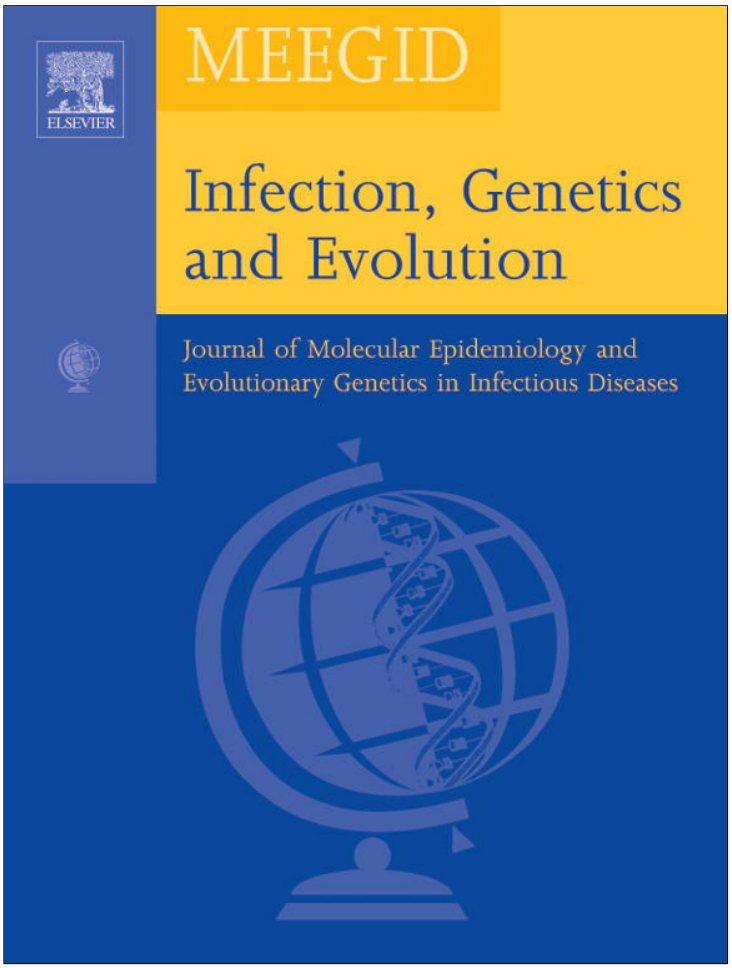

(This is a sample cover image for this issue. The actual cover is not yet available at this time.)

Infection, Genetics and Evolution 14 (2013) 401–405

Contents lists available at SciVerse ScienceDirect

Infection, Genetics and Evolution

journal homepage: www.elsevier.com/locate/meegid

Short communication

Methicillin-resistant *Staphylococcus aureus* ST30-SCC*mec* IVc clone as the major cause of community-acquired invasive infections in Argentina

S. Fernandez [a,1], L. de Vedia [b,1], M.J. Lopez Furst [c], N. Gardella [a], S. Di Gregorio [a], M.C. Ganaha [e], S. Prieto [f], E. Carbone [g], N. Lista [b], F. Rotrying [h], M.E. Stryjewski [d], M. Mollerach [a,*]

[a] *Cátedra de Microbiología, Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires, Argentina*
[b] *Hospital J F Muñiz, CABA, Argentina*
[c] *Sanatorio Mendez, CABA, Argentina*
[d] *Department of Medicine and Division of Infectious Diseases, Centro de Educación Médica e Investigaciones Clínicas "Norberto Quirno" (CEMIC), CABA, Argentina*
[e] *Hospital Vicente Lopez y Planes, Gral Rodríguez, Provincia de Buenos Aires, Argentina*
[f] *Hospital Nuestra Señora de Luján, Provincia de Buenos Aires, Argentina*
[g] *Hospital Aeronáutico, CABA, Argentina*
[h] *Hospital UAI, CABA, Argentina*

ARTICLE INFO

Article history:
Received 5 October 2012
Received in revised form 18 December 2012
Accepted 19 December 2012
Available online 20 January 2013

Keywords:
Methicillin-resistance
Community-associated *Staphylococcus aureus*
CA-MRSA
MRSA invasive infection
Molecular epidemiology

ABSTRACT

Community-acquired methicillin-resistant *Staphylococcus aureus* (CA-MRSA) infections have become a major concern worldwide. We conducted a prospective multicenter study of invasive CA-MRSA to evaluate clinical features and genotype of strains causing invasive infections in Argentina. A total of 55 patients with invasive CA-MRSA infections were included. Most patients (60%) had bloodstream infections, 42% required admission to intensive care unit and 16% died. No CA-MRSA isolates were multiresistant (resistant $\geqslant$3 classes of antibiotics). All isolates carried Panton-Valentine leukocidin (PVL) genes and staphylococcal cassette chromosome (SCC*mec*) type IV. The majority CA-MRSA strains belonged to ST30 and had identical pulsed-field gel electrophoresis (PFGE) patterns, qualifying as a clonal dissemination of a highly transmissible strain. The main clone recovered from patients with CA-MRSA invasive infections was genotyped as pulsed-field gel electrophoresis type C-ST30, SCC*mec* type IVc-*spa* type 019, PVL positive. It has become predominant and replaced the previously described CA-MRSA clone (PFGE type A, ST5, SCC*mec* type IV, *spa* type 311).

