




Draft Genome Sequence of *Magnetospirillum* sp. Strain 15-1, a Denitrifying Toluene Degradator Isolated from a Planted Fixed-Bed Reactor

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ABSTRACT Here, we report the draft genome sequence of *Magnetospirillum* sp. 15-1. This strain was isolated from a planted fixed-bed reactor based on its ability to degrade toluene under anaerobic conditions. The genome assembly consists of 5.4 Mb in 28 contigs and 5,095 coding sequences containing the genes involved in anaerobic toluene degradation.

Anaerobic degradation of toluene has been investigated most intensively among *Betaproteobacteria* strains, represented by *Thauera aromatica* and *Azoarcus* sp. (1–3). Within the *Alphaproteobacteria* group, there are three strains that have been identified as toluene degraders, *Blastochloris sulfovirdis* (4), *Magnetospirillum* sp. TS-6 (5), and *Magnetospirillum* sp. 15-1 (6).

Here, we present the draft genome sequence of *Magnetospirillum* sp.15-1, a non-magnetic strain isolated from a planted fixed-bed reactor (PFR) continuously fed with toluene (6). Sequencing was carried out using the 454-GS Junior system (Roche Applied Science) and MiSeq instrument (Illumina). Illumina libraries were paired-end sequenced with a maximum 300-bp read length (MiSeq reagent kit version 3, 600 cycles, Illumina). Together, sequencing resulted in 1,674,000 reads. *De novo* assembly was conducted with the A5-miseq pipeline (7) and Geneious assembler (<http://www.geneious.com>) (8), generating 527 contigs initially. Contigs shorter than 400 bp with a coverage percentage below 20, an aberrant GC content below 50%, and a low Q score (9) were not considered further. The relative proportion of omitted nucleotides was 1.2%. Five gaps were closed by PCR, resulting in 28 final contigs. The draft genome sequence of *Magnetospirillum* sp. 15-1 consists of 5,422,505 bp with an average G+C content of 65.6%. The genome contains 6 rRNAs, 49 tRNAs, and 5,095 coding sequences (CDSs) (coding percentage 98.82%). The assembled sequences were functionally annotated using the RAST online service (10) and IMG (11). Of 5,095 CDSs, 62.61% were assigned to at least one COG group.

Magnetic strains of *Magnetospirillum* contain a large magnetosome genomic island that harbors a high concentration of insertion sequences and is flanked by repetitive elements (12, 13). This island is absent in strain 15-1. Phylogenetic analyses based on comparisons of average nucleotide identities and 16S rRNA gene alignment showed that strain 15-1 is related more closely to the magnetosome-producing strains *M. magneticum* AMB-1 (87%) (14), *M. magnetotacticum* MS-1 (86%) (15), *Magnetospirillum* sp. XM-1 (16) (88%), *Magnetospirillum* sp. SO-1 (89%) (17), and *M. gryphiswaldense* sp. MSR-1 (76%) (18) than to the nonmagnetic *M. bellicus* VDY (77%) (19) and *Magnetospirillum* sp. WD (77%) (20). This finding suggests that the absence of the magnetosome island in the *Magnetospirillum* sp. 15-1 genome is due to a secondary loss of function.

Received 20 June 2017 Accepted 7 July 2017 Published 10 August 2017

Citation Meyer-Cifuentes I, Fiedler S, Müller JA, Kappelmeyer U, Mäusezahl I, Heipieper HJ. 2017. Draft genome sequence of *Magnetospirillum* sp. strain 15-1, a denitrifying toluene degrader isolated from a planted fixed-bed reactor. *Genome Announc* 5:e00764-17. <https://doi.org/10.1128/genomeA.00764-17>.

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The genes for degradation of toluene under anaerobic conditions were found to be organized in three operons on the chromosome (*bss*, *bbs*, and *bam*), as observed for *Thauera aromatica* K172 (21). Homology searches of the *bss* operon of strain 15-1 against other anaerobic toluene degraders were performed through BLAST (<https://blast.ncbi.nlm.nih.gov/Blast.cgi>). Results showed similarities of 99% to *Magnetospirillum* sp. TS-6 and 80% to both *T. aromatica* K172 (22) and *Azoarcus* sp. (23). Analysis of xenobiotic degradation pathways through the KEGG pipeline to sequenced *Magnetospirillum* strains (excluding strain 15-1) showed that none of these strains contain genes related to aromatic compound degradation pathways.

