



Draft Genome Sequences of *Sulfurovum* spp. TSL1 and TSL6, Two Sulfur-Oxidizing Bacteria Isolated from Marine Sediment

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ABSTRACT *Sulfurovum* spp. TSL1 and TSL6 are sulfur-oxidizing chemolithoautotrophic bacteria isolated from the tsunami-launched marine sediment in the Great East Japan earthquake. This announcement describes the draft genome sequences of the two isolates that possess the gene sets for the sulfur oxidation pathway.

An epsilonproteobacterial genus, *Sulfurovum*, mainly inhabiting deep-sea hydrothermal vents, is a group of sulfur-oxidizing chemolithoautotrophs governing the biogeochemical cycling of elements in seafloor ecosystems (1–5). A large amount of marine sediment was launched on land by a tsunami that occurred in the Great East Japan earthquake, and thereafter, the *Sulfurovum*-affiliated bacteria participated in sulfur oxidation when the marine sediment was exposed to oxic conditions (6). Two sulfur-oxidizing bacteria, TSL1 and TSL6, were isolated from the marine sediment (38° 25' N, 141° 14' E; Higashimatsushima, Miyagi, Japan). Briefly, the sediment was preincubated aerobically for 3 days and then incubated using serial dilution in basal salts of MJ medium (7) supplemented with a vitamin solution containing biotin, folic acid, pyridoxine hydrochloride, thiamine hydrochloride, riboflavin, nicotinic acid, DL-calcium pantothenate, vitamin B₁₂, *p*-aminobenzoic acid, and lipoic acid (8), and thiosulfate or elemental sulfur as an energy source. After repeating serial subcultivation 4 to 7 times, TSL1 and TSL6 were obtained as pure cultures using the thiosulfate and elemental sulfur as energy source, respectively. Based on 16S rRNA gene sequences (Fig. 1), these two bacteria could represent the undescribed *Sulfurovum* species. Here, we sequenced the genomes of these bacteria to reveal the genetic features underlying their biogeochemical activities.

TSL1 and TSL6 were aerobically grown in the above-mentioned medium supplemented with the vitamin solution, PIPES [piperazine-*N,N'*-bis(2-ethanesulfonic acid)] (20 mM), and sodium thiosulfate (20 mM) at room temperature for 2 to 3 weeks. Genomic DNA was isolated using a lysozyme buffer method (9). Paired-end libraries (insert size, ~350 bp) were prepared using a VAHTS universal DNA library prep kit for Illumina (Vazyme Biotech) and sequenced using a HiSeq 2500 system (Illumina), which generated 8.9 and 11.1 million reads (2 × 150 bp) for TSL1 and TSL6, respectively. Adapters were removed using Cutadapt v3.1 (10), while the reads with a Q score of <30 were removed using Sickle v1.33 (<https://github.com/najoshi/sickle>). High-quality reads were preassembled using Unicycler v0.4.8 with default settings (11) and then assembled using SPAdes v3.15.0 (12) with the trusted-contigs tool, as previously described (13). The draft genomes were annotated by the DFAST v1.4.0 (14) and KofamKOALA v2021-07-05 (15). Whole-genome average nucleotide identity (ANI) and digital DNA-DNA hybridization (dDDH) between the two genomes were calculated using the JSpeciesWS (16) and GGDC v2.1 (17), respectively.

TSL1 had a genome size of 2,353,154 bp assembled into 6 contigs (N_{50} value, 1,782,176 bp; GC content, 40.7 mol%; coverage, 500×), while TSL6 had the genome size of

Editor Frank J. Stewart, Montana State University

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The authors declare no conflict of interest.

