ISOTOPE DISCRIMINATION DURING PHOTOSYNTHESIS REMAINED CONSTANT ACROSS THE PHANEROZOIC

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Abstract

Photosynthesis is among the most important evolutionary innovations in Earth history. It has formed the foundation for most life on Earth today, oxygenated our planet, revolutionised the carbon cycle, and enabled aerobic respiration that eventually led to the evolution of large complex organisms. The most widespread oxygenic photosynthesis pathway involves fixing carbon through the enzyme Ribulose 1,5-bisphosphate carboxylase-oxygenase (RuBisCO) that converts CO_2 into sucrose via the Calvin-Benson cycle. During this process, RuBisCO preferentially uptakes $^{12}CO_2$ over $^{13}CO_2$, resulting in stable carbon isotopic fractionation of up to 25-28 ‰.

Despite its fundamental importance to the operation of our planet, it is not clear whether RuBisCO has always operated the same; in other words, has photosynthetic carbon isotope discrimination undergone major evolutionary changes or remained constant across the geologic record? Supporting the argument for change, marine photoautotrophs have experienced multiple step-changes and long-term evolutionary restructurings, atmospheric CO_2/O_2 ratios have fluctuated dramatically, and the carbon cycle has been radically transformed (e.g., by the evolution of land plants) across the Phanerozoic (the last 541 million years), all could theoretically impact the stable carbon isotopic fractionation associated with photosynthesis by RuBisCO. Supporting the argument for no change, the carbon isotopic composition ($\delta^{13}C$) of marine bulk organic matter has remained remarkably consistent over geologic time (e.g., Garcia et al., 2021) and the reconstruction of a Precambrian-age, ancestral RuBisCO gene resurrected in modern cyanobacteria shows isotopic discrimination within modern $\delta^{13}C$ range (Kędzior et al., 2022). However, the $\delta^{13}C$ of bulk organic matter may be influenced by source heterogeneity and differential preservation and is thus too crude to robustly infer changes in the operation of RuBisCO and photosynthetic pathways. Here, we resolve the concerns regarding bulk $\delta^{13}C$ data by applying compound specific isotope analyses of the organic compounds pristane and phytane to determine whether the photosynthetic carbon isotope discrimination has remained constant across the Phanerozoic.

We find that the $\delta^{13}C$ of pristane and phytane in 675 marine oils and sediments parallel one another across the Phanerozoic (Witkowski et al., 2018; Witkowski et al., in preparation). These results provide evidence for three key findings: 1) The remarkable consistency between $\delta^{13}C$ values for pristane and phytane confirms their common source to be chlorophyll, the pigment required for oxygenic photosynthesis, and reveals that oft-invoked alternative sources (e.g., Archaea) are uncommon during the Phanerozoic. 2) Our calculated stable carbon isotopic fractionation associated with CO_2-fixation (ε_p) never surpasses 25 ‰, the maximum fractionation in modern algae, providing evidence for uniformitarian controls for photosynthesis. 3) Our ε_p-based calculations show that atmospheric concentrations of CO_2

($p CO_2$) closely track other proxy-based $p CO_2$ records (Foster et al., 2017; Rae et al., 2021), suggesting that major changes in the stable carbon isotopic composition of photoautotrophs is due to environmental, not evolutionary, factors.

Together, this evidence suggests that controls on isotope discrimination during photosynthesis remained constant over the past 541 million years.

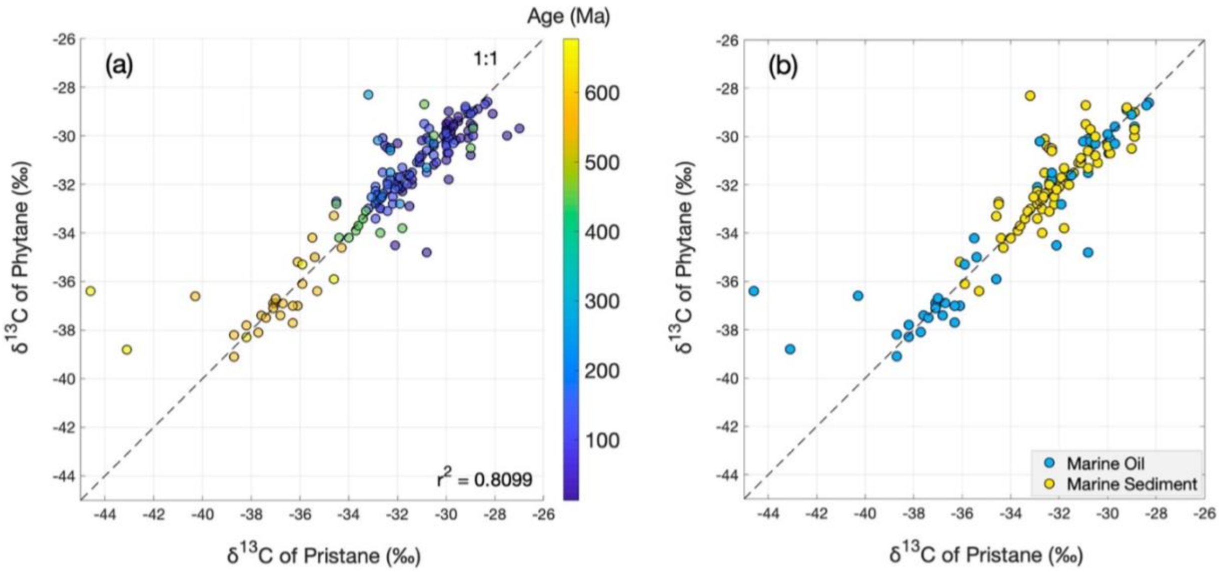

Figure 1 *The $\delta^{13}C_{phytane}$ (x-axis) and $\delta^{13}C_{pristane}$ (y-axis) from the same site and depth, with 1:1 line (dotted) and linear model fitted to the data (solid black line), r^2 value of 0.6013. Samples sorted by (a) age and (b) sample matrix (marine oil in blue vs. marine sediment in yellow).*

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