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Towards improved source apportionment of organic matter in soil and peat using lipid biomarkers and inverse modeling

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1. INTRODUCTION

1.1. SOIL AND PEATLANDS AS CARBON SINKS

Soil plays an essential role in the global carbon cycle, functioning as a net carbon sink, sequestering and storing an estimated 1700 PgC (Jackson et al., 2017), about 25% of which is stored in peatlands (Jackson et al., 2017; Yu et al., 2010). Together, this is about four times greater than the total carbon stock of terrestrial vegetation (450 PgC) (Erb et al., 2018) and more than twice the amount of carbon in the atmosphere (870 PgC) (Canadell et al., 2021). In the Sixth Assessment Report of the Intergovernmental Panel on Climate Change (IPCC), increased soil carbon sequestration and peatland restoration are listed as carbon dioxide removal strategies that could potentially mitigate the effects of anthropogenic climate change (Canadell et al., 2021). To best realize the potential of soil and peatlands as carbon sinks, it is vital to better understand the process of soil organic matter (SOM) formation and stabilization to improve global earth system models (ESM) (Jackson et al., 2017) for accurately quantifying soil carbon stocks and refining predictions related to carbon cycling in response to environmental changes. This applies to both topsoils (<20 cm depth) and the less-investigated subsoils (Angst et al., 2016).

The pool of organic matter is formed in soil through the deposition and incorporation of plant tissues, above- and belowground, as well as animal remains, including those of microorganisms (Kögel-Knabner, 2002; Angst et al., 2016). An interplay of biological, chemical, and physical processes governs the formation and stabilization of organic matter, including factors such as the amount and composition of plant material input and the soil microbial community (Jackson et al., 2017). Investigating the sources of SOM can enhance the understanding of the biotic factors affecting soil carbon sequestration, as the quantity and distribution of SOM in the soil column are heavily influenced by the overlying vegetation (Jobbágy & Jackson, 2000).

As mentioned, peatlands contain nearly 25% of the carbon stored in soils despite only covering just under 3% of the land surface area (Yu et al., 2010; Xu et al., 2018). This is

due to the way in which peatlands form. They contain a large amount of partially decomposed plant material continuously accumulating in waterlogged, anoxic conditions, which are detrimental to microbial activity. Over time, this results in the formation of peat deposits that can store carbon for thousands of years (Barber et al., 1994). However, peatlands have often been disturbed due to human activities such as drainage, peat extraction, and land-use change, thereby potentially becoming a source of carbon emissions rather than a sink (Swindles et al., 2019). To preserve the integrity of current peatlands as well as to better predict their response to climate change, studying their development through time and previous climate changes is essential (e.g., Naafs et al., 2019; Swindles et al., 2019).

1.2. SOIL AND PEAT AS TERRESTRIAL ARCHIVES

Terrestrial archives, such as soil and peat, function not only as essential sinks in the global carbon cycle but also as libraries of data on past vegetation and climate conditions (e.g., Barber et al., 1994; Chambers et al., 2012). Investigating these archives allows us to trace vegetation and climate dynamics on centennial to millennial scales, gaining insight into past environmental changes (McClymont et al., 2023), and can enable the use of these observations to better inform climate models predicting future changes in our warming world. For example, paleovegetation data can validate climate and vegetation models used to predict the future effects of climate change on ecosystems; models that are able to replicate past changes are likely to more reliably predict those in the future (Lavorel et al., 2007).

Several methods have been developed over the past century to elucidate paleoenvironmental conditions through the use of terrestrial archive records as proxies. These methods, including palynology, the characterization of plant macrofossils, geochemical measurements, radiocarbon dating, and the analysis of molecular fossils, known in this context as “biomarkers,” have significantly increased our understanding of past ecosystem and climate interactions.

