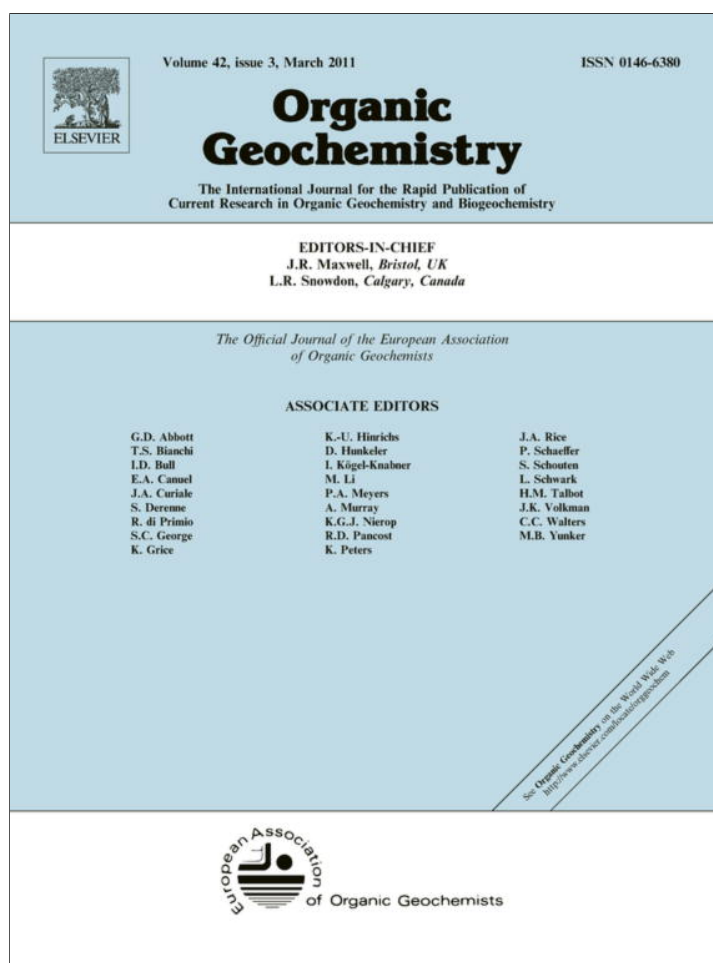


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## Variation in *n*-alkane $\delta D$ values from terrestrial plants at high latitude: Implications for paleoclimate reconstruction

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### ABSTRACT

The molecular hydrogen isotope composition ( $\delta D$ ) of leaf waxes from terrestrial plants is increasingly used to infer hydrological characteristics of ancient high latitude climates. Analysis of the hydrogen isotope composition of *n*-alkanes ( $\delta D_{n\text{-alkane}}$ ) from a global dataset of individual plants growing at low and middle latitudes indicates that plant ecological life form is an important factor in determining the hydrogen isotope fractionation. However, environmental and biological controls of high latitudinal leaf wax  $\delta D$  values are poorly understood because of a lack of  $\delta D$  records from modern flora in these regions. We previously noticed smaller apparent hydrogen isotope fractionations between *n*-alkanes and environmental water ( $\epsilon_{\text{alk-water}}$ ) in deciduous trees growing at high latitudes ( $>59^\circ\text{N}$ ; Liu, W.-G., Yang, H., 2008. Multiple controls for the variability of hydrogen isotopic compositions in higher plant *n*-alkanes from modern ecosystems. *Global Change Biology* 14, 2166–2177.) To further examine these issues, we measured  $\delta D_{n\text{-alkane}}$  from a variety of plants that inhabit high latitude environments and added critically needed leaf wax  $\delta D$  data from grass and herbs to the existing global  $\delta D_{n\text{-alkane}}$  database. Inclusion of these new data with the existing global dataset ( $n = 408$ ) confirms plant ecological life form as an important control for leaf wax  $\delta D$  variation for terrestrial plants living at high latitudes. Our results suggest that, while precipitation  $\delta D$  is captured in these high latitude plants, physiological characters such as leaf area, venation pattern and hydraulic system, that enhance transpiration rate during summer growth, may impart  $\delta D_{n\text{-alkane}}$  differences among plants with different ecological life forms.

