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GENOME SEQUENCES

Complete Genome Sequence of the *Arcobacter trophiarum* Type Strain LMG 25534

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ABSTRACT *Arcobacter* species have been recovered from food and/or food animals, and several of these species are potential human pathogens. *Arcobacter trophiarum* was recovered from fecal samples taken from pigs on two Belgian farms. This study describes the whole-genome sequence of the *A. trophiarum* type strain LMG 25534 (=64^T =CCUG 59229^T).

Arcobacters are routinely recovered from food animals (1, 2); many have either been isolated from cases of human illness (1–3) or are considered to be potential human pathogens (4). *Arcobacter trophiarum* was originally recovered from fecal samples taken from pigs on two Belgian farms (5). *A. trophiarum* is urease negative and nonculturable at 37°C (5), and it is related to other *Arcobacter* species also isolated from food animals, such as *A. cibarius* (6), *A. thereius* (7), and *A. skirrowii* (8). In this study, we report the first closed genome sequence of the *A. trophiarum* type strain LMG 25534 (=64^T =CCUG 59229^T), isolated in 2007 from the feces of a 14-week-old piglet in Belgium (5).

The *A. trophiarum* strain LMG 25534^T genome was completed and closed primarily using the Roche 454 next-generation sequencing platform. Genomic DNA was isolated from a loop of cells using the Wizard genomic DNA purification kit (Promega, Madison, WI), as described previously (9); these cells were taken from cultures grown aerobically on anaerobe basal agar (Oxoid) plus 5% horse blood for 48 h at 30°C. Rapid (shotgun) and mate pair libraries were constructed and sequenced on the GS-FLX+ instrument. Simultaneous assembly of both sets of reads within Newbler (version 2.6; Roche) yielded 53 contigs overall and a single chromosomal scaffold composed of 28 unique contigs; 2 low-quality contigs, each composed of <10 reads, were deleted. A circular contiguous genome sequence for LMG 25534^T was generated by positioning the 23 remaining 454 contigs, many of which were placed at two or more locations into the scaffold, using the Perl script contig_extender3 (9) and verifying the contig placement or closing the gaps via PCR amplification and Sanger sequencing. Manual stepwise assembly of the 454 contigs and Sanger reads into a contiguous sequence was performed using SeqMan Pro (version 8.0; DNASTAR, Madison, WI). The 454 contig assembly was validated using an optical restriction map (restriction enzyme XbaI; OpGen, Gaithersburg, MD). The 454 read coverage across the chromosome was 49×. Base calls within the chromosomal sequence were also validated using Illumina HiSeq reads (SeqWright, Houston, TX). The HiSeq reads were assembled *de novo* within Newbler (version 2.6) using default parameters. As described above, contigs of an overall low quality were deleted. The remaining contigs were trimmed to a Q score of 40; these contigs were assembled into the SeqMan 454 assembly, with 454 base calls adjusted to the Illumina consensus sequence for each contig. Single nucleotide polymorphisms within the repeat contigs and sequences between the Illumina contigs were assessed/verified by assembling the Illumina reads onto the consensus sequence within Geneious (version 8.1; Biomatters, Auckland, NZ). The HiSeq reads yielded a coverage of 981×, for a final coverage across the genome of 1,029×.

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TABLE 1 Genomic features of *Arcobacter trophiarum* strain LMG 25534^T

Feature[a]	Data[b]
Chromosome	
Size (bp)	1,915,613
G+C content (%)	28.22
No. of CDS[c]	
Total	1,888
Assigned function (%)	844 (44.7)
General function annotation (%)	627 (33.2)
Domain/family annotation only (%)	135 (7.2)
Hypothetical (%)	282 (14.9)
Pseudogenes	21
Genomic islands/CRISPR	
No. of prophage/genetic islands	2
No. of CDS in genetic islands	76, [2]
No. of CRISPR/Cas loci	0
Gene content/pathways	
IS elements/mobile elements/transposases	12 (IS*3*); 4, [1] (IS*4*); 1 (other)
Signal transduction	
Che proteins	*cheABCDRVW*$(Y)_2$
No. of methyl-accepting chemotaxis proteins	16, [2]
No. or response regulators	18
No. of histidine kinases	20
No. or response regulator/histidine kinase fusions	0
No. of diguanylate cyclases	11, [1]
No. of diguanylate phosphodiesterases (HD-GYP, EAL)	3, 0
No. of diguanylate cyclase/phosphodiesterases	8
No. of other	6
Motility	
Flagellin genes	*flaAB*
Restriction/modification	
No. of type I (*hsd*) systems	1
No. of type II systems	3, [1]
No. of type III systems	0
Transcription/translation	
No. of transcriptional regulatory proteins	31, [1]
Non-ECF σ factors	σ^{70}
No. of ECF σ factors	0
No. of tRNAs	48
No. of ribosomal loci	5
Nitrogen fixation (*nif*)	No
Pyruvate $\rightarrow$ acetyl-CoA	
Pyruvate dehydrogenase (E1/E2/E3)	Yes
Pyruvate:ferredoxin oxidoreductase	No
Urease	No
Vitamin B_{12} biosynthesis	No

[a]CDS, coding sequences; ECF, extracytoplasmic function; CoA, coenzyme A.
[b]Numbers in brackets indicate pseudogenes/fragments.
[c]Total number does not include pseudogenes.

The genome features for *A. trophiarum* strain LMG 25534^T are presented in Table 1. The LMG 25534^T genome has a circular genome of 1,915,613 bp, with an average GC content of 28.2%. Protein-, rRNA-, and tRNA-encoding genes were identified as described previously (10). The genome is predicted to contain 1,888 putative protein-coding genes, 21 pseudogenes, 5 rRNA operons, and 48 tRNA-encoding genes. Two genomic islands, approximately 48.3 and 23.5 kb in size, were identified in the LMG 25534^T chromosome. The islands are bounded by 17- and 18-bp direct repeats, respectively, with a gene encoding a phage integrase family site-specific tyrosine recombinase present on one end of each island. The largest genomic island encodes a type IIP restriction/modification system and contains genes for a P-type type IV conjugative transfer system. No plasmids or CRISPR/Cas systems were identified in the LMG 25534^T genome.

A noteworthy feature of the *A. trophiarum* strain LMG 25534^T genome is the

presence of multiple insertion sequences. The LMG 25534^T chromosome contains 12 copies of a 1,289-bp IS*3* family mobile element and four copies of a 1,481-bp IS*4* family mobile element. A fifth IS*4* family insertion sequence is disrupted by an IS*3* family mobile element. IS*3* family insertion sequences were also identified within a larger ~22-kb mobile element that encodes four transposition-associated proteins and a type IIG restriction/modification system, as well as within the 23.5-kb genomic island.

Data availability. The complete genome sequence of *A. trophiarum* strain LMG 25534^T has been deposited in GenBank under the accession number CP031367. The 454 and HiSeq sequencing reads have been deposited in the NCBI Sequence Read Archive (SRA; accession number SRP154931).

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