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Genome Sequence of *Listeria monocytogenes* Scott A, a Clinical Isolate from a Food-Borne Listeriosis Outbreak[▽]

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Listeria monocytogenes **is an opportunistic food-borne pathogen and the causative agent of listeriosis in animals and humans. We present the genome sequence of** ***Listeria monocytogenes*** **Scott A, a widely distributed and frequently used serovar 4b clinical isolate from the 1983 listeriosis outbreak in Massachusetts.**

Listeria monocytogenes is an infectious organism and member of the *Firmicutes*. Listeriosis manifests itself as septicemia or meningoencephalitis in elderly and immunocompromised patients, while neonatal listeriosis can lead to abortion and stillbirth. *L. monocytogenes* is transmitted through raw and ready-to-eat foods. Due to changes in food processing and distribution over the last few decades, the rate of listeriosis in Europe is increasing (1, 11).

Since its isolation during the Massachusetts listeriosis outbreak in 1983 (3), the serovar 4b strain Scott A has been widely used as a reference strain for efficacy testing of food processing and preservation techniques (4, 6, 7), establishment of detection methods in foods (12), growth and heat resistance studies (2), and virulence studies (8). Scott A also harbors the temperate phage PSA, which employs unusual decoding mechanisms (14), and its integration into the 3′ end of single-copy $tRNA^{Arg}$ was used for design of a now widely used integration vector (9). However, its genome sequence has not been determined.

A paired-end library (200-bp fragments) was sequenced on an Illumina genome analyzer II, yielding 6,152,268 reads of 31 bp (62-fold total coverage). A high-quality draft genome was assembled with CLC Genomics Workbench 4.6.1 (CLC Bio, Aarhus, Denmark) using a combinatorial approach of reference assembly against *L. monocytogenes* F2365 (GenBank accession number AE017262.2), *de novo* assembly of nonassembled reads, and manual editing. The length of this draft genome is 3,021,822 bp, distributed in 5 contigs (from which the last two form a scaffold), with an average GC content of 37.9%. The final pseudogenome corresponds unequivocally with the physical macrorestriction map of Scott A assembled previously, based on restriction analysis and contour-clamped homogeneous electric field electrophoresis (5). Gene prediction and functional annotation were performed using the DIYA pipeline (13) with manual editing, taking into account the presence of ribosome binding sites, BLASTx results, and the open reading frame (ORF) prediction tool of CLC Genomics Workbench. The chromosome features 2,953 predicted ORFs, at least 65 tRNA genes, two pseudo-tRNAs, six copies of 16S-23S-5S operons, and two prophage sequences.

Scott A possesses a conserved *prfA* virulence gene cluster with high sequence identity (99%) to *Listeria* isolates of lineage I. Comparative analysis of the multilocus sequence typing (MLST) determinants (10) showed that this strain is closely related to lineage I clonal complex 2 (CC2), sequence type 290 (ST290). The only previous isolate of type ST290 is another human clinical serovar 4b strain from Canada (10). Scott A carries *inlA*, *inlB*, *inlD* (2×), *inlE*, *inlI*, *inlJ*, and 18 other leucine-rich internalin-like coding sequences. InlA is produced from a full-length allele type 22-23-24, which has been linked to human illness (10). A gene cluster unique to Scott A (CDS 2150-2470, a putative mobile genetic element from *Enterococcus faecalis*), comprises 32 genes, including a complete arsenic resistance operon and a cadmium-translocating ATPase.

Nucleotide sequence accession number. The sequencing project has been deposited at the Genomes OnLine Database (GOLD) under Gi09588 and at DDBJ/EMBL/GenBank under the accession number AFGI00000000. The version described in this paper is the first version, AFGI01000000. The complete annotation has been assigned GenBank accession number CM001159.

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REFERENCES

1. **Allerberger, F., and M. Wagner.** 2010. Listeriosis: a resurgent foodborne infection. Clin. Microbiol. Infect. **16:**16–23.
2. **Bereksi, N., F. Gavini, T. Benezech, and C. Faille.** 2002. Growth, morphology and surface properties of Listeria monocytogenes Scott A and LO28 under saline and acid environments. J. Appl. Microbiol. **92:**556–565.
3. **Fleming, D. W., et al.** 1985. Pasteurized milk as a vehicle of infection in an outbreak of listeriosis. N. Engl. J. Med. **312:**404–407.
4. **Guenther, S., D. Huwyler, S. Richard, and M. J. Loessner.** 2009. Virulent bacteriophage for efficient biocontrol of Listeria monocytogenes in ready-to-eat foods. Appl. Environ. Microbiol. **75:**93–100.
5. **He, W., and J. B. Luchansky.** 1997. Construction of the temperature-sensitive vectors pLUCH80 and pLUCH88 for delivery of Tn917::NotI/SmaI and use of these vectors to derive a circular map of Listeria monocytogenes Scott A, a serotype 4b isolate. Appl. Environ. Microbiol. **63:**3480–3487.
6. **Ita, P. S., and R. W. Hutkins.** 1991. Intracellular pH and survival of Listeria monocytogenes Scott A in tryptic soy broth containing acetic, lactic, citric, and hydrochloric acids. J. Food Prot. **54:**15–19.
7. **Jin, T.** 2010. Inactivation of Listeria monocytogenes in skim milk and liquid

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egg white by antimicrobial bottle coating with polylactic acid and nisin. J. Food Sci. **75:**M83–M88.
8. **Lammerding, A. M., K. A. Glass, A. Gendron-Fitzpatrick, and M. P. Doyle.** 1992. Determination of virulence of different strains of Listeria monocytogenes and Listeria innocua by oral inoculation of pregnant mice. Appl. Environ. Microbiol. **58:**3991–4000.
9. **Lauer, P., M. Y. N. Chow, M. J. Loessner, D. A. Portnoy, and R. Calendar.** 2002. Construction, characterization, and use of two Listeria monocytogenes site-specific phage integration vectors. J. Bacteriol. **184:**4177–4186.
10. **Ragon, M., et al.** 2008. A new perspective on Listeria monocytogenes evolution. PLoS Pathog. **4:**e1000146. doi:10.1371/journal.ppat.1000146.
11. **Schlech, W. F.** 2000. Foodborne listeriosis. Clin. Infect. Dis. **31:**770–775.
12. **Schmelcher, M., et al.** 2010. Rapid multiplex detection and differentiation of Listeria cells by use of fluorescent phage endolysin cell wall binding domains. Appl. Environ. Microbiol. **76:**5745–5756.
13. **Stewart, A. C., B. Osborne, and T. D. Read.** 2009. DIYA: a bacterial annotation pipeline for any genomics lab. Bioinformatics **25:**962–963.
14. **Zimmer, M., E. Sattelberger, R. B. Inman, R. Calendar, and M. J. Loessner.** 2003. Genome and proteome of Listeria monocytogenes phage PSA: an unusual case for programmed +1 translational frameshifting in structural protein synthesis. Mol. Microbiol. **50:**303–317.