

Complete Genome Sequence of Desulfovibrio desulfuricans IC1, a Sulfonate-Respiring Anaerobe

Leslie A. Day,^a Kara B. De León,^a Megan L. Kempher,^b Jizhong Zhou,^b Judy D. Wall^a

aDepartment of Biochemistry, University of Missouri, Columbia, Missouri, USA bInstitute for Environmental Genomics, Department of Microbiology and Plant Biology, University of Oklahoma, Norman, Oklahoma, USA

ABSTRACT We report the complete genome sequence of the anaerobic, sulfonaterespiring, sulfate-reducing bacterium Desulfovibrio desulfuricans IC1. The genome was assembled into a single 3.25-Mb circular chromosome with 2,680 protein-coding genes identified. Sequencing of sulfonate-metabolizing anaerobes is key for understanding sulfonate degradation and its role in the sulfur cycle.

The anaerobic sulfate-reducing bacterium (SRB) Desulfovibrio desulfuricans IC1 has
been shown to utilize selected sulfonates, compounds that contain an R-C-SO₃moiety in respiration [\(1\)](#page-1-0). Sulfonates are ubiquitous in nature, are synthesized commercially (e.g., detergents), and may be serving as key metabolites for SRB, in sulfate-limited environments.

The genes involved in sulfonate degradation by aerobic bacteria have been extensively investigated [\(2\)](#page-1-1). However, exploration of the genes and biochemistry of sulfonate respiration by anaerobes is incomplete. Therefore, the sequencing and analysis of sulfonate-utilizing anaerobes may reveal novel metabolic pathways for the metabolism of organosulfur compounds.

D. desulfuricans IC1 was isolated with the sulfonate isethionate as the terminal electron acceptor from freshwater marsh mud [\(1\)](#page-1-0). We acquired D. desulfuricans IC1 from Leibniz Institute DSMZ (DSM no. 12129). For sequencing, D. desulfuricans IC1 was grown at 30°C in MOYLS4 medium [\(3\)](#page-1-2). The genomic DNA (gDNA) was extracted with a Wizard genomic DNA purification kit (Promega Corp., Madison, WI, USA) per the manufacturer's guidelines for Gram-negative bacteria, and the DNA was resuspended in Tris-HCl (10 mM, pH 8.5). The DNA was quantified with a Qubit fluorometer and Qubit doublestranded DNA (dsDNA) broad-range (BR) assay kit (Thermo Fisher Scientific, Waltham, MA, USA).

Sample preparation and MiSeq (Illumina, San Diego, CA, USA) sequencing of D. desulfuricans IC1 was performed by the University of Oklahoma Institute for Environmental Genomics as follows: 1 μ g of gDNA was fragmented with a Covaris M220 focused ultrasonicator, and the library was prepared with the Kapa HyperPrep kit (Kapa Biosystems) following the manufacturer's protocol. Paired-end sequencing was performed with the MiSeq v2 500-cycle reagent kit (Illumina). All software used in this study was operated with default parameters. ϕ X174 (GenBank accession no. [NC_001422.1\)](https://www.ncbi.nlm.nih.gov/nuccore/NC_001422.1) reads and adaptor sequences were removed with Bowtie 2 [\(4\)](#page-1-3) and Cutadapt (v1.18) [\(5\)](#page-1-4), respectively, resulting in 4,294,900 paired-end reads with an average length of 163 nucleotides.

A 20-kb SMRTbell long-insert size-selected library was prepared and analyzed on the PacBio (Menlo Park, CA, USA) RS II platform with single-molecule real-time (SMRT) sequencing technology at the University of Maryland Institute for Genome Sciences. Sequencing produced 156,863 raw reads with an average length of 8,203 nucleotides. Reads were trimmed, corrected, and assembled with Canu (v1.7) into one circular **Citation** Day LA, De León KB, Kempher ML, Zhou J, Wall JD. 2019. Complete genome sequence of Desulfovibrio desulfuricans IC1, a sulfonate-respiring anaerobe. Microbiol Resour Announc 8:e00456-19. [https://doi.org/10.1128/](https://doi.org/10.1128/MRA.00456-19) [MRA.00456-19.](https://doi.org/10.1128/MRA.00456-19)

Editor J. Cameron Thrash, University of Southern California

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

Address correspondence to Judy D. Wall, [wallj@missouri.edu.](mailto:wallj@missouri.edu)

Received 1 May 2019 **Accepted** 11 July 2019 **Published** 1 August 2019 contig with an average coverage of 39.1 [\(6\)](#page-1-5). Trimmed Illumina reads were used to correct insertions and deletions in the PacBio assembly with Pilon (v1.21) [\(7\)](#page-1-6).