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### 1. Introduction

The hydrogen isotopic composition of plant leaf waxes preserved in sediments is becoming an increasingly important tool in the reconstruction of ancient hydrologic conditions. However, records of the hydrogen isotope composition of plant *n*-alkanes ( $\delta D_{n\text{-alkane}}$ ) from high latitudes are limited and our understanding of the factors affecting the apparent hydrogen isotope fractionation between *n*-alkanes and environmental water ( $\epsilon_{\text{alk-water}}$ ) is still developing. An improved understanding of  $\epsilon_{\text{alk-water}}$  ultimately comes from the study of the  $\delta D_{n\text{-alkane}}$  variation in identifiable individual plants because the biology and ecology of the plants are known, and physiological experiments can be designed in greenhouses or growth chambers to test the cause and consequence of observed  $\delta D$  variation.

Early studies of  $\delta D_{n\text{-alkane}}$  values of living plants focused on documentation of  $\delta D$  variation from various types of plants, the distribution

of  $\delta D$  values for lipids with different chain length and their relationship with  $\delta^{13}\text{C}$  values for the same compounds (Sessions et al., 1999; Chikaraishi and Naraoka, 2003; Yang and Huang, 2003; Bi et al., 2005). Subsequent studies explored the effects of *n*-alkane synthesis (Chikaraishi et al., 2004, 2005), seasonality (Sessions, 2006; Sachse et al., 2009), plant ecological life forms (Liu et al., 2006), same site variation (Hou et al., 2007a), rooting patterns (Krull et al., 2006) and climate gradient (Sachse et al., 2006; Duan and Wu, 2009). Using the available global *n*-alkane  $\delta D$  record, Liu and Yang (2008) identified and ranked multiple controlling factors and their impact on the variation of  $\delta D$  values of higher plant waxes. They also observed a smaller  $\epsilon_{\text{alk-water}}$  from deciduous trees living at high latitude and anticipated D depleted hydrogen isotope values for grass leaf waxes from the same regions. A recent greenhouse experiment using deciduous conifers grown under a simulated Arctic environment further illustrated high latitude effects on hydrogen isotope fractionation caused by enhanced leaf transpiration under 24 h continuous light illumination (Yang et al., 2009).

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Here, we present  $\delta D_{n\text{-alkane}}$  records of grasses and other plants from high latitude regions ( $>59^\circ\text{N}$ ). These new data, in combination with published records, are used to examine  $\delta D_{n\text{-alkane}}$  differences between plants living at high and mid/low latitudes, with a focus on the influence that ecological life form has on determining  $\delta D_{n\text{-alkane}}$  values. Our results have significant implications for the application of  $n\text{-alkane}$   $\delta D$  data as a paleoclimate proxy and in particular, to high latitude paleohydrology.

## 2. Materials and methods

We analyzed the  $\delta D_{n\text{-alkane}}$  values of 26 leaf samples collected from a range of high latitude environments ( $>59^\circ\text{N}$ ; Table 1). Among these, 15 were collected from Northern Europe (Sweden and Finland) and the rest were from North America (Alaska and Canadian Arctic). Except for one  $C_4$  plant (*Zea mays* L.) from Sweden and one horsetail (*Equisetum pretense* Ehrhart) from Alaska, the samples derive from 16 grass/herb species and 8 tree/shrub species that utilize the  $C_3$  photosynthetic pathway (Table 1). Because of the low occurrence and the difficulty in collecting fresh grass species at high latitude, we supplemented our samples with six herbarium grass specimens collected from Finland in 1983.

Leaves were cleaned with distilled water to remove dust particles, dried at  $40^\circ\text{C}$  and extracted with dichloromethane (DCM). Hydrogen isotope analysis was performed using a HP 6890 gas chromatograph, interfaced via a high temperature conversion interface to a Thermo Finnigan MAT 253 mass spectrometer (Hilkert et al., 1999). The gas chromatography (GC) temperature program was:  $80^\circ\text{C}$  (1 min) to  $180^\circ\text{C}$  at  $3^\circ\text{C}/\text{min}$  and then to  $300^\circ\text{C}$  (held 10 min). Separated components were converted to  $\text{H}_2$  by way of a pyrolysis reactor at  $1445^\circ\text{C}$ . The  $\delta D$  values were calibrated against  $\text{H}_2$  reference gas calibrated via a co-injected laboratory working standard ( $n\text{-C}_{16}$  and  $n\text{-C}_{30}$  alkanes, and  $5\alpha\text{-androstane}$ ; isotopic ratio values determined offline by A. Schimmelmann, Biogeochemical Laboratory at Indiana University). Each sample was analyzed three times;  $\text{H}_3$  factors were calculated daily using the same  $\text{H}_2$  reference gas. The precision of isotopic measurements of  $\text{H}_2$  reference gas after  $\text{H}_3$  factor correction was  $1\text{‰}$  or better. Analytical error was  $<4\text{‰}$  for samples;  $\delta D$  values are expressed relative to the VSMOW standard using Eq. (1).

