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Isolation of a methyl-reducing methanogen outside the Euryarchaeota

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Methanogenic archaea are main contributors to methane emissions, and thus play a crucial role in carbon cycling and global warming. Until recently, methanogens were confined to the phylum Euryarchaeota, but metagenomic studies revealed the presence of genes encoding the methyl coenzyme M reductase complex in other archaeal clades, thereby opening up the premise that methanogenesis is taxonomically more widespread. Nevertheless, laboratory cultivation of these non-Euryarchaeal methanogens was missing to allow the study of their physiology and to corroborate their potential methanogenic capability. Here we describe a thermophilic co-culture from an oil field, containing a single archaeon (strain LWZ-6) belonging to the proposed order *Candidatus* Verstraetearchaeia, together with a H_2-producing *Acetomicrobium* sp. CY-2. Strain LWZ-6, for which we propose the name *Verstraetearchaeum methanopetracarbonis*. Growth, stable labeling tracing experiments, and genomic and transcriptomic analysis demonstrated LWZ-6 is a H_2-dependent methylotrophic methanogen. Nanoscale secondary ion mass spectrometry scanning and lipid stable isotope probing revealed LWZ-6 requires acetate, CO_2, or yeast extract as carbon sources. Although previous metagenomic studies speculated on the fermentative potential of Verstraetearchaeial methanogens, strain LWZ-6 does not ferment sugars, peptides, and amino acids. Its energy metabolism is linked to methanogenesis, with methanol and monomethylamine as electron acceptors and H_2 as electron donor. Comparative (meta)genome analysis revealed that H_2-dependent methylotrophic methanogenesis is a shared trait among Verstraetearchaeia. Our findings corroborate that the diversity of methanogens expands beyond the classical Euryarchaeota and change our current conception of the global carbon cycle.