Microbe of the month: *Methanococcus maripaludis*

Mohd Farid Abdul Halim, Emily H. Hanson, and Kyle C. Costa
Department of Plant and Microbial Biology, University of Minnesota, St. Paul, MN

Methanococcus maripaludis was first isolated in 1983 from the anoxic sediment of a salt marsh in South Carolina, USA. *M. maripaludis* grows by using H_2 or formate to reduce CO_2 to CH_4 and is an autotroph that assimilates CO_2 via the archaeal version of the Wood-Ljungdahl pathway. In some strains, supplementation of growth medium with acetate or propionate can increase biomass yield. This microorganism is strictly anaerobic, has a pleomorphic coccoid shape, and forms translucent, pale yellow, smooth colonies within 3 days on agar plates. The optimum temperature for growth is 38°C with optimal pH between 6.8–7.2.

M. maripaludis has been essential to our understanding of the hydrogenotrophic pathway of methanogenesis and to our understanding of flavin-based electron bifurcation as an energy conservation strategy. In hydrogenotrophic methanogenesis, flavin-based electron bifurcation couples the first, endergonic, CO_2 reducing, step of methanogenesis to the terminal exergonic step of the pathway. ATP generation relies on the production of a Na^+ gradient across the cell membrane during an intermediate step of the pathway, catalyzed by the exergonic transfer of a methyl group by the methyl-tetrahydromethanopterin:CoenzymeM methyltransferase. A Na^+-dependent ATP synthase generates ATP for biosynthesis and other cellular processes.

M. maripaludis grows as single cells or as surface-associated biofilms. The surface layer of *M. maripaludis* is made of N-glycoproteins anchored to the cell membrane. Cells have pili and a polar tuft of archaella, both are glycosylated and structurally similar to the type-IV pilus of bacteria. Archaella allow for weak motility, and pili are necessary for DNA uptake in naturally competent strains. S-layer glycosylation, pili, and archaella are involved in attachment and/or biofilm formation. Most *M. maripaludis* strains exclusively rely on the Sec pathway for translocating proteins across the membrane; however, certain strains that encode extracellular iron-corroding hydrogenases also possess a Tat secretion system.

M. maripaludis is a model organism for studying several additional aspects of archaeal cell physiology. It has a well-established suite of genetic tools including plasmids for markerless mutagenesis, heterologous gene expression and complementation, and CRISPR-based mutagenesis. Tools also exist for inducible gene expression, fluorescent reporters, and transposon mutagenesis. *M. maripaludis* is well-studied as a model of archaeal nitrogen fixation, selenoprotein chemistry, and archaeal sulfur assimilation. *M. maripaludis* has been used to understand basic cellular biology in archaea, such as elucidating the cell surface mechanism of pilus and archaellar assembly, glycosylation, and function.

As a methanogen, *M. maripaludis* is important to the complete degradation of organic matter in anoxic environments. In oxidant deplete sediments, primary and secondary fermentation generate H_2, formate, acetate, and CO_2, the substrates used by methanogens. *M. maripaludis* grows with secondary fermenters to drive forward the thermodynamically unfavorable fermentation of organic acids, a process known as syntrophy. In the absence of a bacterial partner producing H_2, some strains of *M. maripaludis* can grow by the oxidation of Fe^0; this is accomplished by the

extracellular oxidation Fe^0 to Fe^{2+} with the concomitant reduction of H^+ to H_2. Cellular H_2 consumption accelerates this reaction and enhances Fe^0 corrosion in anoxic environments.

Domain: Archaea
Phylum: Euryarchaeota
Class: Methanococci
Order: Methanococcales
Family: Methanococcaceae
Genus: *Methanococcus*
Species: *Methanococcus maripaludis*

Key Facts:

M. maripaludis is a methanogen. Methanogens are strict anaerobes from the domain Archaea and produce methane gas as a metabolic product.

M. maripaludis can be found in anoxic, marine sediments.

M. maripaludis is a polyploid with a single circular chromosome of ~33% G+C content containing ~1,700 protein-coding genes. Some strains contain plasmids.