Received 26 September 2021

Accepted 7 December 2021

Published 6 January 2022

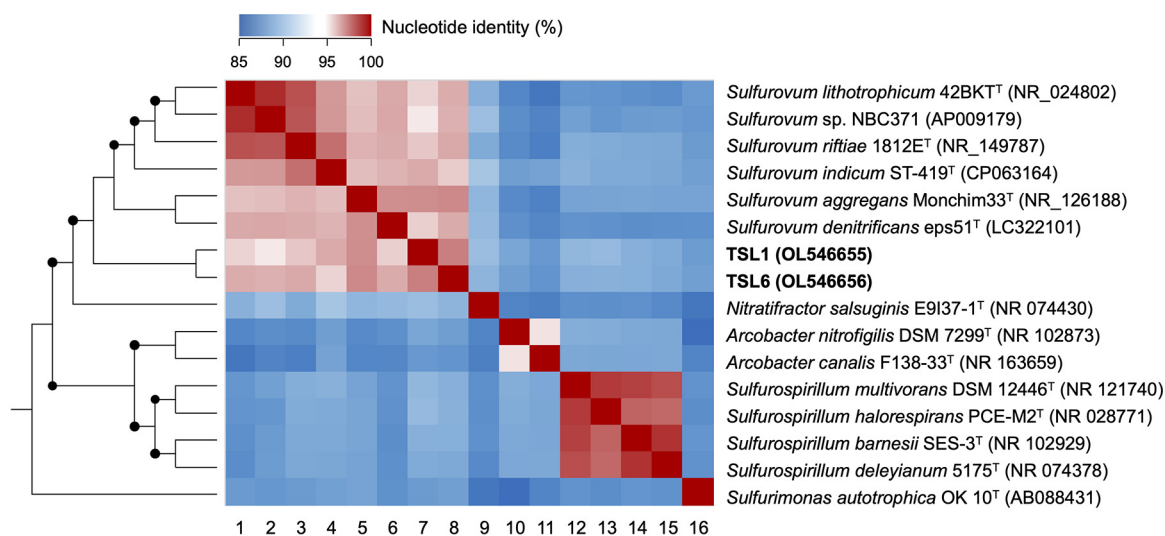


FIG 1 Phylogenetic relationship of TSL1 and TSL6 with the representative strains in the genera *Sulfurovum*, *Nitratifactor*, *Arcobacter*, *Sulfurospirillum*, and *Sulfurimonas*. Neighbor-joining phylogenetic tree (left) is reconstructed using near full-length 16S rRNA gene sequences, and bootstrap values (1,000 replicates) of more than 70% are indicated by solid circles at the branches. Heatmap (right) shows pairwise nucleotide identities of the near full-length 16S rRNA gene sequences. Column numbers below the heatmap indicate the bacterial strains as follows: 1, *Sulfurovum lithotrophicum* 42BKT^T; 2, *Sulfurovum* sp. strain NBC371; 3, *Sulfurovum riftiae* 1812E^T; 4, *Sulfurovum indicum* ST-419^T; 5, *Sulfurovum aggregans* Monchim33^T; 6, *Sulfurovum denitrificans* eps51^T; 7, TSL1; 8, TSL6; 9, *Nitratifactor salsuginis* E9137-1^T; 10, *Arcobacter nitrofigilis* DSM 7299^T; 11, *Arcobacter canalis* F138-33^T; 12, *Sulfurospirillum multivorans* DSM 12446^T; 13, *Sulfurospirillum halorespirans* PCE-M2^T; 14, *Sulfurospirillum barnesii* SES-3^T; 15, *Sulfurospirillum deleyianum* 5175^T; and 16, *Sulfurimonas autotrophica* OK 10^T. TSL1 and TSL6 shared 95.2% to 96.8% of the 16S rRNA gene identities with the type strains of type species in the genus *Sulfurovum*, indicating that the two strains can represent the undescribed species affiliated in the genus *Sulfurovum*.

2,254,663 bp assembled into 7 contigs (N_{50} value, 760,548 bp; GC content, 38.4 mol%; coverage, 650 \times) (Table 1). A total of 2,359 genes were predicted as 3 rRNAs, 42 tRNAs, and 2,314 protein-coding sequences (CDSs) in the TSL1 genome, while there was a total of 2,259 genes as 3 rRNAs, 43 tRNAs, and 2,213 CDSs in the TSL6 genome (Table 1). The ANI and dDDH values between the two isolates were calculated to be 86.5% (aligned coverage, 75.9%) and 32.3%, respectively, which suggests that TSL1 and TSL6 can be classified into different species. Genome annotation revealed that both of the two bacteria possessed a sulfur oxidation system, which would be helpful to understand their roles in the acidification of the tsunami-launched marine sediment (6).

Data availability. The draft genome sequences of *Sulfurovum* spp. TSL1 and TSL6 are available under the DDBJ/ENA/GenBank accession numbers [BPFJ01000001](https://www.ncbi.nlm.nih.gov/nuccore/BPFJ01000001) to [BPFJ01000006](https://www.ncbi.nlm.nih.gov/nuccore/BPFJ01000006) and [BPFJ01000001](https://www.ncbi.nlm.nih.gov/nuccore/BPFJ01000001) to [BPFJ01000007](https://www.ncbi.nlm.nih.gov/nuccore/BPFJ01000007), respectively. The raw reads are available under the DRA accession numbers [DRA011857](https://www.ebi.ac.uk/ena/submit/data/DRA011857) and [DRA011856](https://www.ebi.ac.uk/ena/submit/data/DRA011856) for TSL1 and TSL6, respectively.

TABLE 1 Statistics for genome assemblies of *Sulfurovum* spp. TSL1 and TSL6

Characteristic	Data for:	
	TSL1	TSL6
No. of contigs	6	7
N_{50} value (bp)	1,782,176	760,548
GC content (mol%)	40.7	38.4
Total length (bp)	2,353,154	2,254,663
Genome coverage (\times)	500	650
Genome completeness (%)	99.6	99.6
No. of rRNAs	3	3
No. of tRNAs	42	43
No. of CDSs	2,314	2,213
GenBank accession no.	BPFJ01000001 to BPFJ01000006	BPFJ01000001 to BPFJ01000007
DRA accession no.	DRA011857	DRA011856

ACKNOWLEDGMENTS

This work was supported in part by commissioned research, “Work on the investigation regarding preservation measures for the mural paintings of Takamatsuzuka Tumulus, a national treasure,” from the Agency for Cultural Affairs, Japan.

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