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BRIEF REPORT

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Methicillin-resistant *Staphylococcus aureus* in community-acquired meningitis

Historically, infections caused by methicillin-resistant *Staphylococcus aureus* (MRSA) have been associated with healthcare settings. However, since the 1990s, community-acquired MRSA (CA-MRSA) infections have been increasingly recognized [1]. The microorganisms involved differ from those related to nosocomial infections in the presence of type IV staphylococcal cassette chromosome (SCC) *mec* elements, virulence genes encoding a toxin called Panton–Valentine leukocidin (PVL), which is not found in HA-MRSA isolates [2], and in the lack of the typical multiresistance pattern present in nosocomial staphylococci. Even though most infections caused by CA-MRSA in the community involve skin and soft tissues, and invasive CA-MRSA infections have been reported infrequently until now, their incidence is increasing [3].

Among the life-threatening community-acquired staphylococcal infections, meningitis has fortunately been reported only sporadically, accounting for less than 3% of cases diagnosed as bacterial in origin [4, 5]. Considering the rarity of staphylococcal meningitis cases, it is not surprising that the role of CA-MRSA remains obscure, at least in our region, since almost no data is available on the prevalence of these strains. Nevertheless, hospital-acquired MRSA infections progressing to meningitis or cerebral abscesses are significant [6, 7]. The lack of accompanying resistance in a large series of CNS infections reported by Jones et al. [7] is noteworthy and may suggest the introduction and dissemination of CA-MRSA in the participating hospitals. Here, we report on two children with acute bacterial meningitis due to community-acquired MRSA who were admitted to the Pediatric Hospital of Posadas, a 100-bed tertiary-care hospital located in a region of about 1.5-million inhabitants in northeastern Argentina. As a reference, MRSA accounted for one-third of SA isolates during 2004, and most of them did not exhibit the typical antimicrobial resistance profile associated with nosocomial MRSA.

In December 2004 a 5-year-old girl was admitted with a 5-day history of sacral and lumbar pain, fever and a cough, which were being treated with amoxicillin-clavulanic acid. Diagnosis at admission was meningeal syndrome. Cerebrospinal fluid was purulent and direct Gram stain showed gram-positive cocci. Blood counts showed 15,400 leukocytes/mm^3 and a hematocrit level of 29%. Her C-reactive protein level was 768 mg/l, and she was hematuric. A combined cefotaxime-vancomycin antibiotic treatment was administered immediately. Upon characterization of MRSA without any accompanying resistance, cefotaxime was replaced by rifampin. After treatment for 21 days, clinical evolution was excellent and there was no evidence of neurologic sequelae.

In January 2005, a 6-year-old boy from Posadas City was brought to the hospital with fever and a diffuse petechial rash; the rash had been present for 48 h and he had a 24-h history of headache and urinary incontinence. The severity of symptoms led to an initial diagnosis of anaphylactic shock, and he was admitted directly to the emergency room; he was then transferred to the intensive care unit, where he was treated with diphenhydramine, noradrenaline and corticoids. After medical reanimation was required, the clinical diagnosis was changed to septic shock and meningitis, and he was treated with cefotaxime and acyclovir. Cerebrospinal fluid analysis showed a leukocyte count of 528/mm^3, a glucose level of 0.51 (serum glucose 0.86), and a protein level of 0.31 g/l. Blood analysis showed definite leukopenia (leukocyte count about 3,100/mm^3). From day 3 after admission, when cultures of both blood and cerebrospinal fluid grew MRSA

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with no additional resistance, antimicrobial therapy was changed to vancomycin and rifampin, but the patient died on day 5 of hospitalization.

Both isolates were identified using standard biochemical methods. Antimicrobial susceptibility testing was performed using disk diffusion tests as recommended by the Clinical and Laboratory Standards Institute (formerly National Committee for Clinical Laboratory Standards) [8]. Methicillin resistance was confirmed by detection of the *mec*A gene by PCR [9]. *S. aureus* ATCC 25912 and *S. aureus* ATCC 43300 were used as control strains that were susceptible and resistant to methicillin, respectively.

