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ORIGINAL RESEARCH

Dose-response associations of methicillinresistant *Staphylococcus aureus* between school environmental contamination and nasal carriage by elementary students

Jialing Lin¹ Jianping Liang² Ting Zhang¹ Chan Bai¹ Jiaping Ye¹ Zhenjiang Yao¹

¹Department of Epidemiology and Health Statistics, School of Public Health, Guangdong Pharmaceutical University, Guangzhou, China; ²Department of Health Education, Guangzhou Primary and Secondary School Health Promotion Center, Guangzhou, China

Correspondence: Zhenjiang Yao Department of Epidemiology and Health Statistics, Guangdong Pharmaceutical University, 283 Jianghai Street, Guangzhou 510310, China Tel +86 20 3405 5806 Fax +86 20 3405 5355 Email zhjyao2001@yahoo.com



Background: Methicillin-resistant *Staphylococcus aureus* (MRSA) can cause serious infections in elementary students. Therefore, we aimed to elucidate the quantitative associations of school environmental contamination with nasal carriage of MRSA by students in a dose-response manner. Phenotypic and molecular characterizations were performed to further assess the associations.

Materials and methods: A cross-sectional study was conducted from March to August 2016 in eight elementary schools in Guangzhou, China. A multistage stratified cluster sampling design was used to include Chinese students and selected classes. Nasal swabs from students and environmental swabs from the schools were collected. The dose-response relationship was examined using multivariate logistic regression modeling with cluster-correlated robust variance estimates to account for correlations of isolates between the environment and the students. **Results:** The prevalence rates of MRSA among 1705 students and 1240 environment samples were 10.15% and 3.87%, respectively. There were positive prevalence associations of school (odds ratio 1.09, 95% confidence interval [CI] 1.05–1.13) and class (odds ratio 1.03, 95% CI

1.01–1.06) environmental contamination with nasal carriage of MRSA isolates by students. **Conclusions:** Comparisons of MRSA isolates between the environment and the students in terms of phenotypic and molecular characterization were mostly insignificant. School environmental contamination with MRSA was positively associated with MRSA nasal carriage by elementary students. Disinfection measures and education regarding hand hygiene should be considered to decrease the prevalence of MRSA nasal carriage among students.

Keywords: methicillin-resistant *Staphylococcus aureus*, school environment, contamination, elementary students, nasal carriage

Introduction

Methicillin-resistant *Staphylococcus aureus* (MRSA) can cause infections of the skin and soft tissue,¹ endocarditis,² sepsis,³ and other infections. Nasal carriage of MRSA has been verified to be an important risk factor for MRSA infection.^{4,5} Currently, the number of studies regarding community-associated MRSA (CA-MRSA) of colonization and infection is increasing,^{6,7} and studies have shown high proportions of MRSA nasal carriage in the student population.^{8,9} The majority of recent studies, however, have focused on kindergarten children,^{10,11} high or middle school students,^{9,12} and college students.^{13,14} Few studies have focused on elementary school students.¹⁵ Compared to other student populations, elementary school students may be easily ignored.

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© 2018 Lin et al. This work is published and licensed by Dore Medical Press Limited. The full terms of this license are available at https://www.dorepress.com/terms. php and incorporate the Creative Commons Attribution — Non Commercial (unported, v3.0) License (http://creative.commons.org/licenses/by-nc/3.0/). By accessing the work you hereby accept the Terms. Non-commercial uses of the work are permitted without any further permission from Dove Medical Press Limited, provided the work is properly attributed. For permission for commercial use of this work, please see paragraphs 4.2 and 5 of our Terms (https://www.dovepress.com/terms.php). In addition, results of pulsed-field gel electrophoresis (PFGE) in several studies have qualitatively revealed that the environment might play an important role in the transmission of MRSA among populations. American investigators reported that a homology of MRSA strains existed between dental students and environmental surfaces in the dental school clinic.¹⁶ Mexican investigators found that there were homologous MRSA strains between students and their mobile phones.¹⁷ Portuguese investigators also found that there was possible transmission of MRSA clones between nasal carriage by passengers and contamination of public buses.¹⁸ Therefore, there were possible transmissions between humans and the environment.

