

Community-associated and healthcare-associated methicillin-resistant *Staphylococcus aureus* ~~(CA-MRSA)~~ in children: a microbiological and epidemiological study

Abstract

Aims: Novel methicillin-resistant *Staphylococcus aureus* have been causing infections in the community and are now invading hospitals. In this study we aimed to determine, using risk factors for healthcare associated infections as an epidemiological parameter, and antimicrobial susceptibility profile as a surrogate for SCCmec type and microbiological parameters, the characteristics of circulating *S. aureus* clinical isolates.

Methods: From July 2009 to April 2012, *S. aureus* isolates from children hospitalized in Santa Casa de São Paulo, a tertiary care-center in São Paulo, Brazil, were included. All isolates grew in cultures from sterile sites and we included only one isolate per patient.

Results: Fifty-five isolates were included during the study period, 47 from blood, six from abscesses, one from pleural fluid and one from spinal fluid. Regarding oxacillin resistance Among these isolates, 34 were methicillin susceptible *S. aureus* (MSSA) and 21 were methicillin-resistant *S. aureus* (MRSA). Eleven patients were excluded (5 MSSA and 6 MRSA) because clinical charts were not available for review, reducing the total to 29 MSSA and 15 MRSA isolates. After searching for risk factor for healthcare-associated infections, 11 of the 15 MRSA isolates were epidemiologically considered healthcare-associated MRSA (HCA-MRSA) and 4 community-associated MRSA (CA-MRSA). Using the microbiological classification (multiresistance), five were considered as HCA-MRSA and 10 were considered CA-MRSA. Interestingly, of the 11 isolates considered as epidemiological HCA-MRSA (presence of any risk factor), six had a microbiological profile (non-multiresistant) consistent with CA-MRSA circulating clones. Of the 11 isolates considered epidemiological HCA-MRSA, six had a microbiological profile consistent with CA-MRSA circulating clones.

Conclusion: Our results clearly show that the boundaries between CA-MRSA and HCA- are increasingly difficult to determine.

Keywords

Staphylococcus aureus; MRSA; antimicrobial resistance; oxacillin

Introduction

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Staphylococcus aureus commonly colonizes ~~our species~~ humans in different anatomical sites. Traditionally it is more frequently found in the nose. However, recent evidence suggests that oropharynx could be an even more important site of colonization [1]. Some specific populations, as cystic fibrosis patients, are more commonly colonized than the general population [2]. Depending on host and pathogen factors, *S. aureus* may cause infection. Infectious syndromes include, but are not limited to respiratory, skin and soft tissue, osteoarticular and bloodstream infections. *S. aureus* is an important etiologic agent of both community- and healthcare-associated infections [1-3].

Resistance to antimicrobial agents has complicated the treatment of *S. aureus* infections. After penicillin resistance, methicillin resistance disseminated worldwide, first in the hospitals and eventually in the community [3-6]. These new community-associated methicillin resistant *S. aureus* (CA-MRSA) clones, described originally in the 1990s, have some differences when compared to the traditional healthcare-associated (HCA-MRSA) clones. CA-MRSA are usually resistant to less antimicrobial agents, usually only to beta-lactams and macrolides (and increasingly to clindamycin) [3-5]. These are the phenotypic implications of the presence of different Staphylococcal cassette chromosome *mec* (SCC*mec*) types. SCC*mec* is a mobile genetic element that includes *mecA* gene, responsible for methicillin resistance in *S. aureus*. CA-MRSA most commonly carry SCC*mec* types IV or V [4-6]. Some of these clones also have a specific virulence profile, frequently producing Panton-Valentine leukocidin and other typical virulence factors⁴⁻⁷.

These new clones have also been reported causing nosocomial infections, substituting, at least in part, traditional health-care associated MRSA clones. In ~~Brazil~~ Brazil, although CA-MRSA is still not common, clones carrying SCC*mec* IV have been increasingly found in different hospitals [8-12]. Resistance to oxacillin in hospitals is common in the bacterial isolates from hospitals, varying from 30-60% [9-12]. In the present study, we aimed to characterize the *S. aureus* clinical isolates regarding their epidemiological (presence or absence of risk factors for health-care associated infections) and microbiological (antimicrobial multi-resistance or non-multi-resistance as a surrogate for SCC*mec* type) profiles.

Methods

From July 2009 to April 2012, *S. aureus* isolates from children hospitalized in Santa Casa de São Paulo, a tertiary care-center in São Paulo, Brazil, were included. All isolates grew in cultures from previously sterile sites and we included only one isolate per patient. Bacterial identification was conducted using traditional biochemical methods (Gram stain, catalase, coagulase and DNase) and antimicrobial susceptibility testing was performed according to current Clinical and Laboratory Standards Institute criteria [13].