Detection of *mec*A and *luk*F-PV genes was performed following extraction of genomic DNA. Essentially, bacteria were grown in 4 ml of Luria Bertani broth for 18 h at 37°C; the broth was then centrifuged and resuspended in 100 µl of 25 mM Tris–ClH (pH 8.0) with 50 mM glucose and 10 mM EDTA. Lysostaphin and lysozyme were added (3 and 20 µl of 10 mg/ml solutions, respectively), and the mixture was incubated at 37°C for 1 h. Spheroplasts were lysed by the addition of 40 µl lysis solution (10 mM Tris–ClH pH 8.0, 10% SDS) and 20 µl 0.5 M EDTA (pH 8.0). After treatment with RNAse (10 µg/ml) and proteinase K (0.2 mg/ml), DNA was purified by phenol-chloroform extraction and precipitated with ethanol. Typing of SCC*mec* elements was carried out as described previously [10], and detection of PLV genes was performed using previously described primers [10] and confirmed by sequencing. MRSA belonging to the South American clone [11] and an isolate with the same phenotype as the recently described Cordobés clone [10] were used as SCC*mec* type IIIA [10] and type I (unpublished data) controls, respectively. Both CA-MRSA isolates were susceptible to all non-beta-lactam antibiotics. Both strains harbored the PLV gene and SCC*mec* type IV (Fig. 1).

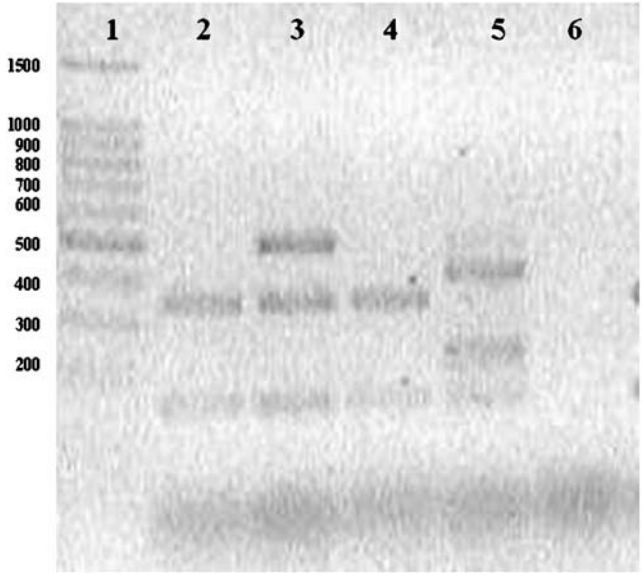

Fig. 1 Structural types of the *mec* elements identified by multiplex PCR. *Lane 1* 100 bp DNA ladder, *lane 2* SCC*mec* type I (first case), *lane 3* SCC*mec* type IV, *lane 4* SCC*mec* type I (second case), *lane 5* SCC*mec* type IIIa, *lane 6* negative control

As suggested previously, MRSA strains carrying PLV might become a severe problem in the future, because of their virulence and successful dissemination [12]. This is supported by a recently described outbreak caused in a neonatal intensive care unit by a PLV-negative MRSA strain harboring SCC*mec* type IV, which was previously found in the community [13]. Considering the rising number of treatment failures for skin infections detected in outpatients attending our hospital, including documented MRSA infections (data not shown), it may be suspected that CA MRSA is increasing in this region. Further studies of the epidemiology of PLV-positive MRSA strains in the community are consequently needed, as are fast diagnostic tools.

Awareness of the local prevalence of these strains, and information about the potential severity of infections they can cause, are necessary in order to implement appropriate intervention measures, including prompt and correct empiric antibiotic treatment. The typical lack of accompanying resistance should, in such cases, be taken into consideration and administration of useful and less toxic antibiotics should not be precluded.

Nosocomial MRSA outbreaks produced after the microorganisms gain access from the community are likely to become more prevalent in the future; in such cases, clear information is essential for evaluating the infection control measures used in each particular setting. Emergence of PLV-positive CA-MRSA causing meningitis in pediatric patients requires further control measures.

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