



PROKARYOTES



Complete Genome Sequence of *Pseudomonas viridiflava* CFBP 1590, Isolated from Diseased Cherry in France

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ABSTRACT *Pseudomonas viridiflava* causes foliar and stem necrosis, as well as stem and root rot on a wide range of plants. We report here the first complete genome of a *P. viridiflava* strain, isolated from diseased tissue of a cherry tree.

The pectinolytic bacterium *Pseudomonas viridiflava* is a multihost pathogen belonging to the *P. syringae* species complex. Phylogeny based on similarity of housekeeping genes placed the *P. viridiflava* species within phylogroup 7 (1, 2), which reflects the genomospecies 6 obtained by DNA-DNA hybridization (3). *P. viridiflava* was originally isolated from a dwarf bean plant in Switzerland and was then reported to be a natural pathogen of a wide range of plants (4), including tomato (5) and the model plant *Arabidopsis thaliana* (6). Recently, strains of *P. viridiflava* were isolated also from nonagricultural habitats (7), thus highlighting the adaptation potential of this species. Previous studies revealed the presence of two distinct phylogenetic clusters within the *P. viridiflava* species (8) and showed the presence of two mutually exclusive configurations of the type III secretion system (T3SS), namely, the tripartite pathogenicity island (T-PAI) and the single pathogenicity island (S-PAI), located in two different genomic locations (9). Recently, Bartoli et al. showed that the T3SS configuration was not linked to pathogenicity and that the only trait found to be correlated with pathogenicity was the presence/absence of the type III effector AvrE (10).

To date, a total of nine *P. viridiflava* whole-genome shotgun sequences are available in GenBank, but no complete genome sequence has been provided yet. In this study, the first complete genome of a member of the *P. viridiflava* species was obtained using PacBio single-molecule real-time (SMRT) read sequencing technology. The selected strain, *P. viridiflava* CFBP 1590, was isolated in 1974 in France from a sour cherry tree displaying symptoms of cortical necrosis.

Genomic DNA for PacBio whole-genome sequencing was extracted following the protocol described by Pitcher et al. (11). PacBio library preparation and sequencing were performed at the Functional Genomic Center Zurich (Zurich, Switzerland). SMRTbells were prepared using the DNA template prep kit version 2.0 (3 kb to 10 kb) (Pacific Biosciences, Menlo Park, CA, USA), and sequencing was performed on a PacBio RSII system (Pacific Biosciences) run with P4/C2 chemistry. Six SMRT cells yielded 464,727 reads, with an average length of 5,901 bp (for a total of 802,154,567 bp). The obtained reads were assembled into one single and circular contig of 6,035,297 bp using the HGAP3 approach on SMRT analysis software version 2.3.0 with manual refinement using BLASTn (12). The sequence was annotated using GenDB (13), which yielded a total of 5,129 genes and a GC content of 59.31%.

The obtained genome possesses the S-PAI T3SS configuration, as well as the AvrE T3E, found also in the draft genomes of *P. viridiflava* strains TA043 (GenBank accession number AVDV01000000), UASWS0038 (GenBank accession number AMQP01000000),

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and CC1582 (GenBank accession number AVDW01000000), isolated from cowslip, rhododendron, and epilithon, respectively (10). In contrast to the draft genome of *P. viridiflava* strain CDRTc14 (GenBank accession number MBPF01000000), no plasmid was found in the strain sequenced in this study.

Accession number(s). The complete genome sequence of *P. viridiflava* strain CFBP 1590 has been deposited in DDBJ/ENA/GenBank under the accession number LT855380.

