SHORT REPORT

Prevalence of community-associated methicillin-resistant Staphylococcus aureus colonization outside the healthcare environment

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SUMMARY

Community-associated methicillin-resistant *Staphylococcus aureus* (CA-MRSA) infections are increasingly recognized in persons without established risk factors. Population-based prevalence studies of CA-MRSA colonization in persons without risk factors are relatively limited. Subjects aged 2–65 years were enrolled from a student recreation centre, public office building, and out-patient clinics. Persons or close contacts with a history of hospitalization, nursing-home residence, surgery, emergency-department visit, or healthcare employment during the previous year and persons with chronic debilitating illness, indwelling catheter, or surgical device were excluded. Swabs of anterior nares were obtained. Demographic and clinical information was collected. During January–June 2005, three (1·2%) of 259 subjects were colonized with MRSA. All three subjects were adults enrolled at the recreation centre. Healthy persons living in households without recent exposure to healthcare environments were at low risk for MRSA colonization. Studies from other geographic locations are needed to elucidate differences in prevalence of CA-MRSA.

Key words: Community-associated, methicillin resistance, prevalence, Staphylococcus aureus.

Methicillin-resistant *Staphylococcus aureus* (MRSA) infections are being increasingly recognized in persons without exposure to factors traditionally associated with these infections (e.g. hospitalization or nursinghome residence, surgery, dialysis, presence of indwelling catheters, or history of such percutaneous devices as tracheostomy tube, gastrostomy tube, or Foley catheter) [1, 2]. These community-associated MRSA (CA-MRSA) infections have been reported in a variety of different populations including prison inmates, Alaskan natives, American Indians, Pacific Islanders, sports participants, and military personnel

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[3–8]. Studies have reported considerable morbidity and mortality associated with CA-MRSA infections with manifestations similar to those from infections caused by susceptible *S. aureus* [2, 9]. Consistent with what is now commonly observed in different regions, a recent survey demonstrated that the majority of community-associated *S. aureus* infections were methicillin-resistant, and the predominant site of involvement is skin and soft tissues [9].

Persons who are carriers of methicillin-susceptible and methicillin-resistant *S. aureus* are at risk for subsequent infections with these organisms [10, 11]. Along with the increasing recognition of symptomatic CA-MRSA infections is the observation that asymptomatic MRSA colonization in persons living in the

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community is increasing as well. Studies performed in healthy children presenting for health maintenance clinic visits at Nashville, Tennessee, demonstrated a MRSA colonization rate of 9.2% in 2004, compared with 0.8% in 2001 [12]. In the National Health and Nutrition Examination Survey (NHANES) conducted between 2001 and 2002, which is a representative sample of civilian and non-institutionalized US population, the prevalence of MRSA nasal colonization was 0.8% [13]. However, studies that have estimated the prevalence of MRSA colonization have typically included subjects with potential exposure to a healthcare environment (e.g. emergency-department visits, hospitalizations, or employment in healthcare), or have included subjects who have household members with frequent healthcare exposure [12–15]. As a result, these analyses might overestimate the prevalence of MRSA associated with contact outside the healthcare environment. We sought to determine the prevalence of MRSA colonization in healthy persons who were living in households without recent exposure to a healthcare environment.

During January–June 2005, a descriptive crosssectional study was implemented in New Orleans, Louisiana. Subjects aged 2-65 years were recruited from selected locations, including a university-based sports recreation centre, a public office building, and two outpatient clinics, including an inner-city paediatric clinic for indigent persons and a group practice serving mainly privately insured adults. Volunteers were excluded if they themselves or someone in whom they were in close contact (defined as a minimum 4 h/day for a minimum of 5 days/week), had within the past 12 months a history of hospitalization, nursing-home residence, in-patient or outpatient surgery, emergency-department visit, or employment in healthcare. Subjects with a history of chronic debilitating illnesses (e.g. cirrhosis, connective tissue disorders, immunosuppression, diabetes mellitus, or burns) and those with a history of dialysis, indwelling intravenous catheters, gastrostomy or tracheostomy tubes, or other surgical devices were also excluded. Persons with asthma or hypertension were not excluded unless they had one of the previously listed exclusion criteria.

