



# Complete Genome Sequence of *Salmonella enterica* Serovar Typhimurium Siphophage Skate

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**ABSTRACT** *Salmonella enterica* serovar Typhimurium is a Gram-negative pathogen and a primary cause of foodborne illnesses worldwide. Here, we present the complete 47,393-bp genome sequence of the siphophage Skate, which was isolated against *S. Typhimurium* strain LT2.

*Salmonella enterica* serovar Typhimurium is a Gram-negative pathogen and a primary cause of foodborne illnesses worldwide (1). *S. Typhimurium* causes acute inflammatory diarrhea that can progress to invasive systemic disease (2), with at least 400 deaths occurring due to acute salmonellosis every year in the United States alone (3). As a control measure for *S. Typhimurium*, bacteriophages have garnered interest in recent years (4, 5).

The phage Skate was isolated from soil in the cattle holding pen of a cattle harvesting facility in Michigan in August 2016 using *S. Typhimurium* strain LT2 (6) as the host. Host bacteria were cultured on tryptic soy broth or agar (Difco) at 37°C with aeration. Phages were cultured and propagated by the soft agar overlay method (7). The phage was identified as a siphophage using negative-stain transmission electron microscopy performed at the Texas A&M University Microscopy and Imaging Center, as previously described (8). Phage genomic DNA was prepared using a modified Promega Wizard DNA cleanup kit protocol, as described previously (9). Pooled indexed DNA libraries were prepared using the Illumina TruSeq Nano LT kit, and the sequence was obtained from the Illumina MiSeq platform using the MiSeq V2 500-cycle reagent kit (2 × 250-bp reads), following the manufacturer's instructions, producing 374,842 reads for the index containing the phage genome. FastQC 0.11.5 (<https://www.bioinformatics.babraham.ac.uk/projects/fastqc/>) was used for quality control of the reads. The reads were trimmed with FASTX-Toolkit 0.0.14 ([http://hannonlab.cshl.edu/fastx\\_toolkit/download.html](http://hannonlab.cshl.edu/fastx_toolkit/download.html)) before being assembled into a single contig at 160.9-fold coverage using SPAdes 3.5.0 (10). Contig completion was confirmed by PCR using primers (5'-GTCGAAGCGCTACGTG AATA-3' and 5'-CTTCCCAGAGATCCTTTGATAC-3') facing off the ends of the assembled contig and Sanger sequencing of the resulting product, with the contig sequence manually corrected to match the resulting Sanger sequencing reads. GLIMMER 3.0 (11) and MetaGeneAnnotator 1.0 (12) were used to predict protein-coding genes with manual correction for appropriate gene starts, and tRNA genes were predicted with ARAGORN 2.36 (13). Rho-independent termination sites were identified with TransTerm (<http://transterm.cbcb.umd.edu/>). Sequence similarity searches by BLASTp 2.2.28 (14) and conserved domain searches with InterProScan 5.15-54.0 (15) were used to predict protein function. All analyses were conducted using default settings via the CPT Galaxy (16) and Web Apollo (17) interfaces (<https://cpt.tamu.edu/>).

The Skate genome was assembled into a complete contig of 47,393 bp at 160.9-fold coverage. It has a GC content of 46%. Genes coding for proteins involved in morphogenesis, such as the major coat, capsid decoration, tail tube, tail spike, terminase large subunit, and portal proteins, were identified. A lysis cassette consisting of a class II

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holin, a transglycosylase-type endolysin, and an embedded i-spanin and o-spanin pair were also identified. Genes linked to DNA replication, such as DNA primase, single-stranded DNA binding protein, DNA polymerase subunit, and an ATP-dependent helicase were found. Skate is highly similar to other *Salmonella* phages, like phage IME207 (GenBank accession number [KX523699](#)) and E1 (GenBank accession number [AM491472](#)) (18, 19), sharing 58% and 55% nucleotide identity, respectively, as determined by progressiveMauve (version 2.4.0) (20).

**Data availability.** The genome sequence of phage Skate was submitted to GenBank as accession number [MH321493](#). The associated BioProject, SRA, and BioSample accession numbers are [PRJNA222858](#), [SRR8787571](#), and [SAMN11259649](#), respectively.