1. Introduction

Infections caused by methicillin-resistant *Staphylococcus aureus* (MRSA) have occurred worldwide, primarily in healthcare settings. Over the last 15 years, there have been increasing reports of patients with little or no healthcare contact suffering from MRSA infection. Defined now as community-acquired MRSA (CA-MRSA) this pathogen has reached epidemic proportions. While most CA-MRSA infections involve skin and skin structures (Stryjewski and Chambers, 2008), the invasive nature of CA-MRSA has also been observed in patients with necrotizing pneumonia, severe sepsis, osteomyelitis, meningitis and death (David and Daum, 2010; Deleo et al., 2010; Deurenberg and Stobberingh, 2009).

CA-MRSA strains are phylogenetically distinct from traditional hospital associated (HA-MRSA) clones. CA-MRSA isolates generally exhibit SCC*mec* IV or V, a narrow range of drug resistance, and commonly carry Panton-Valentine leukocidin (PVL) genes, rarely identified in HA-MRSA isolates (Ma et al., 2002; Naimi et al., 2003). Presence of PVL genes in *S. aureus* isolates has been associated with abscess formation, primary skin infections (Lina et al., 1999), severe necrotizing pneumonia, and increased complications of hematogenous osteomyelitis; however, the role of PVL in the pathogenesis of *S. aureus* infections has not been fully elucidated.

CA-MRSA clones seem to have a delineated (but dynamic) geographical distribution (David and Daum, 2010). In the United States, USA400 clone (ST1-IVa) was the most prevalent clone before 2001 (Stemper et al., 2004) and remains a common cause of community-onset disease among indigenous populations in Alaska and the Pacific Northwest. A second epidemic CA-MRSA clone, the USA300 (ST8-IVa, t008, PVL + and ACME-I) emerged between 1999 and 2001 and now causes most of the CA-MRSA infections in the United States (Kennedy et al., 2008). The European clone (ST80-IVc) was found to be predominant between 2004 and 2006 in Europe. However, the frequency of USA300 and related clones has been increasing in the last two years, representing a changing

* Corresponding author. Address: Cátedra de Microbiología, Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires, Junín 956, 1113 Ciudad Autónoma de Buenos Aires, Argentina. Tel.: +54 11 4964 8285; fax: +54 11 4964 8274.
E-mail address: mmollera@ffyb.uba.ar (M. Mollerach).
[1] These authors contributed equally to this paper.

1567-1348/$ - see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.meegid.2012.12.018

trend in the epidemiology of CA-MRSA in that continent (Rolo et al., 2012). Moreover, recently it was demonstrated that MRSA clones causing invasive infection differ considerably between countries exhibiting a regional distribution in Europe (Grundmann et al., 2010).

The Southwest Pacific clone (ST30-IV) has mostly been described in Australia and New Zealand, where it is also known as the Western Samoan phage pattern (Nimmo et al., 2006; Smith and Cook, 2005). Moreover, 45 distinct clones of CA-MRSA have been identified in Australia; many of these are related to recognized MRSA lineages, but only three clones have successfully adapted to the Western Australian community environment (Coombs et al., 2011).

In Argentina, a PVL-positive epidemic CA-MRSA clone has been previously described (CAA clone: Pulsotype A, ST 5, SCC*mec* IV, *spa* type 311, PVL positive) as the predominant clone causing both invasive and non invasive MRSA infections (Gardella et al., 2008; Sola et al., 2008; von Specht et al., 2006).

However, spread of some epidemic clones into other regions has produced some displacement of previously circulating strains indicating that successful lineages may have competitive advantages which may be key in the evolution of this pathogen (Amorim et al., 2007; Gardella et al., 2005). Recent reports describing CA-MRSA clones invading healthcare settings underscore such potential advantages (Maree et al., 2007; Sola et al., 2012).

In our country we did not have recent, prospective, multicenter, clinically based studies in patients with invasive CA-MRSA infections. The aim of this study was to describe the clinical and molecular epidemiology of current invasive infections caused by CA-MRSA in adolescent and adult patients in Argentina.

2. Material and methods

2.1. Study design

A prospective, multicenter, observational study was designed to evaluate clinical and molecular features of invasive CA-MRSA infections in Argentina between March 2010 and December 2011. Patients were enrolled in 11 participating hospitals located in the central region of the country: Buenos Aires City ($N = 5$), Buenos Aires Province ($N = 5$) and Santa Fe ($N = 1$). The study was reviewed by each Institutional Review Board (IRB) and informed consent form obtained if requested by the IRB.

