



Draft Genome Sequences of Two *Veillonella tobetsuensis* Clinical Isolates from Intraoperative Bronchial Fluids of Elderly Patients with Pulmonary Carcinoma

Izumi Mashima,^a Tohru Miyoshi-Akiyama,^b Junko Tomida,^a Ryo Kutsuna,^a Jumpei Washio,^c Nobuhiro Takahashi,^c Futoshi Nakazawa,^d Takuichi Sato,^e Yoshiaki Kawamura^a

^aDepartment of Microbiology, School of Pharmacy, Aichi-Gakuin University, Nagoya, Aichi, Japan

^bPathogenic Microbe Laboratory, Research Institute, National Center for Global Health and Medicine, Shinjuku, Tokyo, Japan

^cDivision of Oral Ecology and Biochemistry, Tohoku University Graduate School of Dentistry, Sendai, Miyagi, Japan

^dHealth Sciences University of Hokkaido Graduate School of Dentistry, Ishikari-Tobetsu, Hokkaido, Japan

^eDivision of Clinical Chemistry, Department of Medical Technology, Niigata University Graduate School of Health Sciences, Niigata, Japan

ABSTRACT To date, *Veillonella tobetsuensis* has been known as an oral anaerobe and a facilitator of early-stage oral biofilm development with streptococci. Here, we report the draft genome sequences of 2 strains of *V. tobetsuensis* first isolated from intraoperative bronchial fluids of elderly patients with pulmonary carcinoma.

Veillonella tobetsuensis is a strictly anaerobic Gram-negative coccus that was first isolated from a human tongue biofilm in 2013 (1). *V. tobetsuensis* has been isolated frequently from human oral cavities (2, 3), and its ability to form oral biofilms at the early stage with streptococci is significantly higher than that of other *Veillonella* species (4), suggesting that *V. tobetsuensis* has an important role in developing biofilms related to oral infectious diseases (4, 5).

Hasegawa et al. (6) isolated and identified *Veillonella* isolates as predominant anaerobes by 16S rRNA gene sequences from intraoperative bronchial fluids of patients with pulmonary carcinoma. However, 4 unknown isolates (PAGU 1578 to PAGU 1581) belonging to the genus *Veillonella* were isolated from two subjects (PAGU 1578 from a 70-year-old male and PAGU 1579 to PAGU 1581 from an 80-year-old female). These 4 unknown isolates were divided into two clones based on subject sources. Then, draft genome sequences of 2 representative unknown isolates, *Veillonella* sp. strains PAGU 1578 and PAGU 1579, were determined to identify them to the species level in this study.

The genomic DNA of these 2 isolates was extracted from 5-day cultures using phenol-chloroform extraction and ethanol precipitation (7). Further purification was carried out using the Wizard Genomic DNA purification kit (Promega) for high-throughput sequences. DNA libraries were prepared using the Nextera XT library kit (Illumina) according to the manufacturer's instructions. DNA sequencing was performed at the National Center for Global Health and Medicine (Tokyo, Japan) using the HiSeq X Ten system (Illumina) with sequencing runs for paired-end sequencing, which achieved 150-bp read length and over 200-fold genome coverage. In total, 2,878,222 and 2,452,016 reads were obtained for PAGU 1578 and PAGU 1579, respectively, and they were checked for base quality (quality score limit, 0.05; ambiguous limit, 2; minimum number of nucleotides in reads, 15) and *de novo* assembled using CLC Genomics Workbench v. 11.01 (CLCbio). All final assemblies were annotated using the DDBJ Fast Annotation and Submission Tool (DFAST; <https://dfast.nig.ac.jp/>) with default parameters (8).

The draft genome sequences of PAGU 1578 and PAGU 1579 were assembled into 59

Citation Mashima I, Miyoshi-Akiyama T, Tomida J, Kutsuna R, Washio J, Takahashi N, Nakazawa F, Sato T, Kawamura Y. 2019. Draft genome sequences of two *Veillonella tobetsuensis* clinical isolates from intraoperative bronchial fluids of elderly patients with pulmonary carcinoma. *Microbiol Resour Announc* 8:e00397-19. <https://doi.org/10.1128/MRA.00397-19>.

Editor Vincent Bruno, University of Maryland School of Medicine

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Address correspondence to Yoshiaki Kawamura, kawamura@dpc.agu.ac.jp.

Received 7 April 2019

Accepted 3 September 2019

Published 19 September 2019

($N_{50} = 96,198$) and 129 ($N_{50} = 39,212$) contigs and were 1,935,552 bp and 2,151,918 bp in size, respectively. These two draft genomes contained G+C contents of 38.4% and 38.5% and 1,789 and 1,869 coding sequences, respectively.

To investigate genomic comparison among closely related isolates of the genus *Veillonella*, average nucleotide identity (ANI) values were calculated using GENETYX v. 14 software (Tokyo Genetex Co.) with default parameters. The results of BLAST calculation of ANI values between PAGU 1578 and PAGU 1579 or these two clinical isolates and *V. tobetsuensis* ATCC BAA-2400^T, which is the most closely related species, were 96.73% and 96.16% to 97.05%, respectively. These results suggested that PAGU 1578 and PAGU 1579 were the same species, *V. tobetsuensis*.

This is the first report of *V. tobetsuensis* isolated from bronchial fluids, and their draft genome sequences are announced. Streptococci were also isolated from the same clinical samples as predominant bacteria with *Veillonella* species (6), suggesting that *V. tobetsuensis* might have the ability to facilitate biofilm infection in bronchial diseases, as suggested in oral infectious diseases (4, 5).

Data availability. These draft genome sequences of *V. tobetsuensis* PAGU 1578 and PAGU 1579 and the raw sequence data sets have been deposited at DDBJ/EMBL/GenBank under the accession numbers [BJCQ01000001](#) to [BJCQ01000133](#), [BJCR01000001](#) to [BJCR01000129](#), and [DRA008107](#), respectively. The versions described in this paper are the first versions.

ACKNOWLEDGMENTS

This study was supported in part by a grant-in-aid from the Japan Society for Scientific Research from KAKENHI (17K12006 to T.S.) and a Grant for International Health Research (30A1002 to T.M.-A.) from the Ministry of Health, Labor and Welfare of Japan.

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