After administration of an initial set of screening questions, subjects were invited to participate. Subjects or legal guardians were administered additional questions related to demographics, residence, occupation, insurance, child care, sporting activities, incarceration, illicit drug use, and medical history,

including a history of boils, other skin infections, and medications. Subsequently, each participant had the distal part of the anterior nares sampled for culture with the circular movement (five rotations on either side) of a CulturetteTM swab (Becton, Dickinson and Company; USA), moistened with sterile saline. Subjects were informed of the results of their culture when positive for MRSA. This study was approved by the Institutional Review Board of Tulane University Health Sciences Center.

Samples were transported on BD BBLTM Culture-SwabTM Liquid Stuart Transport system (Becton, Dickinson and Company). Remel's mannitol salt agar (Remel, USA) with $4 \mu g/ml$ oxacillin was used as a selective and differential medium for the recovery of staphylococci from primary cultures. Swabs were rolled over a small area of the agar surface and streaked for isolation. Plates were incubated aerobically at 35-37 °C for 24-48 h and observed for characteristic colony morphology and colour change. A coagulase test was performed to differentiate S. aureus from coagulase-negative staphylococci (CNS). The oxacillin salt-agar screening plate procedure was used for detection of MRSA. The test was performed by inoculating a S. aureus isolate onto Mueller-Hinton agar that had been supplemented with NaCl (4% w/v, 0.68% mol/l) and that contained 6 μg oxacillin/ml. The agar was inoculated from a direct colony suspension equivalent to a 0.5 McFarland standard by using either a 1-µl loop or a swab. By using a 1- μ l loop, the inoculum was spread in an area 10-15 mm in diameter. The plate was incubated at temperatures no higher than 35 °C for 24 h and examined carefully with transmitted light for evidence of small colonies (i.e. >1 colony) or a light film of growth indicating oxacillin resistance. Etest® (AB Biodisk, Sweden) was used for antibiotic susceptibility testing of MRSA. Clinical Laboratory Standards Institute (CLSI; formerly known as NCCLS) interpretive criteria for susceptibility categorization were used.

Pulsed-field gel electrophoresis (PFGE) was performed at the Louisiana Office of Public Health microbiology laboratory, by following the recommendations of the Centers for Disease Control and Prevention (CDC) [16]. The banding patterns of chromosomal DNA fragments resulting from restriction endonuclease cleavage were compared to determine the relatedness of MRSA [17].

A total of 490 subjects volunteered to be screened for the study and 259 (52·8%) were enrolled. The remainder (47·2%) met one of the exclusion criteria

as noted below. The majority of the subjects who were screened (233/490, 47.5%) and enrolled (133/259, 51.3%) were interviewed at the student recreation centre. The remainder of subjects screened included 160 (32.6%) subjects screened at the public office building, and 97 (19.8%) at outpatient clinics. Of the 259 subjects enrolled in the study, 83 (32%) were enrolled at the public office building and 43 (16.6%) at the outpatient clinics. The most common personal reason for exclusion from the study was having had an emergency-department visit (43.9%), followed by employment in healthcare (16.3%), chronic illness (14.2%), hospital admission (13.7%), surgical procedure (11.6%), indwelling medical device (4.7%), and nursing-home admission (1.2%), all during the 12 months before enrolment. Overall, 21% of the excluded subjects had more than one personal reason for exclusion; 26% had no personal reason for exclusion but were excluded because of contact with a household member with a healthcare exposure.