2.2. Patient selection and definitions

Patients were enrolled if they were $\geqslant$14 year-old and had invasive MRSA infection (see definition below). Patients were excluded if they had any of the following during the last 12 months: hospitalization, dialysis, surgery, presence of catheters or percutaneous medical devices, or residence in a long-term care facility. Community acquired MRSA infection was defined by a positive MRSA culture at the time of hospital admission or within the 48 h after hospital admission. Invasive MRSA infection was defined by the isolation of MRSA from a normally sterile body site.

2.3. Data collection

Clinical and demographic information was obtained using clinical report forms. Data collected included social–economical variables, comorbidities, use of prior antibiotic, clinical presentation, main laboratory results at baseline, source of isolate (e.g. blood, joint fluid, pleura), and outcomes.

Management decisions were made on an individual basis by physicians at each participating institution, following local policies and standards. Follow-up data was intended to be obtained up to 90 days after the end of treatment.

2.4. Microbiological studies

The isolates were identified at the participating hospitals as *S. aureus* on the basis of conventional diagnostic procedures. All MRSA isolates were stored at −20 °C and shipped to a central laboratory (Cátedra de Microbiología, Facultad de Farmacia y Bioquímica, Universidad de Buenos Aires). Antimicrobial susceptibility testing was performed by diffusion methods according to Clinical and Laboratory Standards Institute guidelines (CLSI, 2009).

2.5. PCR amplification of mecA, lukS/F-PV, SCCmec and agr typing

Detection of the *mec*A and PVL coding genes (*lukS/F-PV*) was performed after extraction of genomic DNA as previously described (von Specht et al., 2006). SCC*mec* types were determined by PCR with a simplified version of Kondo's typing system, including M-PCR-1 and M-PCR-2 (Kondo et al., 2007). SCC*mec* type IV was further sub-typed using published primers (Milheirico et al., 2007). In specific cases, SCC*mec* typing was also performed as recommended by Oliveira et al. (Oliveira and de Lencastre, 2002).

The *agr* locus was genotyped by multiplex PCR as it was described before (Gilot et al., 2002).

2.6. Typing methods

Genotyping analysis was conducted using *spa* typing (Harmsen et al., 2003) and Pulsed-Field Gel Electrophoresis (PFGE) with *Sma*I as previously described (Chung et al., 2000). Comparison of the PFGE fingerprints was performed by the unweighted pair-group method clustering analysis (UPGMA), applying the Dice correlation coefficient. MRSA clones previously described in Argentina were included in PFGE pattern analysis (Gardella et al., 2008). Representative isolates of the major pulsotypes were studied by Multilocus Sequence Typing (MLST) (Enright et al., 2000).

2.7. Statistical analysis

Descriptive statistics were used to summarize the characteristics of the patients. Continuous variables were expressed as medians and interquartile range, and categorical variables as percentages.

3. Results

3.1. Clinical features

A total of 55 patients were included in the study. Most patients were males, the median age was 29 years-old and only 16% had diabetes. More than half of patients had medical evaluation and/or received antibiotics within 30 days prior to the admission. Most common infections were bloodstream and/or pulmonary. Around 40% of patients required ICU. Demographic and clinical variables are displayed in Table 1. Vancomycin was the most commonly prescribed antibiotic and almost two thirds of patients were treated with $\geqslant$2 antibacterials (Table 2). Most patients were cured or improving at the end of therapy and death was documented in 16% of patients.

3.2. Antimicrobial susceptibility

Antibiotic susceptibility testing revealed resistance to oxacillin (100%) Resistance to erythromycin (9%), clindamycin (6%),

Table 1
Demographic and clinical characteristics of patients with invasive infections due to community-acquired methicillin resistant *S. aureus* in Argentina.

Variable	Total ($N = 55$) n (%)[a]
Demographics features	
Age (median) (range)	29 (16–73)
Gender male, n (%)	43 (78)
Predisposing factors	
Prior antibiotic usage (6 months)	20 (36)
Play contact sports	12 (22)
Diabetes mellitus	9 (16)
Household member suffering skin and soft tissue infection	7 (13)
HIV infection	5 (9)
Immunosuppressive therapy	1 (2)
Intravenous drug users	1 (2)
Male to male sex	1 (2)
No predisposing factors	20 (36)
Leading causes of hospitalization	
Skin and skin structures related infection	16 (29)
Fever	15 (27)
Respiratory symptoms	10 (18)
Osteoarticular infections	8 (15)
Others	6 (11)
Medical consultation prior to hospitalization	
Within 30 days prior to admission	40 (73)
Associated to admission diagnosis	36 (66)
Prescribed antibiotics in the previous 30 days	30 (55)
More frequents symptoms	
Fever	45 (82)
Mialgias	23 (42)
Chills	21 (38)
Dysnea	19 (35)
Cough	14 (25)
Rash	6 (11)
Sensory impairment	2 (4)
Sites of infection	
Bloodstream	33 (60)
Lungs	28 (51)
Skin and skin related structures	27 (47)
Muscle	11 (20)
Bone	9 (16)
Pleural	8 (15)
Joints	5 (9)
Kidney/bladder	4 (7)
Heart and/or major vessels	4 (7)
Central Nervous System	2 (4)
Intraabdominal	1 (2)
Primary source of infection	
Skin and skin related structure	26 (47)
Lung	9 (16)
Other/Unknown	20 (36)
Severity Indicators	
Intensive care unit admission	23 (42)
Sepsis	31 (56)
ARDS*	8 (15)
Vasoactive drugs requirement	11 (20)
Mechanical ventilation requirement	13 (24)

