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# D/H Ratios in Lipids as a Tool to Elucidate Microbial Metabolism

Reto S. Wijker and Alex L. Sessions

Division of Geological and Planetary Sciences, California Institute of Technology, Pasadena, United States (rwijker@caltech.edu)

Large D/H fractionations have been observed in the lipids and growth water of most organisms studied today. These fractionations have generally been assumed to be constant across most biota because they originate solely from isotope effects imposed by the highly conserved lipid biosynthetic pathway. Recent data is illustrating this conclusion as incomplete. Lipids from field and laboratory samples exhibit huge variations in D/H fractionation. In environmental samples, lipids vary in $\delta$D by up to 300 ‰ and in laboratory cultures the documented variation is up to 500 ‰ within the same organism. Remarkably, the isotope fractionation appears to be correlated with the type of metabolism employed by the host organism. However, the underlying biochemical mechanisms leading to these isotopic variations are not yet fully understood.

Because the largest proportion of H-bound C in fatty acids is derived directly from NADPH during biosynthesis, the original hypothesis was that large differences in the isotopic composition of NADPH, generated by different central metabolic pathways, were the primary source of D/H variation in lipids. However, recent observations indicate that this cannot be the whole story and lead us to the conclusion that additional processes must affect the isotope composition of NADPH. These processes may include the isotopic exchange of NADPH with water as well as fractionation of NADPH by transhydrogenases, interconverting NADH to NADPH by exhibiting large isotope effects.

In this project, our objective is to ascertain whether D/H fractionation and these biochemical processes are correlated. We investigate correlations between cellular NADPH/$NADP^+$ as well as NADH/$NAD^+$ pool sizes and the D/H fractionation in a set of different microorganisms and will present the trends here. Our results will contribute to a more comprehensive understanding of the basic biological regulations over D/H fractionation and potentially enables their use as tracers and proxies across earth and biological sciences.