EUKARYOTIC STEROL BIOMARKERS: PRODUCTION AND FATE IN A LAMINATED MICROBIAL MAT

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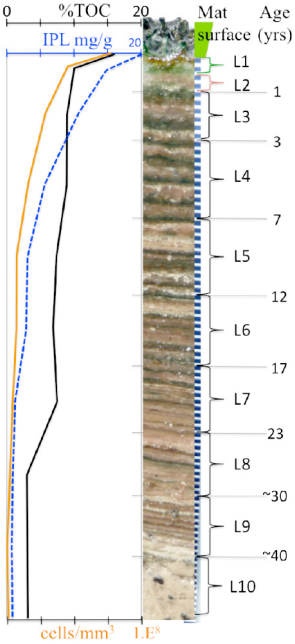
Introduction:

The products of isoprenoid biosynthesis are numerous and serve as important biomarkers for many groups of organisms. Cyclic triterpenoids are particularly well preserved in sediments and provide an organic molecular record, which spans more than two billon years. Sterols and hopanoids are of central importance to the evolution of cellular membrane function [1]. Sterols are synthesized almost exclusively by eukaryotes and therefore are important chemical fossils for this lineage. The hopanoids are equally important as biomarkers for bacteria. Both the hopane and sterol carbon skeleton share a common biosynthetic source, squalene. This C30, acyclic, tail-to-tail triterpene is directly cyclized to hopane (an anaerobic process) in bacteria. However in eukaryotes squalene is first converted to an epoxide (oxygen-requiring) and then cyclized to a C30 proto-sterol, either lanosterol or cycloartenol. Further structural modifications require a total of eleven molecules of oxygen for synthesis of a C27 (cholesterol), C28 (ergosterol) or C29 (sitosterol) molecule [2]. The sterol biosynthetic pathway is well documented, and is a useful tool to understand the emergence and evolution of eukaryotes. Phylogenomic reconstructions from completed eukaryotic and bacterial genomes are now possible [3, 4] and continue to shed light on this evolutionary process. Understanding the role of micro-eukaryotes in sterol synthesis in microbial ecosystems provides a guide to bringing these two records together.

Photosynthetic microbial mats played important roles in the early biosphere. Cyanobacterial oxygenic photosynthesis has dominated global biological productivity for billions of years and had profound consequences for the trajectory of planetary evolution, particularly the evolution of higher plants and animals. Phototrophic mats have probably hosted and recorded many key evolutionary advances in the physiologies of mat-inhabiting microorganisms. Modern microbial mats are dynamic ecosystems. A challenge to interpreting the organic record is to understand the processes of synthesis, modification and deposition of organic biomarkers such as hopanoids and sterols in these microbial ecosystems.

The flat-laminated microbial mats from Pond 4 of the Guerrero Negro saltworks are a well-studied ecosystem [5, 6], but limited information on the eukaryotic population or the expected sterol contribution is available. These mats are primarily composed of prokaryotic microbes and eukaryotes are relatively minor populations. Pennate diatoms of the *Nitzchia* and *Navicula* genera in the water column form a flocculent layer on the mat surface particularly in the summer months. Phylogenetic analysis of eukaryotic sequences have identified abundant nematodes of the *Monhysteridae* and *Rhabdolaimidae* families to a depth of 3.7 cm with some arthropods at the very surface (1-2 mm) [7]. The only photosynthetic group identified was a stramenopile *Nannochloropsis gaditana*.

We have collected a mat with underlying core from Pond 4 for lipid biomarker analysis over the full 8 cm depth. Initial analyses have demonstrated that lipid biomarkers sustained substantial diagenetic alteration throughout a major anoxic portion of the core. In the surface sample (1-2 mm depth), the major sterol was a fully saturated C27 molecule (cholestanol). Other major sterols in this layer are cholesterol (Δ5 monoene), 24-methylcholestanol, 24-methylcholesterol, 24-ethylcholestanol and 24-ethylcholesterol. The relative abundances of the three saturated sterols (C27, C28, C29) increased with depth. Small amounts of cycloartenol and several 4-methylsterols were also present in the surface. However, below the uppermost 4 mm of depth, the abundance of cycloartenol and methylated sterols increased dramatically. The 4-10 mm depth sample (anoxic) contained the highest level of proto-like sterol (4,4,14-trimethylsterols) normally associated with de novo sterol synthesis. Increased levels of sterones and other sterol diagenetic products with depth were also documented.



GN mat and core over 80 mm depth, showing analyzed sample intervals and approximate age. Blk line total organic carbon, Blue membrane acyl/alkyl lipid, red derived cell density ty.

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