




Draft Genome Sequence of Trebouxiphyceae sp. Strain KSI-1, Isolated from an Island Hot Spring

 Hiro Takahashi,^{a,b,c} Satoshi Tanaka,^d Shuhei Hayashi,^e Shido Miyaki,^b Anna Takahashi,^f Shinnosuke Onai,^g Shuichi Fukuyoshi,^h Hitoshi Miyasaka^e

^aGraduate School of Medical Sciences, Kanazawa University, Kanazawa, Ishikawa, Japan

^bGraduate School of Horticulture, Chiba University, Matsudo, Chiba, Japan

^cFundamental Innovative Oncology Core Center, National Cancer Center, Chuo-ku, Tokyo, Japan

^dAdvanced Technology Laboratory, Kansai Electric Power Co., Sourakugun, Kyoto, Japan

^eDepartment of Applied Life Science, Sojo University, Nishiku, Kumamoto, Japan

^fFaculty of Information Technologies and Control, Belarusian State University of Informatics and Radio Electronics, Minsk, Belarus

^gInstitute for Theoretical Physics, Kanazawa University, Kanazawa, Ishikawa, Japan

^hInstitute of Medical, Pharmaceutical, and Health Sciences, Kanazawa University, Kanazawa, Ishikawa, Japan

ABSTRACT Trebouxiphyceae sp. KSI-1 is a green alga isolated from a seashore hot spring on Satsuma Iōjima in Kagoshima, Japan, and is highly tolerant to oxidative stress. Here, we report the draft genome sequence of this strain, thereby providing an insight into the genetic basis for its oxidative stress tolerance.

Reactive oxygen species (ROS) are generated by electron leakage to O₂ in the electron transport system. In oxygen-evolving photosynthetic organisms, various stresses accelerate the generation of ROS generation (1) due to disruption in the balance between photosynthetic excitation energy and terminal energy consumption for CO₂.

Trebouxiphyceae sp. strain KSI-1 was isolated from a seashore hot spring on Satsuma Iōjima, a volcanic island in Kagoshima, Japan. This strain was grouped in a new genus within the class Trebouxiphyceae based on its 18S rRNA sequence (GenBank accession number [LC082307](#)).

Modified Okamoto medium (MOM) (2), containing 3% NaCl, was used for the algal cultures. For the isolation of oxidative stress-tolerant green algae, the aqueous sample was mixed with an equal volume of MOM (pH 8.0) and cultured under continuous illumination at a light intensity of 100 μmol/m²/s of photosynthetically active radiation (PAR), with aeration by bubbling at a rate of 200 ml air/min. After the growth of green algal cells was observed, enrichment culture for the isolation of oxidative stress-tolerant green algae was initiated by culturing the cells under a stressed condition of 50 μM methyl viologen (MV). Passage cultures were repeated three times under the stressed condition, and finally, the stress-tolerant green algal cells were purified from the monoculture by streaking the cells onto the MOM agar plates and isolating the algal colony. Genomic DNA was isolated from the cells (ca. 50 mg) using a DNeasy plant minikit (Qiagen) according to the manufacturer's instructions after being disrupted in liquid nitrogen using a mortar and pestle. A 500-ng aliquot of DNA was sonicated to generate an average of 600-bp DNA fragments for library preparation. End repair, A tailing, adaptor ligation, PCR, and library purification were performed as described in our previous study (3). The sequencing of a TruSeq DNA library (paired-end 2 × 300-bp reads) generated 33,531,306 reads. Removal of the sequencing primers and trimming of the low-quality read regions from the obtained short reads were conducted with the CLC Genomics Workbench version 11.0.1 (Qiagen) with default parameters. *De novo*

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Address correspondence to Hiro Takahashi, takahasi@p.kanazawa-u.ac.jp, or Hitoshi Miyasaka, miyasaka@life.sojo-u.ac.jp. H.T., S.T., and S.H. contributed equally to this work.

assembly was also conducted with the CLC Genomics Workbench. The resulting genome assembly had a length of 44,347,176 bp divided into 22,485 contigs; the N_{50} contig length was 21,380 bp, the GC content was 64.6%, and genome coverage was 194.9 \times .

MV causes severe oxidative stress (4). Trebouxiphyceae sp. KSI-1 shows a surprisingly high tolerance to oxidative stress caused by MV and copper ions. Higher plants can generally tolerate MV and copper ions at concentrations up to 5 and 100 μ M, respectively, while Trebouxiphyceae sp. KSI-1 was tolerant to concentrations of 100 and 10 mM, respectively. Trebouxiphyceae sp. KSI-1 is, therefore, a prominent genetic resource of unique antistress genes for the molecular breeding of stress-tolerant plants (5), and the genomic information of this alga may provide insight into the genetic basis for oxidative stress tolerance in photosynthetic organisms.

Data availability. The draft genome sequences for Trebouxiphyceae sp. KSI-1 have been deposited in DDBJ/ENA/GenBank under accession numbers [BHFV01000001](#) to [BHFV01022485](#). The SRA/DRA/ERA accession number is [DRA007300](#).

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