As dense places, schools could also possibly create the opportunity for the spread of MRSA to some extent.¹⁹ Additionally, the environment of schools could affect the health of students. Current studies, however, are qualitative and cannot reveal the numeric associations of school environmental contamination with nasal carriage of MRSA isolates by elementary students. Therefore, with the aim of elucidating the dose-response associations of school environmental contamination with nasal carriage of MRSA isolates by students, we performed a cross-sectional study to determine the prevalence of MRSA nasal carriage among students, the prevalence of MRSA contamination in the school environment, and the associations of school environmental contamination with MRSA isolates in elementary students by prevalence and phenotypic and molecular characterization.

Materials and methods Ethics statement

This study was approved by the Ethics Committee of Guangdong Pharmaceutical University, and it was performed in accordance with the approved guidelines of the Declaration of Helsinki. All participants were recruited voluntarily after their parents or legal representatives signed an informed consent agreement from.

Study design and population

A cross-sectional study was conducted between March 2016 and August 2016 in eight elementary schools in Guangzhou, China. A multistage, stratified, cluster sampling design was used to obtain a representative sample. In the initial phase, three administrative districts from the urban region and one administrative district from the rural region were selected using simple random sampling. In the second phase, we selected two elementary schools from each administrative district using the simple random sampling technique, and eight schools were included in all. In the third step, we chose students from grades four and five due to their better compliance and lower academic burden. The number of classes in each grade and each school were proportionally and randomly sampled. Finally, all students with consent agreements from each class were included in the study.

Selection of environmental locations

We performed environmental sampling in those classes that we selected above. Three places (classroom, restroom, and common area) in each school were selected for sampling based on previous studies and pre-surveys. Six locations (floor, light switch, door handle, desk, chair, and air) of each classroom, five locations (floor, light switch, door handle, faucet, and toilet flush handle) in the two restrooms (men's and women's), and three locations (handle of stairs, floor of stairs, and floor of corridor) in the common area were sampled. In total, 31 samples of each class were sampled. These locations were frequently touched by people, easily contacted by skin, and amenable to cleaning and disinfecting. With the aim of avoiding bias of sampling time, we performed sampling between 2:00 and 4:00 pm on weekdays.

Data collection and sampling

For students, we collected their demographic information (gender, age, weight, and height) through questionnaires completed by their parents. At the end of every data collection day, each questionnaire was examined for its completeness and consistency. Sterile swabs moistened with sterile saline water were used to sample both nasal vestibules of the students and surfaces of environmental locations, by trained personnel. Approximately 10×10 cm areas of floors, desks, and chairs were wiped because of their regular surfaces. The entire area of light switches, handles, and toilet faucets were wiped because of their irregular surfaces. Swabs were then inoculated in enrichment broth tubes containing 1% tryptone, 7.5% sodium chloride, 1% mannitol, and 0.25% yeast extract.

As for air sampling, we used the natural sedimentation method. We placed five mannitol salt agar plates on five desktops in each classroom during lessons (one plate in the center and four at four corners of the classroom). After 10 minutes, we closed the cover of the plates. All swabs and plates were stored below 4°C and transferred to the laboratory within 4 hours for further analyses.

Isolation and identification

After 24 hours of incubation at $37\pm1^{\circ}$ C, the swabs were transferred to mannitol salt agar plates for another 24 hours

of incubation. Samples, including air samples were identified as *Staphylococcus aureus* (*S. aureus*) isolates according to specific colony morphology and being positive for gram staining, the catalase reaction, hemolysis test, the DNase test, coagulase tests, and *16S rRNA* and *nuc* genes. Those *S. aureus* isolates that were positive for the *mecA* gene and nonsusceptible to cefoxitin were identified as MRSA isolates.

