GENOME SEQUENCES





Complete Genome Sequence of a Mesophilic Obligately Chemolithoautotrophic Hydrogen-Oxidizing Bacterium, *Hydrogenovibrio marinus* MH-110

Hiroyuki Arai,^{a,b} Masaharu Ishii^{a,b}

^aDepartment of Biotechnology, Graduate School of Agricultural and Life Sciences, The University of Tokyo, Bunkyo, Tokyo, Japan ^bCollaborative Research Institute for Innovative Microbiology, The University of Tokyo, Bunkyo, Tokyo, Japan

ABSTRACT *Hydrogenovibrio marinus* is a mesophilic, obligately chemolithoautotrophic, and hydrogen-oxidizing bacterium that uses three different RubisCOs at different carbon dioxide tensions. Here, we report its complete genome sequence, which is 2,491,293 bp long, with an average GC content of 44.1%.

H^{vdrogenovibrio} marinus MH-110 (JCM7688 [https://www.jcm.riken.jp/cgi -bin/jcm_jcm_number?JCM=7688], DSM11271 [https://www.dsmz.de/collection/ catalogue/details/culture/DSM-11271]) is an aerobic, mesophilic, obligately chemolithoautotrophic, and hydrogen-oxidizing bacterium isolated from seawater of the Shonan Coast, Japan (1). It was isolated with hydrogen as an energy source. The bacterium fixes carbon dioxide using three different RubisCOs encoded by different operons (2–5). Carboxysome is produced at low carbon dioxide tensions. Strain MH-110 has an oxygen-tolerant [NiFe]-hydrogenase and accumulates glycogen under nitrogen or magnesium starvation or oxygen limitation (6–8). Two groups have independently reported a draft genome sequence of this strain (DSM11271 [https://www.dsmz.de/ collection/catalogue/details/culture/DSM-11271]). However, the complete genome had not been determined (9, 10). We determined the complete genome sequence in order to facilitate further studies on the application of this bacterium to produce useful materials from carbon dioxide.

The whole genome of strain MH-110, maintained by our laboratory, was sequenced using a Roche 454 GS FLX instrument. Total DNA was isolated by a standard phenolchloroform method from the cells, cultivated autotrophically in an inorganic medium with the gas phase consisting of H₂, O₂, and CO₂ (75:15:10, vol/vol) (1). We constructed the 8-kb mate pair library following the standard procedure (11). The data processing, quality control, and assembly of contigs and scaffolds were performed using the Roche GS FLX software v.2.8 and GS De Novo Assembler v.2.6. Default software parameters were used. The paired-end sequencing yielded 137,935,170 bases from 294,573 reads, with an average read length of 248 bp. After removing adaptors and low-quality reads, the reads were assembled into 54 contigs of >500 bp. The genome coverage was 55.4-fold, and the average contig length and N_{50} size were 45,539 bp and 139,857 bp, respectively. The Q40 plus bases showed 99.92% reliability. Four scaffolds were generated from 36 contigs using the paired-end information. The average scaffold length and N_{50} size were 622,477 bp and 2,478,578 bp, respectively. Gaps between the contigs or scaffolds were closed by sequencing DNA fragments amplified by PCR from genomic DNA with an ABI 3730xl DNA analyzer (Applied Biosystems). Ex Tag or LA Tag DNA polymerase with GC buffer II (TaKaRa) was used for the PCR amplification. The custom primers used for PCR and Sanger sequencing were designed near the ends of the contigs. The genes were annotated using the DDBJ Fast Annotation and Submission Tool (DFAST) (https://dfast.nig.ac.jp/) with ARAGORN to predict tRNA genes.

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Address correspondence to Hiroyuki Arai, aharai@mail.ecc.u-tokyo.ac.jp.

Received 13 September 2019 Accepted 25 September 2019 Published 17 October 2019 The complete genome of strain MH-110 comprises a circular chromosome of 2,491,293 bp with 44.1% GC content. The genome contains 2,322 predicted proteincoding genes, 3 sets of the rRNA genes, and 43 tRNA genes. The scaffolds of approximately 100 kb, presumably derived from a plasmid, reported in the previous draft genome sequences (9, 10) were not identified in the genome of our laboratory strain. A 133.4-kb region between the rRNA gene clusters was inverted compared with the previous draft genome sequences.

Data availability. The complete genome sequence of *H. marinus* MH-110 has been deposited in DDBJ under accession number AP020335. The raw sequence data have been deposited in the DDBJ Sequence Read Archive under accession number DRA008898.