Illumina sequencing reads were mapped to the PacBio assembly in Geneious (v9.0.5; Biomatters, Ltd., Auckland, New Zealand), with an average coverage of 180.2. The chromosome starting position was designated as 150 bases upstream of dnaA. One single-nucleotide polymorphism (SNP) was identified in the population with the Geneious find variations/SNPs function. This SNP is at position 1638748 in the gene for a TetR/AcrR family transcriptional regulator (DDIC_06845) with a 50/50 occurrence of G/T. The BLAST function in Geneious revealed a gene encoding a putative glycyl radical enzyme (DDIC_12990) in D. desulfuricans IC1 which has 77% amino acid identity to the Bilophila wadsworthia protein (GenBank accession no. [EFV45544.1\)](https://www.ncbi.nlm.nih.gov/protein/EFV45544.1) identified in sulfonate respiration [\(8\)](#page-1-7).

The NCBI Prokaryotic Genome Annotation Pipeline identified 2,680 protein-coding genes, 1 clustered regularly interspaced short palindromic repeat (CRISPR) array, 3 rRNA operons, 54 tRNA, and 42 pseudo genes [\(9\)](#page-1-8). The $G+C$ content is 59.1% with a total length of 3,251,440 nucleotides.

Data availability. This whole-genome shotgun project has been deposited in DDBJ/ ENA/GenBank under accession no. [CP036295.](https://www.ncbi.nlm.nih.gov/nuccore/CP036295) This is the first version (CP036295.1). Raw PacBio sequences were deposited in the NCBI SRA database under accession no. [SRR8582205.](https://www.ncbi.nlm.nih.gov/sra/SRR8582205) Raw Illumina sequences were deposited under SRA accession no. [SRR7655955.](https://www.ncbi.nlm.nih.gov/sra/SRR7655955)

ACKNOWLEDGMENTS

We thank Thomas Lie for fruitful discussions.

This material by ENIGMA (Ecosystems and Networks Integrated with Genes and Molecular Assemblies; [http://enigma.lbl.gov\)](http://enigma.lbl.gov), a Scientific Focus Area Program at Lawrence Berkeley National Laboratory, is based upon work supported by the U.S. Department of Energy, Office of Science, Office of Biological and Environmental Research, under contract no. DE-AC02-05CH11231.

REFERENCES

- 1. Lie TJ, Pitta T, Leadbetter ER, Godchaux W, III, Leadbetter JR. 1996. Sulfonates: novel electron acceptors in anaerobic respiration. Arch Microbiol 166:204 –210. [https://doi.org/10.1007/s002030050376.](https://doi.org/10.1007/s002030050376)
- 2. Weinitschke S, Sharma PI, Stingl U, Cook AM, Smits THM. 2010. Gene clusters involved in isethionate degradation by terrestrial and marine bacteria. Appl Environ Microbiol 76:618 – 621. [https://doi.org/10.1128/](https://doi.org/10.1128/AEM.01818-09) [AEM.01818-09.](https://doi.org/10.1128/AEM.01818-09)
- 3. De León KB, Zane GM, Trotter VV, Krantz GP, Arkin AP, Butland GP, Walian PJ, Fields MW, Wall JD. 2017. Unintended laboratory-driven evolution reveals genetic requirements for biofilm formation by Desulfovibrio vulgaris Hildenborough. mBio 8:e01696-17. [https://doi.org/10.1128/mBio](https://doi.org/10.1128/mBio.01696-17) [.01696-17.](https://doi.org/10.1128/mBio.01696-17)
- 4. Langmead B, Salzberg SL. 2012. Fast gapped-read alignment with Bowtie 2. Nat Methods 9:357–359. [https://doi.org/10.1038/nmeth.1923.](https://doi.org/10.1038/nmeth.1923)
- 5. Martin M. 2011. Cutadapt removes adapter sequences from highthroughput sequencing reads. EMBnet J 17:10 –12. [https://doi.org/10](https://doi.org/10.14806/ej.17.1.200) [.14806/ej.17.1.200.](https://doi.org/10.14806/ej.17.1.200)
- 6. Koren S, Walenz BP, Berlin K, Miller JR, Bergman NH, Phillippy AM. 2017. Canu: scalable and accurate long-read assembly via adaptive k-mer weighting and repeat separation. Genome Res 27:722–736. [https://](https://doi.org/10.1101/gr.215087.116) [doi.org/10.1101/gr.215087.116.](https://doi.org/10.1101/gr.215087.116)
- 7. Walker BJ, Abeel T, Shea T, Priest M, Abouelliel A, Sakthikumar S, Cuomo CA, Zeng Q, Wortman J, Young SK, Earl AM. 2014. Pilon: an integrated tool for comprehensive microbial variant detection and genome assembly improvement. PLoS One 9:e112963. [https://doi.org/10.1371/journal.pone](https://doi.org/10.1371/journal.pone.0112963) [.0112963.](https://doi.org/10.1371/journal.pone.0112963)
- 8. Peck SC, Denger K, Burrichter A, Irwin SM, Balskus EP, Schleheck D. 2019. A glycyl radical enzyme enables hydrogen sulfide production by the human intestinal bacterium Bilophila wadsworthia. Proc Natl Acad Sci U S A 116:3171–3176. [https://doi.org/10.1073/pnas.1815661116.](https://doi.org/10.1073/pnas.1815661116)
- 9. 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](https://doi.org/10.1093/nar/gkw569) [.org/10.1093/nar/gkw569.](https://doi.org/10.1093/nar/gkw569)