Accession number(s). This whole-genome shotgun project has been deposited in the European Nucleotide Archive (ENA) under the accession no. [FXXN01000001](https://ena.ebi.ac.uk/ena/record/FXXN01000001) to [FXXN01000028](https://ena.ebi.ac.uk/ena/record/FXXN01000028). The versions described in this paper are the first versions.

ACKNOWLEDGMENTS

This work was funded by CONICYT (grant no. 72140034), Chile, and by the Helmholtz Centre for Environmental Research–UFZ, Leipzig, Germany.

REFERENCES

- Evans PJ, Mang DT, Kim KS, Young LY. 1991. Anaerobic degradation of toluene by a denitrifying bacterium. *Appl Environ Microbiol* 57: 1139–1145.
- Coschigano PW. 2000. Transcriptional analysis of the *tutE* *tutFDGH* gene cluster from *Thauera aromatica* Strain T1. *Appl Environ Microbiol* 66: 1147–1151. <https://doi.org/10.1128/AEM.66.3.1147-1151.2000>.
- Dolfing J, Zeyer J, Binder-Eicher P, Schwarzenbach RP. 1990. Isolation and characterization of a bacterium that mineralizes toluene in the absence of molecular oxygen. *Arch Microbiol* 154:336–341. <https://doi.org/10.1007/BF00276528>.
- Zengler K, Heider J, Rosselló-Mora R, Widdel F. 1999. Phototrophic utilization of toluene under anoxic conditions by a new strain of *Blaschlochloris sulfovoridis*. *Arch Microbiol* 172:204–212. <https://doi.org/10.1007/s002030050761>.
- Shinoda Y, Akagi J, Uchihashi Y, Hiraishi A, Yukawa H, Yurimoto H, Sakai Y, Kato N. 2005. Anaerobic degradation of aromatic compounds by *Magnetospirillum* strains isolation and degradation genes. *Biosci Technol Biochem* 69:1483–1491. <https://doi.org/10.1271/bbb.69.1483>.
- Meyer-Cifuentes I, Martínez-Lavanchy PM, Marin-Cevada V, Bohnke S, Harms H, Müller JA, Heipieper HJ. 2017. Isolation and characterization of *Magnetospirillum* sp. strain 15-1 as a representative anaerobic toluene-degrader from a constructed wetland model. *PLoS One*:e0174750.
- Coil D, Jospin G, Darling AE. 2015. A5-miseq: an updated pipeline to assemble microbial genomes from Illumina MiSeq data. *Bioinformatics* 31:587–589. <https://doi.org/10.1093/bioinformatics/btu661>.
- Kearse M, Moir R, Wilson A, Stones-Havas S, Cheung M, Sturrock S, Buxton S, Cooper A, Markowitz S, Duran C, Thierer T, Ashton B, Meintjes P, Drummond A. 2012. Geneious basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* 28:1647–1649. <https://doi.org/10.1093/bioinformatics/bts199>.
- Ewing B, Green P. 1998. Base-calling of automated sequencer traces using Phred. II. Error probabilities. *Genome Res* 8:186–194. <https://doi.org/10.1101/gr.8.3.186>.
- Aziz RK, Bartels D, Best AA, DeJongh M, Disz T, Edwards RA, Formsma K, Gerdes S, Glass EM, Kubal M, Meyer F, Olsen GJ, Olson R, Osterman AL, Overbeek RA, McNeil LK, Paarmann D, Paccian T, Parrello B, Pusch GD, Reich C, Stevens R, Vassieva O, Vonstein V, Wilke A, Zagnitko O. 2008. The RAST server: rapid annotations using subsystems technology. *BMC Genomics* 9:75. <https://doi.org/10.1186/1471-2164-9-75>.
- Markowitz VM, Chen IMA, Palaniappan K, Chu K, Szeto E, Grechkin Y, Ratner A, Jacob B, Huang J, Williams P, Huntemann M, Anderson I, Mavromatis K, Ivanova NN, Kyrpides NC. 2012. IMG: the integrated microbial genomes database and comparative analysis system. *Nucleic Acids Res* 40:D115–DD122. <https://doi.org/10.1093/nar/gkr1044>.