1.3. PLANT MACROFOSSIL AND POLLEN RECORDS

Pollen and plant macrofossils are traditional paleobotanical approaches used to reconstruct local and regional vegetation patterns through time and to identify climate or human impact

on plant communities and evaluate their response (Mauquoy et al., 2008; Speranza et al., 2000). In peatlands especially, the succession of plant communities through time is often influenced by changes in hydrological regimes, and therefore, pollen and plant macrofossils act as a proxy for paleohydrologic conditions (Andersson et al., 2011). However, there are some limitations to their use. Pollen records often provide a more regional signal due to airborne spread (Farrimond & Flanagan, 1996). Certain taxa may also be over-represented in pollen records due to some species producing more pollen as well as variation in the preservation of pollen grains (Birks & Birks, 2000). While plant macrofossils are more reflective of local vegetation and can provide increased taxonomic precision in comparison to pollen (Birks & Birks, 2000), their use is also dependent on preservation and can be complicated in very humified archives (Andersson et al., 2011; Naafs et al., 2019). Plant wax biomarkers are a comparatively newer vegetation proxy but can be beneficial to either supplement pollen and macrofossil analysis or be analyzed on their own if the more traditional approaches are not feasible (e.g., Ronkainen et al., 2015).

1.4. PLANT WAX BIOMARKERS

A small but important component of soil organic matter originates from plant waxes (Oades, 1988). Plant leaves are protected by a cuticle coated in hydrophobic wax; this wax layer is comprised primarily of lipid and hydrocarbon polymers, including *n*-alkanes, *n*-alcohols (also referred to as *n*-alkanols), and *n*-fatty acids (Chibnall et al., 1934; Eglinton & Hamilton, 1967). Epicuticular wax serves multiple purposes for plants, including reducing transpiration rates and limiting water loss, reflecting and thereby reducing the strength of UV radiation, and impeding the ability of pathogens to adhere to plant tissue (Koch & Ensikat, 2008; Shepherd & Wynne Griffiths, 2006). The constituent compounds of epicuticular wax are environmentally persistent due to their insolubility in water, relative chemical inertness, and resistance to biodegradation (Eglinton & Eglinton, 2008).

These chemicals enter sedimentary archives such as soil, lacustrine or marine sediments, and peat through the deposition of plant tissue (Peters et al., 2005) or as aerosol particles following wind abrasion (Nelson et al., 2018). Multiple studies have investigated the utility of using plant wax biomarkers as chemotaxonomic indicators for their source material with

mixed results (Jansen & Wiesenberg, 2017); these include studies of terrestrial archives as well as studies of archaeological sites and artifacts (e.g., McGovern & Hall, 2016).

1.5. UNRAVELING BIOMARKER COMPOSITIONS

CHALLENGES

While plant wax biomarkers have been used successfully in multiple studies to aid in paleoenvironmental reconstructions and source identification, there are many factors influencing their composition that complicate their use as chemotaxonomic indicators (Jansen and Wiesenberg, 2017). Because the building blocks of plant waxes are ubiquitous across species, there are potential overlaps in the chemical composition and distribution patterns of different species which complicates source assignment. Additionally, while biomarkers are relatively robust following deposition, there are still degradation processes occurring that could potentially obscure the original compositions if certain compounds are preferentially degraded in comparison to others.

Furthermore, due to their purpose as protection for plant leaves, the biomarker composition is affected by environmental factors, such as season, climatic factors such as temperature, and ontological factors such as growth stage (Shepherd and Wynne Griffiths, 2006). Additionally, plant waxes and their constituents are present not only in leaves but in other plant organs, such as roots, with differing compositions (e.g., Jansen et al., 2006). Therefore, terrestrial archives contain a mixture of biomarkers from aboveground and belowground biomass of different species, of various residence times, and growing conditions. Unraveling the archival signal into its various sources is quite a puzzle.

MODELING APPROACHES

As a response to this challenge, the VERHIB (VEgetation Reconstruction with the Help of Inverse modeling and Biomarkers) model was developed (Jansen et al., 2010). It uses the input of biomarker data from plant species as well as from an archive such as a soil or peat core to determine the contribution of individual plant sources to the overall signal.

Other quantitative approaches have been developed as well including the use of mixing models with datasets from lacustrine and marine sediments as well as more recently, the incorporation of Bayesian statistics into these models (Yang and Bowen, 2022) and the

application of machine learning techniques to biomarker source apportionment (Peaple et al., 2021).

