



# Complete Genome Sequence of *Salmonella enterica* Serovar Typhimurium Myophage Mutine

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**ABSTRACT** Mutine is a myophage of *Salmonella enterica* serovar Typhimurium. Here, we present the complete genome of Mutine (161,502 bp) and show that it is similar to that of phage Vi01.

*Salmonella* spp. are major agents causing foodborne illnesses that result in at least 400 deaths per year (1). Strains of *Salmonella enterica* serovar Typhimurium are rapidly gaining antibiotic resistance in both developed and developing countries (2). The study of *S.* Typhimurium phages may offer alternative control means for this pathogen.

Phage Mutine, using *S.* Typhimurium as the host, was isolated from mixed municipal wastewater collected in Brazos County, TX, in 2016. Host bacteria were cultured on tryptic soy broth or agar (Difco) at 37°C with aeration, and phage isolation and propagation were done using the soft agar overlay method (3). Phage genomic DNA was prepared using a modified Promega Wizard DNA cleanup kit protocol, as previously described (4). Pooled indexed DNA libraries were prepared using the Illumina TruSeq Nano low-throughput (LT) kit, and the sequence was obtained using the Illumina MiSeq platform and the MiSeq v2 500-cycle reagent kit, following the manufacturer's instructions, producing 538,626 paired-end reads for the index containing the phage genome. The reads were verified in FastQC 0.11.5 (<https://www.bioinformatics.babraham.ac.uk/projects/fastqc/>), trimmed with FastX-Toolkit 0.0.14 ([http://hannonlab.cshl.edu/fastx\\_toolkit/](http://hannonlab.cshl.edu/fastx_toolkit/)), and built in SPAdes 3.5.0 (5). The genome was closed by PCR using primers 5'-AACACGCTGGGCATACTT-3' and 5'-CGTCAACAAACACCCTATTAC-3' facing towards each end of the assembled contig and by Sanger sequencing of the resulting product, with the contig sequence manually corrected to match the resulting Sanger sequencing read. The genes that coded for proteins were predicted by the software GLIMMER 3.0 (6) and MetaGeneAnnotator 1.0 (7), along with manual correction; the tRNA genes were found using ARAGORN 2.36 (8). To predict the protein functions, BLASTp 2.2.28 (9) was used to find sequence homology, and conserved domain searches were found using InterProScan 5.15-5.40 (10). All analyses were done using default settings via the CPT Galaxy (11) and Web Apollo (12) interfaces ([cpt.tamu.edu](http://cpt.tamu.edu)).

The Mutine genome was assembled into a contig of 161,502 bp with 153-fold coverage. There are in total 218 protein-encoding genes and 3 tRNAs in the Mutine genome. It has a GC content of 44.3% and a coding density of 92.9%. According to a BLASTp search at an expected (E) value of  $<10^{-5}$ , Mutine shares 204 similar proteins with Vi01 (GenBank accession number [FQ312032](https://www.ncbi.nlm.nih.gov/nuccore/FQ312032)) (13), a member of a group of large myophages of the genus *Viunalikevirus* that are distantly related to phage T4 (14). Using the progressiveMAUVE algorithm (version 2.4.0) (15), Mutine shares 87% DNA similarity to *Escherichia* phage vB\_EcoM\_CBA120 (GenBank accession number [NC\\_016570](https://www.ncbi.nlm.nih.gov/nuccore/NC_016570)), another Vi01-like phage. The Mutine genome is presented as syntenic (in the same gene order) to phage T4, with antiabortive infection genes (RIIA or RIIB) at each end of the genome. Mutine has many expected morphogenesis proteins of Vi01-like phages,

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including a cluster of tail spike proteins, an arrangement which might lead to the umbrella-like structures of the tail (13, 14). A superinfection exclusion protein similar to phage P22 gp17 was identified (16). An *N*-acetylmuramidase-like endolysin is identified in Mutine, but no holins or spanin complex could be reliably identified. The tape measure protein was identified without an identifiable chaperone protein.

**Data availability.** The genome sequence of phage Mutine was deposited under GenBank accession number [MG428992](https://ncbi.nlm.nih.gov/nucl/MG428992). The associated BioProject, SRA, and BioSample accession numbers are [PRJNA222858](https://ncbi.nlm.nih.gov/bioproject/PRJNA222858), [SRR8788533](https://ncbi.nlm.nih.gov/sra/SRR8788533), and [SAMN11260686](https://ncbi.nlm.nih.gov/biosample/SAMN11260686), respectively.

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## REFERENCES

- Scallan E, Griffin PM, Angulo FJ, Tauxe RV, Hoekstra RM. 2011. Foodborne illness acquired in the United States—unspecified agents. *Emerg Infect Dis* 17:16–22. <https://doi.org/10.3201/eid1701.P21101>.
- Threlfall EJ. 2002. Antimicrobial drug resistance in *Salmonella*: problems and perspectives in food- and water-borne infections. *FEMS Microbiol Rev* 26:141–148. <https://doi.org/10.1111/j.1574-6976.2002.tb00606.x>.
- Adams MK. 1959. Bacteriophages. Interscience Publishers, Inc., New York, NY.
- 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, Paszkiwicz 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>.
- Pickard D, Toribio AL, Petty NK, van Tonder A, Yu L, Goulding D, Barrell B, Rance R, Harris D, Wetter M, Wain J, Choudhary J, Thomson N, Dougan G. 2010. A conserved acetyl esterase domain targets diverse bacteriophages to the Vi capsular receptor of *Salmonella enterica* serovar Typhi. *J Bacteriol* 192:5746–5754. <https://doi.org/10.1128/JB.00659-10>.
- Adriaenssens EM, Ackermann HW, Anany H, Blasdel B, Connerton IF, Goulding D, Griffiths MW, Hooton SP, Kutter EM, Kropinski AM, Lee JH, Maes M, Pickard D, Ryu S, Seppehrizadeh Z, Shahrbabak SS, Toribio AL, Lavigne R. 2012. A suggested new bacteriophage genus: “Viunalikevirus.” *Arch Virol* 157:2035–2046. <https://doi.org/10.1007/s00705-012-1360-5>.
- 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>.
- Semerjian AV, Malloy DC, Poteete AR. 1989. Genetic structure of the bacteriophage P22 PL operon. *J Mol Biol* 207:1–13. [https://doi.org/10.1016/0022-2836\(89\)90437-3](https://doi.org/10.1016/0022-2836(89)90437-3).