

Genome Sequence of Temperate *Vibrio parahaemolyticus* Bacteriophage vB_VpaS_MAR10

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***Vibrio parahaemolyticus* is recognized as one of the main causes of human gastroenteritis associated with seafood. We have fully sequenced the genome of a newly isolated phage, vB_VpaS_MAR10, which lysed 61.9% of the *V. parahaemolyticus* strains tested. Phage MAR10 is a temperate siphovirus, and its genome consists of double-stranded DNA (dsDNA) with a size of 78,751 bp, a G+C content of 49.70%, and 104 open reading frames. Bioinformatic analysis shows that phage MAR10 is closely related to *Vibrio* phage SSP002.**

Vibrio parahaemolyticus has been distinguished as the most important cause of human gastroenteritis associated with seafood since its initial identification in 1950 (4, 6). Due to the ubiquitous nature of the bacterium, it is difficult to prevent contamination of seafood (9). Therefore, there is a critical need for more accurate, sensitive, and fast detection methods. Current detection methods of *V. parahaemolyticus*, such as the most probable number (MPN) method (FDA standard method) (2, 9), have some disadvantages (e.g., that they are labor-intensive, time-consuming, and lacking in specificity among *Vibrio* strains) (7). Another approach to increase the sensitivity of the detection method of food-borne pathogens is the use of bacteriophages, which are highly host-specific viruses (3). The objectives of the current study were to isolate a novel *V. parahaemolyticus*-specific phage and determine its sequence in order to develop a detection system.

Using the method previously described by Van Twest and Kropinski (8), phages of *V. parahaemolyticus* were isolated from seawater samples (San Felipe, Baja California, Mexico). Phage vB_VpaS_MAR10 (MAR10) is a temperate phage with high specificity to its host and is able to infect 13 out of 21 (61.9%) *V. parahaemolyticus* strains tested. The phage preparation was negatively stained (2% uranyl acetate) and examined by electron microscopy at the University of Guelph. Electron micrographs show that phage MAR10 has a noncontractile tail of 160 nm by 10 nm and an elongated head of 94 nm by 50 nm, revealing that this phage belongs to the family *Siphoviridae* (1).

The Midi Lambda DNA purification kit (Qiagen, Mississauga, Ontario, Canada) was used to extract and purify the DNA of phage MAR10, and the genomic sequence was determined using 454 technology (McGill University and Génome Québec Innovation Centre, Montreal, Quebec, Canada). MyRAST was applied to annotate the genome, and gene calls were verified using Kodon (Applied Maths, Austin, TX). For each protein the number of amino acids, molecular weight, and isoelectric point were determined by means of Batch MW and pI Finder (<http://greengene.uml.edu/programs/FindMW.html>). Homologs were identified using BatchBLAST (http://greengene.uml.edu/programs/NCBI_Blast.html). Protein motifs were predicted using Phobius (<http://phobius.sbc.su.se>), Pfam (<http://pfam.sanger.ac.uk/>), and TMHMM (<http://www.cbs.dtu.dk/services/TMHMM-2.0/>).

Analysis of the sequence reveals that the genome of phage MAR10 is 78,751 bp double-stranded DNA with a G+C content

of 49.70% and encodes 104 open reading frames (ORFs). CoreGenes (10) was used for comparative genomic analysis, which shows that the proteome of phage MAR10 shares 94/102 (92.16%) protein homology with phage SSP002 (5), which infects *Vibrio vulnificus*. It is highly unusual that a temperate member of the *Siphoviridae* has a suite of DNA metabolism and replication genes, including thymidylate kinase and synthase, helicase, DNA polymerase, and DNA ligase. The lack of an integrase and the presence of a parB-like partitioning protein suggest an alternative mechanism for lysogenization. We recommend that MAR10 along with SSP002 be placed into a new genus, the “Ssp002likevirus.”

Nucleotide sequence accession number. The complete genome sequence of temperate phage vB_VpaS_MAR10 is available in GenBank under accession number JX556418.

ACKNOWLEDGMENT

This work was financially supported by the National Sciences and Engineering Research Council of Canada (NSERC).

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Received 26 September 2012 Accepted 27 September 2012

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doi:10.1128/JVI.02666-12

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