$$\delta D = 1000 \times \left[ \frac{(^2\text{H}/^1\text{H})_{\text{sample}}}{(^2\text{H}/^1\text{H})_{\text{VSMOW}}} - 1 \right] \quad (1)$$

Leaf wax D/H ratio values from  $C_{27}$ ,  $C_{29}$  and  $C_{31}$   $n\text{-alkanes}$  are highly correlated (Liu and Yang, 2008), so we used an amount-weighted mean  $\delta D$  value of the three compounds for our interpretations, as practised in other recent studies (e.g. Smith and Freeman, 2006; Feakins and Sessions, 2010). Pearson correlation was conducted to investigate various correlations between  $\delta D$  values of  $n\text{-alkanes}$  and various geographic and environmental factors. One-way ANOVA tests were used to examine the statistical significance of different relationships;  $\epsilon_{\text{alk-water}}$  was calculated from the following relationship:

$$\epsilon_{\text{alk-water}} = 1000 \times \left[ \frac{(\delta D_{n\text{-alkane}} + 1000)}{(\delta D_{\text{water}} + 1000)} - 1 \right] \quad (2)$$

## 3. Results and discussion

### 3.1. Distribution of leaf wax $\delta D$ in high latitude plants

Our new data (Table 1) indicate that  $\delta D_{n\text{-alkane}}$  values from high latitude plants fall into two distinct groups based upon plant ecological life form, including woody trees/shrubs and grasses/herbs. Grasses and herbs tend to possess relatively lower  $\delta D$  values relative to trees/shrubs at given latitudes (Fig. 1). The  $\delta D_{n\text{-alkane}}$  values from grasses and herbs range from  $-171\text{‰}$  to  $-307\text{‰}$ , with the

lowest value for *Alopecurus alpinus* J.E. Smith, a  $C_3$  grass from Axel Heiberg Island in Canada at  $80^\circ\text{N}$ . In contrast, the values for woody plants span a range from  $-151\text{‰}$  in *Castanea sativa* Mill ( $60^\circ\text{N}$ ) to  $-262\text{‰}$  from *Salix arctica* Pall. (woody deciduous shrub from Axel Heiberg Island). Except for one  $C_4$  species (*Zea mays* L.) with a  $\delta D$  value of  $-228\text{‰}$  ( $59^\circ\text{N}$ ), all the species are  $C_3$  plants. Significant  $\delta D$  variation (up to  $89\text{‰}$ ) is evident from the range of plants evaluated at the same site.

These data, in combination with published records from high latitude deciduous plants (Sachse et al., 2006), indicate that  $\delta D_{n\text{-alkane}}$  values track the hydrogen isotope compositions of precipitation as the first order control, which is further modified by plant ecological life form (Fig. 1). While  $\delta D_{n\text{-alkane}}$  signals from plants of both life forms record precipitation  $\delta D$ , these correlations have different slopes and can be clearly separated into two groups: grass vs. woody plants. The pattern and range of  $\delta D_{n\text{-alkane}}$  differences between grass and trees are consistent with our previous observations from middle and low latitudes (Liu et al., 2006; Liu and Yang, 2008). Regression analysis revealed three general patterns: (i) Correlation  $R^2$  values were similar whether modeled annual or summer precipitation  $\delta D$  values were applied; (ii) correlation can be significantly improved if different plant ecological life forms (trees/shrubs vs. grasses/herbs) were considered. (3) A better correlation was obtained between  $\delta D_{n\text{-alkane}}$  values from grasses/herbs and precipitation than trees/shrubs or integrated trees mixed with grasses.