[a] ARDS denotes acute respiratory distress syndrome.

Table 2
Treatment and clinical outcomes.

Treatment variable	Total ($N = 55$) n (%)
Type of antibiotic treatment	
Monotherapy	20 (36)
Combined therapy	35 (64)
More frequents antibiotics used (as mono or combined therapy)	
Vancomycin	30 (55)
Monotherapy	10 (18)
Rifampicin	19 (35)
TMP/SMX	18 (33)
Clindamycin	16 (29)
Any type of surgical treatment	33 (60)
Clinical outcome	
End of treatment	
Clinical cure	30 (55)
Clinical improvement	16 (29)
Clinical failure	9 (16)
Follow up at 90 days	
Clinical success	19/31 (61)[a]
Clinical failure	12/31 (39)[a]
Death	9/55 (16)

TMP/SMX denotes trimethoprim/sulfamethoxazole.
[a] From the total of patient with follow-up at 90 days obtained.

gentamicin (11%), and quinolones (4%) was detected. All MRSA isolates were susceptible to vancomycin, teicoplanin, linezolid, rifampin, doxycyclin and trimethoprim/sulfamethoxazol. None of the isolates were multiresistant (resistant to $\geqslant 3$ classes of antibiotics other than β-lactams).

3.3. Molecular characterization of MRSA isolates

Among the 28 clinical isolates available for molecular analysis, 23 belonged to the clone ST30 with a predominant subtype C1 (16/23) and five minor subtypes (C2 to C6, 7 isolates) (Fig. 1). All isolates of this clone were SCC*mec* type IVc, *spa* t019, PVL positive and were characterized as *agr* III (Table 3). One of the minor clones found in this study ($n = 2$), the CAA clone, (PFGE type A, ST5-SCC*mec* IV-*spa* t311), had been identified as prevalent in Argentina since 2004 (Gardella et al., 2008; Sola et al., 2008). Other minor PFGE types, type F and type E, included 2 and 1 isolates, respectively.

4. Discussion

The emergence and subsequent waves of infections caused by CA-MRSA clones is a major world concern. There is a clear need to better understand the dynamic of CA-MRSA epidemic in each geographic region. In Argentina, only limited information was available on CA-MRSA epidemiology in the adult population. The present prospective study provides data on epidemiology, antimicrobial susceptibility patterns, molecular characteristics and outcomes of adolescents and adults suffering invasive infections due to CA-MRSA.

In our study, skin and skin structure infections were the most common primary source for invasive CA-MRSA infection. This finding is in agreement with both recent (Klevens et al., 2007) and old studies (Skinner and Keefer, 1941) and indicates that certain host-pathogen interactions may result in more aggressive diseases.

Antimicrobial resistance patterns have been used to distinguish between CA-MRSA and HA-MRSA strains, with CA-MRSA strains showing greater susceptibility to several antimicrobial agents (usually gentamicin, clindamycin, and trimethoprim-sulfamethoxazole) than HA-MRSA (Ma et al., 2002; Naimi et al., 2003). In our study, CA-MRSA isolates retained susceptibility to most classes of non β-lactam antibiotics. In our investigation, the antibiotic regimens used were diverse, and almost two thirds of patients received combined therapy. Although available evidence (Korzeniowski and Sande, 1982) and recommendations (Liu et al., 2011) suggest the use of monotherapy with vancomycin to treat serious MRSA infections there are concerns regarding a diminished efficacy of this agent (Deresinski, 2009).

