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Whole-Genome Sequence of Livestock-Associated ST398 Methicillin-Resistant *Staphylococcus aureus* Isolated from Humans in Canada

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Despite reports of high colonization rates of ST398 livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA) among pigs and pig farmers, the incidence of LA-MRSA infection in the general population in Canada appears to be rare in comparison to that in some European countries. In this study, the complete genome sequence of a Canadian representative LA-MRSA isolate (08BA02176) from a human postoperative surgical site infection was acquired and compared to the sequenced genome of an LA-MRSA isolate (S0385) from Europe to identify genetic traits that may explain differences in the success of these particular strains in some locales.

In Canada, livestock-associated methicillin-resistant *Staphylococcus aureus* (LA-MRSA) was first reported in Southwestern Ontario pigs and pig farmers, where prevalence of MRSA colonization was 24.9% (71/285) and 20% (5/25), respectively (7). Whereas LA-MRSA accounted for >20% of all MRSA in 2006 in the Netherlands (14), ST398 is rare (0.14%) among MRSA isolates from humans in Canada, at least those in 2 provinces (3). Recently, the whole-genome sequence of a European LA-MRSA isolate (S0385) was reported (13). In the current study, the complete genome of a Canadian representative LA-MRSA isolate (08BA02176; Ridom *spa* type t034), obtained in 2008 from a human postoperative surgical site infection, was acquired and compared to S0385 to identify genetic traits that may explain the differences in the success of these particular strains. GS FLX titanium pyrosequencing reads were assembled *de novo* by using Newbler version 1.1.03.24 (Roche Diagnostics). Contig gaps in the draft genome were closed with gap-spanning PCR and Sanger sequencing of a fosmid template library. Genome annotation was achieved using an in-house modified version of GenDB (9).

As with S0385 and other characterized ST398 isolates, 08BA2176 lacked many virulence determinants present in other epidemic MRSA strain types, including enterotoxin and exfoliative toxin genes (6, 10–12, 16). Both S0385 and 08BA2176 harbored SAP1-S0385, encoding two putative extracellular proteins with similarity to staphylococcal complement inhibitor and von Willebrand factor-binding protein, which have previously been implicated in ruminant host specificity (4, 13, 15). Genomic comparisons between S0385 and 08BA2176 revealed that 08BA2176 lacked all 3 plasmids, phage phiSa2, and the 3 integrative conjugative elements (ICESa1A, ICESa1B, ICESa2). The ICES have been proposed to enhance the ability of S0385 to acquire foreign DNA (13). Unique to 08BA2176 was a novel staphylococcal cassette chromosome *mec* element type V (SCC*mec* V) subtype (3) and the insertion of Tn5406 into the chromosomal *att554* site, which harbors a variant of *vga*(A) encoding an ABC transporter conferring resistance to pleuromutins, lincosamides, and streptogramin A (5). The SCC*mec* V subtype identified in 08BA2176 included an

ADP-ribosylglycohydrolase, a permease for cytosine/purines, and a ribokinase in the J3 region. A second unique feature present in the SCC*mec* element of 08BA2176 was a clustered regularly interspaced short palindromic repeats (CRISPR) array identified in the J1 region. CRISPRs and associated *cas* genes have been shown to be involved in sequence-directed immunity against phages (1, 2, 8) and plasmids (8). PCR screening revealed that 6/16 Canadian ST398 isolates and 1/29 isolates from a small international collection (6 from the United States, 8 from Belgium, and 15 from Germany) harbored this same CRISPR element, which could provide an additional marker for further delineating these ST398 strains.

We posit that the lack of encoded virulence determinants in the ST398 lineage is likely contributing to the comparatively low incidence of observed human infections in Canada. Further work is required to delineate whether CRISPR element presence confers resistance to plasmids and integrative phages and accounts for why 08BA2176 contains fewer antimicrobial resistance genes and phage-encoded virulence factors relative to S0385 and other epidemic MRSA strains.

Nucleotide sequence accession number. This genome project has been deposited in GenBank under accession no. CP003808.

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