



Complete Genome Sequences of Four *Salmonella enterica* Strains (Including Those of Serotypes Montevideo, Mbandaka, and Lubbock) Isolated from Peripheral Lymph Nodes of Healthy Cattle

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ABSTRACT *Salmonella enterica* serotype Lubbock emerged most likely from a *Salmonella enterica* serotype Mbandaka ancestor that acquired by recombination the *fliC* operon from *Salmonella enterica* serotype Montevideo. Here, we report the complete genome sequence of two *S. Lubbock*, one *S. Montevideo*, and one *S. Mbandaka* strain isolated from bovine lymph nodes.

Salmonella enterica serotype Lubbock (6,7:g,m,s:e,n,z₁₅) has been isolated from peripheral lymph nodes (PLNs) of healthy cattle and bovine liver abscesses (1, 2). This novel antigenic formula was first defined by agglutination and confirmed genetically. This novel serotype most likely emerged from a recombination event transferring the *fliC* operon of *Salmonella enterica* serotype Montevideo into a *Salmonella enterica* serotype Mbandaka ancestor (3). At least two recombination events occurred, given that two *S. Lubbock* lineages represented by the distinct isolates 10TTU468x and 11TTU1590 were recovered from PLNs of healthy cattle. The closed genome sequences are presented here together with the closed genome sequences of one *S. Mbandaka* strain (11TTU1615b) and one *S. Montevideo* strain (11TTUC-046) also isolated from PLNs (4, 5). Whole-genome sequencing of these four strains was performed to inform possible genomic evolutionary events leading to the emergence of the novel mosaic *S. Lubbock* serotype. Genomic DNA was extracted from a single-colony overnight culture at 37°C in tryptic soy broth using Qiagen Genomic-tip 100/G columns and blood and cell culture DNA midi kits (Qiagen, Valencia, CA), following the manufacturer's recommendations. Long-read sequencing was performed by single-molecule real-time technology, using C4/P6 (chemistry/polymerase) on an RS II instrument (Pacific Biosciences, Menlo Park, CA). Raw data were assembled using SMRT Analysis software version 2.3.0 (Pacific Biosciences) and polished with MiSeq-generated paired-end Illumina reads using Pilon version 1.18 (6).

Primary analysis revealed that none of the four strains carried a plasmid and that chromosome size and GC content varied slightly (average coverage, 68×) (Table 1) (6). Default parameters were used for all analyses. Serotypes were confirmed using the SeqSero pipeline (7). The presence of antimicrobial resistance genes was queried using CARD (minimum identity and query coverage minimum, 95%), highlighting the presence of the same single cryptic resistance gene against aminoglycosides [*aac(6′)-Iaa*] in each strain (8, 9). Furthermore, all four genomes contain genes encoding transporters noted for their involvement in resistance phenotypes to various antimicrobials. The presence of virulence genes and *Salmonella* pathogenicity islands (SPI) was investi-

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TABLE 1 Characteristics of the four *S. enterica* strain genomes

Characteristic	Value for strain			
	10TTU468x	11TTU1590	11TTU1615b	11TTUC-046
GC content (%)	52.11	52.11	52.11	52.35
PacBio reads				
No. of reads	96,785	347,976	243,332	274,427
Avg read length (bp)	7,166	10,035	9,873	9,254
Total bases sequenced (bp)	693,598,459	3,491,953,649	2,402,449,873	2,539,597,496
Assembly size (bp)	4,985,863	4,985,874	4,985,867	4,588,222
Illumina reads				
No. of reads	2,838,092	2,567,890	1,040,576	2,341,584
Avg read length (bp)	144	141	222	142
No. of contigs	176	167	201	115
Assembly size (bp)	4,946,889	4,944,961	5,035,112	4,556,712

gated (SPIFinder, version 1.0; <https://cge.cbs.dtu.dk/services/SPIFinder/>) and revealed the presence of C63PI (iron transport in SPI-1), SPI-2, and SPI-4 in each strain. While only the *S. Montevideo* isolate carried SPI-3, SPI-13, and SPI-14, the *S. Lubbock* and *S. Mbandaka* isolates harbored more virulence-associated genes (242 and 245 genes, respectively) than did the *S. Montevideo* isolate (227 genes) (10). This difference may be explained by mobile genetic elements. The *S. Lubbock* and *S. Mbandaka* strains possessed a larger number of mobile elements (87, 88, and 88 for 10TTU468x, 11TTU1590, and 11TTU1615b, respectively) than did the *S. Montevideo* isolate (22 genes) (RASTk; <http://rast.nmpdr.org/>). These data support the hypothesis that *S. Mbandaka* is likely a progenitor of *S. Lubbock* that arose from the acquisition of *flhC_{g,m,s}* in a recombination event with *S. Montevideo*.

