Complete Genome Sequence of *Luteitalea* sp. Strain TBR-22

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**ABSTRACT** We report a complete genome sequence of a novel bacterial isolate, strain TBR-22, belonging to the class *Vicinamibacteria* of the phylum *Acidobacteria*, which was isolated from duckweed fronds. The genome expands our knowledge of the lifestyle of this abundant but rarely characterized phylum.

Duckweeds are commonly observed freshwater aquatic plants and have been shown to harbor unique microbial communities on their body surface (1, 2). With efforts to isolate novel bacterial lineages from a duckweed-associated microbial community, we successfully isolated a bacterial strain, TBR-22, belonging to the phylum *Acidobacteria* (3). Briefly, isolation was performed with wild duckweeds (*Lemnoideae* spp.) collected from rice paddies located in Ibaraki Prefecture, Japan. Duckweeds were washed and then sonicated with sterile distilled water (SDW), followed by serial dilution with SDW and inoculation on 2.0% (wt/vol) agar plates of diluted tryptic soy broth supplemented with phosphate buffer, vitamin mixture, and basal salt solution (modified diluted tryptic soy broth [mDTS]). The phylum *Acidobacteria* is known to be one of the most abundant bacterial lineages in soils and is also found in various natural environments, including terrestrial freshwater, sediments, and terrestrial plants (4, 5). Despite its wide distribution in natural environments, physiological and ecological characterizations of *Acidobacteria* have not been extensively performed due to its fastidious and difficult-to-culture nature, as only 61 species in this phylum have been validly described to date (4, 5).

Genomic DNA was extracted from cells grown in mDTS at 30°C under static conditions by chemical and enzymatic procedures, as described previously (6), or by using the MagAttract high-molecular-weight (HMW) DNA kit (Qiagen). Library preparation and sequencing were performed by using commercial kits according to the manufacturers’ instructions (Table 1). A total of 1.53 million paired-end reads and 6.32 million mate-pair reads were obtained with the Illumina MiSeq system, and 15 thousand single long reads (mean length, 11,446 bp) were obtained with the Oxford Nanopore Technologies MinION system. Read quality control was performed by FastQC version 0.11.5 (7). Hybrid genome assembly was performed by hybridSPAdes version 3.13.0 (8) in KBase (9) with default settings, and a single scaffold was obtained. Closing of two assembly gaps and genome

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TABLE 1 Library preparation kits and sequencing platforms

| Sequencing type | Library prepn kit(s) | Sequencing platform |
|-----------------|----------------------|---------------------|
| Illumina short-read sequencing | KAPA HyperPrep kit (for Illumina) (Kapa Biosciences) and Illumina Nextera mate-pair library prepn kit (Illumina)* | Illumina MiSeq system (paired-end 2 × 300 bp) |
| MinION long-read sequencing | Rapid sequencing kit (Oxford Nanopore Technologies) | MinION system (R9.4 flow cell) |

* Insert lengths for mate-pair libraries were 3 kb and 8 kb.

circularization were performed by Sanger sequencing. The DDBJ Fast Annotation and Submission Tool (DFAST) pipeline version 1.2.13 (10) was used for structural annotation of the TBR-22 genome with following programs: MetaGeneAnnotator version 2008/08/19 (11) for coding sequences, Bambap version 0.8 (12) for rRNAs, ARAGORN version 1.2.38 (13) for tRNAs, and CRT version 1.2 (14) for CRISPRs.

The complete genome of TBR-22 consists of a 6,468,984-bp-long chromosome, with a G+C content of 70.5%; 5,364 predicted protein-coding DNA sequences, 55 tRNAs, a single set of 55/16S55/23S rRNAs, and no CRISPRs were identified. Genome completeness was estimated with CheckM version 1.1.3 (15), and the genome was determined to be 96.56% complete and 6.64% redundant and to have 4.55% strain heterogeneity. Taxonomic assignment by the Genome Taxonomy Database Toolkit (GTBD-Tk) version 0.1.4 (16) placed TBR-22 within the genus Luteitalea in the phylum Acidobacteriota (Acidobacteria), but it was not assigned a species. The genome obtained can largely contribute to understanding of the ecophysiology of Acidobacteria, especially in interactions with aquatic plants, which have been less well investigated than those with terrestrial plants.

Data availability. The genome and raw sequences have been deposited in DDBJ/ENA/GenBank under accession number AP024452 and in the DDBJ Sequence Read Archive (DRA) under accession numbers DRA011791 and DRA013041, respectively.

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11. Noguchi H, Taniguchi T, Itoh T. 2008. MetaGeneAnnotator: detecting species-specific patterns of ribosomal binding site for precise gene prediction in anonymous prokaryotic and phage genomes. DNA Res 15:387–396. https://doi.org/10.1093/dnares/dsn027.

12. Seemann T. 2013. Barmap 0.7: rapid ribosomal RNA prediction. https://github.com/tseemann/barmap.

13. Laslett D, Canback B. 2004. ARAGORN, a program to detect tRNA genes and tmRNA genes in nucleotide sequences. Nucleic Acids Res 32:11–16. https://doi.org/10.1093/nar/gkh152.

14. Bland C, Ramsey TL, Sabree F, Lowe M, Brown K, Kyrpides NC, Hugenholtz P. 2007. CRISPR Recognition Tool (CRT): a tool for automatic detection of clustered regularly interspaced palindromic repeats. BMC Bioinformatics 8:209. https://doi.org/10.1186/1471-2105-8-209.

15. 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.

16. Chaumeil P-A, Mussig AJ, Hugenholtz P, Parks DH. 2019. GTDB-Tk: a tool-kit to classify genomes with the Genome Taxonomy Database. Bioinformatics 36:1925–1927. https://doi.org/10.1093/bioinformatics/btz848.