



Complete Genome Sequence of *Klebsiella pneumoniae* Podophage Patroon

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ABSTRACT *Klebsiella pneumoniae* infection is a serious concern in hospital settings due to the continuing emergence of multidrug-resistant strains. The study of *K. pneumoniae* phages may help the development of new treatment strategies. Here, the complete genome sequence of *K. pneumoniae* phage Patroon, a T3/T7-like phage, is presented.

Klebsiella pneumoniae is a Gram-negative bacterium well known as an opportunistic pathogen that causes pneumonia, septicemia, and urinary tract infection (1, 2). *K. pneumoniae* infection is a serious concern in hospital settings due to the continuing emergence of multidrug-resistant strains carrying the *bla*_{KPC} gene (3). The study of *K. pneumoniae* phages may help us develop new treatment strategies.

Phage Patroon was isolated from influent water from the municipal wastewater treatment plant in Bryan, TX, in 2016, using a carbapenem-resistant (KPC⁺) *K. pneumoniae* clinical isolate of sequence type 258 as the host. Host bacteria were cultured on tryptic soy broth or agar (Difco) at 37°C with aeration. Phages were isolated and propagated by the soft agar overlay method (4). Phage genomic DNA was prepared using a modified Promega Wizard DNA cleanup kit protocol, as described previously (5). Pooled indexed DNA libraries were prepared using the Illumina TruSeq Nano low-throughput (LT) kit, and the sequence was obtained from the Illumina MiSeq platform using the MiSeq V2 500-cycle reagent kit, following manufacturer's instructions, producing 667,982 paired-end reads for the index containing the phage genome. The quality of the reads was checked in FastQC 0.11.5 (<https://www.bioinformatics.babraham.ac.uk/projects/fastqc/>), trimmed with FastX-Toolkit 0.0.14 (http://hannonlab.cshl.edu/fastx_toolkit/download.html), and assembled in SPAdes 3.5.0 (6). The assembled genome was closed with PCR using primers (5'-GCTGGTAAGGAAGTCGGTAAA-3', 5'-GTCGTTAGTTAGGCGGCATAG-3') facing off the ends of the assembled contig and Sanger sequencing of the resulting product, with the contig sequence manually corrected to match the resulting Sanger sequencing read. Protein-coding genes were predicted using GLIMMER 3.0 (7) and MetaGeneAnnotator 1.0 (8) and corrected manually if needed. The tRNA genes were predicted using ARAGORN 2.36 (9). Protein functions were predicted by comparing the sequence homology to proteins found using BLASTp 2.2.28 (10), and conserved domains were analyzed using InterProScan 5.15-5.40 (11). All the analyses were performed under default settings using the CPT Galaxy (12) and Web Apollo (13) interfaces (cpt.tamu.edu).

The Patroon genome was assembled into a complete contig of 39,442 bp at 596.8-fold coverage. It contains 51 predicted coding sequences, with a coding density of 89% and a GC content of 50.54%. At the DNA level, Patroon is most similar (87% to 88%) to other T7-like enterobacterial phages, such as *Escherichia coli* phage ECA2 (GenBank accession number [KX130726](https://www.ncbi.nlm.nih.gov/nuccore/KX130726)), *Yersinia* sp. phage phiYeO3-12 (GenBank accession number [AJ251805](https://www.ncbi.nlm.nih.gov/nuccore/AJ251805)), and *Salmonella* sp. phage phi5G-JL2 (GenBank accession number [EU547803](https://www.ncbi.nlm.nih.gov/nuccore/EU547803)), as determined by the progressiveMauve algorithm (14). Patroon is

Citation Tran R, Kongari R, Lessor L, Gill JJ, Liu M. 2019. Complete genome sequence of *Klebsiella pneumoniae* podophage Patroon. Microbiol Resour Announc 8:e00461-19. <https://doi.org/10.1128/MRA.00461-19>.

Editor John J. Dennehy, Queens College

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Received 22 April 2019

Accepted 30 April 2019

Published 23 May 2019

a T3/T7-like phage with 47 and 45 Patroon proteins matching phages T3 and T7, respectively, determined by BLASTp (E value < 0.001). Genes encoding proteins related to phage morphogenesis, DNA replication, and recombination were identified. The lysis proteins identified consisted of a class II holin, an amidase endolysin, and an embedded i-spanin and o-spanin pair. The phage Patroon tail fiber gp44 (GenBank accession number [QBQ72909](#)) is closely related at its N terminus to other T7-like tail fibers, including the phage T7 tail fiber gp17. The C-terminal receptor-binding domain is related to only a few other phage tail fibers based on BLASTp alignment, including those of coliphage ECA2 (GenBank accession number [ANN86232](#)) and *Yersinia* sp. phage phiYeO3-12 (GenBank accession number [NP_052117](#)).

Data availability. The genome sequence of phage Patroon was submitted to GenBank under accession number [MK608335](#). The associated BioProject, SRA, and BioSample accession numbers are [PRJNA222858](#), [SRR8788210](#), and [SAMN11259695](#), respectively.

ACKNOWLEDGMENTS

This work was supported by funding from the National Science Foundation (awards EF-0949351 and DBI-1565146) and by the National Institutes of Health (NIAID award AI121689). 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 thank Thomas Walsh, Weill Cornell Medical School, for the provision of bacterial isolates. We are grateful for the advice and support of the CPT staff. This announcement was prepared in partial fulfillment of the requirements for BICH464 Phage Genomics, an undergraduate course at Texas A&M University.

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