### 3.2. Global $\delta D_{n\text{-alkane}}$ distribution

Fig. 2 displays the correlations between available  $n\text{-alkane}$   $\delta D$  values from identifiable living plants ( $n = 408$ ) and modeled mean annual precipitation  $\delta D$  values at these sites (Bowen and Revenaugh, 2003). The general pattern is similar to that of an analysis using middle and lower latitude data (Fig. 4b in Liu and Yang, 2008) with grass and woody plants clearly separated. Slopes for both woody trees/shrubs and grasses/herbs have slightly changed, especially for  $\delta D$  values in grasses/herbs which display a lower slope with the addition of new high latitude data. Differences in slopes between the two ecological life forms are distinct and statistically significant ( $p < 0.01$ ). Although both Correlation  $R^2$  values (Fig. 1) are lower (0.31 for grasses and 0.28 for woody plants) compared with the high latitude data alone (0.85 for grasses and 0.53 for woody plants) because of the large variation in  $\delta D_{n\text{-alkane}}$  in the global database, the distinguishable distribution patterns between leaf  $\delta D_{n\text{-alkane}}$  values from grass and woody plants with respect to global precipitation  $\delta D$  remain apparent.

### 3.3. High latitude effect on hydrogen isotope fractionation

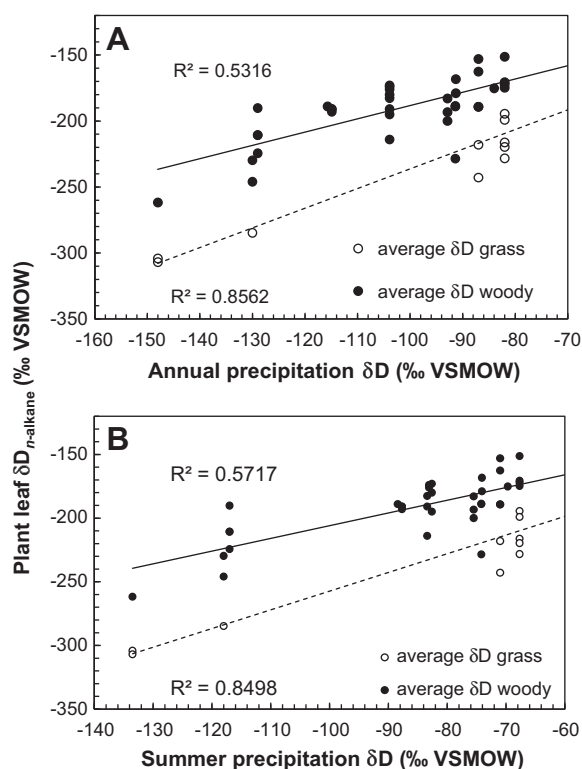
A high latitude effect on  $\epsilon_{\text{alk-water}}$  is apparent in Fig 3. The  $\delta D_{n\text{-alkane}}$  values and  $\epsilon_{\text{alk-water}}$  relationships for both woody deciduous trees and grasses/herbs from middle and low latitudes exhibit a similar linear relationship and  $\delta D_{n\text{-alkane}}$  values from woody plants show slightly lower  $\epsilon_{\text{alk-water}}$  values than those from grasses, as indicated by the trend lines. Correlation  $R^2$  values indicate a tighter correlation between  $\epsilon_{\text{alk-water}}$  and middle and lower latitude  $\delta D_{n\text{-alkane}}$  from grasses (0.94) than that from woody plants (0.88). However,  $\delta D_{n\text{-alkane}}$  signals from high latitude plants deviate from this middle and low latitude linear relationship, the deviation being driven by low  $\epsilon_{\text{alk-water}}$  values. This occurs for woody deciduous trees as well as grasses and herbs at the high latitudes, with woody plants having slightly smaller  $\epsilon_{\text{alk-water}}$  values. We attribute the expression of lower high latitude  $\epsilon_{\text{alk-water}}$  values to higher transpiration rates and other physiological effects caused by increasingly longer daily intervals of photosynthesis associated with increasing latitude.