Antibiotic susceptibility testing

Antibiotic susceptibility testing was conducted using the Kirby-Bauer disk diffusion method, following the Clinical and Laboratory Standards Institute guidelines 2015.²⁰ The reference MRSA strain ATCC43300 was used for quality control. All MRSA isolates underwent phenotype analysis for antibiotic susceptibility to 12 antimicrobial agents: cefoxitin, penicillin, linezolid, gentamicin, teicoplanin, erythromycin, trimethoprim-sulfamethoxazole, moxifloxacin, rifampin, chloramphenicol, tetracycline, and clindamycin. Isolates were defined as non-susceptible if they were resistant or intermediate to an antibiotic. Isolates were classified as multidrug-resistant (MDR) if they were non-susceptible to no less than three antibiotic classes (note that these isolates were already non-susceptible to all beta-lactam antibiotics).²¹

Molecular characterization

The DNA of all MRSA was extracted using the sodium dodecyl sulfate method²² and further tested to confirm the presence of the distinctive genes (*16S rRNA*, *nuc*, and *mecA*),^{23,24} the Panton-valentine leukocidin (*Pvl*) gene,²⁵ immune evasion cluster (IEC) genes (*Hlb*, *Scn*, *Chp*, *Sak*, *Sea*, and *Sep*),²⁶ toxin genes (*Tst*, *Eta*, and *Etb*),²⁷ and resistant genes (penicillin [*BlaZ*], aminoglycosides [*Aac*(6')-*aph*(2')], tetracycline [*Tet*(*M*) and *Tet*(*K*)], macrolides [*Erm*(*A*)and *Erm*(*C*)], and chloramphenicol [*Cat*(*A*)])²⁸ using polymerase chain reaction (PCR) assays as in previous studies.

A multiplex PCR technique was used to confirm and type the staphylococcal cassette chromosome *mec* (SCC-*mec*) gene.²⁴ The results were reported as types I–V and those isolates that were not types I–V were deemed as non-typeable (NT).

Multilocus sequence typing (MLST) PCR assays were conducted using previously published primers and conditions.²⁹ Allelic profiles and sequence types (STs) were assigned using the MLST database (<u>http: //www. mlst.net</u>). Singletons or members of a clonal complex were determined using the eBURST algorithm (<u>http://eburst.mlst.net</u>). Dendrogram analysis was performed based on ST type to determine the clonal relatedness and potential epidemiologic origin.

Definitions of overweight and obesity

The weight of the students was measured using a digital scale designed and manufactured under the guidance of the United Nations International Children's Emergency Fund with a precision of 100 g to measure body weight. Height measurements were taken using a locally produced United Nations International Children's Emergency Fund measuring board with a precision of 0.1 cm. The weight and height of the students were taken twice, and variations between the two measurements of 100 g for weight and 0.1 cm for height were accepted as normal; however, repeated measurements were carried out upon significantly larger variations above 100 g in weight and 0.1 cm in height. The weight scale was calibrated before measuring the students' weights. Repeated measurements were taken and checked by two measurers independently. Body mass index (BMI) was calculated as the weight in kilograms divided by the square of the height in meters (kg/m²). The age- and gender-specific BMI cutoff points recommended by the Working Group on Obesity in China were used to define overweight and obesity.³⁰

Statistical analysis

The data were entered using Epidata 3.1 (EpiData Association, Odense Denmark) and exported to Stata 14.2 (College Station, Texas, USA) software for further analysis. The data were then cleaned using frequencies for logical and consistency errors. Descriptive statistics were carried out to characterize the study population using different variables. Significance of the difference between proportions was determined using Pearson's chi-square tests for categorical variables. Univariate logistic regression and univariate logistic regression modeling with cluster-correlated robust variance estimates were used to analyze the difference of demographic characteristics among students with MRSA nasal carriage. Demographic characteristics independently associated with nasal carriage of MRSA isolates among students were identified as confounding variables in a dose-response manner with multivariate logistic regression modeling with cluster-correlated robust variance estimates to account for the correlations of isolates between the environment and students. There were two clusters in the study. One was a class cluster, and the other was a school cluster. Crude and adjusted odds ratios (ORs) with their 95% confidence interval (CI) were calculated to determine the strength and presence of an association. Moreover, we drew linear prediction plots to intuitively elucidate the dose-response association of MRSA isolates between nasal carriage among students and the environment in classes or schools. All statistical measures were estimated using survey data analysis methods. The data were analyzed using a Stata package. A two-sided *P*-value for statistical significance was defined as P<0.05.

