genomeA
Journals.ASM.org

Draft Genome Sequences for Three Mercury-Methylating, Sulfate-Reducing Bacteria

Steven D. Brown,[a] Richard A. Hurt, Jr.,[a] Cynthia C. Gilmour,[b] Dwayne A. Elias[a]

Biosciences Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee, USA[a]; Smithsonian Environmental Research Center, Edgewater, Maryland, USA[b]

The genetic basis for bacterial mercury methylation has been described recently. For insights into the physiology of mercury-methylating bacteria, we present genome sequences for *Desulfococcus multivorans* strain DSM 2059, *Desulfovibrio alkalitolerans* strain DSM 16529, and *Desulfovibrio* species strain X2.

Received 12 July 2013 **Accepted** 22 July 2013 **Published** 15 August 2013

Citation Brown SD, Hurt RA, Jr, Gilmour CC, Elias DA. 2013. Draft genome sequences for three mercury-methylating, sulfate-reducing bacteria. Genome Announc. 1(4):e00618-13. doi:10.1128/genomeA.00618-13.

Address correspondence to Dwayne A. Elias, eliasda@ornl.gov.

Select members of the sulfate-reducing bacteria (SRB) and Fe(III)-reducing bacteria (IRB) can methylate inorganic mercury [Hg(II)] to methylmercury (MeHg), a more toxic form that bioaccumulates (1). For the first time, recent genetic studies have linked a specific gene cluster (*hgcAB*) to mercury methylation in the model SRB *Desulfovibrio desulfuricans* strain ND132 and IRB *Geobacter sulfureducens* strain PCA (2). The *hgcAB* gene cluster encodes a putative corrinoid-containing CO dehydrogenase/acetyl-coenzyme A (CoA) synthase, HgcA, and a 2[4Fe-4S] ferredoxin, HgcB, and these are predicted to have roles as a methyl carrier and an electron donor, respectively.

The potential involvement of the acetyl-CoA pathway in bacterial Hg methylation was recognized more than 20 years ago (3), although the physiology and MeHg formation in incomplete-oxidizing SRB (conversion of carbon substrates to acetate) and complete-oxidizing SRB (conversion of carbon to CO_2) remain to be fully elucidated (4, 5). The majority of known Hg-methylating *Deltaproteobacteria* are incomplete oxidizers (6). This means that when they reduce sulfate, these organisms incompletely oxidize short-chain fatty acids to acetate and do not utilize C-1 substrates, and thus they do not use the acetyl-CoA pathway as part of their primary substrate utilization machinery during heterotrophic growth (7). Detailed assessments and comparisons of mercury methylation rates for several *Desulfovibrio* species that are incomplete oxidizers, including those investigated in this study, have been reported (6, 8). Genome sequences for several SRBs known or predicted to be mercury methylators have been reported (2, 9–12).

Desulfovibrio multivorans is an example of a complete oxidizer with an acetyl-CoA pathway, and mercury methylation rates have been reported for several strains (4, 5). *D. alkalitolerans* strain DSM 16529 is an example of a complete oxidizer that can generate MeHg (8). *Desulfovibrio* species strain X2 was isolated from soft black mid-Chesapeake Bay bottom sediments sampled in May 1985 (13), and it methylates both Sn(IV) (14) and Hg(II) (6), but its detailed physiology has not been fully assessed.

In this study, we generated draft genomes for *D. multivorans* DSM 2059, *D. alkalitolerans* DSM 16529, and *Desulfovibrio* species strain X2. Sequence data for each genome were generated using an Illumina MiSeq instrument (15) and a paired-end approach with an approximate insert library size of 500 bp and read lengths of 250 bp, according to the manufacturer's instructions. The CLC Genomics Workbench (version 6.0.2) was used to trim and filter reads for quality sequence data and subsequent assemblies. The resulting assemblies generated 149, 32, and 66 DNA contigs that represented the draft genome sequences for strains DSM 2059, DSM 16529, and X2, respectively. The estimated genome sizes were ~4.4, 3.2, and 3.9 Mb and G+C DNA contents were 56.8%, 64.4%, and 67.9%, and there were 3,838, 2,924, and 3,445 putative coding sequences (CDS) for DSM 2059, DSM 16529, and X2, respectively. Sequence depth coverage across the genomes was ~205 to 309 times the genome sizes. Draft genome sequences were annotated as previously described (16).

The draft genome sequences presented here will facilitate comparative studies and assist with investigations into HgcAB native functions, related pathways, and assessments of MeHg potential in different ecological niches.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession numbers ATHJ00000000, ATHI00000000, and ATHV00000000 for *D. multivorans* DSM 2059, *D. alkalitolerans* DSM 16529, and *Desulfovibrio* species strain X2, respectively. The versions described in this paper are the first versions.

ACKNOWLEDGMENTS

We acknowledge Miriam Land (ORNL) and Sagar Utturkar (University of Tennessee) for assistance with annotation and deposition of data into the NCBI GenBank and Sequence Read Archive databases.

This research was supported by the Office of Biological and Environmental Research (OBER), Office of Science, U.S. Department of Energy (DOE), as part of the Mercury Science Focus Area Program at Oak Ridge National Laboratory. Oak Ridge National Laboratory is managed by UT-Battelle, LLC, for the U.S. Department of Energy under contract DE-AC05-00OR22725.

