**NOTES**

**Methanolobus taylorii** sp. nov., a New Methylotrophic, Estuarine Methanogen

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Previously published phylogenetic studies of 16S rRNA showed that methylotrophic, slightly halophilic, methanogenic strain GS-16T (T = type strain) represents a new species of bacterium. We propose the name *Methanolobus taylorii* for this species; strain GS-16 is the type strain.

Strain GS-16T (T = type strain) is a methylotrophic methanogen that was isolated from estuarine sediments from San Francisco Bay (4) and has been deposited in the Oregon Collection of Methanogens (Oregon Graduate Institute, Portland) as strain OCM 58T. This strain was isolated by using dimethyl sulfoxide as the catabolic substrate (4), but it can also grow on methylamines (13) and methanethiol (8, 9) and grew well in this medium when biotin was added. Strain GS-16T did not grow in mineral medium containing methanol as the sole carbon source (4), but it can also grow on dimethylsulfide (16), although they cannot grow on these substrates, and form traces of ethane from ethyl sulfide (15). Methanogenesis from trimethylamine is inhibited by methyl fluoride (11) and methyl bromide (14), but not by dimethyl ether (11).

Strain GS-16T cells did not grow in a mineral medium (MMSHA medium, which was MSHA medium with Trypticase peptones and yeast extract, and coenzyme M deleted and the concentration of Na,S·9H2O increased to 0.5 g/liter) when 20 mM trimethylamine was added as the sole catabolic substrate, but grew well in this medium when biotin was added.

The cells are mesophilic. In the original characterization of strain GS-16T, Oremland et al. (13) reported that this organism grew poorly at temperatures greater than 28°C in mineral medium containing a catabolic substrate (dimethylsulfide) and vitamins as the only organic compounds and grew fastest at 37°C in medium containing Trypticase peptones and yeast extract. However, during our studies of the minimum organic requirements for growth, strain GS-16T cells grew well at 37°C in defined MMSHA medium containing biotin and a catabolic substrate (20 mM trimethylamine). It is possible that since its original isolation strain GS-16T has acquired the ability to grow at 37°C. Similarly, the initial description of *Methanolobus tindarius* Tindari 3T indicated that the optimum growth temperature for this organism was 25°C (5), but cultures of this strain GS-16T now grow fastest at 37°C (18).

Strain GS-16T is slightly halophilic, growing well in the presence of sodium concentrations between 0.2 and 1.0 M and in the presence of Mg2+ concentrations of 15 mM or greater (4). It grows fastest at pH values between 7.0 and 8.8 (4). W. B. Whitman (University of Georgia, Athens) found that the guanine-plus-cytosine content of the DNA is 40.8 ± 0.3 mol% by performing a liquid chromatographic analysis of the bases (7).

The 16S rRNA sequence of strain GS-16T was compared with the 16S rRNA sequences of many other methanogens (3), and the results indicated that strain GS-16T is most closely related to *Methanohalophilus oregonensis* WALI7. However, the evolutionary distances are great enough that the two strains belong in separate species; these distances are as great as distances between other distinct species (3). The 16S rRNA sequences of both of these strains were similar to the sequence of *Methanobacteium tinidarius* Tindari 3T, suggesting that all three species should be assigned to the genus Methanobacterium, as separate species (3). The results of a DNA hybridization study (1) confirmed that strain GS-16T does not belong in the genus *Methanohalophilus* or in the genus *Methanohalobacterium*.

Strain GS-16T is also physiologically distinct from species of the genus Methanobacterium. In 1989, in *Berger's Manual of Systematic Bacteriology*, Stetter recognized three Methanobacterium species: Methanobacterium tindarius, *Methanobacterium siciliae*, and *Methanobacterium vulcani* (18). Since then, *Methanobacterium siciliae* has been transferred to the genus *Methanosarcina* as *Methanosarcina siciliae* (10). The two remaining species group phylogenetically with *Methanohalophilus oregonensis* (6), which has been transferred to the genus *Methanobacterium* (3). Although the proposal for transfer of *Methanohalophilus oregonensis* has not been formalized, we believe that *Methanobacterium tindarius*, *Methanobacterium vulcani*, and *Methanohalophilus oregonensis* are three currently recognized species whose phylogenies indicate that they should be classified as *Methanobacterium* species. Strain GS-16T differs physiologically from these three species (Table 1), although *Methanobacterium vulcani* has not been well characterized (18) relative to minimal standards (2). Strain GS-16T is most similar to *Methanohalophilus oregonensis* WALI7, but it has a lower pH range (pH 6.8 to 9.0 [4]) than *Methanohalophilus oregonensis* WALI7 (pH 7.7 to 9.5 [6]) and it is less halophilic (strain GS-16T does not grow in the presence of NaCl concentrations greater than 1.2 M NaCl [4], whereas *Methanohalophilus oregonensis* WALI7 grows in the presence of 1.6 M NaCl [6]). Because of the phylogenetic and physiological differences of strain GS-16T, we propose that this

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organism should be placed in a new species, Methanolobus taylorii. Strain GS-16 is the type strain of Methanolobus taylorii.

Methanolobus taylorii sp. nov. Methanolobus taylorii (tay'lor. i.i. N. L. gen. masc. n. taylorii, of Taylor; named for Barrie F. Taylor for his contributions to our understanding of the marine organosulfur cycle and for his mentorship of many students and postdoctoral associates in the field of marine microbiology).

Cells are coccoid bodies 0.5 to 1 μm in diameter and sometimes grow in large clumps. Lysed by detergents. Gram negative. Nonmotile.

Cells grow by forming methane from methylamines, methylysulfides, and methanol. They form methane from dimethylselenide and methylmercury and ethane from diethylsulfide, but they cannot grow on any of these substrates, nor can they grow on methionine, dimethyl disulfide, formate, H₂-CO₃, or acetate.

Cells grow well at 37°C, at pH 7.2 to 8.8, and in the presence of 0.2 to 1.2 M NaCl. Very strictly anaerobic. Biotin and catabolic substrate are the only organic compounds required for growth. The guanine-plus-cytosine content of the DNA of the type strain is 40.8 ± 0.3 mol% (as determined by a chromatographic analysis of bases).

The habitat is estuarine sediments.

The type strain is GS-16 (= OCM 58 = DSM 9005).

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REFERENCES


