

First isolation of Methicillin-Resistant Livestock-Associated *Staphylococcus aureus* (LA-MRSA) CC398 and CC1 in intensive pig production farms in Argentina

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Since the mid-2000s, LA-MRSA has been identified among pigs worldwide, being CC398 the most relevant clone. In a previous study we evaluated nasal colonization of MRSA in fattening pigs from four farms located in Buenos Aires, Santa Fe and San Luis provinces. Sixty-four piglets aged 25-154 days were screened for MRSA colonization between 2019 and 2021 and 19 LA-MRSA were isolated. The aim of the present study was to characterize the first LA-MRSA isolated from pigs in Argentina by different molecular techniques, and to determine their clonal diversity.

Antimicrobial resistance was evaluated by disk diffusion according to CLSI. Genetic characterization was assessed by PCR (*mecA* and *sau1-hsdS1* genes), SmaI-PFGE and WGS by Illumina.

The 19 isolates showed resistance to penicillin, ceftiofur, tetracycline, chloramphenicol and ciprofloxacin, and susceptibility to rifampicin, nitrofurantoin, vancomycin and linezolid. 79% were resistant to clindamycin and lincomycin, 68% to erythromycin, 58% to gentamicin and 37% to trimethoprim/sulfamethoxazole. All the isolates were MDR, being resistant to five to seven different antimicrobial families.

All the isolates carried the *mecA* gene and the *sau1-hsdS1* gene specific sequence characteristic of CC398 was detected in 11/19. All the isolates were SCC*mecV* and PVL negative. By SmaI-PFGE, 12/19 isolates were differentiated in 3 clonal types and the remaining isolates were repeatedly untypeable. Four representative isolates selected according to PFGE clonal types were WGS.

All the sequenced isolates carried *mecA*, *bla_Z*, *fexA* and at least one gene encoding tetracycline resistance (*tet(M)*, *tet(L)*, *tet(K)* and/or *tet38*) and presented mutations in ParC and GyrA. Gentamicin resistant isolates harbored the *aac(6′)-Ie-aph(2′′)-Ia* gene and trimethoprim/sulfamethoxazole resistant isolates the *dfpG* gene. Complete correlation between phenotype and genotype was observed for all the antibiotic tested. 11/19 isolates from Buenos Aires and Santa Fe were ST398 and 8/19 isolates from Buenos Aires and San Luis were ST9, CC1.

LA-MRSA from Argentinian fattening pigs belonged to two genetic lineages, CC398 and CC1, frequently associated with pigs in other countries. Our study alerts about the emergence of CC398 in Argentina and underscores the importance of monitoring the evolution of LA-MRSA in pig farms in order to implement control measures and reduce the risk of spread in animal and human population.

Keywords: livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA); pigs; ST398; whole-genome sequencing; zoonosis