2. OBJECTIVES AND RESEARCH QUESTIONS

This thesis was completed as part of the "IQ-SASS - Improved Quantitative Source Assessment of organic matter in Soils and Sediments using molecular markers and inverse modeling" project, the goals of which include developing a VERHIB 2.0 model that will improve upon the first version and will run in R, rather than MATLAB, as R is open-source and more accessible.

As mentioned in the Introduction, plant-derived biomarkers have been used frequently for studies of soil organic matter and in the context of paleoecological reconstructions. Of these, *n*-alkanes are the most commonly used and well-studied. Consequently, *n*-alkanes offer the best starting point for improving quantitative source assessment. However, there has not been a systematic assessment aimed at deriving quantitative degradation parameters of *n*-alkanes, which limits the potential to develop a universally applicable approach to source assessment of organic matter using biomarkers. Therefore, the first objective of this research was as follows:

Objective I: Identify processes and parameters that should be considered in the improved model

Research questions: What data is available about changes in biomarker composition along the plant to soil continuum and to what extent can this data be readily quantified into a widely applicable parameter for source assessment?

Hypothesis: Compiling data across available studies of *n*-alkanes, as the most frequently used biomarker class, will allow for the calculation of a degradation coefficient that could be implemented into the VERHIB model to widen its applicability.

n-Alkanols and *n*-fatty acids are also plant-derived straight chain lipid biomarkers but have been much less frequently used than *n*-alkanes. Including these measurements in biomarker studies theoretically has the potential to triple the information regarding organic matter sources, depending on how complementarity the data is. However, very few studies include

these three classes simultaneously in high-resolution multi proxy paleoecological reconstructions. Therefore, to qualitatively assess the possibility of improving source apportionment using multiple lipid classes, the joint applicability of *n*-alkanes, *n*-alkanols, and *n*-fatty acids should be assessed by reconstructing the paleoecology of a suitable terrestrial archive with available fossil pollen and plant macrofossil data as a comparison. The second objective of the research was as follows:

Objective II: Paleoecological reconstruction of the Beerberg peatland using plant-derived biomarkers and traditional paleobotanical proxies

Research questions: How did the climate and vegetation develop over time at the Beerberg peatland?

What biomarker ratios are most indicative for paleo transitions at the Beerberg peatland?

Hypothesis: A high-resolution biomarker record including *n*-alkanes, *n*-alkanols, and *n*-fatty acids will enable a more robust reconstruction than traditional proxies alone and ratios indicative of vegetation transitions can be derived for *n*-alkanols and *n*-fatty acids, as these are not typically measured in such studies.

To have a truly systematic and quantitative approach to source apportionment and reconstruction, the VERHIB model must be optimized and better calibrated. To evaluate the added value of the additional lipid classes and test the potential for expanding VERHIB, the third objective was developed as follows:

Objective III: Evaluate VERHIB (MATLAB) performance using Beerberg peatland biomarker dataset

Research questions: How does the biomarker composition of modern vegetation samples vary across plant organs and groups?

What combination of parameters provides the best model fit for the Beerberg sequence?

What does the inclusion of *n*-alkanols and *n*-fatty acids add to the analysis?

Hypothesis: Combining multiple plant-derived molecular compounds, i.e., *n*-alkanes, *n*-alkanols, and *n*-fatty acids, to the VERHIB model will enable more precise identification of plant species in the reconstructions.

3. LIST OF MANUSCRIPTS

The objectives and research questions were addressed in three research manuscripts.

Manuscript I

Thomas, C. L., Jansen, B., van Loon, E. E., and Wiesenberg, G. L. B., 2021. Transformation of *n*-alkanes from plant to soil: a review. *SOIL*, 7, 785–809, <https://doi.org/10.5194/soil-7-785-2021>.

Authors' contributions: **CLT: Conceptualization, Methodology, Formal analysis, Data curation, Writing Original draft, Writing–Review & Editing, Visualization**; **BJ: Conceptualization, Writing–Review & Editing, Supervision**; **EEvL: Conceptualization, Writing–Review & Editing, Supervision**; **GLBW: Conceptualization, Writing–Review & Editing, Supervision, Project administration, Funding acquisition**.

