



Complete Genome Sequence of *Streptomyces* Siphophage Sitrop

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ABSTRACT Streptomyces sp. strain Mg1 is a Gram-positive soil bacterium capable of causing cell lysis and degradation of *Bacillus subtilis* colonies. Here, we report the 48,481-bp genome of *Streptomyces* sp. Mg1 siphophage Sitrop. With 77 predicted protein-coding genes and one tRNA, Sitrop shares 77% nucleotide sequence identity with the *Streptomyces* phage Verse.

S treptomyces sp. strain Mg1 is a Gram-positive, saprotrophic bacterium predominantly found in soil (1, 2). This filamentous organism is known to produce the antibiotic chalcomycin A, which plays a role in competition with and inhibition of *Bacillus subtilis* (2). Streptomycetes produce several useful secondary metabolites, including approximately 80% of today's antibiotics (3). Studying the genomes of phages of industrially important bacterial species, such as *Streptomyces* sp. Mg1 siphophage Sitrop, may be useful for improving bioproduction technologies.

Bacteriophage Sitrop was isolated in February 2019 from a soil sample taken from Lincoln, Nebraska. Sitrop was plaque purified as described elsewhere (4) using Streptomyces sp. Mg1 as its host and cultured at 30°C on nutrient broth or agar supplemented with 10 mM MgCl₂, 8 mM Ca(NO₃)₂, and 0.5% glucose. Sitrop was found to be chloroform sensitive. DNA was purified using a modified Wizard DNA clean-up kit (Promega) protocol (5) and prepared as Illumina TruSeq libraries. Sequencing was performed on an Illumina iSeq 100 instrument with paired-end 300-bp reads using a TruSeq Nano DNA kit. Quality control of the 254,194 resulting sequence reads was done using FastQC (http://www.bioinformatics.babraham.ac.uk/projects/fastqc) and trimmed manually with FastX 0.0.14 (http://hannonlab.cshl.edu/fastx_toolkit/download.html). The genome was assembled into a single contig at 109.8-fold coverage using SPAdes v3.5.0 (6) and closed by PCR and Sanger sequencing of the resulting product using primers GGACGTTGAACTTGTTGAGGA (forward) and GTCCTCCAGGTGTGAAGAAG (reverse). Structural annotations of protein-coding genes were initially predicted by GLIMMER v3 (7) and MetaGeneAnnotator v1.0 (8), while tRNAs were found using ARAGORN v2.36 (9). Conserved domains, sequence similarity, and transmembrane domains were found using InterProScan v5.33 (10), BLAST v2.9.0 (11), and TMHMM v2.0 (12), respectively, to predict gene function. BLAST similarity searches used a 0.001 maximum expectation value cutoff against the NCBI nonredundant, Swiss-Prot, and TrEMBL databases (13) (accessed 8 April 2020). The DNA sequence similarity of the entire genome was calculated with progressiveMauve v2.4 (14). Annotation tools used in Galaxy and Web Apollo are hosted at https://cpt.tamu.edu/galaxy-pub (15) by the Center for Phage Technology. Phage virion morphology was visualized as 2% (wt/vol) uranyl acetate negatively stained samples by transmission electron microscopy at the Texas A&M Microscopy and Imaging Center and determined to be a siphovirus (data not shown). All tools were run with default parameters unless otherwise specified.

Sitrop is a siphophage with a genome of 48,481 bp and a G+C content of 65.6%,

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Received 23 November 2020 Accepted 3 December 2020 Published 7 January 2021 compared to 72.17% of its host. The genome was composed of 1 tRNA gene and 77 predicted protein-coding genes, 41 being assigned putative functions, with a total coding density of 89.7%. Sitrop is most closely related to *Streptomyces* phage Verse (GenBank accession number KT186229.1), having 76.93% nucleotide identity and 67 shared proteins (16). Sitrop also shared close similarity to other *Streptomyces* phages within the *Camvirus* genus. Most of the protein-coding genes predicted to be tail proteins appear to be novel, sharing significant amino acid sequence similarity only with *Streptomyces* phages Alsaber (MG298964.1) and Saftant (MN204498.1), two novel phages with close similarity to Sitrop overall. There were four genes predicted to be involved in lysis, including an endolysin endopeptidase, a holin, and a separated two-component spanin complex gene (17).

Data availability. The genome of Sitrop is available in GenBank under accession number MT701598.1. The associated BioProject, SRA, and BioSample accession numbers are PRJNA222858, SRR11558341, and SAMN14609631, respectively.