The subjects' baseline characteristics are detailed in Table 1. The majority of the subjects were female (54·4%), white (62·2%), aged 18–65 years (87·3%), and medically insured (92·7%). The most common occupation was student (42·9%). Approximately one third had taken an antibiotic, and 7·7% reported having had boils or sores during the previous year. Three subjects reported having used illicit intravenous (i.v.) drugs, two within the previous year, and two had been incarcerated in prison. The majority had visited a doctor, and one third reported having had an infection during the previous year. Approximately one fifth had participated in team sports at least once a week.

Only 3/259 subjects (1·2%, 95% confidence interval 0·2–3·3) were colonized with MRSA. All three were enrolled at the student recreation centre. One subject was a male aged 20 years who had used illicit i.v. and non-i.v. drugs during the previous year and had boils and sores during the previous 6 months. Another subject reported having taken antibiotics during the previous year. The third had no identifiable risk factor. Seventy-two (27·8%) subjects were colonized with methicillin-resistant CNS.

All three MRSA isolates were sensitive to vancomycin, linezolid, clindamycin, and trimethoprim sulfamethoxazole, and two of three were sensitive to erythromycin. PFGE patterns of the three isolates revealed that they were unrelated.

CA-MRSA infections have emerged worldwide with noticeable differences from hospital-associated

Table 1. Baseline characteristics of subjects (n = 259), New Orleans, Louisiana, 2005

Characteristic	No. (%)
Sex	
Female	141 (54·4)
Male	117 (45·2)
Age (yr)	
2–17	33 (12·7)
18–65	226 (87·3)
Race	
White	161 (62·2)
Black	65 (25·1)
Other	33 (12·7)
Occupation	444 (42.0)
Student	111 (42.9)
Professional	77 (29·7)
Annual salary (\$)	
< 18 000	81 (31·3)
> 72 000	71 (27·4)
Medically insured	240 (92·7)
Number in household	
1–4	215 (83)
5–8	33 (12·7)
Child care	10 (30·3)*
Residence	
House	137 (53·1)
Apartment	100 (38.8)
Homeless	0
Antibiotics (during previous year)	94 (36.6)
Boils or sores (during previous year)	20 (7.7)
Infection (during previous year)	87 (33.6)
Doctor visit (during previous year)	205 (79·5)
Illicit non-intravenous drug use	
Ever	79 (30.6)
During previous year	38 (14·7)
Illicit intravenous drug use	
Ever	3 (1·2)
During previous year	2 (0.8)
Prison	
Ever	2 (0.8)
During previous year	0
Team sport participant	
Daily or more than once a week	55 (21·3)
Never	113 (43.8)

^{*} Percentage of paediatric subjects (n=33).

MRSA infections with respect to the majority of common clinical presentations, antibiotic susceptibilities, and molecular analyses [1, 18]. This has alerted public health professionals and infection-control practitioners to assess the prevalence of these infections in the community. A recently published

population-based study conducted to assess the burden of CA-MRSA disease reported an annual incidence of 25·7 cases/100 000 population in Atlanta, Georgia, and 18·0 cases/100 000 in Baltimore, Maryland [1]. Children aged <2 years at both geographic locations and blacks in Atlanta had substantially higher incidence of disease, compared with persons aged >2 years and whites, respectively.

In the majority of the studies conducted to assess asymptomatic carriage of MRSA in otherwise healthy persons, cultures were not obtained from sites outside the healthcare environment, possibly because of the lack of a clear definition of what should be labelled community-associated or community-acquired [12–15]. A pooled analysis of the studies that performed surveillance cultures for community-acquired MRSA carriage demonstrated that persons from whom samples had been obtained within the healthcare environment (e.g. emergency departments, clinics, or hospitals) were 2·35 times more likely to be colonized with MRSA, compared with those from whom cultures had been obtained outside the healthcare environment [15].

In this study, we identified a low overall prevalence of 1·2% in these subjects, which is contrary to the common belief among healthcare practitioners. Because of the detection of nasal carriage in only three subjects, we were unable to identify any risk factors for colonization with MRSA in this study. Interestingly, however, MRSA colonization was detected in one of the two subjects who reported i.v. drug use during the previous year. Injecting drug use has been cited as a risk factor for both colonization as well as disease with CA-MRSA [14].