Manuscript I addresses Objective I and the related research questions through a systematic literature review examining the behavior of *n*-alkanes along the plant-to-soil continuum. Many studies have investigated plant-derived biomarkers at individual sites or regions and available review papers are qualitative in nature. There has not been a concentrated focus on quantifying degradation or transformation processes for use in widely applicable models. The manuscript compiles and analyzes available data on the fate of *n*-alkanes in soil to improve mechanistic understanding.

Manuscript II

Thomas, C. L., Jansen, B., Czerwiński, S., Gałka, M., Knorr, K.-H., van Loon, E. E., Egli, M., and Wiesenberg, G. L. B., 2023. Comparison of paleobotanical and biomarker records of mountain peatland and forest ecosystem dynamics over the last 2600 years in central Germany, *Biogeosciences*, 20, 4893–4914, <https://doi.org/10.5194/bg-20-4893-2023>.

Authors' contributions: **CLT: Conceptualization, Formal analysis, Investigation, Data curation, Writing–Original draft, Writing–Review & Editing, Visualization**; **BJ: Writing–Review & Editing, Supervision**; **SC: Formal analysis, Investigation, Writing–Review & Editing, Visualization**; **MG: Methodology, Formal analysis, Investigation, Writing–Review & Editing, Visualization**; **K-HK: Conceptualization, Methodology,**

Writing–Review & Editing; EEvL: Writing–Review & Editing, Supervision; ME: Formal analysis, Investigation, Writing–Review & Editing; GLBW: Conceptualization, Methodology, Resources, Writing–Review & Editing, Supervision, Project administration, Funding acquisition.

Manuscript II addresses Objective II through a high-resolution multi-proxy study of a core from the Beerberg peatland including radiocarbon dating, elemental analysis, pollen and plant macrofossil identification, and biomarker analysis. Although the multi-proxy approach is not new, there are very few studies that include multiple lipid classes as most focus on only *n*-alkanes. The extensive dataset developed from the Beerberg core was used to assess the qualitative contributions that biomarker analyses add to paleoecological reconstructions.

Manuscript III

Thomas, C. L., Jansen, B., van Loon, E. E., and Wiesenberg, G. L. B. Evaluating the applicability of the VERHIB model to a 2600-year peat sequence from central Germany. Will be submitted to *Palaeogeography, Palaeoclimatology, Palaeoecology*, 2024.

Authors' contributions: **CLT: Conceptualization, Investigation, Formal analysis, Data curation, Writing–Original draft, Writing–Review & Editing, Visualization**; BJ: Conceptualization, Writing–Review & Editing, Supervision; EEvL: Conceptualization, Methodology, Writing–Review & Editing, Supervision; GLBW: Conceptualization, Methodology, Resources, Writing–Review & Editing, Supervision, Project administration, Funding acquisition.

Manuscript III addresses Objective III through the application of the previously published VERHIB model to the Beerberg dataset, including a biomarker analysis of modern vegetation samples from the Beerberg site to be used as model input. This study also tests the use of *n*-fatty acid data in the model for the first time and evaluated the best combination of parameters using the Beerberg dataset.

4. SUMMARY OF METHODOLOGY

This section describes the methods used in the three manuscripts as well as further work that will be published following thesis submission.

Table 1. Summary of methods used for each manuscript.

Manuscript	Research type	Methods and Section #
I	Literature review	Literature review and meta-analysis (3.1)
II	Field and laboratory analysis	Sampling (3.2), Laboratory analysis (3.3.1-3.3.5), Data analysis (3.4.1-3.4.3)
III	Field and laboratory analysis and modeling (MATLAB)	Sampling (3.2), Laboratory analysis (3.3.1-3.3.4), Data analysis (3.4.2, 3.4.4), Modeling (3.5)

4.1. LITERATURE REVIEW AND META-ANALYSIS

To gain insight into potential processes or parameters that should be considered in an updated model, Manuscript I was a quasi-systematic literature review performed to quantitatively investigate the fate of *n*-alkanes from plant source to deposition and incorporation into soil and peat. *n*-Alkanes were chosen because they are by far the most common plant-derived biomarker studied.

