linezolid-resistant *Staphylococcus*. J Antimicrob Chemother **2013**; 68:4–11.
36. Kahl BC, Duebbers A, Lubritz G, et al. Characteristics of *Staphylococcus aureus*, isolated from airways of cystic fibrosis patients, and their small colony variants. J Clin Microbiol **2003**; 41:4424–7.
37. Maslow JN, Brecher S, Gunn J, et al. Variation and persistence of methicillin-resistant *Staphylococcus aureus* strains among individual patients over extended periods of time. Eur J Clin Microbiol Infect Dis **1995**; 14:282–90.
38. Branger C, Gardye C, Lambert-Zechovsky N. Persistence of *Staphylococcus aureus* strains among cystic fibrosis patients over extended periods of time. J Med Microbiol **1996**; 45: 294–301.