

Complete Genome Sequence of *Caulobacter crescentus* Bacteriophage φ CbK

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φ CbK is a B3 morphotype bacteriophage of the *Siphoviridae* family that infects *Caulobacter crescentus*, the preeminent model system for bacterial cell cycle studies. The last 4 decades of research with φ CbK as a genetic and cytological tool to study the biology of the host warrant an investigation of the phage genome composition. Herein, we report the complete genome sequence of φ CbK and highlight unusual features that emerged from its annotation. The complete genome analysis of the φ CbK phage provides new insight into its characteristics and potential interactions with its *Caulobacter crescentus* host, setting the stage for future functional studies with φ CbK.

The aquatic alphaproteobacterium *Caulobacter crescentus* divides asymmetrically at each cell cycle into two progeny cells with distinct morphologies and fates: a sessile and replicative stalked (St) cell and a motile swarmer (Sw) cell with a polar flagellum and polar pili. The Sw cell resides in a G₁-like nonreplicative state and must differentiate into the St cell for division to occur (11).

The dimorphism of *Caulobacter* is exploited by certain bacteriophages (caulophages), most of which are *Siphoviridae* family members (B3 morphotype) with a prolate cylindrical head and a long, flexible, noncontractile tail (12). This morphotype is relatively rare, possibly comprising ~1% of the total number of characterized phages (1).

φ CbK, a virulent B3 morphotype caulophage with a double-stranded DNA genome, infects Sw cells (2, 3) by elaborating a head filament that wraps around the flagellum (6), presumably to help position the phage tail toward the pilus portals, the site for irreversible attachment and subsequent DNA injection. While the Sw tropism of φ CbK has been exploited to uncover cell cycle and polarity genes in *Caulobacter* (5, 8, 9, 13), including the master cell cycle transcriptional regulator GcrA (7), the φ CbK genome sequence had not been determined.

After high-throughput sequence analysis with an Illumina HiSeq 2000 sequencer (3,399 \times coverage) of φ CbK genomic DNA (extracted using the Norgen Biotek phage DNA extraction kit), we assembled quality-filtered reads (Velvet 01.01.04 software), predicted coding sequences (pCDS), and transfer RNAs (tRNAs) using Glimmer3.02 (4), FgenesV (Softberry, Inc., Mount Kisco, NY), and tRNAscan-SE (10) software. Functional assignments of pCDS were based on a best-hit query (*E* value of < 0.001) to the UniProtKB (Swiss-Prot+TrEMBL, release 2012_04) database using NCBI BLAST v2.2.22.

The φ CbK linear genome is 205,204 bp in length with 319 pCDS and 24 tRNAs. The codon usage and G+C content (66.1%) of the φ CbK genome match those of its host (67.2%). The pCDS cover 90.8% of the genome, with 13.8% of the pCDS resembling genes of known function, 11% resembling genes of unknown function (hypothetical conserved), and 75.2% being distinct from genes in current databases (new hypotheticals).

The pCDS for phage packaging and assembly (one terminase, one major capsid, two tail fibers, and one tail length tape measure protein), host lysis (one endolysin), and phage DNA replication/

modification and repair (one DNA polymerase III, two DNA polymerase I, one helicase, two HNH (His-Asn-His) endonucleases, one cytosine-specific methyltransferase, one DNA ligase, one exodeoxyribonuclease V, one crossover junction endodeoxyribonuclease, and one DNA lyase protein) are scattered over the φ CbK genome. Surprisingly, although φ CbK is known to be virulent (12), its genome harbors pCDS for an integrase and a Cro/CI transcriptional regulator, raising the possibility that φ CbK can enter a lysogenic state. Moreover, we identified a pCDS for a GcrA homolog that might coordinate transcription of the host and φ CbK replication cycles. In sum, the complete analysis of the φ CbK genome extends the repertoire of novel alphaproteobacterial genes while also hinting at the genetic interactions of a cell cycle-specific virus with a dimorphic bacterium.

Nucleotide sequence accession number. The caulophage φ CbK genome sequence was deposited in GenBank (accession number [JX163858](#)).

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ADDENDUM IN PROOF

After submission of this genome announcement, we learned that Jason J. Gil and Ry Young (Texas A&M University) also determined the φ CbK genome sequence (unpublished data; accession number [JX100813](#)).

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