To be as systematic as possible, a Boolean search string was devised to query the databases included in the Web of Science: (“leaf wax*” OR “lipid biomarker*” OR “alkane*” OR “*n*-alkane*” OR “chemical fossil*” OR “epicuticular wax*” OR “molecular prox*”) AND (“soil*” OR “peat*” OR “topsoil*” OR “litter*”). To screen the results, the following selection criteria were adopted:

1. Published in a peer-reviewed journal.
2. Contains primary data or observations on the degradation of the distribution patterns, concentration, or index measurements (such as Carbon Preference Index (CPI), Odd-over-Even Predominance (OEP), or Average Chain Length (ACL)) of *n*-alkanes.

3. Conducted in natural soil or peat with minimal contamination, or in laboratory conditions if microbial degradation of *n*-alkanes in natural soils was investigated.
4. No enhanced or artificial measures taken to promote degradation.
5. Includes data on modern soils.

From the studies that met these criteria, the following types of information were extracted when available:

1. Molecular data (*n*-alkane concentration, CPI, OEP, ACL)
2. Environmental data (elevation, mean annual air temperature (MAAT), mean annual precipitation (MAP))
3. Soil data (soil type, pH, total organic carbon (TOC))
4. Vegetation data (species, general vegetation)

A meta-analysis was performed on the extracted data by grouping the studies into six biome types based on dominant vegetation: coniferous forest, deciduous forest, mixed forest, grassland or shrubland, peatland, and steppe.

4.2. STUDY SITE AND SAMPLING PROCEDURES

The chosen site for the test dataset developed in Manuscripts II and III was the Beerberg peatland (50° 39' 32" N, 10° 44' 36" E, 983 m), located in the Thuringian Forest in central Germany. This location was chosen due to the scarcity of paleoenvironmental records from the Thuringian Forest and the peatland is part of a nature reserve and is therefore likely more intact than most other regional bogs. Beerberg peatland sits atop the Grosser Beerberg mountain and is ombrotrophic ("rain-fed"), meaning that it receives water and nutrients only from precipitation (Andersson et al., 2011). Estimated annual precipitation is 1300 mm (Görner et al., 1984) with a mean annual temperature of 4°C (Jeschke and Paulson, 2000). The current vegetation assemblage at the site is consistent with typical bog species including *Sphagnum* mosses, a few dwarf shrubs, and young trees scattered across the peatland.

Sampling took place in October 2019. Peat cores were extracted using two Russian peat corers (5 cm diameter, Eijkelkamp, Giesbeck, The Netherlands; 7 cm diameter, self-made). Two hummocks approximately 20 cm apart were alternately cored to a depth of 340 cm.

Plants within a five-meter radius of the coring sites were sampled, with multiple specimens per species being mixed into one sample.

4.3. LABORATORY ANALYSIS

SAMPLE PREPARATION

Both the peat core and plant samples were kept cool with ice packs until they could be transported to the Physical Geography lab at the University of Zurich and stored at -20 C. From the peat core, bulk samples were taken at 5 cm intervals, with additional samples taken where there was a layer with a distinct color or texture change, including 10-12 cm, 170-172 cm, 270-272 cm, 325-327 cm, 327-328 cm, and 337.5-340 cm. Plant samples were separated as well as possible into their various parts: non-woody aboveground biomass (e.g., leaves or needles), woody aboveground biomass (hereafter referred to as stems), and roots. Moss species samples were left intact as separating above- and belowground biomass would be very difficult and time-consuming. All of the samples were then freeze-dried until they reached a constant weight and homogenized using a horizontal ball mill.