**Table 1**  
Location, systematic and ecological life form of fresh and herbarium plants from high latitudes and hydrogen isotope composition of their leaf *n*-alkanes (n.d., not determined).<sup>a</sup>

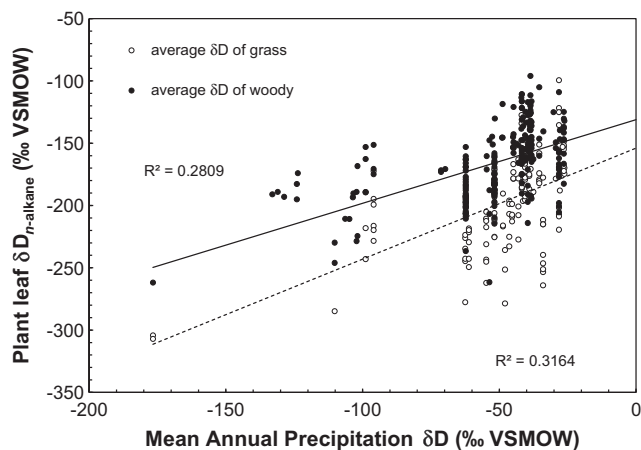
Taxon	Family	Life Form	Location	Coordination	C <sub>27</sub>		C <sub>29</sub>		C <sub>31</sub>		Mean δD
					δD	SD <sup>b</sup>	δD	SD <sup>b</sup>	δD	SD <sup>b</sup>	
<i>Cotinus coggyria</i> Scop.	Anacardiaceae	C <sub>3</sub> woody shrub	Stockholm, Sweden	59°20'N; 18°02'E	-168	1.1	-182	0.1	n.d.	n.d.	-175
<i>Castanea sativa</i> Mill.	Fagaceae	C <sub>3</sub> woody tree	Stockholm, Sweden	59°20'N; 18°02'E	n.d.	n.d.	-151	0.4	-152	1.1	-151
<i>Triticum aestivum</i> L.	Poaceae	C <sub>3</sub> grass	Stockholm, Sweden	59°20'N; 18°02'E	-177	1.3	-171	0.1	-170	0.6	-172
<i>Thamnochlamys spathaceus</i> (Franch.) Söderstr.	Poaceae	C <sub>3</sub> woody grass	Stockholm, Sweden	59°20'N; 18°02'E	-177	1.2	-162	n.d.	-173	n.d.	-171
<i>Zea mays</i> L.	Poaceae	C <sub>4</sub> grass	Stockholm, Sweden	59°20'N; 18°02'E	-228	1.6	n.d.	n.d.	n.d.	n.d.	-228
<i>Phragmites australis</i> (Cav.) Trin. ex Steud.	Poaceae	C <sub>3</sub> herb	Stockholm, Sweden	59°20'N; 18°02'E	-190	1.9	-199	0.6	n.d.	n.d.	-195
<i>Luzula sylvatica</i> (Hudson) Gaudin	Juncaceae	C <sub>3</sub> grass	Stockholm, Sweden	59°20'N; 18°02'E	-216	n.d.	n.d.	n.d.	n.d.	n.d.	-216
<i>Brachypodium sylvaticum</i> (Huds.) Beauv.	Poaceae	C <sub>3</sub> grass	Stockholm, Sweden	59°20'N; 18°02'E	-211	2.4	-224	0.9	-224	1.9	-220
<i>Arundo donax</i> L.	Poaceae	C <sub>3</sub> grass	Stockholm, Sweden	59°20'N; 18°02'E	-202	1.8	-197	0.0	n.d.	n.d.	-199
<i>Mercurialis glyptostrobiloides</i> Hu & Cheng	Cupressaceae	C <sub>3</sub> woody tree	Uppsala, Sweden	59°55'N; 17°38'E	-176	2.0	-178	1.0	-173	2.0	-176
<i>Hierochloa hirta</i> (Schrank) Borbas	Poaceae	C <sub>3</sub> grass	Helsinki, Finland	60°10'N; 24°56'E	n.d.	n.d.	-213	n.d.	-223	n.d.	-218
<i>Poa pratensis</i> L.	Poaceae	C <sub>3</sub> grass	Helsinki, Finland	60°10'N; 24°56'E	n.d.	n.d.	-244	1.1	-241	1.7	-243
<i>Solidago virgaurea</i> L.	Asteraceae	C <sub>3</sub> herb	Helsinki, Finland	60°10'N; 24°56'E	n.d.	n.d.	-188	7.2	-190	4.7	-189
<i>Antennaria dioica</i> (L.) Gaertn.	Asteraceae	C <sub>3</sub> herb	Helsinki, Finland	60°10'N; 24°56'E	n.d.	n.d.	-189	1.3	n.d.	n.d.	-189
<i>Marricaria matricarioides</i> (Less.) Porter	Asteraceae	C <sub>3</sub> herb	Helsinki, Finland	60°10'N; 24°56'E	-158	1.1	-156	1.9	-145	1.4	-153
<i>Lonicera tatarica</i> L.	Caprifoliaceae	C <sub>3</sub> woody shrub	Helsinki, Finland	60°10'N; 24°56'E	-164	1.3	-161	1.2	n.d.	n.d.	-163
<i>Salix arctica</i> Pall.	Salicaceae	C <sub>3</sub> woody shrub	Axel Heiberg Island, Canada	79°54'N; 89°01'W	-273	0.7	-251	0.4	n.d.	n.d.	-262
<i>Puccinellia angustata</i> (R. Br.) Rand & Redf.	Poaceae	C <sub>3</sub> grass	Axel Heiberg Island, Canada	79°54'N; 89°01'W	-299	1.1	-301	1.0	-312	0.7	-304
<i>Alopecurus alpinus</i> J. E. Smith	Poaceae	C <sub>3</sub> grass	Axel Heiberg Island, Canada	79°54'N; 89°01'W	n.d.	n.d.	-298	1.3	-316	2.8	-307
<i>Salix alaxensis</i> (Andersson) Coville	Salicaceae	C <sub>3</sub> woody shrub	Sutton, Alaska	61°44'N; 148°56'W	-224	1.0	n.d.	n.d.	n.d.	n.d.	-224
<i>Achillea millefolium</i> L.	Asteraceae	C <sub>3</sub> herb	Yarrow, Alaska	61°44'N; 148°56'W	-193	1.7	-190	0.6	-187	0.1	-190
<i>Equisetum pratense</i> Ehrhart	Equisetaceae	C <sub>3</sub> horsetail	Sutton, Alaska	61°44'N; 148°56'W	-219	n.d.	-202	n.d.	n.d.	n.d.	-211
<i>Alnus viridis</i> ssp. <i>Sinuata</i> (Regel) A.&D. Löve	Betulaceae	C <sub>3</sub> woody tree	Sutton, Alaska	61°44'N; 148°56'W	-211	1.4	n.d.	n.d.	n.d.	n.d.	-211
<i>Picea glauca</i> (Moench) Voss	Pinaceae	C <sub>3</sub> woody tree	Denali, Alaska	63°43'N; 148°54'W	-230	2.1	-229	2.2	n.d.	n.d.	-230
<i>Rhododendron groenlandicum</i> (Oeder) Kron&Judd	Ericaceae	C <sub>3</sub> woody shrub	Denali, Alaska	63°43'N; 148°55'W	-248	0.6	-248	0.6	-243	2.4	-246
<i>Poa trivialis</i> L.	Poaceae	C <sub>3</sub> grass	Denali, Alaska	63°43'N; 148°54'W	-263	0.4	-294	0.2	-297	0.1	-285

