




The Draft Genome Sequence of *Pseudomonas frederiksbergensis* Strain 11-D3 Reveals Its Ability To Mobilize Phosphorus

 Bang-Xiao Zheng,^{a,b} Huang-Kai Zhang,^c Yu-Sen Zhang,^a Kai Ding^a

^aKey Laboratory of Urban Environment and Health, Institute of Urban Environment, Chinese Academy of Sciences, Xiamen, People's Republic of China

^bFaculty of Biological and Environmental Sciences, Ecosystems and Environment Research Programme, University of Helsinki, Lahti, Finland

^cXiamen Anjie Medical Data Technology Co. Ltd., Xiamen, People's Republic of China

ABSTRACT *Pseudomonas frederiksbergensis* strain 11-D3 is a Gram-negative, nonmotile, aerobic bacterium isolated from a managed maize field in North China. This strain displays high efficiency for solubilization of inorganic phosphorus (P). We present the draft genome sequence of strain 11-D3 with P cycling genes which indicate the presence of a probable mechanism for P mobilization.

Species of the genus *Pseudomonas* are commonly considered important decomposers of organic compounds in soil and water (1). Certain *Pseudomonas* strains have biotechnological importance due to their significant plant growth promotion effects, including control of fungal disease and mobilization of phosphorus (P) (2). *Pseudomonas frederiksbergensis* was first reported in 2000 (3) and then was proved to have the ability to oxidate organic sulfides (4). Strain 11-D3 was isolated from a managed maize field in Hailun, Heilongjiang Province, China (47°26'N, 126°38'E), by using a rapid isolation method (5). This bacterium is of interest because similar strains have shown the ability to solubilize P from diverse inorganic sources, including calcium, aluminum, and ferric phosphate (6).

Strain 11-D3 is an aerobic bacterium which survives best at 30°C and is viable in tested pH ranges from 5.5 to 8.0 (5). To taxonomically identify the affiliation of strain 11-D3, the 16S rRNA sequence (NCBI accession number [KU647205](https://www.ncbi.nlm.nih.gov/nuccore/KU647205)) was amplified with the bacterial universal primers 27F and 1492R (7), obtained by Sanger sequencing (Invitrogen, Shanghai, China), and submitted to EzBioCloud for alignment with the type strain database (8). This sequence contains 1,444 bp (99.0% of the full-length 16S rRNA), and the highest sequence identity match of 98.68% was with *P. frederiksbergensis* strain JAJ28^T. Hence, we propose the affiliation of strain 11-D3 with *P. frederiksbergensis*.

Strain 11-D3 was cultured on Pikovskaya (PVK) medium (9) at 30°C for 24 h, and a single colony was picked for culture in liquid PVK medium for another 24-h cultivation period. The strain cells were collected by centrifugation at 5,000 × *g* for 15 min, and the genomic DNA was extracted by using a genomic DNA extraction kit for bacteria (Bioteck, Beijing, China) according to the manufacturer's instructions. The draft genome was sequenced by using an Illumina HiSeq 2000 platform (Majorbio, Shanghai, China). Trim Galore v0.3.2 (https://www.bioinformatics.babraham.ac.uk/projects/trim_galore/) was used to remove adapters and low-quality bases and reads. A 1,139-Mb sequence, with 1,223 Mb of total reads, was assembled by using the Short Oligonucleotide Analysis Package (SOAPdenovo v2.01) with all parameters set to default (10). The submitted genome sequence was autoannotated by using the NCBI Prokaryotic Genome Annotation Pipeline with default settings (11). The draft genome assembly of strain 11-D3 contains 35 contigs in 35 scaffolds, and the total genome size is 6,524,485 bp. The G+C content is 59.44%. Genome annotation produced 6,211 genes, of which 5,839 (94.01%) are protein-coding genes. Two gene clusters (*pqqEDCBA* and

Citation Zheng B-X, Zhang H-K, Zhang Y-S, Ding K. 2019. The draft genome sequence of *Pseudomonas frederiksbergensis* strain 11-D3 reveals its ability to mobilize phosphorus. *Microbiol Resour Annot* 8:e01321-18. <https://doi.org/10.1128/MRA.01321-18>.