The severity of invasive infections associated with CA-MRSA infections was reflected in the high proportion of subjects who

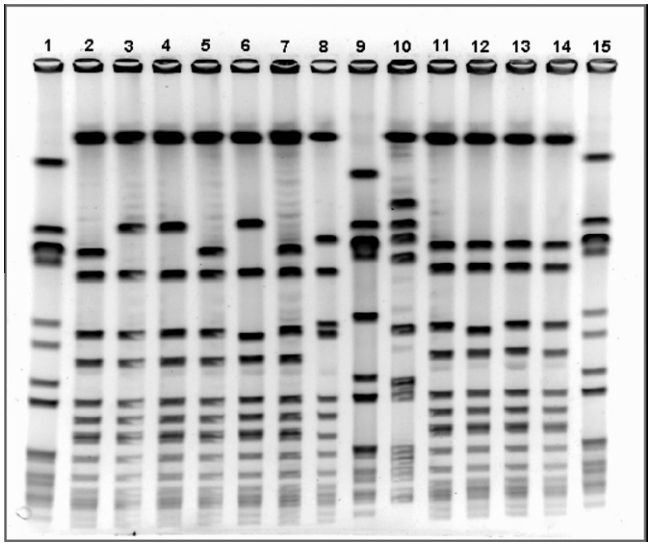

Fig. 1. Smal restriction patterns of prevalent clones in Argentina and representative isolates. Lane 1: Pulsotype A control (CAA clone), lane 2: 11-I-05 (subtype C1), lane 3: 11-I-04 (subtype C3) lane 4: 10-I-05 (subtype C3), lane 5: 10-I-03 (subtype C1), lane 6: Tan 104 (subtype C4), lane 7: Pulsotype C control, lane 8: Pulsotype D control, lane 9: 18-I-01 (subtype A), lane 10: 03-I-04 (subtype F2), lane 11: 03-I-05 (subtype C1), lane 12: 03-I-06 (subtype C5), lane13: 02-I-06 (subtype C1), lane 14: 12-I-02 (subtype C1), lane 15: Pulsotype A control (CAA clone).

were admitted to ICU, with vasoactive drugs or mechanical ventilation. The mortality in our population was relatively high (16%). This finding was in agreement with other studies (Hageman et al., 2006; Hidron et al., 2009). Importantly, this study provides key information to trace the evolution of CA-MRSA clones over the time. The major clone that had been dominant in Argentina during the past 6 years (CAA clone: ST5-SCC*mec* IV-*spa* t311), has been replaced by the ST30- SCC*mec* IVc-*spa* t019 clone.

CAA clone emerged in 2004 and existed at a high prevalence rate until now. In a multicenter study conducted in children, this CA-MRSA clone predominated in community-associated cases (82%) and healthcare-associated, community-onset cases (57%) in hospitals of northern, eastern and central regions of Argentina during 2007 and 2008 (Sola et al., 2012). Recently, we have demonstrated that this clone was also widely distributed in colonization of healthy children (Gardella et al., 2011). By contrast, in this study conducted between March 2010 and December 2011 the CA-MRSA lineage ST30, SCC*mec* IVc, *spa* t019 has been detected as the dominant clone in adolescent and adult patients with invasive diseases. The molecular characteristics shared by the isolates belonging to this major clone largely corresponded to those reported for a minor clone described in our previous study, among patients with skin and soft tissue infections in 2004–2006 (Gardella et al., 2008). The pattern of clonal lineages is now similar to that observed in neighboring countries and is clearly different from our previous findings. The clonal replacement is strongly supported by results obtained by our group in a prospective observational multicenter study of acute bacterial skin and related structure CA-MRSA infections conducted in the same period that this study: MRSA was isolated in 70% of patients (218/311) and ST30 clone was predominant (68%) (unpublished data).

The reasons to explain this replacement have to be explored, but ST30 lineage is now apparently well adapted to the Argentinean community as a successful CA-MRSA clone, that has undergone clonal expansion. We speculate that a combination of virulence factors and epidemic features including a higher ability to survive on human skin and mucosa would allow ST30 CA-MRSA/PFGE C to be a successful clone.

Table 3
Molecular characteristics of dominant CA-MRSA clones causing invasive infections in Argentina.

PFGE type	No. of isolates	ST	SCC*mec* type	*spa* type	*agr* allelic group
C	23	30	IVc	t019	III
A	2	5	IVa	t311, t2724	II
F	2	72	IV, IVvar	t1364, t148[a]	I
E	1	ND	IV	t002	II

[a] Within pulsotype F, the isolate t148 was PVL$^-$ and exhibited SCC*mec* IV variant) (*tnp20* was detected integrated into the class B *mec* complex).

The clonal complex 30 currently represents one of the major among HA-MRSA and CA-MRSA. In contrast to other clones which have been described to have a certain continent specificity, ST30 is distributed worldwide (Deleo et al., 2010; Vandenesch et al., 2003). In our region, ST30-SCC*mec* IV, PVL positive was the most disseminated clone in the large outbreak reported in Uruguay (limiting to the East of Argentina) (Ma et al., 2005).

This study provides an overview of the invasive CA-MRSA clones currently circulating in Argentina, but was limited by the availability of isolates for molecular studies, as they were sent by institutions belonging to Buenos Aires and Santa Fe states only, representing a limited geographic area (central) of our country. Given the large population served by the enrolled hospitals, our results are a good indicator of the evolving epidemiology of MRSA in our country.

