

Complete Genome Sequence of T4-Like *Escherichia coli* Bacteriophage HX01

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Phage T4 is among the best-characterized biological systems (S. Kanamaru and F. Arisaka, Seikagaku 74:131-135, 2002; E. S. Miller et al., Microbiol. Mol. Biol. Rev. 67:86-156, 2003; W. B. Wood and H. R. Revel, Bacteriol. Rev. 40:847-868, 1976). To date, several genomes of T4-like bacteriophages are available in public databases but without any APEC bacteriophages (H. Jiang et al., Arch. Virol. 156:1489-1492, 2011; L. Kaliniene, V. Klausa, A. Zajanckauskaite, R. Nivinskas, and L. Truncaite, Arch. Virol. 156:1913-1916, 2011; J. H. Kim et al., Vet. Microbiol. 157:164-171, 2012; W. C. Liao et al., J. Virol. 85:6567-6578, 2011). We isolated a bacteriophage from a duck factory, named HX01, that infects avian pathogenic *Escherichia coli* (APEC). Sequence and morphological analyses revealed that phage HX01 is a T4-like bacteriophage and belongs to the family *Myoviridae*. Here, we announce the complete genome sequence of phage HX01 and report the results of our analysis.

A vian pathogenic *Escherichia coli* (APEC), an important etiologic infection and among the most significant infectious diseases, causes colibacillosis, airsacculitis, and associated pericarditis, with perihepatitis and peritonitis being the most typical diseases, resulting in severe economic losses to the poultry industry worldwide (2). Bacteriophage therapy is considered to be an alternative method for the treatment of bacterial infection (1). To date, there is no report of an APEC phage genome sequence (3, 4, 6, 7). Here, we report the full genome sequence and organization of novel virulent bacteriophage HX01, which infects APEC.

The DNA of phage HX01 was extracted as described previously (9) and was sequenced on a GS-FLX (454 Life Sciences). The 454 reads were assembled with Newbler (version 2.0) (Roche) using default assembly parameters. Open reading frames (ORFs) were predicted with Glimmer3.0 (http://www.ncbi.nlm.nih.gov/projects/gorf/) software. BLASTX and BLASTP (http://www.ncbi.nlm.nih.gov/blast/Blast.cgi) were used to search for homologous proteins. tRNA were identified with the tRNA-scan-SE program (http://lowelab.ucsc.edu/tRNAscan-SE/).

Electron microscopy showed that the phage had an icosahedral head of 95 by 70 nm and a contractile tail of 100 by 130 nm with long tail fibers. The genome of HX01 consists of 169,158 bp in length, with an average GC content of 37.59%, 269 open reading frames (ORFs), and 2 tRNA genes. Of the 269 predicted ORFs, more than 95% possessed defined functions and were highly homologous to those of T4 or T4-like phages (5, 8, 10) in the *Myoviridae* family. Structural proteins included head protein, tail protein, tail fiber protein, baseplate protein, and portal protein, while nonstructural proteins included a putative DNA replication enzyme, DNA polymerase, endonuclease VII, putative rIIA protector, putative rIIB protector, and putative single-strand binding protein.

The genome sequence showed that the DNA shared greatest identity, up to 91% of the base pairs, with phage RB69, which is a pathogenic *E. coli* phage isolated in the United States and belongs to the T4-like group (11). This is the first report of the complete genome sequence of a T4-like phage that infects APEC. Studies investigating the complete genome of phage HX01 would provide novel information about APEC-targeting phage.

Nucleotide sequence accession number. The complete ge-

nome sequence of phage HX01 is available in GenBank under accession number JX536493.

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REFERENCES

- Brussow H. 2005. Phage therapy: the Escherichia coli experience. Microbiology 151:2133–2140.
- 2. Dho-Moulin M, Fairbrother JM. 1999. Avian pathogenic Escherichia coli (APEC). Vet. Res. 30:299–316.
- 3. Jiang H, et al. 2011. The complete genome sequence of a novel T4-like bacteriophage, IME08. Arch. Virol. 156:1489–1492.
- Kaliniene L, Klausa V, Zajanckauskaite A, Nivinskas R, Truncaite L. 2011. Genome of low-temperature T4-related bacteriophage vB_EcoM-VR7. Arch. Virol. 156:1913–1916.
- Kanamaru S, Arisaka F. 2002. The structural biology and infection mechanism of bacteriophage T4. Seikagaku 74:131–135. (In Japanese.)
- Kim JH, et al. 2012. Complete genome sequence and characterization of a broad-host range T4-like bacteriophage phiAS5 infecting Aeromonas salmonicida subsp. salmonicida. Vet. Microbiol. 157:164–171.
- 7. Liao WC, et al. 2011. T4-like genome organization of the Escherichia coli O157:H7 lytic phage AR1. J. Virol. 85:6567–6578.
- 8. Miller ES, et al. 2003. Bacteriophage T4 genome. Microbiol. Mol. Biol. Rev. 67:86–156.
- Sillankorva S, Neubauer P, Azeredo J. 2008. Isolation and characterization of a T7-like lytic phage for Pseudomonas fluorescens. BMC Biotechnol. 8:80.
- Wood WB, Revel HR. 1976. The genome of bacteriophage T4. Bacteriol. Rev. 40:847–868.
- 11. Yeh LS, Hsu T, Karam JD. 1998. Divergence of a DNA replication gene cluster in the T4-related bacteriophage RB69. J. Bacteriol. 180:2005–2013.

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