



Complete Genome Sequence of a Lytic T7-Like Phage, *Escherichia* Phage vB_EcoP-Ro45lw, Isolated from Nonfecal Compost Samples

Yen-Te Liao,^a Fang Liu,^{a,b}  Vivian C. H. Wu^a

^aProduce Safety and Microbiology Research Unit, U.S. Department of Agriculture, Agricultural Research Service, Western Regional Research Center, Albany, California, USA

^bCollege of Food Science and Engineering, Ocean University of China, Qingdao, China

ABSTRACT Although isolation of the bacteriophages (phages) specific to Shiga toxin-producing *Escherichia coli* (STEC) is increasing, the number of those specific to STEC non-O157 strains, instead of STEC O157, with whole-genome sequencing characterization is relatively low. Here, we announce the complete genome sequence of a T7-like lytic phage against STEC O45.

Shiga toxin-producing *Escherichia coli* (STEC), the major cause of hemolytic uremic syndrome, has been frequently associated with produce contamination, including the recent romaine lettuce outbreak in California (1, 2). Lytic bacteriophages are increasingly considered promising naturally occurring agents, alternative to antibiotics, to combat bacterial pathogens (3). With the bacteria constantly evolving, the isolation of novel and diverse bacteriophages specific to the pathogens is needed. In this study, the complete genome sequence of a STEC O45-specific phage is described.

Escherichia phage vB_EcoP-Ro45lw, or Ro45lw, was isolated from nonfecal compost samples which were enriched with a cocktail of STEC O157 and the top six non-O157 strains in tryptic soy broth with 10% CaCl₂ and were incubated at 37°C for 24 h. After a spot test against the strains used for enrichment, the phage purification process was performed by picking a single plaque and plating with the STEC O145 host using a double-layer plaque assay, as previously described (4). Phage DNA was extracted using a Norgen phage DNA extraction kit (Norgen Biotek, Ontario, Canada). The DNA library was constructed using a TruSeq nano DNA library prep kit (Illumina, San Diego, CA) and sequenced with an Illumina MiSeq sequencer, resulting in approximately 1 million 2 × 250-bp paired-end reads. The quality control of raw sequence reads was performed using FASTQC and Trimmomatic (5), with the setting of average quality at Q30. *De novo* assembly of the remaining quality reads was performed using Unicycler version 0.4.1 (SPAdes) on the Galaxy server (6). Genome annotation was subjected to Prokka workflow (7) and confirmed using Artemis (8), open reading frame (ORF) finder (Geneious version 11.0.4), and BLASTP (<https://blast.ncbi.nlm.nih.gov/Blast.cgi?PAGE=Proteins>). The packaging mechanisms and genome termini were determined using PhageTerm (9). PromoterHunter in the phiSITE database was used to predict regulatory genes, such as promoter and regulator (10). tRNA was predicted using tRNAscan-SE search server (11). The ResFinder (version 3.0) database was used to screen for antibiotic resistance genes (12).

Phage Ro45lw has a double-stranded DNA with a genome size of 39,793 bp (2,352-fold coverage), with an average G+C content of 52.2%, and belongs to T7-like phages under subfamily *Autographivirinae* in *Podoviridae*. There are 50 ORFs, 6 promoters, and 1 terminator, but no tRNA, predicted in the genome. Ro45lw has a 190-bp terminal repeat region at both sides of the genome end, with the packaging mecha-

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Address correspondence to Vivian C. H. Wu, vivian.wu@ars.usda.gov.

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nism belonging to direct terminal repeats (DTR) (13). A total of 25 ORFs were annotated with functional proteins, such as RNA polymerase, endonuclease, DNA primase/helicase, collar, and tail tubular proteins A and B, which are highly related to T7-like phages. None of the ORFs were found to contain lysogenic, virulence, or antibiotic resistance sequences. Ro45lw is closely related to the T7-like *Cronobacter* phage Dev2 (GenBank accession number [HG813241](https://doi.org/10.1017/S0950268815000047)), with an average nucleotide identity (ANIb) of 89.18% (77.31% query coverage) calculated by JSpeciesWS (14). Furthermore, three proteins of Ro45lw containing DNA-directed RNA polymerase (ORF_06), head-to-tail joining protein (ORF_32), and capsid and scaffold protein (ORF_34) show 99% identity to the amino acid sequences of *Cronobacter* phage Dev2. The findings of this study provide valuable insight into the diversity of T7-like phages specific to STEC O45 strains.

Data availability. The complete genome sequence of *Escherichia* phage vB_EcoP-Ro45lw has been deposited in GenBank under the accession number [MK301532](https://doi.org/10.1017/S0950268815000047). The sequencing reads have been deposited in SRA under the accession number [PRJNA509478](https://doi.org/10.1017/S0950268815000047). The version of the phage genome described in this paper is the first version.

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