Results

Prevalence of MRSA nasal carriage among students

Overall, 1705 students from 40 classes in eight elementary schools were included. The prevalence of MRSA nasal carriage among the students was 10.15% (173/1705). There were statistically significant differences of MRSA nasal carriage among students in different schools (χ^2 =31.211, *P*<0.001) and classes (χ^2 =94.749, *P*<0.001).

Prevalence of MRSA contamination in the environment

The prevalence of MRSA contamination in the school environment was 3.87% (48/1240). There were statistically significant differences of MRSA environmental contamination in different schools (χ^2 =15.345, *P*=0.018) and classes (χ^2 =59.011, *P*<0.001); however, there were no statistically significant differences of MRSA environmental contamination in different places (χ^2 =0.720, *P*=0.698) and locations (χ^2 =18.259, *P*=0.148). There were also no statistically significant differences of MRSA environmental contamination in different locations of classrooms, restrooms, or common areas. More details can be found in Table 1.

Association of prevalence between the environment and students

Grade (χ^2 =4.443, *P*=0.035) had a statistically significant effect on MRSA nasal carriage among students; however, after adjusting for school and class cluster, there was no significant difference. More details can be found in Table 2.

There were positive prevalence associations of school or class environment with MRSA isolates in students. The ORs and 95% CIs between the school or class environment and students in terms of the prevalence of MRSA isolates were 1.09 (95% CI, 1.05–1.13; P<0.001) and 1.03 (95% CI, 1.01–1.06; P=0.018), respectively. More details can be found in Figure 1.

Association of phenotypic characterization between the environment and students

The most predominant non-susceptible antibiotic among elementary students was penicillin (100.00%, 173/173), followed by erythromycin (98.27%, 170/173), clindamycin

 Table I Distribution of MRSA isolates in the elementary school

 environment in Guangzhou, China, 2016

Location	N	Positive (%)	χ²	P-value
Classroom	760	32 (4.21)	1.238	0.457
Floor	40	3 (7.50)		
Light switch	80	6 (7.50)		
Door handle	40	l (2.50)		
Desk	200	13 (6.50)		
Chair	200	5 (2.50)		
Air	200	4 (2.00)		
Restroom	320	10 (3.13)	1.325	0.441
Floor	40	l (2.50)		
Light switch	80	0 (0.00)		
Door handle	40	l (2.50)		
Faucet	80	6 (7.50)		
Toilet flush handle	80	2 (2.50)		
Common area	160	6 (3.75)	0.060	0.909
Handle of stairs	80	3 (3.75)		
Floor of stairs	40	l (2.50)		
Floor of corridor	40	2 (5.00)		
Total	1240	48 (3.87)		

Abbreviations: MRSA, methicillin-resistant *Staphylococcus aureus*; N, number of isolates.

(95.95%, 166/173), teicoplanin (91.91%, 159/173), tetracycline (82.66%, 143/173), rifampin (80.35%, 139/173), linezolid (72.25%, 125/173), chloramphenicol (34.68%, 60/173), trimethoprim-sulfamethoxazole (28.90%, 50/173), moxifloxacin (23.70%, 41/173), and gentamicin (10.40%, 18/173). The most predominant non-susceptible antibiotic among the school environment was penicillin (100.00%, 48/48) and erythromycin (100.00%, 48/48), followed by teicoplanin (95.83%, 46/48), tetracycline (95.83%, 46/48), rifampin (91.67%, 44/48), clindamycin (87.50%, 42/48), linezolid (85.42%, 41/48), moxifloxacin (29.17%, 14/48), chloramphenicol (25.00%, 12/48), trimethoprimsulfamethoxazole (25.00%, 12/48), and gentamicin (14.58%, 7/48). When comparing the proportions of antibiotic nonsusceptibility among students and the environment in terms of MRSA isolates, only tetracycline and clindamycin were significant. Note that the proportions of MDR MRSA were over 95% in both students and the environment. More details can be found in Table 3.