Further surveillance in the region will be required to obtain a complete description of MRSA clones distribution and to determine whether colonization by these strains is becoming also prevalent.

Molecular epidemiological research is highly relevant for identifying the genetic variation that underlies changes in clinical behavior and to improve our understanding of the pathogenic performance of a particular clone.

Acknowledgments

We are very grateful to all the participants of the Study Group for CA-MRSA, SADI: Ameri D, Duarte A, Rolón MJ, Sisto A (Hospital Fernández); Nadal P, Salvador P, Targa L (Hospital Aeronáutico Central); Alfonso C, D'Alessandro D, Scapelatto P (Hospital Tornú) Burgos M, Gil D, Longo L, Rodríguez V (Hospital Santojani); Cantarella S, Gutiérrez M, Landaburu F, Puentes T, Zarlenga L, Torres C (Sanatorio Julio Méndez); Stern L, Tanco A (Hospital Español); Gullo H (HIGA Gral. Rodríguez); Ruzo G (Hospital NS de Luján); Prieto R, Cisneros J, Di Virgilio E, Rollet R (Hospital Muñiz), Costa R, Moyano M (Hospital Evita Pueblo, Berazategui); Méndez E, Morera G. (Hospital Cullen, Santa Fe).

This work was supported in part by grants from University of Buenos Aires, Argentina (20020100100510, 2011–2014)) and Agencia Nacional de Promoción Científica y Tecnológica (PICT 1634) to MM. M.M is member of "Carrera del Investigador" of CONICET.

References

Amorim, M.L., Faria, N.A., Oliveira, D.C., Vasconcelos, C., Cabeda, J.C., Mendes, A.C., Calado, E., Castro, A.P., Ramos, M.H., Amorim, J.M., de Lencastre, H., 2007. Changes in the clonal nature and antibiotic resistance profiles of methicillin-resistant *Staphylococcus aureus* isolates associated with spread of the EMRSA-15 clone in a tertiary care Portuguese hospital. J. Clin. Microbiol. 45, 2881–2888.

Chung, M., de Lencastre, H., Matthews, P., Tomasz, A., Adamsson, I., Aires de Sousa, M., Camou, T., Cocuzza, C., Corso, A., Couto, I., Dominguez, A., Gniadkowski, M., Goering, R., Gomes, A., Kikuchi, K., Marchese, A., Mato, R., Melter, O., Oliveira, D., Palacio, R., Sa-Leao, R., Santos Sanches, I., Song, J.H., Tassios, P.T., Villari, P., 2000. Molecular typing of methicillin-resistant *Staphylococcus aureus* by pulsed-field gel electrophoresis: comparison of results obtained in a multilaboratory effort using identical protocols and MRSA strains. Microb. Drug Resist. 6, 189–198.

CLSI, 2009. Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. Approved standard, seventh ed. CLSI document M7–A8. CLSI, Wayne, PA.

Coombs, G.W., Monecke, S., Pearson, J.C., Tan, H.L., Chew, Y.K., Wilson, L., Ehricht, R., O'Brien, F.G., Christiansen, K.J., 2011. Evolution and diversity of community-associated methicillin-resistant *Staphylococcus aureus* in a geographical region. BMC Microbiol. 11, 215.

David, M.Z., Daum, R.S., 2010. Community-associated methicillin-resistant *Staphylococcus aureus*: epidemiology and clinical consequences of an emerging epidemic. Clin. Microbiol. Rev. 23, 616–687.

Deleo, F.R., Otto, M., Kreiswirth, B.N., Chambers, H.F., 2010. Community-associated meticillin-resistant *Staphylococcus aureus*. Lancet 375, 1557–1568.

Deresinski, S., 2009. Vancomycin in combination with other antibiotics for the treatment of serious methicillin-resistant *Staphylococcus aureus* infections. Clin. Infect. Dis. 49, 1072–1079.

Deurenberg, R.H., Stobberingh, E.E., 2009. The molecular evolution of hospital – and community-associated methicillin-resistant *Staphylococcus aureus*. Curr. Mol. Med. 9, 100–115.

Enright, M.C., Knox, K., Griffiths, D., Crook, D.W., Spratt, B.G., 2000. Molecular typing of bacteria directly from cerebrospinal fluid. Eur. J. Clin. Microbiol. Infect. Dis. 19, 627–630.

Gardella, N., Picasso, R., Predari, S.C., Lasala, M., Foccoli, M., Benchetrit, G., Famiglietti, A., Catalano, M., Mollerach, M., Gutkind, G., 2005. Methicillin-resistant *Staphylococcus aureus* strains in Buenos Aires teaching hospitals: replacement of the multidrug resistant South American clone by another susceptible to rifampin, minocycline and trimethoprim-sulfamethoxazole. Rev. Argent. Microbiol. 37, 156–160.