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REFERENCES

- Hoefler BC, Konganti K, Straight PD. 2013. De novo assembly of the Streptomyces sp. strain Mg1 genome using PacBio single-molecule sequencing. Genome Announc 1:e00535-13. https://doi.org/10.1128/genomeA .00535-13.
- Barger SR, Hoefler BC, Cubillos-Ruiz A, Russell WK, Russell DH, Straight PD. 2012. Imaging secondary metabolism of Streptomyces sp. Mg1 during cellular lysis and colony degradation of competing Bacillus subtilis. Antonie Van Leeuwenhoek 102:435–445. https://doi.org/10.1007/s10482-012 -9769-0.
- de Lima Procópio RE, da Silva IR, Martins MK, de Azevedo JL, de Araújo JM. 2012. Antibiotics produced by Streptomyces. Braz J Infect Dis 16:466–471. https://doi.org/10.1016/j.bjid.2012.08.014.
- van Charante F, Holtappels D, Blasdel B, Burrowes B. 2019. Isolation of bacteriophages. In Harper D, Abedon S, Burrowes B, McConville M (ed), Bacteriophages. Springer, Cham, Switzerland.
- Summer EJ. 2009. Preparation of a phage DNA fragment library for whole genome shotgun sequencing, p 27–46. *In* Clokie MRJ, Kropinski AM (ed), Bacteriophages: methods and protocols, volume 2: molecular and applied aspects. Humana Press, Totowa, NJ.
- Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. J Comput Biol 19:455–477. https://doi.org/10.1089/cmb.2012.0021.
- Delcher AL, Harmon D, Kasif S, White O, Salzberg SL. 1999. Improved microbial gene identification with GLIMMER. Nucleic Acids Res 27:4636–4641. https://doi.org/10.1093/nar/27.23.4636.
- Noguchi H, Taniguchi T, Itoh T. 2008. MetaGeneAnnotator: detecting species-specific patterns of ribosomal binding site for precise gene prediction in anonymous prokaryotic and phage genomes. DNA Res 15:387–396. https:// doi.org/10.1093/dnares/dsn027.
- Laslett D, Canback B. 2004. ARAGORN, a program to detect tRNA genes and tmRNA genes in nucleotide sequences. Nucleic Acids Res 32:11–16. https://doi.org/10.1093/nar/gkh152.

- Jones P, Binns D, Chang H-Y, Fraser M, Li W, McAnulla C, McWilliam H, Maslen J, Mitchell A, Nuka G, Pesseat S, Quinn AF, Sangrador-Vegas A, Scheremetjew M, Yong S-Y, Lopez R, Hunter S. 2014. InterProScan 5: genome-scale protein function classification. Bioinformatics 30:1236–1240. https://doi.org/10.1093/bioinformatics/btu031.
- Camacho C, Coulouris G, Avagyan V, Ma N, Papadopoulos J, Bealer K, Madden TL. 2009. BLAST+: architecture and applications. BMC Bioinformatics 10:421–421. https://doi.org/10.1186/1471-2105-10-421.
- Krogh A, Larsson B, von Heijne G, Sonnhammer ELL. 2001. Predicting transmembrane protein topology with a hidden Markov model: application to complete genomes. J Mol Biol 305:567–580. https://doi.org/10 .1006/jmbi.2000.4315.
- The UniProt Consortium. 2018. UniProt: the universal protein knowledgebase. Nucleic Acids Res 46:2699–2699. https://doi.org/10.1093/nar/gky092.
- Darling AE, Mau B, Perna NT. 2010. progressiveMauve: multiple genome alignment with gene gain, loss and rearrangement. PLoS One 5:e11147. https://doi.org/10.1371/journal.pone.0011147.
- Afgan E, Baker D, Batut B, van den Beek M, Bouvier D, Cech M, Chilton J, Clements D, Coraor N, Grüning BA, Guerler A, Hillman-Jackson J, Hiltemann S, Jalili V, Rasche H, Soranzo N, Goecks J, Taylor J, Nekrutenko A, Blankenberg D. 2018. The Galaxy platform for accessible, reproducible and collaborative biomedical analyses: 2018 update. Nucleic Acids Res 46:W537–W544. https://doi.org/10.1093/nar/gky379.
- 16. Layton SR, Hemenway RM, Munyoki CM, Barnes EB, Barnett SE, Bond AM, Narvaez JM, Sirisakd CD, Smith BR, Swain J, Syed O, Bowman CA, Russell DA, Bhuiyan S, Donegan-Quick R, Benjamin RC, Hughes LE. 2016. Genome sequences of Streptomyces phages Amela and Verse. Genome Announc 4:e01589-15. https://doi.org/10.1128/genomeA.01589-15.
- 17. Kongari R, Rajaure M, Cahill J, Rasche E, Mijalis E, Berry J, Young R. 2018. Phage spanins: diversity, topological dynamics and gene convergence. BMC Bioinformatics 19:326–326. https://doi.org/10.1186/s12859-018 -2342-8.