- Fukuda Y, Okamura Y, Takeyama H, Matsunaga T. 2006. Dynamic analysis of a genomic island in *Magnetospirillum* sp. strain AMB-1 reveals how magnetosome synthesis developed. *FEBS Lett* 580:801–812. <https://doi.org/10.1016/j.febslet.2006.01.003>.
- Schübbe S, Kube M, Scheffel A, Wawer C, Heyen U, Meyerdierks A, Madkour MH, Mayer F, Reinhardt R, Schüler D. 2003. Characterization of a spontaneous nonmagnetic mutant of *Magnetospirillum gryphiswaldense* reveals a large deletion comprising a putative magnetosome island. *J Bacteriol* 185: 5779–5790. <https://doi.org/10.1128/JB.185.19.5779-5790.2003>.
- Matsunaga T, Okamura Y, Fukuda Y, Wahyudi AT, Murase Y, Takeyama H. 2005. Complete genome sequence of the facultative anaerobic magnetotactic bacterium *Magnetospirillum* sp. strain AMB-1. *DNA Res* 12: 157–166. <https://doi.org/10.1093/dnares/dsi002>.
- Smalley MD, Marinov GK, Bertani LE, DeSalvo G. 2015. Genome sequence of *Magnetospirillum magnetotacticum* strain MS-1. *Genome Announc* 3(2):e00233-15. <https://doi.org/10.1128/genomeA.00233-15>.
- Wang Y, Zhang T, Lin W, Zhang B, Cai Y, Yang C, Li J, Xu H, Pan Y. 2016. Complete genome sequence of *Magnetospirillum* sp. strain XM-1, isolated from the Xi'an-City-Moat, China. *Genome Announc* 4(5):e01171-16. <https://doi.org/10.1128/genomeA.01171-16>.
- Grouzdev DS, Dziuba MV, Sukhacheva MS, Mardanov AV, Beletskiy AV, Kuznetsov BB, Skryabin KG. 2014. Draft genome sequence of *Magnetospirillum* sp. strain SO-1, a freshwater magnetotactic bacterium isolated from the Ol'khovka River, Russia. *Genome Announc* 2(2):e00235. <https://doi.org/10.1128/genomeA.00235-14>.
- Wang X, Wang Q, Zhang W, Wang Y, Li L, Wen T, Zhang T, Zhang Y, Xu J, Hu J, Li S, Liu L, Liu J, Jiang W, Tian J, Li Y, Schüler D, Wang L, Li J. 2014. Complete genome sequence of *Magnetospirillum gryphiswaldense* MSR-1. *Genome Announc* 2(2):e00171-14. <https://doi.org/10.1128/genomeA.00171-14>.
- Thrash JC, Ahmadi S, Torok T, Coates JD. 2010. *Magnetospirillum bellicus* sp. nov., a novel dissimilatory perchlorate-reducing alphaproteobacterium isolated from a bioelectrical reactor. *Appl Environ Microbiol* 76: 4730–4737. <https://doi.org/10.1128/AEM.00015-10>.
- Michaelidou U, Achenbach LA, Coates JD. 2000. Isolation and characterization of two novel (Per)chlorate-reducing bacteria from swine waste lagoons, p 271–283. *In* Urbansky ET (ed), *Perchlorate in the environment*. Springer, Boston, MA. https://doi.org/10.1007/978-1-4615-4303-9_25.
- Hermuth K, Leuthner B, Heider J. 2002. Operon structure and expression of the genes for benzylsuccinate synthase in *Thauera aromatica* strain K172. *Arch Microbiol* 177:132–138. <https://doi.org/10.1007/s00203-001-0375-1>.
- Leuthner B, Leutwein C, Schulz H, Hörth P, Haehnel W, Schiltz E, Schagger H, Heider J. 1998. Biochemical and genetic characterization of benzylsuccinate synthase from *Thauera aromatica*: a new glycol radical enzyme catalyzing the first step in anaerobic toluene metabolism. *Mol Microbiol* 28: 615–628. <https://doi.org/10.1046/j.1365-2958.1998.00826.x>.
- Achong GR, Rodriguez AM, Spormann AM. 2001. Benzylsuccinate synthase of *Azoarcus* sp. strain T: cloning, sequencing, transcriptional organization and its role in anaerobic toluene and *m*-xylene mineralization. *J Bacteriol* 183:6763–6770. <https://doi.org/10.1128/JB.183.23.6763-6770.2001>.