



Complete Genome Sequence of Bacteriophage MA12, Which Infects both *Campylobacter jejuni* and *Salmonella enterica* Serovar Enteritidis

Sunjin Lee,^a Taesoo Kwon,^b Su-Jin Chae,^a Jong-Hyun Kim,^a Yeon Ho Kang,^{a,b} Gyung Tae Chung,^a Dae-Won Kim,^b Deog-Yong Lee^a

Division of Enteric Diseases, Center for Infectious Diseases, National Institute of Health, KCDC, Heungdeok-gu, Cheongju-si, Chungcheongbuk-do, Republic of Korea^a; Division of Biosafety Evaluation and Control, National Institute of Health, KCDC, Heungdeok-gu, Cheongju-si, Cheongbuk-do, Republic of Korea^b

S.L. and T.K. contributed equally to the work.

Here, we announce the complete genome sequence of *Salmonella enterica* serovar Enteritidis (*S.* Enteritidis) bacteriophage MA12, a 41-Kb chromosome. The strain can infect both *Campylobacter jejuni* (*C. jejuni*) and *S.* Enteritidis and can be used in phage therapy experiments with poultry and poultry meat.

Received 15 June 2016 Accepted 20 October 2016 Published 8 December 2016

Citation Lee S, Kwon T, Chae S-J, Kim J-H, Kang YH, Chung GT, Kim D-W, Lee D-Y. 2016. Complete genome sequence of bacteriophage MA12, which infects both *Campylobacter jejuni* and *Salmonella enterica* serovar Enteritidis. Genome Announc 4(6):e00810-16. doi:10.1128/genomeA.00810-16.

Copyright © 2016 Lee et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Dae-Won Kim, todaewon@gmail.com, or Deog-Yong Lee, leedy0610@korea.kr.

Campylobacter spp. and *Salmonella* spp. are common causes of gastrointestinal disease and are typically transmitted by food-products derived from the poultry industry (1, 2). These pathogens can be controlled by specific phages applied throughout the food chain (3).

Bacteriophage MA12 was isolated from a surface water sample using *Salmonella enterica* serovar Enteritidis (*S*. Enteritidis) PT1b as a host, and it was also able to infect *C. jejuni*. The phage was purified using the top agar method and polyethylene glycol (PEG)-precipitation. Morphological analysis by TEM revealed that phage MA12 belongs to the family *Siphoviridae* (4), it has an icosahedral head of 81 nm, and a long tail of 123 nm by 14 nm.

Genomic DNA was concentrated using PEG 8000 precipitation and isolated using the Sambrook's phenol extraction method (5). The GS FLX Titanium rapid library preparation kit (Roche Diagnostics, Germany) was used for a sequencing library construction by following the manufacturer's instructions. The sequence reads were generated using GS FLX platform and de novo assembled using GS De Novo Assembler (version 2.9). A total of 15,674 reads (6,964,979 bp) were generated and the average read length was 444,366 bp. The sequencing coverage was 169.8×. A total of 15,667 reads were used for de novo assembly and 15,209 reads were assembled into a contig after removing singletons and partial reads. After the de novo assembly, the physical ends of the genome were not determined (6). The size of the MA12 genome was 41,224 bp and G+C content was 49.88%. When the genome was compared to the reference genomes, Salmonella phage Jersey, the nucleotide homology was approximately 47.2%. The nucleotide homology of MA12 was calculated using EMBOSS Stretcher (EMBOSS version 6.5.7.0) (7). Whole-genome based phylogenetic analysis revealed that the closest neighbor of MA12 is Salmonella phage wksl3 and Salmonella phage SS3e. The three strains, MA12, wksl3, and SS3e were classified into an independent clade set apart from Jerseylikevirus (8). A multiple sequence alignment was performed using MAFFT (version 7.221) (9) and the whole-genome based phylogenetic tree was inferred using RAxML (version 8.2.0) (10) with the Gamma GTR model. The annotation of the genome

was performed using the RAST (Rapid Annotation using Subsystem Technology) server (11). From the annotation, 59 protein-coding sequences were identified. Of those protein-coding sequences, 54 matched identified phage genes, including 47 genes involved in phage structure, six genes involved in DNA replication, one gene of late protein, and one homing endonuclease and the others are protein-coding sequences that encoded hypothetical proteins.

Accession number(s). The genome sequence of MA12 was deposited in DDBJ/EMBL/GenBank under the accession number KX245013.

ACKNOWLEDGMENTS

This work was supported by grants from the Division of Enteric Diseases (KNIH 4800-4837-301) and the Division of Biosafety Evaluation and Control (KNIH 4800-4847-311) of the National Institute of Health, Korea Centers for Disease Control and Prevention.

FUNDING INFORMATION

This work, including the efforts of Jong-Hyun Kim, was funded by MOHW | Korea Centers for Disease Control & Prevention (KCDC) (4837-301-210 and 4847-311-210).

REFERENCES

- Skarp CP, Hänninen ML, Rautelin HI. 2016. Campylobacteriosis: the role of poultry meat. Clin Microbiol Infect 22:103–109. http://dx.doi.org/ 10.1016/j.cmi.2015.11.019.
- Antunes P, Mourão J, Campos J, Peixe L. 2016. Salmonellosis: the role of poultry meat. Clin Microbiol Infect 22:110–121. http://dx.doi.org/ 10.1016/j.cmi.2015.12.004.
- Hong SS, Jeong J, Lee J, Kim S, Min W, Myung H. 2013. Therapeutic effects of bacteriophages against *Salmonella gallinarum* infection in chickens. J Microbiol Biotechnol 23:1478–1483. http://dx.doi.org/10.4014/ jmb.1304.04067.
- Ackermann HW, Prangishvili D. 2012. Prokaryote viruses studied by electron microscopy. Arch Virol 157:1843–1849. http://dx.doi.org/ 10.1007/s00705-012-1383-y.
- 5. Sambrook J, Russell DW. 2001. Molecular cloning: a laboratory manual. Cold Spring Harbor Laboratory Press, New York, NY.
- 6. Li W, Cowley A, Uludag M, Gur T, McWilliam H, Squizzato S, Park YM, Buso N, Lopez R. 2015. The EMBL-EBI bioinformatics web and

programmatic tools framework. Nucleic Acids Res 43:W580–W584. http://dx.doi.org/10.1093/nar/gkv279.

- Casjens SR, Gilcrease EB. 2009. Determining DNA packaging strategy by analysis of the termini of the chromosomes in tailed-bacteriophage virions. Methods Mol Biol 502:91–111. http://dx.doi.org/10.1007/978-1 -60327-565-1_7.
- Anany H, Switt AI, De Lappe N, Ackermann HW, Reynolds DM, Kropinski AM, Wiedmann M, Griffiths MW, Tremblay D, Moineau S, Nash JH, Turner D. 2015. A proposed new bacteriophage subfamily: "Jerseyvirinae." Arch Virol 160:1021–1033. http://dx.doi.org/10.1007/ s00705-015-2344-z.
- 9. Katoh K, Standley DM. 2013. MAFFT multiple sequence alignment soft-

ware version 7: Improvements in performance and usability. Mol Biol Evol **30:**772–780. http://dx.doi.org/10.1093/molbev/mst010.

- Stamatakis A. 2006. RAxML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. Bioinformatics 22:2688–2690. http://dx.doi.org/10.1093/bioinformatics/btl446.
- 11. Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paczian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: Rapid Annotations using Subsystems Technology. BMC Genomics 9:75. http://dx.doi.org/10.1186/ 1471-2164-9-75.