Gardella, N., von Specht, M., Cuirolo, A., Rosato, A., Gutkind, G., Mollerach, M., 2008. Community-associated methicillin-resistant *Staphylococcus aureus*, Eastern Argentina. Diagn. Microbiol. Infect. Dis. 62, 343–347.

Gardella, N., Murzicato, S., Di Gregorio, S., Cuirolo, A., Desse, J., Crudo, F., Gutkind, G., Mollerach, M., 2011. Prevalence and characterization of methicillin-resistant *Staphylococcus aureus* among healthy children in a city of Argentina. Infect. Genet. Evol. 11, 1066–1071.

Gilot, P., Lina, G., Cochard, T., Poutrel, B., 2002. Analysis of the genetic variability of genes encoding the RNA III-activating components Agr and TRAP in a population of *Staphylococcus aureus* strains isolated from cows with mastitis. J. Clin. Microbiol. 40, 4060–4067.

Grundmann, H., Aanensen, D.M., van den Wijngaard, C.C., Spratt, B.G., Harmsen, D., Friedrich, A.W., 2010. Geographic distribution of *Staphylococcus aureus* causing invasive infections in Europe: a molecular-epidemiological analysis. PLoS Med. 7, e1000215.

Hageman, J.C., Liedtke, L.A., Sunenshine, R.H., Strausbaugh, L.J., McDonald, L.C., Tenover, F.C., 2006. Management of persistent bacteremia caused by methicillin-resistant *Staphylococcus aureus:* a survey of infectious diseases consultants. Clin. Infect. Dis. 43, e42–e45.

Harmsen, D., Claus, H., Witte, W., Rothganger, J., Turnwald, D., Vogel, U., 2003. Typing of methicillin-resistant *Staphylococcus aureus* in a university hospital setting by using novel software for *spa* repeat determination and database management. J. Clin. Microbiol. 41, 5442–5448.

Hidron, A.I., Low, C.E., Honig, E.G., Blumberg, H.M., 2009. Emergence of community-acquired meticillin-resistant *Staphylococcus aureus* strain USA300 as a cause of necrotising community-onset pneumonia. Lancet Infect. Dis. 9, 384–392.

Kennedy, A.D., Otto, M., Braughton, K.R., Whitney, A.R., Chen, L., Mathema, B., Mediavilla, J.R., Byrne, K.A., Parkins, L.D., Tenover, F.C., Kreiswirth, B.N., Musser, J.M., DeLeo, F.R., 2008. Epidemic community-associated methicillin-resistant *Staphylococcus aureus*: recent clonal expansion and diversification. Proc. Natl. Acad. Sci. USA 105, 1327–1332.

Klevens, R.M., Morrison, M.A., Nadle, J., Petit, S., Gershman, K., Ray, S., Harrison, L.H., Lynfield, R., Dumyati, G., Townes, J.M., Craig, A.S., Zell, E.R., Fosheim, G.E., McDougal, L.K., Carey, R.B., Fridkin, S.K., 2007. Invasive methicillin-resistant *Staphylococcus aureus* infections in the United States. JAMA 298, 1763–1771.

Kondo, Y., Ito, T., Ma, X.X., Watanabe, S., Kreiswirth, B.N., Etienne, J., Hiramatsu, K., 2007. Combination of multiplex PCRs for staphylococcal cassette chromosome *mec* type assignment: rapid identification system for *mec, ccr*, and major differences in junkyard regions. Antimicrob. Agents Chemother. 51, 264–274.

Korzeniowski, O., Sande, M.A., 1982. Combination antimicrobial therapy for *Staphylococcus aureus* endocarditis in patients addicted to parenteral drugs and in nonaddicts: a prospective study. Ann. Intern. Med. 97, 496–503.

Lina, G., Piemont, Y., Godail-Gamot, F., Bes, M., Peter, M.O., Gauduchon, V., Vandenesch, F., Etienne, J., 1999. Involvement of Panton-Valentine leukocidin-producing *Staphylococcus aureus* in primary skin infections and pneumonia. Clin. Infect. Dis. 29, 1128–1132.

Liu, C., Bayer, A., Cosgrove, S.E., Daum, R.S., Fridkin, S.K., Gorwitz, R.J., Kaplan, S.L., Karchmer, A.W., Levine, D.P., Murray, B.E., M, J.R., Talan, D.A., Chambers, H.F., 2011. Clinical practice guidelines by the infectious diseases society of America for the treatment of methicillin-resistant *Staphylococcus aureus* infections in adults and children: executive summary. Clin. Infect. Dis. 52, 285–292.

Ma, X.X., Ito, T., Tiensasitorn, C., Jamklang, M., Chongtrakool, P., Boyle-Vavra, S., Daum, R.S., Hiramatsu, K., 2002. Novel type of staphylococcal cassette chromosome *mec* identified in community-acquired methicillin-resistant *Staphylococcus aureus* strains. Antimicrob. Agents Chemother. 46, 1147–1152.