We explored multiple factors that have been associated with colonization or infection with CA-MRSA. Investigators have examined previous antibiotic use and have identified conflicting results. Multiple studies have reported previous antibiotic use as a risk factor for MRSA colonization and infection [19]. Conversely, others have identified no association with prior antibiotic use [8]. Approximately one third of our subjects reported having taken an antibiotic in the previous year. In the worldwide dissemination of CA-MRSA, the relative significance of prior antibiotic therapy might be minimal.

Outbreaks of MRSA infections have occurred in persons participating in competitive and team sports, and team members have been identified as having asymptomatic colonization with MRSA [7]. Approximately half of the subjects in this study were

enrolled at the sports recreation centre, and one fifth had participated in team sports at least once a week. Of the 55 subjects who reported participating in team sports at least once a week, two (3.6%) were colonized with MRSA, a rate lower than the 8% that had been reported in a previous study of professional football players [7]. Possibly, the close clustering and specific practices of sharing utensils did not exist in our population, who would be expected to participate in team sports in a less structured environment.

Using a stringent screening criterion enabled us to estimate with greater authority the prevalence of MRSA that might have been acquired with contact in the community. Conversely, we might have underestimated the true prevalence of CA-MRSA or community-acquired MRSA by excluding approximately half of the subjects in this study.

Another explanation for a low prevalence of MRSA in our study might be inappropriate culture technique employed in the study. Broth enrichment along with agar plating might have increased detection rates of MRSA from swab specimens. Culturing sites in addition to nares might have increased detection rates of MRSA colonization [20]. Although we did not culture for methicillin-susceptible *S. aureus*, we identified 27·8% of subjects to be colonized with methicillin-resistant CNS. This is consistent with the results of a limited number of studies assessing methicillin-resistant CNS colonization rates [21].

This study had certain limitations. First, confirmation of resistance to methicillin and other β -lactam antibiotics by testing for the mecA gene on isolates was not performed in the study. Approximately half of the subjects were enrolled at the sports recreation centre, thereby representing young adults who were mainly college students, a group that might not be representative of the general population. Second, we enrolled subjects from clinics; however, only 10 adults and 33 children were enrolled from clinic sites. A paediatric clinic was chosen as an enrolment site for convenience and to avoid logistic and legal hurdles associated with enrolling children at more open sites, where identification and verification of subjects legally authorized to sign consent for minors may not be entirely possible. Using strict screening minimized the possibility of enrolling subjects with hospital exposure. On the basis of previous experience, we had expected to identify a higher colonization rate in persons enrolled in the study at the clinics. Last, a larger sample size might have enabled us to narrow the confidence interval of our results; however, this was beyond the resources available for this study.

In conclusion, we identified a low prevalence of asymptomatic colonization in healthy children and adults in New Orleans prior to hurricane Katrina. The Louisiana Office of Public Health plans to continue with similar studies in specific population groups, at potentially elevated risk (e.g. prison inmates, police officers, armed forces, school personnel) to more accurately describe the prevalence of MRSA in New Orleans. Additional population-based sampling from other geographic locations is needed to elucidate the differences in regional prevalence.

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DECLARATION OF INTEREST

None.

