





Genome Sequence of the *Pseudomonas protegens* Phage **•GP100**

DJordan Vacheron, a Peter Kupferschmied, a* Grégory Resch, a Christoph Keela

^aDepartment of Fundamental Microbiology, University of Lausanne, Lausanne, Switzerland

ABSTRACT We report here the complete annotated genome sequence of Φ GP100, a lytic bacteriophage of the *Podoviridae* family. Φ GP100 was isolated from rhizosphere soil in Switzerland and infects specifically strains of *Pseudomonas protegens* that are known for their plant-beneficial activities. Phage Φ GP100 has a 50,547-bp genome with 76 predicted open reading frames.

In 2002, Keel and colleagues isolated a lytic bacteriophage belonging to the *Podoviridae* family from the rhizosphere of cucumber plants (1). The phage, named Φ GP100, was found to infect specifically *Pseudomonas protegens* CHA0 and related strains of the same species. *P. protegens* strains are highly competitive root colonizers and are studied for their biocontrol effects against plant pathogens (2, 3, 4) and herbivorous insect pests (5, 6).

We sequenced and annotated the full genome of Φ GP100. Extraction of Φ GP100 DNA was done from a purified suspension of the phage containing 10^9 PFU \cdot ml $^{-1}$ using a standard phenol-chloroform extraction procedure. The phage DNA was sequenced at the Lausanne Genomic Technologies Facility in Switzerland. Sequencing libraries were prepared using the TruSeq Nano DNA LT library preparation kit (Illumina, San Diego, CA, USA) and sequenced with the HiSeq 2500 platform, generating an output of 100-bp paired-end reads. Reads were assembled into contigs with the Edena v3 *de novo* short read assembler (7). Annotation of open reading frames (ORFs) was done with Rapid Annotations using Subsystems Technology (RAST) (8) and PHAge Search Tool Enhanced Release (PHASTER) (9). Each predicted ORF was further examined using BLAST and Conserved Domain database searches on the NCBI website (https://www.ncbi.nlm.nih .gov). tRNAs were predicted using ARAGORN (10).

A total of 20,139,130 paired-end reads were obtained, leading to a coverage exceeding 39,500×. The assembly generated a single contig of 50,547 bp with a G+C content of 51% corresponding to the entire phage genome, which is in agreement with the genome size previously determined by restriction analysis (1). Seventy-six potential ORFs were predicted. In particular, we found structural genes coding for phage tail fiber protein (GenBank accession number SPF82154), phage terminase large subunit (SPF82151), phage portal protein (SPF82150), and phage capsid protein (SPF82132). We also found genes encoding proteins potentially involved in phage DNA replication, notably a DNA helicase (GenBank accession number SPP13286), a polymerase (SPF82110), and a lysin for phage release (SFP82136). Two tRNA sequences were predicted, one of which is a 73-nucleotide (nt)-long tRNA for which the anticodon reads a stop codon (TAA), suggesting that it may act as a nonsense suppressor (11). The anticodon of the second predicted tRNA reads an Asn codon (GTT).

The best nucleotide BLAST hits for the whole genome were *Pseudomonas* phage IME180 (GenBank accession number MF788075) and *Pseudomonas* phage O4 (NC_031274), which shared less than 70% identity on maximally 32% of their genome lengths with the Φ GP100 genome. All of these phages infect *P. aeruginosa* strains, unlike phage Φ GP100, which seems to be specific to a subset of *P. protegens* strains (1;

Received 12 March 2018 **Accepted** 7 May 2018 **Published** 21 June 2018

Citation Vacheron J, Kupferschmied P, Resch G, Keel C. 2018. Genome sequence of the *Pseudomonas protegens* phage ФGP100. Genome Announc 6:e00261-18. https://doi.org/10.1128/qenomeA.00261-18.

Copyright © 2018 Vacheron et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Jordan Vacheron, jordan.vacheron@unil.ch, or Christoph Keel, christoph.keel@unil.ch.

* Present address: Peter Kupferschmied, Federal Office for Agriculture FOAG, Plant Health and Varieties Unit, Bern, Switzerland. our unpublished data). Phages can be considered a major driving force influencing microbial diversity in soil (12), and the ecological study of this phage-*Pseudomonas* model may thus lead, in a larger perspective, to an improved understanding of phage-bacterium interactions in complex environments such as the rhizosphere.