<sup>a</sup> δD values are expressed in ‰ relative to VSMOW.

<sup>b</sup> SD = standard deviation of three measurements.



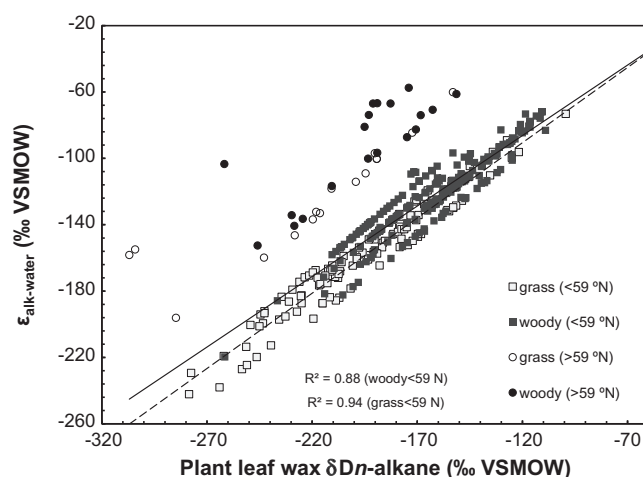
**Fig. 1.** Variation in  $\delta D_{n\text{-alkane}}$  from high latitude plants showing separate distributions of  $\delta D_{n\text{-alkane}}$  for grass (open circles) and woody (solid circles) plants vs.  $\delta D$  values for modeled precipitation.  $\delta D_{n\text{-alkane}}$  for grass ( $y = 1.4927x - 87.013$ ,  $R^2 = 0.8562$ ) and woody ( $y = 1.0071x - 87.549$ ,  $R^2 = 0.5316$ ) plants vs. average mean annual precipitation (1A) and  $\delta D_{n\text{-alkane}}$  for grass ( $y = 1.4687x - 110.6$ ,  $R^2 = 0.8498$ ) and woody ( $y = 0.9985x - 106.18$ ,  $R^2 = 0.5717$ ) plants vs. mean summer precipitation (1B). Precipitation  $\delta D$  data were obtained from the Online Isotopes in Precipitation Calculator, version 2.2 (Bowen and Revenaugh, 2003).



