

Draft Genome Sequence of the Bacterium Delftia acidovorans Strain D4B, Isolated from Soil

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ABSTRACT Delftia acidovorans strain D4B is an aerobic bacterium within the Betaproteobacteria lineage that was isolated from soil. The genome size is 6.26 Mbp, with a $G+C$ content of 67%. The genome encodes enzymes potentially involved in the degradation of fluorinated compounds.

There, we describe the draft genome sequence of *Delftia acidovorans* strain D4B,
which was isolated from soil contaminated with perfluorinated compounds (PFCs). Delftia acidovorans strains are environmental bacteria with diverse metabolic pathways that make them strong candidates for bioremediation ([1\)](#page-1-0). D. acidovorans strain D4B grows aerobically in the presence of either 1 ppm or 100 ppm of the perfluoroalkyl substance (PFAS) perfluorooctanoic acid (PFOA) in Raymond's medium ([2](#page-1-1)) supplemented with 1,000 ppm glucose at 30°C with shaking. The sequenced genome of D. acidovorans strain D4B provides the first genome for an isolate of this genus, which will further our understanding of how this organism could be leveraged for bioremediation.

D4B was isolated from a Winogradsky column with soil from a PFC-contaminated site. The soil was incubated with paper, hard-boiled egg yolk, and lake water artificially contaminated with either perfluorooctane sulfonate (PFOS) (100 ppm or 666 ppm) or PFOA (100 ppm or 750 ppm). Columns were grown for 1 month, and the top of the culture was swabbed. Unique colonies were restreaked on 100 ppm PFOA-agar plates, and D4B was isolated as a single colony. D4B was grown in tryptic soy broth at 32°C for 18 h with shaking. The culture was diluted, grown for \sim 8 h under identical conditions, and harvested in the mid-logarithmic growth phase. Genomic DNA was extracted from the harvested cell pellet using the Qiagen DNeasy UltraClean microbial silica spin column kit. The library was made with 155 ng of DNA using Illumina DNA Prep, (M) Tagmentation (24 sample, product number 20018704; Illumina) and IDT for Illumina DNA UD indexes. The library was sequenced using the NovaSeq 6000 SP reagent kit v1.5 (200 cycles) (product number 20040719; Illumina) on an Illumina NovaSeq 6000 system run using 101-bp paired-end (PE) reads.

Default parameters were used for all software unless otherwise specified. Sequencing resulted in 4,482,539 raw PE reads, which were trimmed and quality-filtered with Trimmomatic v0.39 ([3\)](#page-1-2). The 4,472,394 (894,478 total) trimmed, quality-filtered PE and unpaired reads (average read length, 101 bp) were assembled using SPAdes 3.13.1 [\(4\)](#page-1-3) in careful mode, resulting in \sim 140 \times coverage and 52 contigs $(\geq 200$ bp), with a G+C content of 67%, N₅₀ value of 334,654 bp, L₅₀ value of 6, and genome size of 6,259,045 Mb, as calculated by QUAST v5.0.2 ([5](#page-1-4)). D4B genome completeness was estimated to be 99.85%, with $<1\%$ contamination, using CheckM v1.1.3 ([6\)](#page-1-5). Citation Harris J, Gross M, Kemball J, Farajollahi S, Dennis P, Sitko J, Steel JJ, Almand E, Kelley-Loughnane N, Varaljay VA. 2021. Draft genome sequence of the bacterium Delftia acidovorans strain D4B, isolated from soil. Microbiol Resour Announc 10:e00635-21. [https://doi.org/10](https://doi.org/10.1128/MRA.00635-21) [.1128/MRA.00635-21.](https://doi.org/10.1128/MRA.00635-21)

Editor David Rasko, University of Maryland School of Medicine

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Received 22 June 2021 Accepted 12 October 2021 Published 4 November 2021 Whole-genome, de novo gene prediction and annotation were performed with Prokka v1.12 ([7](#page-1-6)), which resulted in 5,484 putative genes of $>$ 50 amino acids. RNAmmer v1.2 ([8](#page-1-7)) was used to extract the full-length 16S rRNA gene, which was 100% identical to other Delftia acidovorans sequences using BLASTN against the NCBI nonredundant database.

Based on the Prokka annotations, BLASTP searches with two Delftia acidovorans haloacid dehalogenases from the NCBI database (GenBank accession numbers [WP](https://www.ncbi.nlm.nih.gov/protein/WP_011137954.1) [_011137954.1](https://www.ncbi.nlm.nih.gov/protein/WP_011137954.1) and [PZP66635.1](https://www.ncbi.nlm.nih.gov/protein/PZP66635.1)), and Phyre2 homology-based structural predictions ([9](#page-1-8)) with 100% confidence scores, we identified two putative haloacid dehalogenases and a fluoroacetate dehalogenase, which could be involved in the degradation of fluorinated compounds. This sequenced genome should provide critical insights into the metabolism and physiology leading to degradation and remediation of fluorinated compounds by this ubiquitous soil bacterium.