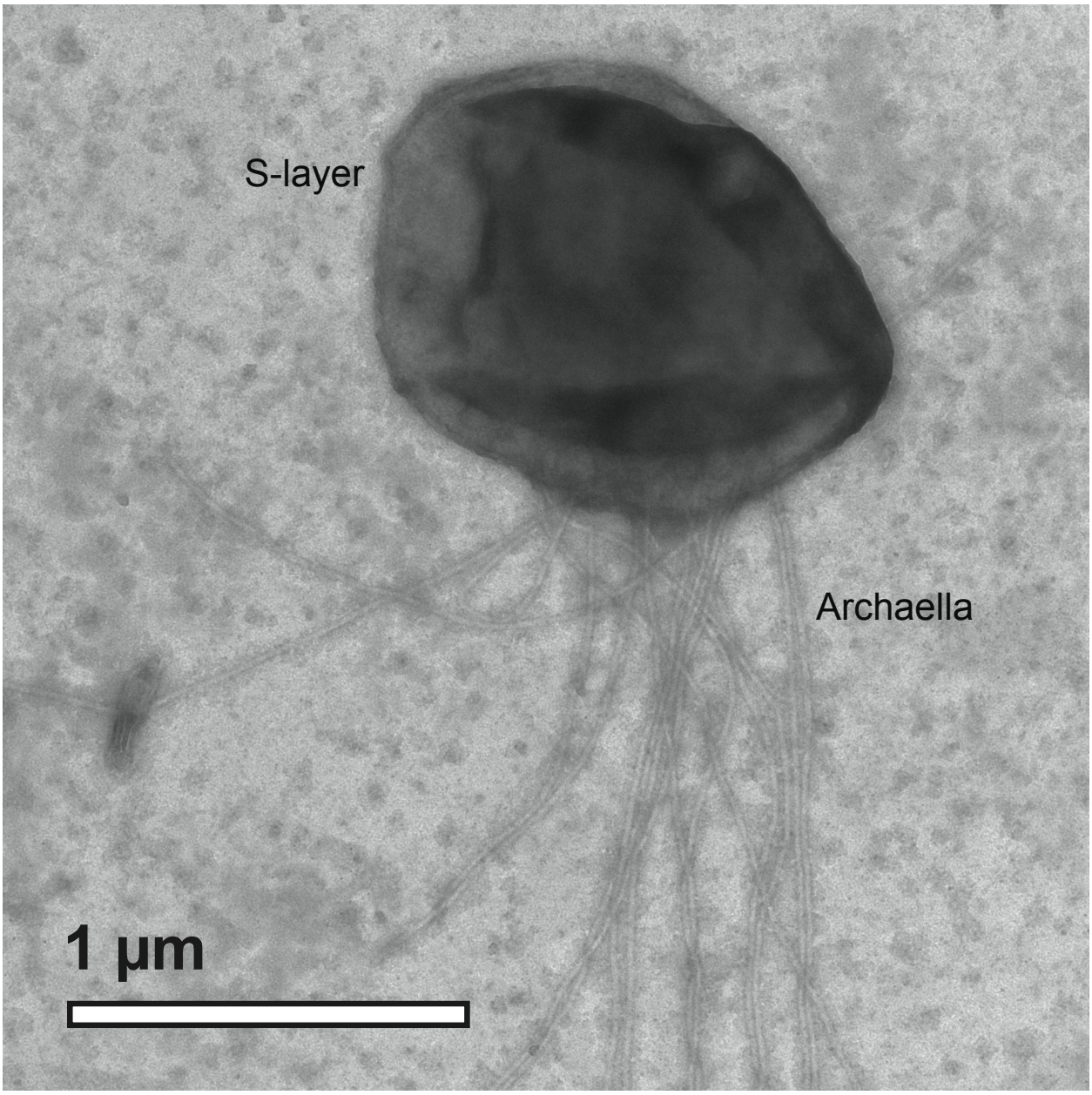
M. maripaludis is a model organism to study archaea due to its genetic tractability, fast and reproducible growth, and ability to form colonies on agar plates.

