

Complete Genome Sequence of Escherichia coli Myophage Minorna

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ABSTRACT The Gram-negative bacterium Escherichia coli causes many diseases, and antibiotic resistance has become a problem for their treatment. Bacteriophages may present a viable treatment alternative. Here, the complete genome sequence of E. coli-infecting myophage Minorna is presented. Proteins needed for replication, morphogenesis, and lysis were identified in the Minorna coding sequence.

The Gram-negative bacterium *Escherichia coli* causes serious human diseases, including bloodstream and urinary tract infections [\(1\)](#page-1-0). The rate of antibiotic resistance in E. coli strains is rapidly rising [\(2\)](#page-1-1), and bacteriophage therapeutics are the new frontier in combating this threat [\(3\)](#page-1-2). Here, we describe the complete genome of a new E. coli myophage, Minorna.

Bacteriophage Minorna, against E. coli JE-1 carrying the plasmid pRA1 (RA1::Tn5 Sq^r), was isolated from the creek near Northgate Park in College Station, TX [\(4\)](#page-1-3). The water sample was processed through a 0.22- μ m filter. Luria broth or agar (BD) was used for growing the host and phage under aerobic conditions at 37°C, as described by Adams [\(5\)](#page-1-4). To determine phage morphology, samples were negatively stained with 2% (wt/vol) uranyl acetate and viewed by transmission electron microscopy at the Texas A&M Microscopy and Imaging Center. Minorna genomic DNA was prepared as previously described [\(6\)](#page-1-5) and sequenced on an Illumina MiSeq instrument with 250-bp paired-end reads using v2 500-cycle chemistry from an Illumina TruSeq Nano LT kit library. The 339,988 sequence reads from the index containing the phage genome were quality controlled using FastQC [\(http://www.bioinformatics](http://www.bioinformatics.babraham.ac.uk/projects/fastqc/) [.babraham.ac.uk/projects/fastqc/\)](http://www.bioinformatics.babraham.ac.uk/projects/fastqc/) and assembled using SPAdes v3.5.0 [\(7\)](#page-1-6) with 687 fold contig coverage after trimming using the FastX toolkit 0.0.14 [\(http://hannonlab](http://hannonlab.cshl.edu) [.cshl.edu\)](http://hannonlab.cshl.edu). PCR (with primers 5'-CGCGCAGCGTAGCATATAAT-3' and 5'-GAGTTACCTGA ACGTAGCGAC-3') and Sanger sequencing confirmed that the contig was complete. Genes were called using GLIMMER v3.0 and MetaGeneAnnotator v1.0 [\(8,](#page-1-7) [9\)](#page-1-8), and tRNA genes were screened with ARAGORN v2.36 [\(10\)](#page-1-9). The gene functions were predicted with InterProScan v5.22 [\(11\)](#page-1-10), BLAST v2.2.31 against the NCBI nr [\(12\)](#page-1-11) and UniProt Swiss-Prot and TrEMBL databases [\(13\)](#page-1-12), LipoP [\(14\)](#page-1-13), and TMHMM v2.0 [\(15\)](#page-1-14). Rhoindependent termination sites were annotated using TransTermHP [\(http://transterm](http://transterm.cbcb.umd.edu/) [.cbcb.umd.edu/\)](http://transterm.cbcb.umd.edu/). All analyses were run using default parameters in the Galaxy [\(16\)](#page-1-15) and Web Apollo [\(17\)](#page-1-16) instances hosted by the Center for Phage Technology [\(https://cpt](https://cpt.tamu.edu/galaxy) [.tamu.edu/galaxy\)](https://cpt.tamu.edu/galaxy).

Bacteriophage Minorna is a myophage with a 43,264-bp genome. The G+C content is 51%, matching the G-C content of its host. There are 57 protein-coding genes, 26 with predicted functions, and no tRNAs, with an overall 94% coding density. Minorna was compared with other phages by progressiveMauve2.4.0 [\(18\)](#page-1-17) and shares similarity with many Klebsiella phages, including its highest nucleotide sequence identity, at 80%, with KPV811 (GenBank accession number [KY000081\)](https://www.ncbi.nlm.nih.gov/nuccore/KY000081) and its highest amino acid similarity at 84% identity to Klebsiella phages KpV48 [\(KX237514\)](https://www.ncbi.nlm.nih.gov/nuccore/KX237514), both T1-like phages.

Citation Rogers K, Min L, Newkirk H, Liu M, Ramsey J. 2019. Complete genome sequence of Escherichia coli myophage Minorna. Microbiol Resour Announc 8:e00533-19. [https://doi.org/10.1128/MRA.00533-19.](https://doi.org/10.1128/MRA.00533-19)

Editor John J. Dennehy, Queens College

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Received 6 May 2019 **Accepted** 13 May 2019 **Published** 6 June 2019 Consistent with the packaging mechanism of phage T1, headful packaging for Minorna was predicted by PhageTerm [\(19\)](#page-2-0).

A complete lysis cassette is present at the beginning of the genome, including an endolysin with two predicted transmembrane domains (GenBank accession number [QBP07053\)](https://www.ncbi.nlm.nih.gov/protein/QBP07053), a holin with one transmembrane domain [\(QBP07054\)](https://www.ncbi.nlm.nih.gov/protein/QBP07054), and a unimolecular spanin [\(QBP07055\)](https://www.ncbi.nlm.nih.gov/protein/QBP07055) similar to the T1 phage unimolecular spanin, which is associated with both bacterial membranes [\(20,](#page-2-1) [21\)](#page-2-2). The morphogenesis proteins identified are also grouped together and span from the large [\(QBP07058\)](https://www.ncbi.nlm.nih.gov/protein/QBP07058) and small terminase subunits through tail proteins, internal core protein, major capsid protein, and portal protein [\(QBP07071\)](https://www.ncbi.nlm.nih.gov/protein/QBP07071). The internal core protein [\(QBP07063\)](https://www.ncbi.nlm.nih.gov/protein/QBP07063) is the largest protein, at 1,232 residues. Encoded proteins for replication and transcription include DNA helicase [\(QBP07093\)](https://www.ncbi.nlm.nih.gov/protein/QBP07093), DNA primase [\(QBP07096\)](https://www.ncbi.nlm.nih.gov/protein/QBP07096), DNA polymerase [\(QBP07090\)](https://www.ncbi.nlm.nih.gov/protein/QBP07090), and RNA polymerase [\(QBP07075\)](https://www.ncbi.nlm.nih.gov/protein/QBP07075). There are no identified introns or frameshifts.

Data availability. The genome sequence and associated data for phage Minorna were deposited under GenBank accession number [MK598851,](https://www.ncbi.nlm.nih.gov/nuccore/MK598851) BioProject accession number [PRJNA222858,](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA222858) SRA accession number [SRR8869232,](https://www.ncbi.nlm.nih.gov/sra/SRR8869232) and BioSample accession number [SAMN11360441.](https://www.ncbi.nlm.nih.gov/biosample/SAMN11360441)

ACKNOWLEDGMENTS

This work was supported by funding from the National Science Foundation (awards EF-0949351 and DBI-1565146). Additional support came from the Center for Phage Technology (CPT), an Initial University Multidisciplinary Research Initiative supported by Texas A&M University and Texas AgriLife, and from the Department of Biochemistry and Biophysics at Texas A&M University.

We are grateful for the advice and support of the CPT staff.

This announcement was prepared in partial fulfillment of the requirements for BICH464 Bacteriophage Genomics, an undergraduate course at Texas A&M University.

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