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Determining controls on hydrogen isotope fractionation in archaeal tetraether lipids in a thermoacidophilic archaeal heterotroph

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Hydrogen from water plays a critical role in nearly all metabolic processes and is incorporated into all biomolecules, often with distinctive isotope fractionations that track processes. The hydrogen isotope composition (δ^2H) of biomolecules can be used as proxy for past hydrology and ecology. While most research has focused on plant waxes, microbial lipids are of interest because lipid-H can retain its original isotopic composition for up to 10^8 years [1]. Bacterial lipids, however, are unreliable proxies for paleohydrology because their lipid-water fractionation varies widely (from -150 to +380‰) and their δ^2H composition mainly reflects central metabolism [2]. The potential for archaeal lipid δ^2H to indicate the δ^2H of water during growth is understudied. Here, we examine the H isotope fractionation between glycerol dialkyl glycerol tetraether membrane lipids (GDGTs) and water with the model thermoacidophilic archaeal heterotroph, *Sulfolobus acidocaldarius*, over different pH, temperature, and aeration rates [3]. Separately, we vary the H-isotope composition of the growth water and of the electron donor and carbon source (D-glucose), which allows us to estimate the relative contributions of H from protons in water and organic substrates to the final tetraether lipid products. The ether bonds in harvested GDGTs were chemically cleaved and the resulting biphytanal chains were analyzed for δ^2H via GC-pyrolysis-IRMS. We show that biphytane δ^2H is strongly correlated water δ^2H and all biphytanes are ^2H-depleted relative to growth water. We will discuss factors contributing to variation in fractionation factors including culture conditions (i.e., temp, pH, and O_2). Ultimately, these experiments help develop an interpretive framework for the δ^2H of archaeal lipid biomarkers in modern and ancient systems that can be validated with environmental samples across similar gradients in major geochemical parameters.

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[3] Cobban et al. (2020), *Environmental Microbiology* 22.9: 4046-4056.