**Fig. 2.** Global analysis of  $\delta D_{n\text{-alkane}}$  ( $n = 408$ ) and  $\delta D$  of mean annual precipitation in the Northern Hemisphere. Solid circles and solid line trend represent woody plants ( $y = 0.61716x - 131.06$ ,  $R^2 = 0.2809$ ), whereas open circles and dash line trend annotate grasses ( $y = 0.8909x - 153.92$ ,  $R^2 = 0.3164$ ). Average mean annual precipitation  $\delta D$  values are based upon data from the International Atomic Energy Agency and the World Meteorological Organization (IAEA, 2001).

#### 4. Implications

The apparent hydrogen isotope fractionation between plant leaf wax and environmental water is controlled by multiple environmental and biological factors (Sachse et al., 2006; Liu and Yang,



**Fig. 3.** Plot between plant leaf  $\delta D_{n\text{-alkane}}$  ( $n = 408$ ) and apparent hydrogen isotope fractionation factors ( $\epsilon_{\text{alk-water}}$ ), showing deviation of  $\epsilon_{\text{alk-water}}$  from high latitude plants from those in middle and low latitudes in the Northern Hemisphere. Note the linear relationship for middle and low latitude plants (square symbols) and the detrended  $\epsilon_{\text{alk-water}}$  from both woody trees and grasses inhabited at high latitudes (circle symbols). Solid line and dash line trend represent middle to low latitude woody plants ( $y = 0.8499x + 15.736$ ,  $R^2 = 0.88$ ) and grasses ( $y = 0.9072x + 18.289$ ,  $R^2 = 0.94$ ) respectively.

2008). Our new data and published information demonstrate that the  $\delta D$  of precipitation and plant physiology exerts primary influences on  $\epsilon_{\text{alk-water}}$ . Previous studies have shown that evaporation of soil water (Hou et al., 2008), temperature (Duan and Wu, 2009), rooting pattern (Krull et al., 2006), plant systematics (angiosperms vs. gymnosperms; Pedentchouk et al., 2008), water use efficiency (Hou et al., 2007b) and rate of leaf transpiration (Yang et al., 2009; Feakins and Sessions, 2010) also influence the distribution of  $\delta D_{n\text{-alkane}}$  variation at a given site. While the amount of lipid  $\delta D$  variation among plants growing at the same high latitude site is consistent with previous reports (Chikaraishi and Naraoka, 2003; Liu et al., 2006; Hou et al., 2007a; Feakins and Sessions, 2010), our new  $n\text{-alkane}$   $\delta D$  data provide additional support for the link between  $\delta D$  variation and plant ecological life form (Liu et al., 2006). As Fig. 1 illustrates, the distribution of  $\delta D$  values from grasses and herbs can be separated from that of shrubs and trees, with a lack of evidence for  $\delta D$  variation as a result of plant systematics (i.e. angiosperms vs. gymnosperms) or photosynthetic pathways.

We posit that higher  $\delta D_{n\text{-alkane}}$  values of woody plants reflect higher rates of leaf water transpiration during high latitude summer months than for grasses, preliminarily because of their characteristics by way of leaf area and denser and more orderly venation patterns that increase hydraulic conductance (Brodrribb et al., 2007; McKown et al., 2010). Hydrogen isotope enrichment in leaf water during transpiration is well known (Leaney et al., 1985, see also Farquhar et al., 2007 and Barbour, 2007 for recent reviews), but there is a lack of understanding for how the D-enriched leaf water influences  $\delta D_{n\text{-alkane}}$  signatures. In a greenhouse experiment Yang et al. (2009) detected that, under 24 h simulated arctic environment, some deciduous conifers can lose up to 2.4 times more water per day than that under regular diurnal light at lower latitudes, and this transpiration effect resulted in D enrichment of leaf water attributed to have produced up to 40‰ heavier  $\delta D_{n\text{-alkane}}$  signals in the leaf waxes. However, a recent growth chamber experiment with grasses has shown that  $\epsilon_{\text{alk-water}}$  in grasses is insensitive to relative humidity, suggesting evaporation rather than transpiration has more influence on leaf  $\delta D_{n\text{-alkane}}$  signatures in grasses (McInerney et al., 2011, but see Brader et al., 2010 for

