Journal of Virology

Complete Genome Sequence of Bacillus cereus Bacteriophage PBC1

Minsuk Kong, Minsik Kim and Sangryeol Ryu
J. Virol. **2012, 86(11):6379. DOI: 10.1128/JVI.00706-12.**

JVI Journals.ASM.org

GENOME ANNOUNCEMENT

Complete Genome Sequence of *Bacillus cereus* Bacteriophage PBC1

Minsuk Kong, Minsik Kim, and Sangryeol Ryu

Department of Food and Animal Biotechnology, Department of Agricultural Biotechnology and Center for Agricultural Biomaterials, Seoul National University, Seoul, South Korea

***Bacillus cereus* is a ubiquitous, spore-forming bacterium associated with food poisoning cases. To develop an efficient biocontrol agent against *B. cereus*, we isolated lytic phage PBC1 and sequenced its genome. PBC1 showed a very low degree of homology to previously reported phages, implying that it is novel. Here we report the complete genome sequence of PBC1 and describe major findings from our analysis.**

Bacillus cereus is an opportunistic human pathogen responsible for 2 to 5% of the food-borne illnesses reported worldwide (2, 8). Although outbreaks caused by *B. cereus* are relatively infrequent, concerns about *B. cereus* have not diminished because it is commonly isolated from foods and food additives due to its abilities to form endospores and to grow at a broad range of temperatures (5 to 55°C) (5, 9, 10, 15). With the rapid emergence of antibiotic-resistant bacteria, the use of bacteriophages has regained attention as an efficient alternative method for their control (7, 14). Although many phages infecting various strains of *B. cereus* have been isolated in our lab (11, 16), only one phage, PBC1, can infect *B. cereus* strain ATCC 21768. PBC1 belongs to the *Siphoviridae* family and forms clear plaques (data not shown).

Phenol-chloroform extraction was used to isolate the phage's genomic DNA, and it was sequenced using the Genome Sequencer FLX Titanium by Macrogen, Seoul, South Korea. The assembly of quality filtered reads was performed using GS De Novo Assembler (v. 2.6), and the open reading frames (ORFs) were predicted using the Glimmer 3.02 (4), GeneMark.hmm (3), and FgenesB (Softberry, Inc., Mount Kisco, NY) software. The ORFs were limited to those encoding proteins of more than 50 amino acids. Conserved-domain analysis of the predicted ORFs was carried out using BLASTP (1), InterProScan (17), and the NCBI Conserved Domain Database (13). Searches for tRNAs were conducted using the tRNAscan-SE program (12).

Genomic analysis revealed that PBC1 contains 41,164 bp of linear double-stranded DNA with a G+C content of 41.7 mol%. PBC1 has terminally redundant and partially permuted genomes, suggesting that PBC1 uses a headful packaging mechanism. We identified 50 predicted ORFs, all of which were transcribed in the same direction, and found no tRNAs. Of the 50 predicted ORFs, 28 were identified as encoding hypothetical proteins. Homology searches identified packaging and structural proteins (a terminase, a portal protein, major/minor capsid proteins, and a tail length measure protein), host lysis proteins (a holin and an endolysin), and DNA replication and modification proteins (a thymidylate synthase, a nucleoside triphosphatase, a DNA polymerase, a resolvase, a glutaredoxin-like protein, a nuclease, and a helicase). We could not find any lysogeny-related proteins, such as an integrase or repressors, supporting the notion that PBC1 is a virulent phage. Interestingly, PBC1 has a putative YD repeat protein at its tail that is known to be involved in extracellular carbohydrate binding (6). We speculate that the host, *B. cereus* ATCC 21768, may have a unique carbohydrate structure on the cell surface and that PBC1 specifically recognizes and binds the host receptor by using the YD repeat protein. The analysis of the complete PBC1 genome not only facilitates its development as a biocontrol agent against *B. cereus* but also improves our understanding of the bacteriophage host range.

Nucleotide sequence accession number. The complete genome sequence of *B. cereus* phage PBC1 is available in GenBank under accession number JQ619704.

ACKNOWLEDGMENT

This work was supported by the Agriculture Research Center program of the Ministry for Food, Agriculture, Forestry and Fisheries, Republic of Korea.

REFERENCES

1. **Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ.** 1990. Basic local alignment search tool. J. Mol. Biol. **215**:403–410.
2. **Anonymous.** 2011. Surveillance for foodborne disease outbreaks—United States, 2008. MMWR Morb. Mortal. Wkly. Rep. **60**:1197–1202.
3. **Besemer J, Lomsadze A, Borodovsky M.** 2001. GeneMarkS: a self-training method for prediction of gene starts in microbial genomes. Implications for finding sequence motifs in regulatory regions. Nucleic Acids Res. **29**:2607–2618.
4. **Delcher AL, Bratke KA, Powers EC, Salzberg SL.** 2007. Identifying bacterial genes and endosymbiont DNA with Glimmer. Bioinformatics **23**:673–679.
5. **de Sarrau B, et al.** 2012. Influence of anaerobiosis and low temperature on *Bacillus cereus* growth, metabolism, and membrane properties. Appl. Environ. Microbiol. **78**:1715–1723.
6. **Feulner G, et al.** 1990. Structure of the *rhsA* locus from *Escherichia coli* K-12 and comparison of *rhsA* with other members of the *rhs* multigene family. J. Bacteriol. **172**:446–456.
7. **Hagens S, Loessner MJ.** 2007. Application of bacteriophages for detection and control of foodborne pathogens. Appl. Microbiol. Biotechnol. **76**: 513–519.
8. **Ivanova N, et al.** 2003. Genome sequence of *Bacillus cereus* and comparative analysis with *Bacillus anthracis*. Nature **423**:87–91.
9. **Kailas L, et al.** 2011. Surface architecture of endospores of the *Bacillus cereus/anthracis/thuringiensis* family at the subnanometer scale. Proc. Natl. Acad. Sci. U. S. A. **108**:16014–16019.
10. **Kim SK, et al.** 2009. Prevalence and toxigenic profiles of *Bacillus cereus* isolated from dried red peppers, rice, and Sunsik in Korea. J. Food Prot. **72**:578–582.
11. **Lee JH, Shin H, Son B, Ryu S.** 2012. Complete genome sequence of *Bacillus cereus* bacteriophage BCP78. J. Virol. **86**:637–638.

Received 21 March 2012 **Accepted** 21 March 2012

Address correspondence to Sangryeol Ryu, sangryu@snu.ac.kr.

doi:10.1128/JVI.00706-12

12. **Lowe TM, Eddy SR.** 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic sequence. Nucleic Acids Res. **25**: 955–964.
13. **Marchler-Bauer A, et al.** 2007. CDD: a conserved domain database for interactive domain family analysis. Nucleic Acids Res. **35**:D237–D240.
14. **Matsuzaki S, et al.** 2005. Bacteriophage therapy: a revitalized therapy against bacterial infectious diseases. J. Infect. Chemother. **11**:211–219.
15. **Okanlawon BM, Ogunbanwo ST, Okunlola AO.** 2010. Growth of *Bacillus cereus* isolated from some traditional condiments under different regimens. Afr. J. Biotechnol. **9**:2129–2135.
16. **Shin H, Bandara N, Shin E, Ryu S, Kim KP.** 2011. Prevalence of *Bacillus cereus* bacteriophages in fermented foods and characterization of phage JBP901. Res. Microbiol. **162**:791–797.
17. **Zdobnov EM, Apweiler R.** 2001. InterProScan—an integration platform for the signature-recognition methods in InterPro. Bioinformatics **17**: 847–848.