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REFERENCES

- Parkinson N, Bryant R, Bew J, Elphinstone J. 2011. Rapid phylogenetic identification of members of the *Pseudomonas syringae* species complex using the *rpoD* locus. Plant Pathol 60:338–344. https://doi.org/10.1111/ j.1365-3059.2010.02366.x.
- Berge O, Monteil CL, Bartoli C, Chandeysson C, Guilbaud C, Sands DC, Morris CE. 2014. A user's guide to a data base of the diversity of *Pseudomonas syringae* and its application to classifying strains in this phylogenetic complex. PLoS One 9:e105547. https://doi.org/10.1371/ journal.pone.0105547.
- Gardan L, Shafik H, Belouin S, Broch R, Grimont F, Grimont PA. 1999. DNA relatedness among the pathovars of *Pseudomonas syringae* and description of *Pseudomonas tremae* sp. nov. and *Pseudomonas cannabina* sp. nov. (ex Sutic and Dowson 1959). Int J Syst Bacteriol 49:469–478. https:// doi.org/10.1099/00207713-49-2-469.
- Goumans DE, Chatzaki AK. 1998. Characterization and host range evaluation of *Pseudomonas viridiflava* from melon, blite, tomato, chrysanthemum and eggplant. Eur J Plant Pathol 104:181–188. https://doi.org/ 10.1023/A:1008675704834.
- Goumas DE, Malathrakis NE, Chatzaki AK. 1999. Characterization of *Pseudomonas viridiflava* associated with a new symptom on tomato fruit. Eur J Plant Pathol 105:927–932. https://doi.org/10.1023/A:1008725818334.
- Jakob K, Goss EM, Araki H, Van T, Kreitman M, Bergelson J. 2002. Pseudomonas viridiflava and P. syringae—natural pathogens of Arabidop- sis thaliana. Mol Plant-Microbe Interact 15:1195–1203. https://doi.org/10 .1094/MPMI.2002.15.12.1195.
- Morris CE, Sands DC, Vanneste JL, Montarry J, Oakley B, Guilbaud C, Glaux C. 2010. Inferring the evolutionary history of the plant pathogen *Pseudomonas syringae* from its biogeography in headwaters of rivers in

North America, Europe, and New Zealand. mBio 1:e00107-10. https://doi .org/10.1128/mBio.00107-10.

- Goss EM, Kreitman M, Bergelson J. 2005. Genetic diversity, recombination and cryptic clades in *Pseudomonas viridiflava* infecting natural populations of *Arabidopsis thaliana*. Genetics 169:21–35. https://doi.org/ 10.1534/genetics.104.031351.
- Araki H, Tian D, Goss EM, Jakob K, Halldorsdottir SS, Kreitman M, Bergelson J. 2006. Presence/absence polymorphism for alternative pathogenicity islands in *Pseudomonas viridiflava*, a pathogen of *Arabidopsis*. Proc Natl Acad Sci U S A 103:5887–5892. https://doi.org/10.1073/ pnas.0601431103.
- Bartoli C, Berge O, Monteil CL, Guilbaud C, Balestra GM, Varvaro L, Jones C, Dangl JL, Baltrus DA, Sands DC, Morris CE. 2014. The *Pseudomonas viridiflava* phylogroups in the *P. syringae* species complex are characterized by genetic variability and phenotypic plasticity of pathogenicityrelated traits. Environ Microbiol 16:2301–2315. https://doi.org/10.1111/ 1462-2920.12433.
- 11. Pitcher DG, Saunders NA, Owen RJ. 1989. Rapid extraction of bacterial genomic DNA with guanidium thiocyanate. Lett Appl Microbiol 8:151–156. https://doi.org/10.1111/j.1472-765X.1989.tb00262.x.
- Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ. 1990. Basic local alignment search tool. J Mol Biol 215:403–410. https://doi.org/10.1016/ S0022-2836(05)80360-2.
- Meyer F, Goesmann A, McHardy AC, Bartels D, Bekel T, Clausen J, Kalinowski J, Linke B, Rupp O, Giegerich R, Pühler A. 2003. GenDB - an open source genome annotation system for prokaryote genomes. Nucleic Acids Res 31:2187–2195. https://doi.org/10.1093/nar/gkg312.