

## Clarifying the influence of water availability and plant types on carbon isotope discrimination by C3 plants

We are pleased that the results of Kohn (1) so strongly support our findings published earlier in PNAS (2). Both studies (1, 2) analyzed published measurements of the carbon isotope composition of plants and quantified relationships between isotopic fractionation and environmental factors at large spatial scales. These relationships will be useful when interpreting archives of plant-derived carbon, such as fossils, soil organic matter, and plant biomarkers. Kohn (1) emphasized differences between the two papers, particularly the coefficients and proportion of variance explained in regression models, but the major findings were the same: water availability [estimated by mean annual precipitation (MAP)] explains most of the observed variability in  $\delta^{13}\text{C}$  values for modern C3 plant leaves. We disagree with Kohn (1) on two aspects of data analysis: (i) including data from very arid sites in the regression and (ii) using mean carbon isotope values for each site.

We reanalyzed the data from Kohn (1) and found that the multiple regression results are very similar to our own results (Table 1). We use  $\log_{10}\text{MAP}$  instead of Kohn's (1) predictor [ $\log_{10}(\text{MAP} + 300)$ ], which condensed and obscured variation at low MAP. We transformed altitude to its square root to improve normality, a key assumption of linear regression. Very arid regions were not included in our study. In Kohn's data (1), very arid regions (<90 mm/y MAP) behaved as statistical outliers (i.e., >1.5 times the interquartile range from the lowest quartile), consistently with positive residuals. MAP only approximates water availability during plant growth in all settings (2), and water availability in dry regions is strongly influenced by soil moisture, topographic exposure, plant water, and life strategies (3), factors that are independent of MAP. Alternatively, very arid regions can be identified in the past using floral, faunal, biomarker, and pedological data. For all models from both papers, when full uncertainties in data, slope, and intercept were considered, propagated errors in MAP predictions were too large to be of practical use in paleoenvironment or climate reconstructions (1, 2).

By combining data for all plant types into a single mean  $\delta^{13}\text{C}$  value for each site, Kohn (1) assumed that available data for a geographic locality accurately represented both community composition and diversity across the wide range of MAP reported. However, sources of ancient plant carbon are not equally represented in mineral and organic archives. Fossil tooth enamel reflects the weighted values of an animal's diet, including different plant functional types, tissues, and canopy position. Soil organic matter is dictated by litter flux, which is dominated by the canopy (4), and it can reflect differences in canopy closure (5). We reported isotopic data along with plant taxa and functional types. Such an approach permits broader applications of leaf  $\delta^{13}\text{C}$ -MAP relationships to different plant carbon archives and strengthens interpretations when fossil evidence indicates changes in plant community. We disagree with Kohn's (1) use of mean values; our information on vegetation and plant functional type allowed greater insights to plant fractionation and ultimately, enabled more detailed reconstructions of paleodiet, paleovegetation, and paleoclimate.

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**Table 1. Multiple regression analysis results comparing data from Diefendorf et al. (2) with data from Kohn (1)**

Data source	Model type ( <i>n</i> )	$R^2$	Intercept	Log (MAP) coefficient	Altitude <sup>1/2</sup> coefficient
Kohn (1)	Site means (480)	0.50	13.07 ( $\pm 0.38$ )	2.60 ( $\pm 0.13$ )	-0.02 ( $\pm 0.003$ )
Kohn (MAP > 90 mm/y) (1)	Site means (461)	0.55	9.18 ( $\pm 0.53$ )	3.91 ( $\pm 0.18$ )	-0.01 ( $\pm 0.003$ )
Diefendorf et al. (2)	Species site means (502)	0.61	9.31 ( $\pm 0.90$ )	4.20 ( $\pm 0.26$ )	-0.06 ( $\pm 0.007$ )
Diefendorf et al. (2)	Site means (69)	0.73	10.03 ( $\pm 1.39$ )	3.94 ( $\pm 0.45$ )	-0.06 ( $\pm 0.009$ )