Association of molecular characterization between the environment and students

When comparing the proportions of IEC genes in the MRSA isolates among students and the environment, there was no significant difference. Among the proportions of all toxin genes, only the *Etb* gene was significantly different between MRSA isolates in students and the environment. There

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Characteristics	N	Positive (%)	OR (95% CI)	AOR (95% CI) ^a	AOR (95% CI) ^b
Gender					
Female	789	73 (9.25)	Reference	Reference	Reference
Male	916	100 (10.92)	1.20 (0.87-1.65)	1.20 (0.93-1.56)	1.20 (0.93-1.55)
Age, years					
≤10	1033	115 (11.13)	Reference	Reference	Reference
>10	672	58 (8.63)	0.75 (0.54-1.05)	0.75 (0.40-1.41)	0.75 (0.45-1.26)
BMI group		, , ,	· · · ·	· · · ·	· · · · ·
Normal weight	1213	120 (9.89)	Reference	Reference	Reference
Overweight	171	12 (7.02)	0.69 (0.37-1.27)	0.69 (0.51-0.93)	0.69 (0.39-1.23)
Obese	114	16 (14.04)	1.49 (0.85-2.60)	1.49 (0.86-2.58)	1.49 (0.80-2.76)
Grade					
Four	792	98 (12.37)	Reference	Reference	Reference
Five	913	75 (8.21)	0.63 (0.46–0.87)	0.63 (0.36-1.12)	0.63 (0.36-1.10)
Region					
Urban	1399	139 (9.94)	Reference	Reference	Reference
Rural	306	75 (8.21)	1.13 (0.76–1.69)	1.13 (0.61–2.10)	1.13 (0.67–1.93)

Notes: ^aAdjusted for school cluster; ^bAdjusted for class cluster.

Abbreviations: MRSA, methicillin-resistant *Staphylococcus aureus*; N, number of isolates; BMI, body mass index, weight (kg)/height (m)²; OR, odds ratio; AOR, adjusted OR; CI, confidence interval.

were insignificant differences in the resistant genes between students and the environment for MRSA isolates, with the exception of the Tet(K) and Cat(A) genes. More details can be found in Table 3.

Of the MRSA isolates among the students, the most predominant SCC*mec* type was type IV (69.36%, 120/173), followed by NT (11.56, 20/173), type V (8.09%, 14/173), type I (5.20%, 9/173), type III (4.05%, 7/173), and type II (1.73%, 3/173). Of the MRSA isolates in the environment, the most predominant SCC*mec* type was type IV (58.33%, 28/48), followed by NT (12.50, 6/48), type I (8.33%, 4/48), type II (8.33%, 4/48), type III (6.25%, 3/48), and type V (6.25%, 3/48). There was a significant difference of SCC*mec* type II in MRSA isolates between the students and environment.

There were 26 CC types and 77 STs of MRSA isolates in this study. The five most predominant CC types of MRSA isolates among the students were CC45 (26.01%, 45/173), CC59 (20.81%, 36/173), CC1 (8.09%, 14/173), CC121 (8.09%, 14/173), and CC30 (5.78%, 10/173). The five most predominant CC types of MRSA isolates in the environment were CC45 (37.50%, 18/48), CC30 (18.75%, 9/48), CC6 (12.50%, 6/48), CC59 (8.33%, 4/48), and CC121 (8.33%, 4/48). The statistically significant CC types of MRSA isolates between the students and environment were CC6, CC59, and CC188. Note that ST5, ST6, ST30, ST45, ST59, ST120, ST488, ST944, ST1478, ST1891, ST2791, and ST3191 were found in MRSA isolates from both the students and environment. More details can be found in Figure 2 and Figure 3.

