Genome Sequences of Livestock-Associated Methicillin-Resistant \textit{Staphylococcus aureus} \textit{spa} Type t899 Strains Belonging to Three Different Sequence Types (ST398, ST9, and ST4034)

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ABSTRACT Livestock-associated methicillin-resistant \textit{Staphylococcus aureus} (LA-MRSA) is an emerging MRSA lineage rapidly evolving in the community. In this report, we present the draft genome sequences of nine LA-MRSA strains. These strains were isolated from meat and a human nasal swab sample and belong to one unique \textit{spa} type (t899), but to three different sequence types, ST398, ST9, and ST4034.

Livestock-associated methicillin-resistant \textit{Staphylococcus aureus} (LA-MRSA) is the largest MRSA pool in humans outside the hospital setting, with livestock as a primary reservoir (1, 2). This lineage is predominantly represented by clonal complex 398 (CC398), but it also comprises other clonal complexes, including CC9 and CCS (2–4).

\textit{Staphylococcus aureus} protein A (\textit{spa}) typing has a remarkable predictive power over clonal relatedness (5, 6). In most instances, a single \textit{spa} type is strictly associated with a specific multilocus sequence type (MLST). However, some exceptions do exist, such as \textit{spa} type t899, which is reported in multiple sequence types, namely, ST398 and ST9. In this report, we present the draft genome sequences of nine LA-MRSA strains, all belonging to \textit{spa} type t899 but clustering in three different sequence types, ST398 (3), ST9 (5), and ST4034 (1) (Table 1).

Isolates were obtained from meat samples collected in retail markets and from a nasal swab sample from a dialysis patient that was taken during hospital screening in the Czech Republic. The meat from which the samples were drawn was produced in different countries and sold in the Czech Republic. The samples were primarily enriched in buffered peptone water and cultured on Baird-Parker agar. Presumptive \textit{S. aureus} colonies were transferred to blood agar and confirmed using matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) (7). All MRSA isolates were identified using PCR detection of the \textit{S. aureus}-specific fragment SA442 and the \textit{mecA} gene (8). MLST (https://cge.cbs.dtu.dk/services/MLST) (9) and \textit{spa} typing (https://www.spaserver.ridom.de) (10) were performed prior to the whole-genome sequence run.

Total genomic DNA was extracted using a DNeasy blood and tissue kit (Qiagen, Valencia, CA) from pure culture colonies cultivated on Columbia sheep blood agar (Bio-Rad Laboratories, Temse, Belgium). Whole-genome sequencing was performed with a MiSeq sequencing platform (Illumina, San Diego, CA). Library preparation was performed with the Nextera XT DNA sample preparation kit (Illumina). The libraries were then sequenced using a 250-bp paired-end protocol (MiSeq reagent kit v3, Illumina) according to the manufacturer’s instructions. Data analysis was performed...
TABLE 1 Livestock-associated methicillin-resistant *Staphylococcus aureus* type 1899 isolates

| Isolate IDa | Isolate source | Sample originb | Year of isolation | STc | No. of raw reads | Genome coverage (x) | No. of contigs | $N_{50}$ (bp)d | GC content (%)d | Genome size (bp) | GenBank accession no. |
|-------------|----------------|----------------|-------------------|-----|-----------------|---------------------|---------------|---------------|----------------|------------------|----------------------|
| SAV0154     | Pork           | CZ             | 2013              | 103 | 772,052         | 64.34               | 178           | 39,112        | 32.91          | 2,781,027        | QYAQ00000000         |
| SAV0987     | Human          | CZ             | 2017              | 398 | 1,109,012       | 92.42               | 196           | 37,830        | 32.79          | 2,922,579        | QYAX00000000         |
| SAV1035     | Poultry meat   | PO             | 2017              | 9   | 1,062,670       | 88.56               | 123           | 85,312        | 32.71          | 2,783,450        | QYAW00000000         |
| SAV1109     | Poultry meat   | PO             | 2017              | 398 | 699,934         | 58.33               | 303           | 20,030        | 32.82          | 2,857,636        | QYAV00000000         |
| SAV1146     | Poultry meat   | DE             | 2017              | 9   | 675,512         | 56.29               | 131           | 55,180        | 32.85          | 2,756,154        | QYAS00000000         |
| SAV1149     | Poultry meat   | DE             | 2017              | 9   | 893,060         | 74.42               | 277           | 21,660        | 32.79          | 2,749,354        | QYAT00000000         |
| SAV1150     | Poultry meat   | DE             | 2017              | 9   | 808,170         | 67.35               | 99            | 78,511        | 32.75          | 2,896,873        | QYAV00000000         |
| SAV1158     | Poultry meat   | DE             | 2017              | 9   | 1,231,020       | 102.58              | 64            | 135,732       | 32.73          | 2,896,873        | QYAV00000000         |
| SAV1228     | Pork           | CZ             | 2017              | 9   | 2,321,580       | 193.47              | 116           | 80,797        | 32.71          | 2,730,307        | QYAQ00000000         |

#D. Identification.
#C. CZ, Czech Republic; DE, Germany; PO, Poland.
#ST. Sequence type.
#N50 value and GC percentage were calculated based on contigs of ≥500 bp.

using an in-house instance of the Galaxy workflow management system (11). Sequencing yielded a total of 9,573,010 reads with 35- to 251-bp read lengths. Raw reads were quality checked with FastQC v.0.65, and low-quality reads were trimmed using Trimmomatic v.0.36.4 (12). Subsequently, assemblies were generated using the SPAdes v.3.1.3 algorithm (13). Contigs ≥200 bp long were retained in the assembly. The genome sizes ranged from 2,730,307 to 2,922,579 bp. The average GC content and the $N_{50}$ value were 32.8% and 61,574 bp, respectively. Final assemblies consisted of 64 to 303 contigs with an average coverage of 88.64× (Table 1). Annotation was carried out using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (https://www.ncbi.nlm.nih.gov/genome/annotation_prok/) (14).

**Data availability.** The genome sequences reported here have been deposited at DDBJ/ENA/GenBank under the accession numbers QYAQ00000000 to QYAY00000000. The versions described in this paper are the first versions, QYAQ01000000 to QYAY01000000 (Table 1). Raw sequences are available under the SRA study accession number SRP161670.

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