Editor Jason E. Stajich, University of California, Riverside

Copyright © 2019 Zheng et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Kai Ding, kding@iue.ac.cn.

Received 26 September 2018

Accepted 12 February 2019

Published 14 March 2019

pqqFECBA) involved in pyrroloquinoline quinone (PQQ) biosynthesis were identified from the genome of strain 11-D3, which have been proven to be related to its capacity to solubilize inorganic phosphate (12). According to previous studies, the *pqq* operon is a key regulation factor for the expression of gluconic acid and 2-keto-D-gluconic acid, which are important for high-efficiency inorganic P solubilization in the genus *Pseudomonas* (13–15). Two alkaline phosphatase genes (one belongs to phosphodiester superfamily and the other is *phoD*) were also found. Three phosphatase genes, *phoA*, *phoX*, and *phoD*, have been reported to be widely distributed in diverse habitats (16), and *phoD* is commonly considered a gene marker for bacteria that mineralize soil P (17, 18). The findings reported here may indicate the high efficiency of strain 11-D3 for inorganic P solubilization.

Data availability. This whole-genome shotgun project was deposited at DDBJ/ENA/GenBank under the accession number [PUIN00000000](https://pub.ncbi.nlm.nih.gov/record/PUIN00000000). The version described in this paper is version PUIN01000000. The raw sequences were deposited in the Sequence Read Archive ([SRX3744610](https://www.ncbi.nlm.nih.gov/sra/SRX3744610)).

ACKNOWLEDGMENTS

This study was financially supported by the Strategic Priority Research Program of the Chinese Academy of Sciences (XDB15020402), the Natural Science Foundation of China (41771285, 41430858, and 41701299), and the National Key Research and Development Program of China (2017YFD0200201).

We acknowledge Jian-Qiang Su (Institute of Urban Environment, Chinese Academy of Sciences) and Yu Yan (College of Chemical Engineering, Huaqiao University) for their helpful suggestions.