Discussion

To the authors' knowledge, this is the first study on the dose-response prevalence associations of MRSA isolates between nasal carriage by elementary students and school environmental contamination. This study found that there were positive prevalence associations of school or class environmental contamination with nasal carriage of MRSA isolates by elementary students. The ORs of the MRSA isolates in school environments was higher than that in class environments, which may demonstrate a more severe effect of school environmental contamination on the MRSA nasal carriage by students. Therefore, these findings may suggest that school environmental contamination with MRSA plays a role in MRSA nasal colonization of students.

Our study contributes to the existing literature because we assessed the quantitative associations of MRSA isolates between the environment and students, which had not been done in other published studies. Moreover, the phenotypic characterization of MRSA isolates between the students and environment was statistically insignificant with the exception of two or three antibiotics, which meant that the phenotypic characterization of the isolates between the environment and students was similar. Regarding the molecular characterization of the MRSA isolates between the students and environment, differences observed in the majority of genes were insignificant, which also suggests that the molecular characterization of isolates between the environment and students was similar. From these three aspects of similar characterization, it is implied that there was transmission



Figure I The relationship of the prevalence rates of MRSA isolates between the environment and students in Guangzhou, China, 2016. Notes: (A) Linear prediction plot between the predicted prevalence of MRSA among students and the prevalence of MRSA isolates in the school environment. (B) Linear prediction plot between the predicted prevalence of MRSA among students and the prevalence of MRSA isolates in the class environment. Abbreviation: MRSA, methicillin-resistant *Staphylococcus aureus*.

Table 3 Antibiotic resistance an	genetic characteristics of MRSA isolates from elementary	/ students in Guangzhou, 2016 (r	າ [%])
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ltem	Student (N=173)	Environment (N=48)	χ ²	P-value
Antibiotic (non-susceptible)	1			
FOX	151 (87.28)	40 (83.33)	0.500	0.480
Р	173 (100.00)	48 (100.00)	_	_
LZD	125 (72.25)	41 (85.42)	3.482	0.062
CN	18 (10.40)	7 (14.58)	0.654	0.419
TEC	159 (91.91)	46 (95.83)	-	0.532
E	170 (98.27)	48 (100.00)	-	1.000
SXT	50 (28.90)	12 (25.00)	0.283	0.594
MXF	41 (23.70)	14 (29.17)	0.601	0.438
RD	139 (80.35)	44 (91.67)	3.382	0.066
С	60 (34.68)	12 (25.00)	1.604	0.205
TE	143 (82.66)	46 (95.83)	5.267	0.022
DA	166 (95.95)	42 (87.50)	-	0.039
MDR	169 (97.69)	48 (100.00)	_	0.579
Gene (positive)				
IEC genes				
HIb	126 (72.83)	33 (68.75)	0.310	0.578
Scn	38 (21.97)	6 (12.50)	2.111	0.146
Chp	95 (54.91)	27 (56.25)	0.027	0.869
Sak	167 (96.53)	46 (95.83)	0.053	0.819
Sea	115 (66.47)	38 (79.17)	2.842	0.092
Sep	I (0.58)	0 (0.00)	_	1.000
Toxin genes				
Tst	42 (24.28)	9 (18.75)	0.647	0.421
Eta	18 (10.40)	2 (4.17)	-	0.258
Etb	7 (4.05)	7 (14.58)	-	0.015
Pvl	10 (5.78)	I (2.08)	-	0.463
Resistant genes				
BlaZ	169 (97.69)	48 (100.00)	-	0.579
Aac(6')-aph(2')	15 (8.67)	5 (10.42)	-	0.776
Tet(M)	113 (65.32)	34 (70.83)	0.513	0.474
Tet(K)	84 (48.55)	31 (64.58)	3.868	0.049
Erm(A)	I (0.58)	0 (0.00)	-	1.000
Erm(C)	3 (1.73)	2 (4.17)	-	0.298
Cat(A)	0 (0.00)	48 (100.00)	221.000	<0.001

Note: -, no estimate is provided owing to the lack of occurrence of the outcome of interest in at least one group.

Abbreviations: N/n, number of isolates; FOX, cefoxitin; P, penicillin; LZD, linezolid; CN, gentamicin; TEC, teicoplanin; E, erythromycin; SXT, trimethoprim-sulfamethoxazole; MXF, moxifloxacin; RD, rifampin; C, chloramphenicol; TE, tetracycline; DA, clindamycin; MDR, multidrug resistant; IEC, immune evasion cluster.

of MRSA between the school environment and elementary students.

