



Complete Genome Sequence of *Latilactobacillus* sp. Strain WDN19, a High-D-Aspartate-Producing Lactic Acid Bacterium Isolated from a Japanese Pickle

Kengo Kajitani,^a  Shouji Takahashi^a

^aDepartment of Bioengineering, Nagaoka University of Technology, Nagaoka, Niigata, Japan

ABSTRACT We report here the complete genome sequence of *Latilactobacillus* sp. strain WDN19, isolated from a Japanese pickle. This strain can produce a large amount of D-aspartate in the culture broth. The genome consists of a circular chromosome (1,967,462 bp; GC content, 41.88%) and a circular plasmid (66,648 bp; GC content, 35.08%).

The amino acid D-aspartate (D-Asp) is an important intermediate for synthetic penicillin (1). Some lactic acid bacteria (LAB) produce D-Asp as a component of cell wall peptidoglycan (2). LAB strains with greater D-Asp production would therefore be valuable for industrial D-Asp production. We isolated a novel high-D-Asp-producing strain, *Latilactobacillus* sp. strain WDN19, from a Japanese pickle purchased at a market in Niigata Prefecture, Japan (3). Here, we report the complete genome sequence of strain WDN19. This genome sequence will provide an avenue for the biotechnological manipulation of strain WDN19 for increased production of D-Asp.

Strain WDN19 was isolated from a Japanese pickle as described previously (3) and stored as glycerol stock at -80°C . To prepare the genomic DNA of strain WDN19, a single colony grown on MRS agar medium was transferred to 14 ml MRS medium (BD Difco, Sparks, MD, USA), statically cultured until the early stationary phase, approximately 8 h at 30°C , and pelleted. Genomic DNA was isolated from the pelleted cells using the Genomic-tip 100/G kit (Qiagen, Hilden, Germany) according to the manufacturer's protocol with modifications for Gram-positive bacteria. A SMRTbell library was prepared and sequenced using a single-molecule real-time (SMRT) cell in a PacBio RS II sequencer at Macrogen Corp. Japan (Tokyo, Japan) for long-read sequencing. A total of 1.21 Gbp sequence data (coverage, $597\times$) in 121,749 filtered subreads was obtained. The average length and N_{50} value of the subreads were 9,972 and 13,416 bp, respectively. The PacBio data were assembled using the Hierarchical Genome Assembly Process 3 (HGAP3) in the PacBio SMRT Analysis version 2.3.0 platform (4). Gene prediction and annotation of the generated contigs were performed using the DDBJ Fast Annotation and Submission Tool (DFAST) version 1.2.13 (<https://dfast.ddbj.nig.ac.jp/>). A BLASTn search of the 16S rRNA gene sequence of strain WDN19 was performed against the NCBI rRNA/ITS databases (<https://blast.ncbi.nlm.nih.gov/Blast.cgi>). The average nucleotide identity (ANI) was calculated using JSpeciesWS (5). Default parameters were used for all software unless otherwise specified.

The complete genome of strain WDN19 consists of a single circular chromosome 1,967,462 bp long with a GC content of 41.88% and a single circular plasmid 66,648 bp long with a GC content of 35.08%. DFAST predicted that the chromosome contains 2,055 protein-coding sequences (CDSs), 64 tRNA genes, and 18 rRNA genes and the plasmid sequence contains 70 CDSs and 1 tRNA gene. The 16S rRNA gene sequence of strain WDN19 had the highest similarity to that of *Latilactobacillus curvatus* DSM 20019 (=NBRC 15884^T) (GenBank accession number [AB626051](https://ncbi.nlm.nih.gov/nucl/AB626051)), with 99.86% identity. Furthermore, ANI analysis showed that the chromosome sequence of strain WDN19 has the highest similarity to that of strain DSM 20019 ([GCA_001435495.1](https://ncbi.nlm.nih.gov/nucl/GCA_001435495.1)), with 98.5% identity, which is above the

Citation Kajitani K, Takahashi S. 2021. Complete genome sequence of *Latilactobacillus* sp. strain WDN19, a high-D-aspartate-producing lactic acid bacterium isolated from a Japanese pickle. Microbiol Resour Announc 10:e00568-21. <https://doi.org/10.1128/MRA.00568-21>.

Editor David A. Baltrus, University of Arizona

Copyright © 2021 Kajitani and Takahashi. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Shouji Takahashi, shoutaka@vos.nagaokaut.ac.jp.

Received 29 June 2021

Accepted 30 July 2021

Published 26 August 2021

threshold value of 95 to 96% ANI for species delineation (6), suggesting that strain WDN19 is a member of *L. curvatus*.

Data availability. The complete genome sequence of strain WDN19 has been deposited in the DDBJ/EMBL/GenBank databases under accession numbers [AP024685](#) for the chromosome and [AP024686](#) for the plasmid. The raw reads were deposited under BioProject accession number [PRJDB11678](#), BioSample accession number [SAMD00323104](#), and DDBJ Sequence Read Archive (DRA) accession number [DRA012183](#).

ACKNOWLEDGMENT

This study was supported in part by a Grant-in-Aid for Scientific Research (C) (19K05765) to S. Takahashi from the Japan Society for the Promotion of Science.

REFERENCES

1. Senuma M, Otsuki O, Sakata N, Furui M, Tosa T. 1989. Industrial production of D-aspartic acid and L-alanine from DL-aspartic acid using a pressurized column reactor containing immobilized *Pseudomonas dacunhae* cells. *J Ferment Bioeng* 67:233–237. [https://doi.org/10.1016/0922-338X\(89\)90224-9](https://doi.org/10.1016/0922-338X(89)90224-9).
2. Chapot-Chartier M-P, Kulakauskas S. 2014. Cell wall structure and function in lactic acid bacteria. *Microb Cell Fact* 13 Suppl 1:S9. <https://doi.org/10.1186/1475-2859-13-S1-S9>.
3. Kajitani K, Ishikawa T, Shibata K, Kouya T, Kera Y, Takahashi S. 2021. Development of an enzymatic screening method for D-aspartate-producing lactic acid bacteria. *Enzyme Microb Technol* 149:109835. <https://doi.org/10.1016/j.enzmictec.2021.109835>.
4. Chin C-S, Alexander DH, Marks P, Klammer AA, Drake J, Heiner C, Clum A, Copeland A, Huddleston J, Eichler EE, Turner SW, Korlach J. 2013. Nonhybrid, finished microbial genome assemblies from long-read SMRT sequencing data. *Nat Methods* 10:563–569. <https://doi.org/10.1038/nmeth.2474>.
5. Richter M, Rosselló-Móra R, Oliver Glöckner F, Peplies J. 2016. JSpeciesWS: a Web server for prokaryotic species circumscription based on pairwise genome comparison. *Bioinformatics* 32:929–931. <https://doi.org/10.1093/bioinformatics/btv681>.
6. Richter M, Rosselló-Móra R. 2009. Shifting the genomic gold standard for the prokaryotic species definition. *Proc Natl Acad Sci U S A* 106:19126–19131. <https://doi.org/10.1073/pnas.0906412106>.