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REFERENCES

- Nishihara H, Igarashi Y, Kodama T. 1991. *Hydrogenovibrio marinus* gen. nov., sp. nov., a marine obligately chemolithoautotrophic hydrogenoxidizing bacterium. Int J Syst Bacteriol 41:130–133. https://doi.org/10 .1099/00207713-41-1-130.
- Hayashi NR, Oguni A, Yaguchi T, Chung SY, Nishihara H, Kodama T, Igarashi Y. 1998. Different properties of gene products of three sets ribulose 1,5-bisphosphate carboxylase/oxygenase from a marine obligately autotrophic hydrogen-oxidizing bacterium, *Hydrogenovibrio marinus* strain MH-110. J Ferment Bioeng 85:150–155. https://doi.org/10 .1016/S0922-338X(97)86759-1.
- Yoshizawa Y, Toyoda K, Arai H, Ishii M, Igarashi Y. 2004. CO₂-responsive expression and gene organization of three ribulose-1,5-bisphosphate carboxylase/oxygenase enzymes and carboxysomes in *Hydrogenovibrio marinus* strain MH-110. J Bacteriol 186:5685–5691. https://doi.org/10 .1128/JB.186.17.5685-5691.2004.
- Toyoda K, Yoshizawa Y, Arai H, Ishii M, Igarashi Y. 2005. The role of two CbbRs in the transcriptional regulation of three ribulose-1,5-bisphosphate carboxylase/oxygenase genes in *Hydrogenovibrio marinus* strain MH-110. Microbiology 151:3615–3625. https://doi.org/10.1099/mic.0.28056-0.
- Toyoda K, Ishii M, Arai H. 2018. Function of three RuBisCO enzymes under different CO₂ conditions in *Hydrogenovibrio marinus*. J Biosci Bioeng 126:730–735. https://doi.org/10.1016/j.jbiosc.2018.06.005.
- Shomura Y, Yoon K-S, Nishihara H, Higuchi Y. 2011. Structural basis for a [4Fe-3S] cluster in the oxygen-tolerant membrane-bound [NiFe]hydrogenase. Nature 479:253–256. https://doi.org/10.1038/nature10504.
- Nishihara H, Miyata Y, Miyashita Y, Bernhard M, Pohlmann A, Friedrich B, Takamura Y. 2001. Analysis of the molecular species of hydrogenase in the cells of an obligately chemolithoautotrophic, marine hydrogen-

oxidizing bacterium, *Hydrogenovibrio marinus*. Biosci Biotechnol Biochem 65:2780–2784. https://doi.org/10.1271/bbb.65.2780.

- Nishihara H, Igarashi Y, Kodama T, Nakajima T. 1993. Production and properties of glycogen in the marine obligate chemolithoautotroph, *Hydrogenovibrio marinus*. J Ferment Bioeng 75:414–416. https://doi.org/ 10.1016/0922-338X(93)90087-0.
- Jo BH, Hwang BH, Cha HJ. 2014. Draft genome sequence of *Hydrog-enovibrio marinus* MH-110, a model organism for aerobic H₂ metabolism. J Biotechnol 185:37–38. https://doi.org/10.1016/j.jbiotec.2014.06.009.
- 10. Scott KM, Williams J, Porter CMB, Russel S, Harmer TL, Paul JH, Antonen KM, Bridges MK, Camper GJ, Campla CK, Casella LG, Chase E, Conrad JW, Cruz MC, Dunlap DS, Duran L, Fahsbender EM, Goldsmith DB, Keeley RF, Kondoff MR, Kussy BI, Lane MK, Lawler S, Leigh BA, Lewis C, Lostal LM, Marking D, Mancera PA, McClenthan EC, McIntyre EA, Mine JA, Modi S, Moore BD, Morgan WA, Nelson KM, Nguyen KN, Ogburn N, Parrino DG, Pedapudi AD, Pelham RP, Preece AM, Rampersad EA, Richardson JC, Rodgers CM, Schaffer BL, Sheridan NE, Solone MR, Staley ZR, Tabuchi M, Waide RJ, Wanjugi PW, Young S, Clum A, Daum C, Huntemann M, Ivanova N, Kyrpides N, Mikhailova N, Palaniappan K, Pillay M, Reddy TBK, Shapiro N, Stamatis D, Varghese N, Woyke T, Boden R, Freyermuth SK, Kerfeld CA. 2018. Genomes of ubiquitous marine and hypersaline Hydrogenovibrio, Thiomicrorhabdus and Thiomicrospira spp. encode a diversity of mechanisms to sustain chemolithoautotrophy in heterogeneous environments. Environ Microbiol 20:2686-2708. https://doi.org/ 10.1111/1462-2920.14090
- Mardis E, McCombie WR. 2017. Preparation of an 8-kb mate-pair library for Illumina sequencing. Cold Spring Harb Protoc 2017:pdb prot094664. https://doi.org/10.1101/pdb.prot094664.