The prevalence in students was 10.15% (173/1705) and this rate is higher than that observed in other studies.^{31–34} Likewise, the prevalence of MRSA in the environment was 3.87% (48/1240) and this rate is higher than that observed in other studies.^{35–37} The results of the prevalence demonstrate a higher burden of MRSA isolates among the environment and students, which should be paid greater attention.

The proportions of antibiotic non-susceptibility in MRSA isolates among students were higher than in other studies,^{12,38} which should be noted by clinical workers. A majority of antibiotic non-susceptibility of MRSA isolates between the environment and students was statistically insignificant,

which may suggest that isolates transmit between the environment and students. Studies have revealed that isolates with the same PFGE types have similar or the same patterns of antibiotic susceptibility.^{36,39}

IEC genes are becoming increasingly important because they can affect transmission ability⁴⁰ and the epidemic characteristics of isolates.²⁶ The proportions of IEC genes in this study were similar to other studies,⁴¹ as were the proportions of toxin genes and resistant genes.^{16,19,42} The main SCC*mec* types in this study were type IV and V, but the proportion of types I–III was over 10%, which may demonstrate that there was cross-transmission between the communities and hospitals.^{16,36} The distribution of CC types demonstrated the phylogenetic relatedness of isolates



Figure 2 Distribution of clonal complex types of MRSA isolates from students and the environment in Guangzhou, China, 2016. **Note:** *There was a statistically significant difference, *P*<0.05.

Abbreviations: MRSA, methicillin-resistant Staphylococcus aureus; MLST, multilocus sequence typing.



Figure 3. The sequence type relationship of MRSA isolates between the students and school environment in Guangzhou, China, 2016. Abbreviation: MRSA, methicillin-resistant Staphylococcus aureus.

between the environment and students. Therefore, with regard to the molecular characterization of MRSA isolates between the environment and students, differences in the majority of genes were insignificant, which may also suggest that isolates transmit between the environment and students. Studies have revealed that isolates with the same PFGE types have similar or the same molecular characterization.^{16,34}

Our findings suggest the importance of considering school environmental contamination when designing interventions aimed at reducing MRSA nasal carriage in elementary students. In hospitals, studies have shown that increased environmental cleaning reduces the proportions of MRSA isolates in the environment.⁴³ Therefore, strengthening environmental cleaning in communities may also reduce the proportions of MRSA isolates. Further research is needed to determine whether effective decontamination of the school environment reduces the nasal carriage by elementary students.

This study has several limitations. First, we could not establish temporality because of the cross-sectional design. Second, because we could not collect the height and weight data on the spot, we used self-reported height and weight to calculate BMI, which could have resulted in misclassification; however, parents did not know the colonization status of their children, so any misclassification was likely non-differential. Third, we did not offer decolonization to students and whether such intervention would improve the health of the students is scientifically unproven.

Despite these limitations, the study had several strengths. First, the sample size of this study was large. Second, to our knowledge, our study was the first to elucidate the quantitative association of isolates between the environment and students. Third, we performed phenotypic and molecular characterization of the isolates to further assess the associations.

Conclusion

In conclusion, school environmental contamination with MRSA was positively associated with MRSA nasal carriage in elementary students. Environmental decontamination should be considered as a strategy to prevent MRSA nasal carriage. Therefore, disinfection measures of the school environment and education regarding hand hygiene for elementary students should be considered to decrease the prevalence of MRSA nasal carriage among students.

Availability of data and materials

Due to ethical restrictions related to protecting participant confidentiality, underlying data cannot be made publicly available. These data are available upon request from the principal investigator, Zhenjiang Yao (e-mail: zhjyao2001@ yahoo.com).

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Author contributions

All authors contributed towards data analysis, drafting and critically revising the paper and agree to be accountable for all aspects of the work.

Disclosure

The authors report no conflicts of interest in this work.

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