Methicillin-resistant isolates resistant to four or more different classes of antibiotics were considered multi-resistant (HCA-MRSA by microbiological classification), whereas isolates resistant to three or less classes were considered non-multi-resistant (CA-MRSA by microbiological classification) [4-6].

Comment [D2]: Species or surface? We chose humans.

Comment [D3]: It should be [1]. Please modify all the citations according to the author guid. Done.

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Comment [D4]: This phrase is not clear. These are previously sterile sites, sites that are not usually colonized by bacteria. We only included isolates from blood, subcutaneous abscess, etc, and not from skin, stools, upper respiratory tract, etc. We inserted the word "previously".

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Comment [D5]: Do you have some references for this classification of HCA-MRSA and CA-MRSA? This is the classification routinely used in the literature. We included the references in the paragraph. (publications already in our reference list)

Clinical charts were reviewed and searched for the presence of any risk factors for health-care associated infections, including hospitalization within the previous year, chronic disease that implicates in frequent contact (≥ 6 /year) with hospital environment, or current hospitalization for at least 48 hours. Isolates were considered as HCA-MRSA (epidemiologically) if any of these risk factors were present, and were considered as CA-MRSA (epidemiologically) if they were all absent [4-6, 9-12]. The study was approved by the institutional ethics committee.

Comment [D6]: Please check and complete this sentence. Completed.

Results and Discussion

Fifty-five isolates were included during the study period, 47 from blood, six from abscesses, one from pleural fluid and one from spinal fluid. Regarding oxacillin resistance, 34 were MSSA and 21 were MRSA. Eleven patients were excluded (5 MSSA and 6 MRSA) because clinical charts were not available for review, reducing the total to 29 MSSA and 15 MRSA isolates.

Comment [D7]: Here, the classification of HCA-MRSA and CA-MRSA is different from that presented above. What is the connection of the two definitions? Both are used in the literature, one is an epidemiological classification and the other is a microbiological classification. An isolate may be epidemiologically HCA but microbiologically CA (for example, although the patient where *S. aureus* was isolated had risk factors for HCA infection, the susceptibility pattern could be of a CA isolate – non-multiresistant) and vice-versa. This is largely recognized by the available literature on the subject. We also includes references for this paragraph (publications already in our reference list).

After searching for risk factor for healthcare-associated infections, 11 of the 15 MRSA isolates were epidemiologically considered HCA-MRSA and 4 CA-MRSA. Using the microbiological classification (multiresistance), five were considered HCA-MRSA and 10 were considered CA-MRSA. Interestingly, of the 11 isolates considered epidemiological HCA-MRSA (presence of one or more any risk factors), six had a microbiological profile (non-multiresistant) consistent with CA-MRSA circulating clones.

Comment [D8]: Revise this phrase. “any” is usually used in negative sentence. Revised.

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Although these novel SCCmec type IV MRSA clones were first reported in the community, there is increasing evidence that they are replacing traditional hospital circulating clones [8-10]. In Brazil, the Brazilian clone (SCCmec type III) have been disseminated nationally in hospitals, but recent evidence shows that different clones carrying a type IV cassette are disseminating as a cause of healthcare-associated infections [11,12].

Comment [D9]: Clones or isolates? These are indeed clones, groups of isolates that share the same SCCmec type.

Comment [D10]: Why clones, but not strains? These are indeed clones, groups of isolates that share the same SCCmec type.

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Although the number of isolates included in our study is low, and we have not performed molecular typing, phenotypic susceptibility testing served as a surrogate for the SCCmec type, since we have previously and consistently reported the strong correlation between these parameters [11,12]. In addition, its the results in the present study clearly show that the boundaries between CA-MRSA and HCA-MRSA are increasingly difficult to determine.

Comment [D11]: Why is the relationship between clone and type? Clones are groups of isolates that share molecular characteristics, as PFGE profile or SCCmec type.

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Nosocomial isolates cause infection in patients without risk factors and community isolates invade the hospital causing health-care associated infections. In these challenging epidemiological and microbiological situation, knowledge of local and global circulating clones is paramount to ensure the appropriate preventive and therapeutic approaches.

References

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Table 1. Epidemiological and microbiological classification of MRSA isolates from children hospitalized at Santa Casa de São Paulo, Brazil, 2009-2012.

Classification		Epidemiological		
		CA	HCA	Total
Microbiological	CA	4	6	10
	HCA	0	5	5
	Total	4	11	15