Ma, X.X., Galiana, A., Pedreira, W., Mowszowicz, M., Christophersen, I., Machiavello, S., Lope, L., Benaderet, S., Buela, F., Vincentino, W., Albini, M., Bertaux, O., Constenla, I., Bagnulo, H., Llosa, L., Ito, T., Hiramatsu, K., 2005. Community-acquired methicillin-resistant *Staphylococcus aureus*, Uruguay. Emerg. Infect. Dis. 11, 973–976.

Maree, C.L., Daum, R.S., Boyle-Vavra, S., Matayoshi, K., Miller, L.G., 2007. Community-associated methicillin-resistant *Staphylococcus aureus* isolates causing healthcare-associated infections. Emerg. Infect. Dis. 13, 236–242.

Milheirico, C., Oliveira, D.C., de Lencastre, H., 2007. Multiplex PCR strategy for subtyping the staphylococcal cassette chromosome *mec* type IV in methicillin-resistant *Staphylococcus aureus*: 'SCC*mec* IV multiplex'. J. Antimicrob. Chemother. 60, 42–48.

Naimi, T.S., LeDell, K.H., Como-Sabetti, K., Borchardt, S.M., Boxrud, D.J., Etienne, J., Johnson, S.K., Vandenesch, F., Fridkin, S., O'Boyle, C., Danila, R.N., Lynfield, R., 2003. Comparison of community-and health care-associated methicillin-resistant *Staphylococcus aureus* infection. JAMA 290, 2976–2984.

Nimmo, G.R., Coombs, G.W., Pearson, J.C., O'Brien, F.G., Christiansen, K.J., Turnidge, J.D., Gosbell, I.B., Collignon, P., McLaws, M.L., 2006. Methicillin-resistant *Staphylococcus aureus* in the Australian community: an evolving epidemic. Med. J. Aust. 184, 384–388.

Oliveira, D.C., de Lencastre, H., 2002. Multiplex PCR strategy for rapid identification of structural types and variants of the *mec* element in methicillin-resistant *Staphylococcus aureus*. Antimicrob. Agents Chemother. 46, 2155–2161.

Rolo, J., Miragaia, M., Turlej-Rogacka, A., Empel, J., Bouchami, O., Faria, N.A., Tavares, A., Hryniewicz, W., Fluit, A.C., de Lencastre, H., 2012. High genetic diversity among community-associated *Staphylococcus aureus* in Europe: results from a multicenter study. PLoS ONE 7, e34768.

Skinner, D., Keefer, C.S., 1941. Significance of bacteremia caused by *Staphylococcus aureus*. Arch. intern. med. 68, 851–875.

Smith, J.M., Cook, G.M., 2005. A decade of community MRSA in New Zealand. Epidemiol. Infect. 133, 899–904.

Sola, C., Saka, H.A., Vindel, A., Bocco, J.L., 2008. Emergence and dissemination of a community-associated methicillin-resistant Panton-Valentine leucocidin-positive *Staphylococcus aureus* clone sharing the sequence type 5 lineage with the most prevalent nosocomial clone in the same region of Argentina. J. Clin. Microbiol. 46, 1826–1831.

Sola, C., Paganini, H., Egea, A.L., Moyano, A.J., Garnero, A., Kevric, I., Culasso, C., Vindel, A., Lopardo, H., Bocco, J.L., 2012. Spread of epidemic MRSA-ST5-IV clone encoding PVL as a major cause of community onset Staphylococcal infections in Argentinean children. PLoS ONE 7, e30487.

Stemper, M.E., Shukla, S.K., Reed, K.D., 2004. Emergence and spread of community-associated methicillin-resistant *Staphylococcus aureus* in rural Wisconsin, 1989 to 1999. J. Clin. Microbiol. 42, 5673–5680.

Stryjewski, M.E., Chambers, H.F., 2008. Skin and soft-tissue infections caused by community-acquired methicillin-resistant *Staphylococcus aureus*. Clin. Infect. Dis. 46 (Suppl. 5), S368–S377.

Vandenesch, F., Naimi, T., Enright, M.C., Lina, G., Nimmo, G.R., Heffernan, H., Liassine, N., Bes, M., Greenland, T., Reverdy, M.E., Etienne, J., 2003. Community-acquired methicillin-resistant *Staphylococcus aureus* carrying Panton-Valentine leukocidin genes: worldwide emergence. Emerg. Infect. Dis. 9, 978–984.

von Specht, M., Gardella, N., Tagliaferri, P., Gutkind, G., Mollerach, M., 2006. Methicillin-resistant *Staphylococcus aureus* in community-acquired meningitis. Eur. J. Clin. Microbiol. Infect. Dis. 25, 267–269.