Data availability. Whole-genome sequences have been deposited in DDBJ/ENA/GenBank under the accession and BioProject numbers CP032814 and PRJNA494676 (10TTU468x), CP032817 and PRJNA494681 (11TTU1590), CP032815 and PRJNA494678 (11TTU1615b), and CP032816 and PRJNA494679 (11TTUC-046), respectively. Whole-genome shotgun projects have been deposited in DDBJ/ENA/GenBank under the accession and BioProject numbers JXYU00000000 and PRJNA274416 (10TTU468x), JXYV00000000 and PRJNA274419 (11TTU1590), QYWD00000000 and PRJNA491195 (SRA number SRX5075396) (11TTU1615b), and QYWT00000000 and PRJNA491195 (SRA number SRX5075395) (11TTUC-046), respectively.

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REFERENCES

- Amachawadi RG, Nagaraja TG. 2015. First report of anaerobic isolation of *Salmonella enterica* from liver abscesses of feedlot cattle. *J Clin Microbiol* 53:3100–3101. <https://doi.org/10.1128/JCM.01111-15>.
- Amachawadi RG, Thomas M, Nagaraja TG, Scaria J. 2016. Genome sequences of *Salmonella enterica* subsp. *enterica* serovar Lubbock strains isolated from liver abscesses of feedlot cattle. *Genome Announc* 4:e00319-16.
- Bugarel M, den Bakker HC, Nightingale KK, Brichta-Harhay DM, Edrington TS, Loneragan GH. 2015. Two draft genome sequences of a new serovar of *Salmonella enterica*, serovar Lubbock. *Genome Announc* 3:e00215-15.
- Gragg SE, Loneragan GH, Brashears MM, Arthur TM, Bosilevac JM, Kalchayanand N, Wang R, Schmidt JW, Brooks JC, Shackelford SD, Wheeler TL, Brown TR, Edrington TS, Brichta-Harhay DM. 2013. Cross-sectional study examining *Salmonella enterica* carriage in subiliac lymph nodes of cull and feedlot cattle at harvest. *Foodborne Pathog Dis* 10:368–374. <https://doi.org/10.1089/fpd.2012.1275>.
- Gragg SE, Loneragan GH, Nightingale KK, Brichta-Harhay DM, Ruiz H, Elder JR, Garcia LG, Miller MF, Echeverry A, Ramirez Porras RG, Brashears MM. 2013. Substantial within-animal diversity of *Salmonella* isolates from lymph nodes, feces, and hides of cattle at slaughter. *Appl Environ Microbiol* 79:4744–4750. <https://doi.org/10.1128/AEM.01020-13>.

6. Walker BJ, Abeel T, Shea T, Priest M, Abouelliel A, Sakthikumar S, Cuomo CA, Zeng Q, Wortman J, Young SK, Earl AM. 2014. Pilon: an integrated tool for comprehensive microbial variant detection and genome assembly improvement. *PLoS One* 9:e112963. <https://doi.org/10.1371/journal.pone.0112963>.
7. Zhang S, Yin Y, Jones MB, Zhang Z, Deatherage Kaiser BL, Dinsmore BA, Fitzgerald C, Fields PI, Deng X. 2015. *Salmonella* serotype determination utilizing high-throughput genome sequencing data. *J Clin Microbiol* 53:1685–1692. <https://doi.org/10.1128/JCM.00323-15>.
8. Jia B, Raphenya AR, Alcock B, Waglehner N, Guo P, Tsang KK, Lago BA, Dave BM, Pereira S, Sharma AN, Doshi S, Courtot M, Lo R, Williams LE, Frye JG, Elsayegh T, Sardar D, Westman EL, Pawlowski AC, Johnson TA, Brinkman FSL, Wright GD, McArthur AG. 2017. CARD 2017: expansion and model-centric curation of the comprehensive antibiotic resistance database. *Nucleic Acids Res* 45:D566–D573. <https://doi.org/10.1093/nar/gkw1004>.
9. Salipante SJ, Barlow M, Hall BG. 2003. GeneHunter, a transposon tool for identification and isolation of cryptic antibiotic resistance genes. *Antimicrob Agents Chemother* 47:3840–3845. <https://doi.org/10.1128/AAC.47.12.3840-3845.2003>.
10. Chen L, Xiong Z, Sun L, Yang J, Jin Q. 2012. VFDB 2012 update: toward the genetic diversity and molecular evolution of bacterial virulence factors. *Nucleic Acids Res* 40:D641–D645. <https://doi.org/10.1093/nar/gkr989>.