minimal evaporation effect on *Sphagnum*). In the above cases, leaf water was not measured, leaving room for a direct assessment of the impact of D-enriched leaf water on  $\delta D_{n\text{-alkane}}$  due to transpiration. It is possible that, as a result of different physiological characteristics, such as leaf area, venation pattern, hydraulic system and different timing for lipid synthesis between woody plants and grasses (Koch et al., 2004; Richardson et al., 2005), the impact of transpiration on  $\delta D_{n\text{-alkane}}$  signals is different between these two major plant groups with different ecological life forms. If this is true, then it may help explain the profound and consistent pattern of heavier  $\delta D_{n\text{-alkane}}$  in woody plants relative to that in grasses at a given site (Fig. 2).

Our results have several implications for high latitude paleoclimate interpretations based on  $\delta D_{n\text{-alkane}}$  values. First, as apparent  $\epsilon_{\text{alk-water}}$  for plants living at high latitude is 20–50‰ smaller than at lower latitudes,  $\delta D$  in ancient precipitation can be overestimated if a globally averaged  $\epsilon_{\text{alk-water}}$  value is applied. Second, as grasses and woody trees fall into distinct isotopic fields, it is important to understand the specifics of the ecosystem in order to distinguish whether  $\delta D$  change was due to a change of vegetation or climate. Thus, paleovegetation information obtained from traditional paleobotanic and palynological methods should be consulted when interpreting  $\delta D$  data from sediments. Third, as both  $\delta D_{n\text{-alkane}}$  and  $\epsilon_{\text{alk-water}}$  vary across different plant taxa, specific fractionation factors calibrated for an individual taxon may be used in collaboration with the reconstruction of paleo-precipitation  $\delta D$  based upon lipids from sediments, especially in high latitude regions. It has been noticed (Sachse et al., 2004; Hou et al., 2008; Polissar and Freeman, 2010) that  $\delta D_{n\text{-alkane}}$  values for lipids extracted from sediments and soils usually have a closer correlation with  $\delta D$  in precipitation, presumably because of an ecosystem-wide averaging effect of lipid isotopic signals. While the range and mechanism for the isotopic averaging effect are not well understood, an ecosystem-wide difference in  $\epsilon_{\text{alk-water}}$  complicates the reconstruction of  $\delta D$  in regional precipitation (Polissar and Freeman, 2010; McInerney et al., 2011). Taxon-specific  $\delta D$  studies based upon taxon-specific fractionation factors calculated from controlled experiments (Yang et al., 2009; Nichols et al., 2009; Brader et al., 2010; McInerney et al., 2011) should shed more light on the mechanism of the process. Finally, the application of  $\delta D$  values generated from herbarium specimens for studies of recent climate changes has not been exploited and our study demonstrates the feasibility of utilizing herbarium specimens, collected from historical times or regions that are now difficult to access, as a viable resource for compound-specific hydrogen isotope analysis.

## 5. Conclusions

Leaf  $\delta D_{n\text{-alkane}}$  data obtained from plants living at high latitude (>59°N) provide further evidence for multiple controls on D/H composition of plant leaf waxes and  $\epsilon_{\text{alk-water}}$ , extending the general trend predicted by Liu and Yang (2008) to higher latitudes. Our results further demonstrate that plant ecological life forms play an important role in controlling  $\delta D_{n\text{-alkane}}$  values. Physiological characteristics that impact on leaf transpiration rates are likely responsible for some of the observed  $\delta D$  variation in woody plants, as well as for the appearance of smaller  $\epsilon_{\text{alk-water}}$  values for high latitude plants. Our results point toward the importance of understanding ecosystem changes when interpreting *n*-alkane  $\delta D$  data from sediments.

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