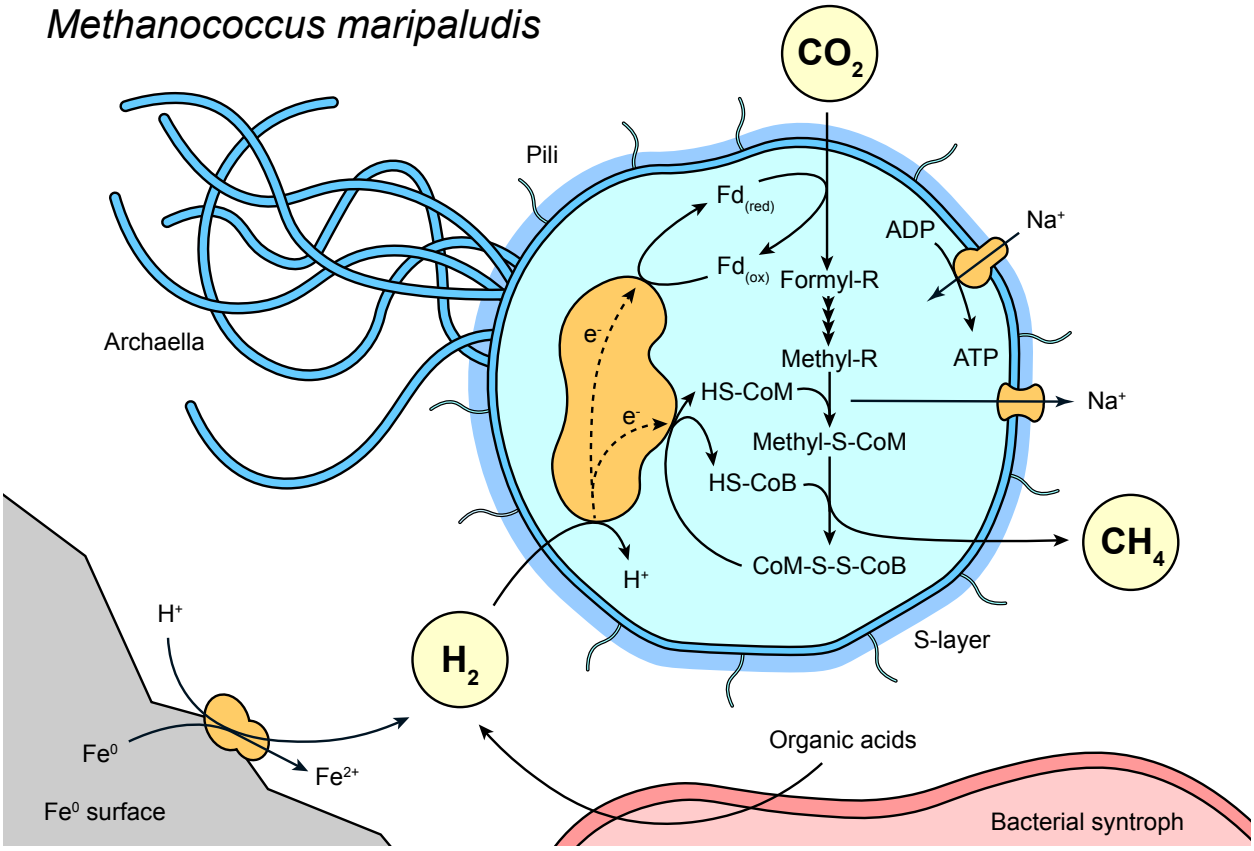
M. maripaludis is capable of nitrogen fixation.

Acknowledgements: Research in the author's lab is supported funded by grants from the U.S. Department of Energy, Office of Science, Basic Energy Sciences (DE-SC0019148) and the U.S. National Science Foundation (MCB-2148165).

References:

1. Jones, W.J. *et al.* (1983) Characterization of *Methanococcus maripaludis* sp. nov., a new methanogen isolated from salt marsh sediment. *Arch. Microbiol.* 135, 91–97
2. Rother, M. and Quitzke, V. (2018) Selenoprotein synthesis and regulation in Archaea. *Biochim. Biophys. Acta* 1862, 2451–2462
3. Pohlschroder, M. *et al.* (2018) Archaeal cell surface biogenesis. *FEMS Microbiol. Rev.* 42, 694–717
4. Fonseca, D.R. *et al.* (2023) Random transposon mutagenesis identifies genes essential for transformation in *Methanococcus maripaludis*. *Mol. Genet. Genomics* 298, 537–548
5. Leigh, J.A. and Dodsworth, J.A. (2007) Nitrogen regulation in bacteria and archaea. *Annu. Rev. Microbiol.* 61, 349–377
6. Tsurumaru, H. *et al.* (2018) An extracellular [NiFe] hydrogenase mediating iron corrosion is encoded in a genetically unstable genomic island in *Methanococcus maripaludis*. *Sci. Rep.* 8, 15149
7. Costa, K.C. and Whitman, W.B. (2023) Model organisms to study methanogenesis, a uniquely archaeal metabolism. *J. Bacteriol.* 205, e0011523
8. Milton, R.D. *et al.* (2018) *Methanococcus maripaludis* employs three functional heterodisulfide reductase complexes for flavin-based electron bifurcation using hydrogen and formate. *Biochemistry* 57, 4848–4857
9. Li, J. *et al.* (2023) Genetic and metabolic engineering of Methanococcus spp. *Current Research in Biotechnology* 5, 100115
10. Zhang, W. *et al.* (2023) Internal transcription termination widely regulates differential expression of operon-organized genes including ribosomal protein and RNA polymerase genes in an archaeon. *Nucleic Acids Res.* 51, 7851–7867


S-layer
Archaella
1 µm


Methanococcus maripaludis
CO2
Pili
Archaella
Fd(red)
Fd(ox)
Formyl-R
ADP
Na+
ATP
Methyl-R
HS-CoM
Na+
Methyl-S-CoM
HS-CoB
CoM-S-S-CoB
CH4
e-
e-
H+
H2
S-layer
H+
Fe0
Fe2+
Fe0 surface
Organic acids
Bacterial syntroph