Accession number(s). The complete genome sequence of Φ GP100 was deposited at the European Nucleotide Archive as BioProject ID PRJEB24648, sample ERS2161702. The assembled genome sequence was deposited at DDBJ/EMBL/GenBank under the accession number LT986460.

ACKNOWLEDGMENTS

This study was supported by Swiss National Science Foundation grant 31003A-159520 to C.K.

We thank Aurélie Marchet, Shawna McCallin, and Frank Oechslin for technical expertise and advice.

REFERENCES

- Keel C, Ucurum Z, Michaux P, Adrian M, Haas D. 2002. Deleterious impact of a virulent bacteriophage on survival and biocontrol activity of *Pseu-domonas fluorescens* strain CHA0 in natural soil. Mol Plant Microbe Interact 15:567–576. https://doi.org/10.1094/MPMI.2002.15.6.567.
- Haas D, Défago G. 2005. Biological control of soil-borne pathogens by fluorescent pseudomonads. Nat Rev Microbiol 3:307–319. https://doi .org/10.1038/nrmicro1129.
- Kupferschmied P, Maurhofer M, Keel C. 2013. Promise for plant pest control: root-associated pseudomonads with insecticidal activities. Front Plant Sci 4:287. https://doi.org/10.3389/fpls.2013.00287.
- Vacheron J, Desbrosses G, Bouffaud M-L, Touraine B, Moënne-Loccoz Y, Muller D, Legendre L, Wisniewski-Dyé F, Prigent-Combaret C. 2013. Plant growth-promoting rhizobacteria and root system functioning. Front Plant Sci 4:356. https://doi.org/10.3389/fpls.2013.00356.
- Flury P, Aellen N, Ruffner B, Péchy-Tarr M, Fataar S, Metla Z, Dominguez-Ferreras A, Bloemberg G, Frey J, Goesmann A, Raaijmakers JM, Duffy B, Höfte M, Blom J, Smits THM, Keel C, Maurhofer M. 2016. Insect pathogenicity in plant-beneficial pseudomonads: phylogenetic distribution and comparative genomics. ISME J 10:2527–2542. https://doi.org/10.1038/ismej.2016.5.
- Keel C. 2016. A look into the toolbox of multi-talents: insect pathogenicity determinants of plant-beneficial pseudomonads. Environ Microbiol 18:3207–3209. https://doi.org/10.1111/1462-2920.13462.
- 7. Hernandez D, François P, Farinelli L, Østerås M, Schrenzel J. 2008. De

- novo bacterial genome sequencing: millions of very short reads assembled on a desktop computer. Genome Res 18:802–809. https://doi.org/10.1101/gr.072033.107.
- Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the Rapid Annotation of microbial genomes using Subsystems Technology (RAST). Nucleic Acids Res 42:D206–D214. https://doi.org/10.1093/nar/gkt1226.
- Arndt D, Grant JR, Marcu A, Sajed T, Pon A, Liang Y, Wishart DS. 2016. PHASTER: a better, faster version of the PHAST phage search tool. Nucleic Acids Res 44:W16–W21. https://doi.org/10.1093/nar/gkw387.
- 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.
- Pedulla ML, Ford ME, Houtz JM, Karthikeyan T, Wadsworth C, Lewis JA, Jacobs-Sera D, Falbo J, Gross J, Pannunzio NR, Brucker W, Kumar V, Kandasamy J, Keenan L, Bardarov S, Kriakov J, Lawrence JG, Jacobs WR, Jr, Hendrix RW, Hatfull GF. 2003. Origins of highly mosaic mycobacteriophage genomes. Cell 113:171–182. https://doi.org/10.1016/S0092-8674 (03)00233-2.
- De Smet J, Hendrix H, Blasdel BG, Danis-Wlodarczyk K, Lavigne R. 2017. Pseudomonas predators: understanding and exploiting phage-host interactions. Nat Rev Microbiol 15:517–530. https://doi.org/10.1038/ nrmicro.2017.61.

Volume 6 Issue 25 e00261-18 genomea.asm.org **2**