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PROKARYOTES

Full-Genome Sequence of *Listeria monocytogenes* Strain H34, Isolated from a Newborn with Sepsis in Uruguay

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ABSTRACT The foodborne pathogen *Listeria monocytogenes* causes severe disease mainly in the vulnerable populations of the young, old, pregnant, and immunocompromised. Here, we present the genome sequence of *L. monocytogenes* H34, a serotype 1/2b, lineage I, sequence type 489 (ST489) strain, isolated from a neonatal sepsis case in Uruguay.

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In August 2016, 10 human cases of listeriosis were observed in Uruguay. As part of a subcluster consisting of 2 strains with similar pulsed-field gel electrophoresis (PFGE) patterns (ApaI and AscI), *L. monocytogenes* strain H34 was isolated from blood and lung cultures from a newborn with sepsis.

Serology (using antisera from Denka Seiken, Dagmersellen, Switzerland) and 7-gene multilocus sequence typing (MLST) (http://bigsdb.pasteur.fr/) identified H34 as a serotype 1/2b, lineage I, sequence type 489 (ST489), clonal complex 489 (CC489) strain. While lineage I, serotype 1/2b strains are commonly associated with human clinical cases (1), there is currently only 1 CC489 entry in the Pasteur MLST database.

For sequencing, DNA was isolated using the GenElute bacterial genomic DNA kit (Sigma, Buchs, Switzerland). The DNA was sheared to 10 kb in size and size-selected, and PacBio libraries were prepared using C4-P6 sequencing chemistry (Pacific Biosciences, Menlo Park, CA, USA). A total of 4.5×10^9 bp of reads were obtained using 2 single-molecule real-time (SMRT) cells in a PacBio RSII device (Pacific Biosciences). A *de novo* assembly was performed with the HGAP_3 algorithm (2), resulting in a single contig with a 945× coverage. Genome closure was confirmed by PCR over the ends of the contig. Annotation was performed through the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) (3).

The H34 genome is 2,892,474 bp in size, with a G+C content of 38.0%, and it carries no plasmids. PGAP annotated 2,823 coding sequences and 89 RNAs, and RAST (http://rast.nmpdr.org) determined the lab strain Scott A to be the closest relative to H34.

The H34 sequence was further analyzed with a focus on the virulence genes. All genes associated with the *Listeria* pathogenicity islands 1 (LIPI-1) (4) and 3 (LIPI-3) (5) were present. Compared to the *L. monocytogenes* reference strains EGDe and 10403S, two variations in H34 stand out: (i) a deletion in the *actA* coding sequence (CDS) and (ii) several single nucleotide polymorphisms within the phospholipase C (*plcB*) CDS and its 5′ untranslated region (UTR).

The H34 *actA* CDS has a 105-nucleotide (nt) in-frame deletion in the P-region of the ActA protein. While the P-region is not required for actin polymerization, it has a role in determining the length of the actin tails and the speed of bacterial motility (6), and

its deletion has led to significantly reduced cellular aggregation (7). Within the remaining portion of *actA*, there is a considerable mismatch on the protein level between H34, EGDe, and 10403S. To test the hypothesis that H34 might have an aggregation defect, we performed an aggregation assay that showed no differences between H34, EGDe, and 10403S (data not shown).

There are several nonsense mutations within the first 246 bp of the *plcB* CDS, leading to either a nonfunctional or shortened PlcB protein (85 amino acids), and H34 contains the same mutations in the 5′ UTR of *plcB*, as described for strain H4 by Jiang et al. (8).

The H34 genome contains 2 incomplete prophages, as determined by Phaster (9); none of them are integrated into the *comK* CDS.

Accession number(s). The complete H34 genome has been deposited in the NCBI GenBank under accession number CP020774.

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