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## REFERENCES

- Scallan E, Hoekstra RM, Angulo FJ, Tauxe RV, Widdowson MA, Roy SL, Jones JL, Griffin PM. 2011. Foodborne illness acquired in the United States—major pathogens. *Emerg Infect Dis* 17:7–15. <https://doi.org/10.3201/eid1701.P11101>.
- Anderson CJ, Kendall MM. 2017. *Salmonella enterica* serovar Typhimurium strategies for host adaptation. *Front Microbiol* 8:1983. <https://doi.org/10.3389/fmicb.2017.01983>.
- Fàbrega A, Vila J. 2013. *Salmonella enterica* serovar Typhimurium skills to succeed in the host: virulence and regulation. *Clin Microbiol Rev* 26:308–341. <https://doi.org/10.1128/CMR.00066-12>.
- Carvalho C, Costa AR, Silva F, Oliveira A. 2017. Bacteriophages and their derivatives for the treatment and control of food-producing animal infections. *Crit Rev Microbiol* 43:583–601. <https://doi.org/10.1080/1040841X.2016.1271309>.
- Kahn LH, Bergeron G, Bourassa MW, De Vegt B, Gill J, Gomes F, Malouin F, Opengart K, Ritter GD, Singer RS, Storrs C, Topp E. 2019. From farm management to bacteriophage therapy: strategies to reduce antibiotic use in animal agriculture. *Ann N Y Acad Sci* 1441:31–39. <https://doi.org/10.1111/nyas.14034>.
- McClelland M, Sanderson KE, Spieth J, Clifton SW, Latreille P, Courtney L, Porwollik S, Ali J, Dante M, Du F, Hou S, Layman D, Leonard S, Nguyen C, Scott K, Holmes A, Grewal N, Mulvaney E, Ryan E, Sun H, Florea L, Miller W, Stoneking T, Nhan M, Waterston R, Wilson RK. 2001. Complete genome sequence of *Salmonella enterica* serovar Typhimurium LT2. *Nature* 413:852–856. <https://doi.org/10.1038/35101614>.
- Adams MK. 1959. Bacteriophages. Interscience Publishers, Inc., New York, NY.
- Summer EJ, Liu M, Gill JJ, Grant M, Chan-Cortes TN, Ferguson L, Janes C, Lange K, Bertoli M, Moore C, Orchard RC, Cohen ND, Young R. 2011. Genomic and functional analyses of *Rhodococcus equi* phages ReqiPep6, ReqiPoco6, ReqiPine5, and ReqiDocB7. *Appl Environ Microbiol* 77:669–683. <https://doi.org/10.1128/AEM.01952-10>.
- Gill JJ, Berry JD, Russell WK, Lessor L, Escobar-Garcia DA, Hernandez D, Kane A, Keene J, Maddox M, Martin R, Mohan S, Thorn AM, Russell DH, Young R. 2012. The *Caulobacter crescentus* phage phiCbK: genomics of a canonical phage. *BMC Genomics* 13:542. <https://doi.org/10.1186/1471-2164-13-542>.
- 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>.
- Camacho C, Coulouris G, Avagyan V, Ma N, Papadopoulos J, Bealer K, Madden TL. 2009. BLAST+: architecture and applications. *BMC Bioinformatics* 10:421. <https://doi.org/10.1186/1471-2105-10-421>.
- Jones P, Binns D, Chang HY, Fraser M, Li W, McAnulla C, McWilliam H, Maslen J, Mitchell A, Nuka G, Pesseat S, Quinn AF, Sangrador-Vegas A, Scheremetjew M, Yong SY, Lopez R, Hunter S. 2014. InterProScan 5: genome-scale protein function classification. *Bioinformatics* 30:1236–1240. <https://doi.org/10.1093/bioinformatics/btu031>.
- Cock PJ, Gruning BA, Paszkiewicz K, Pritchard L. 2013. Galaxy tools and workflows for sequence analysis with applications in molecular plant pathology. *PeerJ* 1:e167. <https://doi.org/10.7717/peerj.167>.
- Lee E, Helt GA, Reese JT, Munoz-Torres MC, Childers CP, Buels RM, Stein L, Holmes IH, Elisk CG, Lewis SE. 2013. Web Apollo: a Web-based genomic annotation editing platform. *Genome Biol* 14:R93. <https://doi.org/10.1186/gb-2013-14-8-r93>.
- Liu Y, Mi L, Mi Z, Huang Y, Li P, Zhang X, Tong Y, Bai C. 2016. Complete genome sequence of IME207, a novel bacteriophage which can lyse multidrug-resistant *Klebsiella pneumoniae* and *Salmonella*. *Genome Announc* 4:e01015-16. <https://doi.org/10.1128/genomeA.01015-16>.
- Pickard D, Thomson NR, Baker S, Wain J, Pardo M, Goulding D, Hamlin N, Choudhary J, Threfall J, Dougan G. 2008. Molecular characterization of the *Salmonella enterica* serovar Typhi Vi-typing bacteriophage E1. *J Bacteriol* 190:2580. <https://doi.org/10.1128/JB.01654-07>.
- 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>.