REFERENCES

- 1. **Fridkin SK**, *et al*. Methicillin-resistant *Staphylococcus aureus* disease in three communities. *New England Journal of Medicine* 2005; **352**: 1436–1444.
- 2. Naimi TS, et al. Comparison of community- and health care-associated methicillin-resistant Staphylococcus aureus infection. Journal of the American Medical Association 2003; 290: 2976–2984.
- Centers for Disease Control and Prevention. Methicillinresistant Staphylococcus aureus infections in correctional facilities – Georgia, California, and Texas, 2001– 2003. Morbidity and Mortality Weekly Report 2003; 52: 992–996.
- Baggett HC, et al. Community-onset methicillinresistant Staphylococcus aureus associated with antibiotic use and the cytotoxic panton-valentine leukocidin during a furunculosis outbreak in rural Alaska. Journal of Infectious Diseases 2004; 189: 1565– 1573.
- Centers for Disease Control and Prevention. Methicillinresistant Staphylococcus aureus infections in Pacific Islanders – Hawaii, 2001–2003. Morbidity and Mortality Weekly Report 2004; 53: 767–770.
- 6. Nguyen DM, Mascola L, Bancroft E. Recurring methicillin-resistant *Staphylococcus aureus* infections in a football team. *Emerging Infectious Diseases* 2005; 11: 526–532.

- Ellis MW, et al. Natural history of community-acquired methicillin-resistant Staphylococcus aureus colonization and infection in soldiers. Clinical Infectious Diseases 2004; 39: 971–979.
- 8. Gonzalez BE, *et al.* Severe staphylococcal sepsis in the era of community-acquired methicillin-resistant *Staphylococcus aureus*. *Pediatrics* 2005; 115: 642–648.
- Kaplan SL, et al. Three-year surveillance of communityacquired Staphylococcus aureus infections in children. Clinical Infectious Diseases 2005; 40: 1785–1791.
- Eiff CV, et al. Nasal carriage as a source of Staphylococcus aureus bacteremia. New England Journal of Medicine 2001; 344: 11–16.
- Huang SS, Platt R. Risk of methicillin-resistant Staphylococcus aureus infection after previous infection or colonization. Clinical Infectious Diseases 2003; 36: 281–285.
- Creech II CB, et al. Increasing rates of nasal carriage of methicillin-resistant Staphylococcus aureus in healthy children. Pediatric Infectious Disease Journal 2005; 24: 617–621
- Kuehnert MJ, et al. Prevalence of Staphylococcus aureus nasal colonization in the United States, 2001– 2002. Journal of Infectious Diseases 2006; 193: 172– 179.
- 14. Charlebois ED, et al. Population based community prevalence of methicillin-resistant Staphylococcus aureus in the urban poor of San Francisco. Clinical Infectious Diseases 2002; 34: 425–433.
- Salgado CD, Farr BM, Calfee DP. Communityacquired methicillin-resistant *Staphylococcus aureus*: a meta-analysis of prevalence and risk factors. *Clinical Infectious Diseases* 2003; 36: 131–139.
- 16. Centers for Disease Control and Prevention. Oxacillinresistant *Staphylococcus aureus* on PulseNet (OPN): laboratory protocol for molecular typing of *S. aureus* by pulsed-field gel electrophoresis (PFGE). Atlanta, GA: US Department of Health and Human Services, Centers for Disease Control and Prevention, 2001.
- Tenover FC, et al. Interpreting chromosomal DNA restriction patterns produced by pulsed-field gel electrophoresis: criteria for bacterial strain typing. *Journal of Clinical Microbiology* 1995; 33: 2233–2239.
- 18. Vandenesch F, et al. Community-acquired methicillinresistant Staphylococcus aureus carrying pantonvalentine leukocidin genes: worldwide emergence. Emerging Infectious Diseases 2003; 9: 978–984.
- Baggett HC, et al. An outbreak of communityonset methicillin-resistant Staphylococcus aureus skin infections in southwestern Alaska. Infection Control and Hospital Epidemiology 2003; 24: 397–402.
- Suggs AH, et al. Methicillin-resistant and borderline methicillin-resistant Staphylococcus aureus colonization in children without identifiable risk factors. Pediatric Infectious Disease Journal 1999; 18: 410– 414.
- 21. **Hisata K**, *et al.* Dissemination of methicillin-resistant *Staphylococci* among healthy Japanese children. *Journal of Clinical Microbiology* 2005; **43**: 3364–3372.