REFERENCES

- Palleroni NJ. 1993. *Pseudomonas* classification. A new case history in the taxonomy of Gram-negative bacteria. *Antonie Van Leeuwenhoek* 64: 231–251.
- Nielsen MN, Sørensen J, Fels J, Pedersen HC. 1998. Secondary metabolite- and endochitinase-dependent antagonism toward plant-pathogenic microfungi of *Pseudomonas fluorescens* isolates from sugar beet rhizosphere. *Appl Environ Microbiol* 64:3563–3569.
- Andersen SM, Johnsen K, Sørensen J, Nielsen P, Jacobsen CS. 2000. *Pseudomonas frederiksbergensis* sp. nov., isolated from soil at a coal gasification site. *Int J Syst Evol Microbiol* 50:1957–1964. <https://doi.org/10.1099/00207713-50-6-1957>.
- Adam W, Heckel F, Saha-Möller CR, Taupp M, Schreier P. 2004. A highly enantioselective biocatalytic sulfoxidation by the topsoil bacterium *Pseudomonas frederiksbergensis*. *Tetrahedron Asymmetry* 15:983–985. <https://doi.org/10.1016/j.tetasy.2003.12.030>.
- Zheng B-X, Ibrahim M, Zhang D-P, Bi Q-F, Li H-Z, Zhou G-W, Ding K, Peñuelas J, Zhu Y-G, Yang X-R. 2018. Identification and characterization of inorganic-phosphate solubilizing bacteria from agricultural fields with a rapid isolation method. *AMB Express* 8:47. <https://doi.org/10.1186/s13568-018-0575-6>.
- Zeng QW, Wu XQ, Wen XY. 2016. Effects of soluble phosphate on phosphate-solubilizing characteristics and expression of *gcd* gene in *Pseudomonas frederiksbergensis* strain JW-SD2. *Curr Microbiol* 72: 198–206. <https://doi.org/10.1007/s00284-015-0938-z>.
- Galkiewicz JP, Kellogg CA. 2008. Cross-kingdom amplification using bacteria-specific primers: complications for studies of coral microbial ecology. *Appl Environ Microbiol* 74:7828–7831. <https://doi.org/10.1128/AEM.01303-08>.
- Yoon S-H, Ha S-M, Kwon S, Lim J, Kim Y, Seo H, Chun J. 2017. Introducing EzBioCloud: a taxonomically united database of 16S rRNA gene sequences and whole-genome assemblies. *Int J Syst Evol Microbiol* 67: 1613–1617. <https://doi.org/10.1099/ijsem.0.001755>.
- Nautiyal CS. 1999. An efficient microbiological growth medium for screening phosphate solubilizing microorganisms. *FEMS Microbiol Lett* 170: 265–270. [https://doi.org/10.1016/S0378-1097\(98\)00555-2](https://doi.org/10.1016/S0378-1097(98)00555-2).
- Luo R, Liu B, Xie Y, Li Z, Huang W, Yuan J, He G, Chen Y, Pan Q, Liu Y. 2012. SOAPdenovo2: an empirically improved memory-efficient short-read de novo assembler. *Gigascience* 1:18. <https://doi.org/10.1186/2047-217X-1-18>.
- Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Nawrocki EP, Zaslavsky L, Lomsadze A, Pruitt KD, Borodovsky M, Ostell J. 2016. NCBI Prokaryotic Genome Annotation Pipeline. *Nucleic Acids Res* 44: 6614–6624. <https://doi.org/10.1093/nar/gkw569>.
- Zheng B-X, Ding K, Yang X-R, Wadaan MAM, Hozzein WN, Peñuelas J, Zhu Y-G. 2019. Straw biochar increases the abundance of inorganic phosphate solubilizing bacterial community for better rape (*Brassica napus*) growth and phosphate uptake. *Sci Total Environ* 647:1113–1120. <https://doi.org/10.1016/j.scitotenv.2018.07.454>.
- Grissa I, Vergnaud G, Pourcel C. 2007. The CRISPRdb database and tools to display CRISPRs and to generate dictionaries of spacers and repeats. *BMC Bioinformatics* 8:172. <https://doi.org/10.1186/1471-2105-8-172>.
- Hwangbo H, Park RD, Kim YW, Rim YS, Park KH, Kim TH, Suh JS, Kim KY. 2003. 2-Ketogluconic acid production and phosphate solubilization by *Enterobacter intermedium*. *Curr Microbiol* 47:87–0092. <https://doi.org/10.1007/s00284-002-3951-y>.
- Rodríguez H, Fraga R, Gonzalez T, Bashan Y. 2006. Genetics of phosphate solubilization and its potential applications for improving plant growth-promoting bacteria. *Plant Soil* 287:15–21. <https://doi.org/10.1007/s11104-006-9056-9>.
- Kageyama H, Tripathi K, Rai AK, Cha-Um S, Waditee-Sirisattha R, Takabe T. 2011. An alkaline phosphatase/phosphodiesterase, PhoD, induced by salt stress and secreted out of the cells of *Aphanethece halophytica*, a halotolerant cyanobacterium. *Appl Environ Microbiol* 77:5178–5183. <https://doi.org/10.1128/AEM.00667-11>.
- Fraser T, Lynch DH, Entz MH, Dunfield KE. 2015. Linking alkaline phosphatase activity with bacterial *phoD* gene abundance in soil from a long-term management trial. *Geoderma* 257:115–122. <https://doi.org/10.1016/j.geoderma.2014.10.016>.
- Ragot SA, Kertesz MA, Bünemann EK. 2015. *phoD* alkaline phosphatase gene diversity in soil. *Appl Environ Microbiol* 81:7281–7289. <https://doi.org/10.1128/AEM.01823-15>.