

## Reply to E. Schefuß (SC3: Vegetation effects on sedimentary organic isotope records)

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*I disagree with the other comments that this is a novel approach. It is advocated by the authors since several years with various applications.*

→ We acknowledge that our coupled  $\delta^2\text{H}_{n\text{-alkane}}-\delta^{18}\text{O}_{\text{sugar}}$  paleohygrometer approach was first published five years ago by Zech et al. (2013). A validation study using topsoils along a climate transect followed two years later by Tuthorn et al. (2015); in the same year, a modified version of the coupled approach was applied to lacustrine sediments (Hepp et al., 2015); and an application to a terrestrial paleosol sequence followed one year ago by Hepp et al. (2017).

Whether this approach is novel or not, should be seen in our opinion against the background that compound-specific  $\delta^2\text{H}$  analysis using GC-Py-IRMS came up twenty years ago and meanwhile hundreds of papers dealing with  $\delta^2\text{H}$  of leaf wax compounds or sedimentary hydrocarbons have been published. For recent reviews please see e.g. Pedentchouk and Zhou (2018) and Sessions (2016). In any case, we readily leave it to every reader to make his own opinion concerning the innovation of our approach.

*My comment on it, however, remains the same each time: The approach cannot be done in soils or sediments as it compares apples with pears. This is due to two reasons.*

→ Both *n*-alkane and sugar biomarkers that are produced in leaves reflect the isotopic composition of precipitation/plant source water modified by (primarily RH-dependent) evapotranspirative enrichment of leaf water. Therefore, it is difficult for us to follow/understand the argumentation of E. Schefuß concerning apples and pears.

→ We furthermore disagree with E. Schefuß that our coupled  $\delta^2\text{H}_{n\text{-alkane}}-\delta^{18}\text{O}_{\text{sugar}}$  paleohygrometer approach cannot be applied to soils and sediments. Please allow us to refer once again to the validation paper of Tuthorn et al. (2015). In that study, *n*-alkane and sugar biomarkers were extracted from topsoils along a climate gradient in S-America

covering different vegetation types. Reconstructed RH values based on our coupled  $\delta^2\text{H}_{n\text{-alkane}}-\delta^{18}\text{O}_{\text{sugar}}$  paleohygrometer approach correlated highly significantly with actual RH values ( $R = 0.79$ ,  $p < 0.001$ ,  $n = 20$ ).

*First, plants incorporate the leaf water enrichment signal to variable degrees in their waxes and hemicellulose (Kahmen et al., 2013, Zech et al., 2014). Leaf water is not the sole source of the hydrogen in waxes and oxygen in hemicellulose but a leaf water – xylem water mixture which is different between plants. It is not only grasses versus other plants as suggested here but various plants to a variable degree.*

→ This comment and statement are surprising and puzzling to us, because E. Schefuß is co-author of Kahmen et al. (2013). In that publication, the abstract reads “For dicotyledonous plants we found that the full extent of leaf water evaporative D-enrichment is recorded in leaf wax *n*-alkane  $\delta\text{D}$  values. For monocotyledonous plants [such as grasses], we found that between 18% and 68% of the D-enrichment in leaf water was recorded in the  $\delta\text{D}$  values of their *n*-alkanes.” Concerning Zech et al. (2014), that paper dealt with stem material not leaf material. While the former does show a dampening effect, the latter doesn't. Hence, neither evidence nor literature are provided by E. Schefuß supporting his statement that other plants than grasses (for which we included correction calculations in our manuscript) incorporate noteworthy amounts of the xylem water signal in their leaf biomarkers.

*Second, plants produce waxes and hemicellulose in highly variable amounts (e.g. Diefendorf & Freimuth, 2017) depending on plant type and not correlated with each other, i.e. higher wax content is not necessarily associated to higher hemicellulose content. In sedimentary archives or soils this means that the hydrogen isotope signal of leaf waxes is a wax-production weighted signal of the primary signal (temperature, amount, source effect) overprinted to a certain degree by evapo-transpiration and the hemicellulose oxygen isotope signal is a hemicellulose-production weighted signal of the same primary signal but affected to a different degree by evapo-transpiration due to different vegetation contributions to both parameters. Both  $\delta\text{D}$  of wax lipids and  $\delta^{18}\text{O}$  of hemicellulose are thus qualitative hydrologic parameters that are not directly correlated and comparable. The position of the data points in  $\delta^{18}\text{O}-\delta\text{D}$  space is thus dependent on vegetation composition and changes thereof and cannot be interpreted as reflecting leaf water*

*isotopic enrichment in a quantitative approach. Application of such approach to sediments or soils will lead to erroneous and misleading interpretations.*

→ As long as only dicotyledonous plants are investigated or their leaves contributed primarily to a sedimentary archive, E. Schefuß is wrong in his statement that the data points in a  $\delta^2\text{H} - \delta^{18}\text{O}$  diagram are noteworthy affected by vegetation changes. Why should a variable production of *n*-alkane or sugar biomarkers affect the  $\delta^2\text{H}/\delta^{18}\text{O}$  values? E. Schefuß does not provide any evidence or literature supporting this statement.

→ The issue raised by E. Schefuß would become only relevant when grasses/monocotyledonous plants and at the same time coniferous trees are the primary sources of biomarkers to a sedimentary archive/soil (this does not apply to the Gemündener Maar according to the pollen results indicating strong presence of *Betula* during the Late Glacial). This assessment is based on the notion that except for *Juniperus*, conifers produce very low amounts of *n*-alkanes (e.g. Zech et al., 2012). In such cases, sugars will show a mixed  $\delta^{18}\text{O}$  signal of conifer needles and grasses (and thus partly a dampened leaf water enrichment signal), whereas *n*-alkanes will show the dampened  $\delta^2\text{H}$  signal of the grasses. As a result, reconstructed RH values will underestimate actual RH values. This explanation is corroborated by data obtained for topsoils along a European climate transect; the respective manuscript will be submitted during the next weeks.

*Diefendorf, A.F., Freimuth, E.J., 2017. Extracting the most from terrestrial plant-derived n-alkyl lipids and their carbon isotopes from the sedimentary record: A review. Organic Geochemistry 103, 1-21.*

*Kahmen, A., Schefuß, E., Sachse, D., 2013. Leaf water deuterium enrichment shapes leaf wax n-alkane  $\delta^2\text{H}$  values of angiosperm plants I: Experimental evidence and mechanistic insights. Geochimica et Cosmochimica Acta 111, 39-49.*

*Zech, M., Mayr, C., Tuthorn, M., Leiber-Sauheitl, K., Glaser, B., 2014. Oxygen isotope ratios ( $^{18}\text{O}/^{16}\text{O}$ ) of hemicellulose-derived sugar biomarkers in plants, soils and sediments as paleoclimate proxy I: Insight from a climate chamber experiment. Geochimica et Cosmochimica Acta 126, 614-623.*

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