Draft Genome Sequence of n-Alkane-Utilizing Acinetobacter sp. Strain BS1, Isolated from Ethane Oxidation Culture

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ABSTRACT Here, we report the draft whole-genome sequence of a bacterial strain, Acinetobacter sp. strain BS1, isolated from black soil during ethane oxidation culture. Medium- or long-chain alkane oxidation-related genes were identified; however, the short-chain alkane monooxygenase was not detected.

Microbes of the genus Acinetobacter have been involved in the bioremediation of petroleum (1) because of their high efficiency at degrading different types of alkanes (2). Several pathways and functional genes for the degradation of medium- or long-chain alkanes have been identified for the strains of this genus (3–5). However, the ability to carry out gaseous oxidation has never been reported for these organisms.

To isolate ethane oxidation microbes, a floating-filter method (6) was applied on nitrate mineral salts (NMS) medium (7). Acinetobacter sp. strain BS1 was isolated from colonies on a floating filter through which diluted black soil of the Changbai Mountains in China was filtered and was purified with R2A agar medium. Genomic DNA was extracted using a PowerSoil DNA isolation kit (Mo Bio, USA). The Kapa LTP library kit (Kapa Biosystems, USA) was used to construct the library, and whole-genome sequencing was performed on an Illumina HiSeq 2500 platform using a 2 × 250 protocol. The reads were quality trimmed using Sickle software version 1.33 (8), and bases with a quality above 20 (Q20) were used for assembly by Velvet (version 1.2.10) (9), resulting in 128 scaffolds (including plasmid sequences) with a maximum length of 513 kb and an N50 value of 138 kb. According to the analysis using CheckM (version 1.0.5) (10), the size of the draft genome was 3,929,328 bp (G+C content, 38.8%), with a coverage of 93.6-fold and completeness of 100%.

Annotation and identification of metabolic pathways were carried out using the NCBI Prokaryotic Genome Annotation Pipeline (PGAP) server. A total of 3,637 coding sequences or open reading frames (ORFs), as well as 64 tRNA genes, were identified. Based on the sequence coverage, about 9 copies of 23S rRNA and 16S rRNA genes were identified, with at least 3 copies of 5S-23S-16S rRNA operons. The draft genome does not encode monooxygenases that play important roles in the oxidation of short-chain (C1 to C4) n-alkanes. However, it encodes a cytochrome P450 hydroxylase that has been reported to be involved in medium-chain n-alkane degradation (4). In addition to the genes encoding rubredoxin (rubA) and rubredoxin reductase (rubB), genes of the AlkB family alkane hydroxylases (11) were also identified and scattered throughout the whole chromosome, which include alkB (encoding alkane-1 monoxygenase), alkJ (encoding alcohol dehydrogenase), alkH (encoding aldehyde dehydrogenase), and alkK (encoding acyl-coenzyme A [acyl-CoA] synthetase). Additionally, AlmA- and LadA-encoding genes, involved in long-chain n-alkane oxidation, were also identified by the probing of complete protein sequences from Acinetobacter sp. strain DSM 17874 (12) and Geobacillus thermodenitrificans (13), respectively. The utilization of medium- or

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long-chain alkanes by strain BS1 was confirmed with nonane (C9), tridecane (C13), or triacontane (C30) as a sole carbon source (data not shown).

Strain BS1 can use methanol or ethanol as a sole carbon source but cannot directly use ethane (data not shown). This indicates that strain BS1 may grow during the ethane oxidation process based on the ethanol produced by other ethane-oxidizing microbes, as well as with high contents of methanol-oxidizing bacteria during methane oxidation (14).

Accession number(s). This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession no. NPMK00000000. The version described in this paper is the first version, NPMK01000000.

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REFERENCES

1. Aislabie J, Saul DJ, Foght JM. 2006. Bioremediation of hydrocarbon-contaminated polar soils. Extremophiles 10:171–179. https://doi.org/10.1007/s00792-005-0498-4.
2. Luo Q, Zhang J-G, Shen X-R, Fan Z-Q, He Y, Hou D-Y. 2013. Isolation and characterization of marine diesel oil-degrading Acinetobacter sp. strain Y2. Ann Microbiol 63:633–640. https://doi.org/10.1007/s13213-012-0513-9.
3. Geissdorfer W, Kok RG, Ratajczak A, Hellingwerf KJ, Hillen W. 1999. The genes rubA and rubB for alkane degradation in Acinetobacter sp. strain ADP1 are in an operon with estB, encoding an esterase, and oxyR. J Bacteriol 181:4292–4298.
4. Ji Y, Mao G, Wang Y, Bartlam M. 2013. Structural insights into diversity and n-alkane biodegradation mechanisms of alkane hydroxylases. Front Microbiol 4:58. https://doi.org/10.3389/fmicb.2013.00058.
5. Ratajczak A, Rfer WGD, Hillen W. 1998. Alkane hydroxylase from Acinetobacter sp. strain ADP1 is encoded by alkM and belongs to a new family of integral-membrane hydrocarbon hydroxylases. Appl Environ Microbiol 64:1175–1179.
6. de Bruyn JC, Boogerd FC, Bos P, Kuenen JG. 1990. Floating filters, a novel technique for isolation and enumeration of fastidious, acidophilic, iron-oxidizing, autotrophic bacteria. Appl Environ Microbiol 56:2891–2894.
7. Bowman JP, Jimenez L, Rosorio I, Hazen TC, Sayler GS. 1993. Characterization of the methanotrophic bacterial community present in a trichloroethylene-contaminated subsurface groundwater site. Appl Environ Microbiol 59:2380–2387.
8. Joshi NA, Fass JN. 2011. Sickle: a sliding-window, adaptive, quality-based trimming tool for FastQ files (version 1.33). https://github.com/najoshi/sickle.
9. Zerbino DR, Birney E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res 18:821–829. https://doi.org/10.1101/gr.074492.107.
10. 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://doi.org/10.1101/gr.186072.114.
11. van Beilen JB, Panke S, Lucchini S, Franchini AG, Röthlisberger M, Witholt B. 2001. Analysis of Pseudomonas putida alkane-degradation gene clusters and flanking insertion sequences: evolution and regulation of the alk genes. Microbiology 147:1621–1630. https://doi.org/10.1099/00221287-147-6-1621.
12. Throne-Holst M, Wentzel A, Ellingsen TE, Kotlar H-K, Zotchev SB. 2007. Identification of novel genes involved in long-chain n-alkane degradation by Acinetobacter sp. strain DSM 17874. Appl Environ Microbiol 73:3327–3332. https://doi.org/10.1128/AEM.00064-07.
13. Feng L, Wang W, Cheng J, Ren Y, Zhao G, Gao C, Tang Y, Liu X, Han W, Peng M, Liu R, Wang L. 2007. Genome and proteome of long-chain alkane degrading Geobacillus thermodenitrificans NG80-2 isolated from a deep-subsurface oil reservoir. Proc Natl Acad Sci U S A 104:5602–5607. https://doi.org/10.1073/pnas.0609650104.
14. Beck DAC, Kalyuzhnaya MG, Malfatti S, Tringe SG, Glavina del Rio T, Ivanova N, Lidstrom ME, Chistoserdova L. 2013. A metagenomic insight into freshwater methane-utilizing communities and evidence for cooperation between the Methylococcaceae and the Methylophilaceae. Peer J 1:e23. https://doi.org/10.7717/peerj.23.