Cholesterol synthesis by a bacterium

PAULA V. WELANDER[1*], ALYSHA K. LEE[1], AND JEREMY H. WEI[1]

[1]Department of Earth System Science, Stanford University, 473 Via Ortega, Stanford, CA 94305, USA (*correspondence: welander@stanford.edu)

Sterol lipids are cyclic triterpenoids that can be preserved in ancient sedimentary rocks as sterane hydrocarbons. The majority of sterane hydrocarbons detected in ancient sediments are typically the diagenetic products of complex steroids such as cholesterol or those with modified side chains such as stigmasterol. To date, no bacterium has been shown to produce cholesterol or sterols with side-chain alkylations and, therefore, these complex sterols are thought to be unique to eukaryotes. Thus, sterane hydrocarbons are considered robust geological proxies, or biomarkers, for the occurrence of microbial eukaryotes throughout Earth's history. However, here we show the production of one complex sterol, cholesterol, by a δ-Proteobacterium, *Enhygromyxa salina*. *E. salina* is a marine, aerobic heterotroph that was isolated off the coast of Japan. Like many myxobacteria, it can differentiate into complex fruiting bodies under certain conditions and grows primarily as a gliding swarm on solidified agar medium. Lipid analyses of this organism revealed the production of zymosterol, desmosterol, and cholesterol. Bioinformatics analyses of the *E. salina* genome revealed the presence of several cholesterol biosynthesis genes although the eukaryotic genes for demethylation at the C-4 position were missing. Using comparative genomics and heterologous expression we identified C-4 sterol demethylase genes in this organisms that are distinct from what is observed in eukaryotes. This study highlights the power of linking genomics and lipid analyses to identify lipid biomarkers and their biosynthetic pathways in diverse organisms. In addition, this finding raises the possibility that the production of complex sterols may not be exclusive to eukaryotes and may occur in other uncultured marine bacteria.