**On the origins of crenarchaeol:**

*Environmental factors controlling distribution in hot springs*

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Glycerol dibiphytanyl glycerol tetraethers (GDGTs) are membrane-spanning lipids of Archaea that are ubiquitous in hot spring, ocean, lake, and soil environments.¹,² These lipids allow for microbial acclimitization to environmental stress and serve as the basis for important paleotemperature proxies.¹ The number of cyclopentyl rings in the core structure of archaeal GDGTs change in response to temperature, pH, and oxidant load in both environmental samples and in cultured marine and acidophilic hot spring strains.¹,² Archaea from acidic hot springs can produce GDGT moieties with up to eight cyclopentyl rings (GDGT-0 to -8). Biophysical models show that synthesizing more cyclopentyl rings promotes tighter membrane packing and decreased permeability, enabling archaea to occupy hotter, more acidic, and more oxidizing environments.³ A unique GDGT found in both marine and hot spring archaea, *crenarchaeol*, contains four cyclopentyl and one cyclohexyl ring. While the function of this lipid in archaeal membranes is not well understood, it has been proposed that the cyclohexyl ring allowed archaea adapted to acidic, high temperature hot springs to adapt to cooler and circumneutral waters, eventually allowing for diversification into oceanic environments.⁴ To improve the understanding of crenarchaeol’s function in membranes, its role in archaeal evolution, and its distribution in the geologic record, we are quantifying the relationship between pH, temperature, redox, and other parameters with crenarchaeol abundance in the natural thermal springs of Yellowstone National Park where these parameters vary widely. We evaluate our data in the context of a comprehensive compilation of previously reported GDGT data from hot spring environments, focusing particularly on crenarchaeol, to examine the environmental factors that likely control its distribution. These results shed new light on the role of crenarchaeol in the diversification of archaea into lower temperature, circumneutral environments.