



Draft Genome Sequences of Two Gammaproteobacterial Methanotrophs Isolated from Rice Ecosystems

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ABSTRACT The genomes of the aerobic methanotrophs “*Methylothericola oryzae*” strain 73a^T and *Methylothericola ishizawai* strain 175 were sequenced. Both strains were isolated from rice plants. *Methylothericola oryzae* strain 73a^T represents the first isolate of rice paddy cluster I, and strain 175 is the second representative of the recently described genus *Methylothericola*.

Aerobic methanotrophic bacteria play a key role in controlling global climate by reducing the emission of the greenhouse gas methane in ecosystems such as paddy fields (1). Gammaproteobacterial methanotrophs are common inhabitants of rice fields (2, 3). We sequenced the genomes of two gammaproteobacterial isolates from rice plants (4). “*Methylothericola oryzae*” strain 73a^T (=LMG 29185=VKM-B-2986) is currently the only cultivated representative of rice paddy cluster I (3, 5), and *Methylothericola ishizawai* strain 175 (= LMG 28717=VKM-B-2989) is the second representative of the genus (6).

Genomic DNA was extracted from bacterial cultures using a phenol-chloroform method (7), and draft genome sequences were generated at the DOE Joint Genome Institute. The genome of strain 73a^T was sequenced using an Illumina HiSeq 2000, which generated 11,844,428 reads (1.79 Gb). The Pacific Biosciences RS was used for strain 175, and 230,505 filtered subreads (0.73 Gb) were generated. Sequence filtering, genome assembly, and gene annotation were performed as described earlier (8, 9). The final draft of strain 73a^T had 302.0× read coverage, contained 74 contigs in 73 scaffolds, was 4.9 Mb in size, and had an average GC content of 61.1%. The draft of strain 175 had 140.8× read coverage, contained 8 contigs in 8 scaffolds, was 5.5 Mb in size, and had an average GC content of 63.0%.

Both strains encode metabolic inventory typical for type I methanotrophs (10). They harbor genes encoding a particulate methane monooxygenase (*pmoCAB*) and a methane/

Received 28 April 2017 Accepted 2 May 2017 Published 17 August 2017

Citation Frindte K, Kalyuzhnaya MG, Bringel F, Dunfield PF, Jetten MSM, Khmelenina VN, Klotz MG, Murrell JC, Op den Camp HJM, Sakai Y, Semrau JD, Shapiro N, DiSpirito AA, Stein LY, Svenning MM, Trotsenko YA, Vuilleumier S, Woyke T, Knief C. 2017. Draft genome sequences of two gammaproteobacterial methanotrophs isolated from rice ecosystems. *Genome Announc* 5:e00526-17. <https://doi.org/10.1128/genomeA.00526-17>.

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This is contribution 13 from the Organization for Methanotroph Genome Analysis (OMeGA).

ammonia monooxygenase-related protein (*pxmABC*) (11). Additionally, the genome of strain 175 encodes a soluble methane monooxygenase (*mmoXYBZDCGR*). Gene clusters for PQQ-dependent methanol dehydrogenases and PQQ biosynthesis were found in both strains (*mxafJGIRSACKLD*, *soxFJ*, *pqqABCDE*, and *pqqFG*). Formaldehyde oxidation is predicted to proceed via the tetrahydromethanopterin (H₄MPT) pathway (presence of *fae*, *mtdB*, *mch*, *fhcABCD*, and H₄MPT biosynthesis genes) (12). Additionally, both strains contained genes encoding the tetrahydrofolate-dependent pathway (presence of *mtdA*, *fchA*, *fhs*, and, additionally, a *folD* gene in strain 73a^T). Formate can potentially be oxidized via one (strain 175) or two (strain 73a^T) types of formate dehydrogenase.

Both strains may assimilate formaldehyde via the ribulose monophosphate pathway. The cleavage cascade can be realized via fructose-1,6-bisphosphate, and in strain 73a^T additionally via 2-keto-3-deoxy-6-phosphogluconate. Rearrangement of ribulose-5-phosphate can occur by transketolase and transaldolase reactions. A complete serine cycle is unlikely to be present. Both strains have the genes necessary for operational oxidative pentose phosphate and TCA cycle pathways, whereas a complete glycolysis cascade is encoded only in strain 73a^T. RubisCO genes are present (*cbbL* and *cbbS* in strain 175; *cbbM* in strain 73a^T), as well as genes for a complete Calvin-Benson-Bassham cycle.

For nitrogen acquisition, both strains possessed genes encoding ammonium (*amtB*), nitrate (*nasA*), and urea (*urtABCDE*) transporters, as well as urease genes (*ureABCDEFG*). Moreover, *nif* genes were present, suggesting the potential for dinitrogen fixation. The strains may form polyphosphate (*ppk*) and glycogen (*glgAB*, *glgC*, *glgP*, *glgX*, *malQ*, and *pgm*) as storage compounds. Strain 175, in addition, can potentially produce polyhydroxybutyrate (*phbAB* and *phbC*), a characteristic not yet known for type I methanotrophs (13).

Accession number(s). The genome sequences have been deposited in GenBank under the accession numbers [JYNS00000000](https://doi.org/10.1093/genbank/JYNS00000000), for *Methyloterricola oryzae* strain 73a^T, and [FXAM00000000](https://doi.org/10.1093/genbank/FXAM00000000), for *Methylomagnum ishizawai* strain 175.

ACKNOWLEDGMENTS

We thank all members of the Organization for Methanotroph Genome Analysis (OMeGA) and Genoscope (France) for access to its MicroScope platform for comparative genome analysis (<http://www.genoscope.cns.fr/agc/microscope/home>). The work conducted by the DOE Joint Genome Institute was supported by the Office of Science of the U.S. Department of Energy under contract no. DE-AC02-05CH11231.

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