Abstract:
Fossilized sterols, or steranes, can be found in rocks up to 1.6 billion years old and are used as biomarkers for past eukaryotic life. Specific modifications to the sterol side chain, such as alkylation at the C-24 position, remain unique to certain eukaryotic lineages and function as more specific biomarkers. One such side chain alkylated sterane, 24-isopropylcholestane (24-ipc), has been used as a biomarker for demosponges as they are the only extant organisms known to produce large amounts of 24-isopropyl sterols. However, this interpretation has been challenged as other potential sources of 24-ipc have been recently identified and the biochemical mechanism behind the propylation at C-24 remains unknown. In this study, we experimentally characterized a variety of sterol 24-C-methyltransferase (SMT) homologs, the only enzymes known to alkylate at the C-24 position. We show that sponge SMTs are functional and can promiscuously alkylate the sterol side chain. However, no analyzed sponge SMTs were capable of producing 24-propyl sterols. Further, we demonstrate that SMTs from bacterial sources are functional and can produce 24-propyl sterols - the first instance demonstrating alkylation at the C-24 position by bacterial proteins. Taken together, our study indicates that alternative sources of 24-ipc are possible and that a more nuanced interpretation of 24-ipc biomarkers in the rock record is necessary.

Title: Synthesis of sponge sterol lipid fossils: Who makes what and why does it matter?

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