Data availability. This whole-genome shotgun project has been deposited in DDBJ/ ENA/GenBank under the accession number [JAHLGQ000000000.](https://www.ncbi.nlm.nih.gov/nuccore/JAHLGQ000000000) The version described in this paper is version [JAHLGQ000000000.2.](https://www.ncbi.nlm.nih.gov/nuccore/JAHLGQ000000000.2) The raw reads have been deposited in the Sequence Read Archive (SRA) database under SRA accession number [SRP323477](https://trace.ncbi.nlm.nih.gov/Traces/sra/?run=SRR14773506) and BioProject accession number [PRJNA736486](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA736486).

ACKNOWLEDGMENTS

The research reported in this publication has been cleared for public release under reference numbers USAFA-DF-2021-CSURF-63, AFRL-2021-1725, and AFRL-2021-1718.

We acknowledge the University of Kansas Medical Center Genomics Core (University of Kansas Medical Center, Kansas City, KS) for core support services in generating the genome sequencing data under University of Kansas Medical Center NIH supporting grants, i.e., Kansas Intellectual and Developmental Disabilities Research Center (grant NIH U54 HD 090216), Molecular Regulation of Cell Development and Differentiation, COBRE (grant P30 GM122731- 03), NIH S10 high-end instrumentation grant (grant S10OD021743), and Frontiers CTSA grant (grant UL1TR002366). This work was supported by funding from the Office of the Under-Secretary for Research and Engineering ARAP Program and the Defense Health Agency.

We thank Nereyda Sevilla (Defense Health Agency Clinical Investigations Program), Don Veverka (Life Sciences Research Center at the U.S. Air Force Academy), and Colonel Steven Hasstedt (Department of Biology, U.S. Air Force Academy).

REFERENCES

- 1. Braña V, Cagide C, Morel MA. 2016. The sustainable use of Delftia in agriculture, bioremediation, and bioproducts synthesis, p 227–247. In Castro-Sowinski S (ed), Microbial models: from environmental to industrial sustainability, vol 1. Springer, Singapore. [https://doi.org/10.1007/978](https://doi.org/10.1007/978-981-10-2555-6_11) [-981-10-2555-6_11.](https://doi.org/10.1007/978-981-10-2555-6_11)
- 2. Chetverikov SP, Sharipov DA, Korshunova TY, Loginov ON. 2017. Degradation of perfluorooctanyl sulfonate by strain Pseudomonas plecoglossicida 2.4-D. Appl Biochem Microbiol 53:533–538. [https://doi.org/10.1134/](https://doi.org/10.1134/S0003683817050027) [S0003683817050027.](https://doi.org/10.1134/S0003683817050027)
- 3. Bolger AM, Lohse M, Usadel B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics 30:2114–2120. [https://doi.org/10](https://doi.org/10.1093/bioinformatics/btu170) [.1093/bioinformatics/btu170.](https://doi.org/10.1093/bioinformatics/btu170)
- 4. Prjibelski A, Antipov D, Meleshko D, Lapidus A, Korobeynikov A. 2020. Using SPAdes de novo assembler. Curr Protoc Bioinformatics 70:e102. <https://doi.org/10.1002/cpbi.102>.
- 5. Mikheenko A, Prjibelski A, Saveliev V, Antipov D, Gurevich A. 2018. Versatile genome assembly evaluation with QUAST-LG. Bioinformatics 34: i142–i150. [https://doi.org/10.1093/bioinformatics/bty266.](https://doi.org/10.1093/bioinformatics/bty266)
- 6. Parks DH, Imelfort M, Skennerton CT, Hugenholtz P, Tyson GW. 2015. CheckM: assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes. Genome Res 25:1043–1055. [https://](https://doi.org/10.1101/gr.186072.114) [doi.org/10.1101/gr.186072.114.](https://doi.org/10.1101/gr.186072.114)
- 7. Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. Bioinformatics 30:2068–2069. [https://doi.org/10.1093/bioinformatics/btu153.](https://doi.org/10.1093/bioinformatics/btu153)
- 8. Lagesen K, Hallin P, Rødland EA, Staerfeldt H-H, Rognes T, Ussery DW. 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. Nucleic Acids Res 35:3100–3108. <https://doi.org/10.1093/nar/gkm160>.
- 9. Kelley LA, Mezulis S, Yates CM, Wass MN, Sternberg MJE. 2015. The Phyre2 web portal for protein modeling, prediction and analysis. Nat Protoc 10: 845–858. [https://doi.org/10.1038/nprot.2015.053.](https://doi.org/10.1038/nprot.2015.053)