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GENOME ANNOUNCEMENT

Biology of a Novel Mycobacteriophage, SWU1, Isolated from Chinese Soil as Revealed by Genomic Characteristics

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Mycobacteriophage SWU1 is a newly isolated phage from a soil sample collected at Gongping village, Pingchang County, Sichuan Province, China, using *Mycobacterium smegmatis* mc^{2}155 as a host. Plaques of SWU1 appear as a unique bull's-eye on an *M. smegmatis* lawn. In this paper, we report the complete genome sequence of SWU1 and some major findings from the analysis result.

Bacteriophages represent unprecedented bioresources and an indispensable quarry for novel genetic tools. This holds true for mycobacteriophages, a magic toolkit for *Mycobacterium* molecular manipulation and tuberculosis control (7–10, 13, 14). However, most mycobacteriophages are isolated from limited regions, and many prophages and the CRISPR spacers within mycobacterial genomes fail to find matching phages (4), suggesting that more work on the isolation and description of mycobacteriophages remains necessary.

Using *Mycobacterium smegmatis* mc^{2}155 as the host organism, a novel mycobacteriophage (SWU1) was isolated from a soil sample collected at Sichuan Province, China. Plaques of SWU1 appear a characteristic bull's-eye with a halo, which is thought an indicator for the appearance of phage-associated depolymerases. In order to understand the formation mechanism of halo, we sequenced and analyzed the genome of SWU1.

We used phenol to break up SWU1 particles and isolate the DNA. Purified phage DNA was sheared with a HydroShear DNA-fragmenting system (Gene Machines). The 1.5-to-3-kb fragmented DNAs were purified, repaired, and cloned into PUC118 to create a shotgun library. The resulting ligation mixture was used to transform *Escherichia coli* DH10B. Recovered plasmid was sequenced on an ABI 3730XL DNA Sequencer (Applied Biosystem) by GBI in China (approximately 4.8× coverage). Predicted genes were subjected to database searches using Glimmer software (3). The putative functions of the open reading frames (ORFs) were determined by BLASTP searches (1). The use of a previously constructed L5 codon usage table assisted the identification of SWU1 predicted genes (6).

The SWU1 genome was found to be 52,474 bp in length with 94 putative protein-coding genes and 3 tRNA genes with a G+C content of 62.4%. SWU1 belongs to cluster A2 using the previously stated criteria (5). Among those genes, 91 have sequence similarity to other genes and 22 have been assigned functions, although 3 fail to match any genes. Many of the genes (genes 6, 13, 14, 16, 17, 23, 26, 27, and 28) are presumably involved in phage structure and assembly. Several genes (genes 44, 49, and 50) might encode proteins involved in phage DNA metabolism. Genes 10, 11, and 12 encode LysA, Holin, and LysB, which are components of the lytic cassette of SWU1. The phage integrase (gp33), the phage attachment site (bp 25,583 to 25,625), and the excisionase (gp36) form an integration cassette. We note that gp45 (encoding a single-stranded DNA [ssDNA] annealing protein, SSAP), gp60 (a recombination endonuclease VII), and gp72 (a RecB family exonuclease) are functional genes of homologous recombination. Genes 84, 85, and 86 are highly similar to genes encoding three cytotoxic early proteins of L5 (11). Phage-associated depolymerases have not been identified in the SWU1 genome. But we found that SWU1 gene 59 has weak sequence similarity to a leishmanolysin-like peptidase of *Clonorchis sinensis* and the putative head-binding domain of a phage tailspike protein of *Escherichia* sp. 4_1_40B. The tailspike protein can recognize and bind the receptor of the host cell and can destroy the macromolecular structure of the bacterial capsule (2, 12). So our preliminary analysis suggests that SWU1 gp59 might be related to the observed novel plaque of SWU1.

Nucleotide sequence accession number. The GenBank accession number for SWU1 is JF946695.

ACKNOWLEDGMENTS

This work was supported by National Megaproject for Key Infectious Disease (grants 2008ZX10003-006 and 2012ZX10003-003), the Fundamental Research Funds for the Central Universities (grants XDJK2009A003 and XDJK2011D006), National Natural Science Foundation (grant 81071316), Excellent Ph.D. Thesis Fellowship of Southwest University (grants kb2010017 and ky2011003), and New Century Excellent Talents in Universities (NCET-11-0703).

We thank Graham F. Hatfull for excellent technical assistance and inspiration for the projectwork.

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Received 20 June 2012 **Accepted** 25 June 2102

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doi:10.1128/JVI.01568-12

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