REFERENCES

1. **Hsu-Kim H, Kucharzyk KH, Zhang T, Deshusses MA.** 2013. Mechanisms regulating mercury bioavailability for methylating microorganisms in the aquatic environment: a critical review. Environ. Sci. Technol. **47:** 2441–2456.
2. **Parks JM, Johs A, Podar M, Bridou R, Hurt RA, Smith SD, Tomanicek SJ, Qian Y, Brown SD, Brandt CC, Palumbo AV, Smith JC, Wall JD, Elias DA, Liang L.** 2013. The genetic basis for bacterial mercury methylation. Science **339:**1332–1335.
3. **Choi SC, Bartha R.** 1993. Cobalamin-mediated mercury methylation by *Desulfovibrio desulfuricans* LS. Appl. Environ. Microbiol. **59:**290–295.
4. **Ekstrom EB, Morel M, FM, Benoit JM.** 2003. Mercury methylation independent of the acetyl-coenzyme a pathway in sulfate-reducing bacteria. Appl. Environ. Microbiol. 69:5414–5422.
5. **King JK, Kostka JE, Frischer ME, Saunders FM.** 2000. Sulfate-reducing bacteria methylate mercury at variable rates in pure culture and in marine sediments. Appl. Environ. Microbiol. **66:**2430–2437.
6. **Gilmour CC, Elias DA, Kucken AM, Brown SD, Palumbo AV, Schadt CW, Wall JD.** 2011. Sulfate-reducing bacterium *Desulfovibrio desulfuricans* ND132 as a model for understanding bacterial mercury methylation. Appl. Environ. Microbiol. **77:**3938–3951.
7. **Keller KL, Wall JD.** 2011. Genetics and molecular biology of the electron flow for sulfate respiration in *Desulfovibrio*. Front. Microbiol. **2:**135.
8. **Graham AM, Bullock AL, Maizel AC, Elias DA, Gilmour CC.** 2012. Detailed assessment of the kinetics of Hg-Cell Association, Hg methylation, and methylmercury degradation in several Desulfovibrio species. Appl. Environ. Microbiol. **78:**7337–7346.
9. **Brown SD, Gilmour CC, Kucken AM, Wall JD, Elias DA, Brandt CC, Podar M, Chertkov O, Held B, Bruce DC, Detter JC, Tapia R, Han CS, Goodwin LA, Cheng JF, Pitluck S, Woyke T, Mikhailova N, Ivanova NN, Han J, Lucas S, Lapidus AL, Land ML, Hauser LJ, Palumbo AV.** 2011. Genome sequence of the mercury-methylating strain *Desulfovibrio desulfuricans* ND132. J. Bacteriol. **193:**2078–2079.
10. **Brown SD, Wall JD, Kucken AM, Gilmour CC, Podar M, Brandt CC, Teshima H, Detter JC, Han CS, Land ML, Lucas S, Han J, Pennacchio L, Nolan M, Pitluck S, Woyke T, Goodwin L, Palumbo AV, Elias DA.** 2011. Genome sequence of the mercury-methylating and pleomorphic *Desulfovibrio africanus* strain Walvis Bay. J. Bacteriol. **193:**4037–4038.
11. **Pagani I, Lapidus A, Nolan M, Lucas S, Hammon N, Deshpande S, Cheng JF, Chertkov O, Davenport K, Tapia R, Han C, Goodwin L, Pitluck S, Liolios K, Mavromatis K, Ivanova N, Mikhailova N, Pati A, Chen A, Palaniappan K, Land M, Hauser L, Chang YJ, Jeffries CD, Detter JC, Brambilla E, Kannan KP, Djao ODN, Rohde M, Pukall R, Spring S, Goker M, Sikorski J, Woyke T, Bristow J, Eisen JA, Markowitz V, Hugenholtz P, Kyrpides NC, Klenk HP.** 2011. Complete genome sequence of *Desulfobulbus propionicus* type strain (1pr3). Stand. Genomic Sci. **4:**100–110.
12. **Brown SD, Utturkar SM, Arkin AP, Deutschbauer AM, Elias DA, Hazen TC, Chakraborty R.** 2013. Draft genome sequence for *Desulfovibrio africanus* strain PCS. Genome Announc. **1**(2):e00144-13. doi:10.1128/genomeA.00144-13.
13. **Gilmour CC, Tuttle JH, Means JC.** 1987. Anaerobic microbial methylation of inorganic tin in estuarine sediment slurries. Microb. Ecol. **14:** 233–242.
14. **Gilmour CC, Tuttle JH, Means JC.** 1985. Tin methylation in sulfide bearing sediments, p 239–258. In Sigleo AC, Hattori A (ed), Marine and estuarine geochemistry. Lewis Publishers, Ann Arbor, MI.
15. **Quail MA, Smith M, Coupland P, Otto TD, Harris SR, Connor TR, Bertoni A, Swerdlow HP, Gu Y.** 2012. A tale of three next generation sequencing platforms: comparison of Ion Torrent, Pacific Biosciences and Illumina MiSeq sequencers. BMC Genomics **13:**341. doi:10.1186/1471-2164-13-341.
16. **Brown SD, Klingeman DM, Lu TY, Johnson CM, Utturkar SM, Land ML, Schadt CW, Doktycz MJ, Pelletier DA.** 2012. Draft genome sequence of *Rhizobium* sp. strain PDO1-076, a bacterium isolated from *Populus deltoides*. J. Bacteriol. **194:**2383–2384.