Complete Genome Sequence of a Channel Catfish Epidemic Isolate, Aeromonas hydrophila Strain ML09-119

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Aeromonas hydrophila is a Gram-negative, rod-shaped, mesophilic bacterium that infects both aquatic poikilothermic animals and mammals, including humans. Here, we present the complete genome sequence of Aeromonas hydrophila strain ML09-119, which represents a clonal group of A. hydrophila isolates causing outbreaks of bacterial septicemia in channel catfish since 2009.

Aeromonas species are Gram-negative facultative anaerobes that are ubiquitous in aquatic environments and cause infections in several host species, including humans, invertebrates, reptiles, and amphibians (1–5). In particular, many of the Aeromonas species are pathogenic to fish, causing septicemia in carp, tilapia, perch, salmon, catfish, and other species (6). In channel catfish aquaculture, Aeromonas hydrophila is historically considered an opportunistic pathogen. However, since 2009 a clonal group of A. hydrophila isolates have been causing large-scale disease outbreaks in Alabama and Mississippi (7). Strain ML09-119 is an isolate from a disease outbreak on a commercial catfish farm, and it is representative of this clonal group.

The genome sequence of Aeromonas hydrophila ML09-119 was completed using a combination of Illumina Genome Analyzer IIx next-generation sequencing (a total of 4,077,018 reads, with 104X coverage) (Illumina, Inc., San Diego, CA) (M. J. Hossain, G. C. Waldbieser, D. Sun, N. K. Capps, W. B. Hemstreet, K. Carlisle, M. J. Griffin, L. Khoo, A. E. Goodwin, T. S. Sonstegard, S. Schroeder, K. Hayden, J. C. Newton, J. S. Terhune, and M. R. Liles, submitted for publication) and the 454 GS-FLX titanium platform (a total of 96,601 reads with 308X coverage) (Roche Applied Science). Sequences from both platforms were assessed for errors and trimmed for quality using CLC workbench 5.0.1 (CLC Bio) and Sequencher 5.1 (Gene Codes Corporation). Assembly was performed by CLC workbench 5.0.1. Scaffolded gaps were closed by Sanger sequencing of PCR amplicons. Un scaffolded gaps were closed by sequencing single-primer PCR amplicons (8). rRNA operons and other repeat regions were amplified and sequenced to resolve misassemblies.

The final closed-circle version of the A. hydrophila ML09-119 genome sequence was submitted to the NCBI's Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) (9) for annotation, followed by submission to GenBank. The total A. hydrophila genome comprises 5,024,500 bp with 60.8% GC content. It contains 4,577 predicted genes, of which 4,434 are protein-coding sequences. A total of 112 tRNAs and 10 rRNA operons were predicted by using tRNAscan-SE (10) and RNAmmer 1.2 (11), respectively.

The ML09-119 reads were assembled against the A. hydrophila ATCC 7966T genome (NC_008570.1) in CLC workbench 5.0.1 to identify contiguous regions of the ML09-119 genome that are not present in the ATCC 7966T genome. Functional analysis of predicted open reading frames (ORFs) in these unique contigs indicated that strain ML09-119 has a complete inositol utilization pathway that is not present in ATCC 7966T. More than 20 unique prophage-linked ORFs and several transposons were identified, and several putative virulence loci appear to be linked to prophage elements. Relative to strain ATCC 7966T, ML09-119 contains a unique 33-kb O polysaccharide biosynthesis gene cluster with 29 total predicted ORFs. Twenty-four of these do not have any similarity to ATCC 7966T genes. Thus, it appears that ML09-119 has a different O antigen serotype than ATCC 7966T.

In summary, the A. hydrophila ML09-119 genome encodes putative proteins suggesting that it has unique biochemical and serological features relative to strain ATCC 7966T. Further analysis of these unique predicted ORFs (Hossain et al., submitted) showed that they are consistently present in other epidemic isolates and absent from A. hydrophila isolates that were not associated with epidemic outbreaks.

Nucleotide sequence accession numbers. The complete genome sequence of A. hydrophila ML09-119 was deposited in GenBank under the accession number CP005966, version CP005966.1.

ACKNOWLEDGMENTS

This work was supported by the Mississippi State University College of Veterinary Medicine, the USDA Agricultural Research Service CRIS project 6402-31000-009-00D, and the Alabama Agricultural Experiment Station (Hatch project number ALA021-1-09005).

We thank Michelle Banes for technical assistance.
REFERENCES