LIPID BIOMARKER EXTRACTION AND PREPARATION

The milled samples were subjected to Soxhlet extraction following the method by Wiesenberg and Gocke (2015). In short, the samples were placed in the Soxhlet apparatus for ca. 30 hours with a solvent mixture of dichloromethane (DCM): methanol (MeOH) (93:7, v/v) to obtain the total lipid extracts. Afterwards, these total extracts were sequentially divided into three fractions: the first comprising neutral components like *n*-alkanes and *n*-alkanols, the second containing *n*-fatty acids, and the third consisting of polar and compounds with high molecular weights. Separation was performed using a glass column packed with Silica 60 + 5% potassium hydroxide (KOH), utilizing the solvents DCM, DCM: formic acid (99:1, v/v), and DCM: MeOH (1:1, v/v). The neutral fraction was further separated into aliphatic, aromatic, and heterocompound fractions using a Pasteur pipette filled with activated silica gel and the solvents *n*-hexane, *n*-hexane: DCM (1:1, v/v), and DCM: MeOH (93:7, v/v). Further details may be found in Manuscript II (Thomas et al., 2023).

IDENTIFICATION AND QUANTIFICATION USING GAS-CHROMATOGRAPHY

Quantification of *n*-alkanes, *n*-alkanols, and *n*-fatty acids was performed using an Agilent 7890B GC equipped with a multimode inlet and flame ionization detector (FID). Identification of biomarker compounds was conducted with an Agilent 6890N GC fitted with a split-splitless injector and an Agilent 5973 mass selective detector (MS).

Both instruments utilized a DB-5MS column (50 m x 0.2 mm x 0.33 μm) and a 1.5 m deactivated pre-column, with helium as the carrier gas flowing at 1 ml min⁻¹. The same heating program was also used for each instrument. The temperature program for *n*-alkanes maintained a starting temperature of 70°C for 4 min, then increased to 320°C at a rate of 5°C min⁻¹ held for 50 min. For *n*-fatty acids and *n*-alkanols, the temperature started at 50°C for 4 min, rose to 150°C at a rate of 4°C min⁻¹, and ramped up to 320°C at 3°C min⁻¹ held for 40 min. Each sample (1 μl) was injected into the GCs in splitless mode. The GC-MS operated in electron ionization mode at 70 eV, scanning a range from *m/z* 50–550. Mass spectra of individual biomarker compounds were compared with external standards and entries from the NIST and Wiley mass spectra library to facilitate identification.

RADIOCARBON DATING

The core from the Beerberg peatland was dated using hand-picked plant remains. First, these remains underwent an acid-alkali-acid treatment then were combusted at 900°C to generate carbon dioxide, which was subsequently reduced to graphite. Finally, the carbon isotope composition was determined using Accelerator Mass Spectrometry (AMS) at the Institute of Ion Beam Physics at the Swiss Federal Institute of Technology (Zurich, Switzerland) with the 0.2 MV MICADAS facility.

MACROFOSSIL ANALYSIS

Plant macrofossils in the core were examined at 4 cm intervals, using samples 1 cm thick with a volume of approximately 8 cm³. Before analysis, the samples were washed with warm water over 0.20 mm mesh screens. Comparison with recently collected specimens and identification keys (Smith, 2004; Mauquoy and Van Geel, 2007) were used to estimate the percentage of fossils from vascular plants and brown mosses and to identify carpological remains and vegetative fragments, including rootlets, leaves, and epidermis. For a detailed explanation of the classification of *Sphagnum* mosses, see Manuscript II (Thomas et al., 2023).

POLLEN ANALYSIS

Samples of the core were taken at depths 2.5 cm, 6.5 cm, 9.5 cm, and then every 5 cm beginning at a depth of 12.5 cm, resulting in 69 samples to be used for pollen analysis. Each sample had a volume of 2 cm³ and was prepared according to Berglund and Ralska-Jasiewiczowa (1986). Briefly, carbonates were dissolved using 10% hydrochloric acid (HCl), humic compounds were eliminated by heating the samples in 10% potassium hydroxide (KOH), and the mineral fraction was removed by soaking the samples in 40% hydrofluoric acid (HF) for 24 h. Finally, a *Lycopodium* tablet (10679 spores; produced by Lund University) was added as a marker (Stockmarr, 1971). Pollen grains were counted using an Eclipse 50i upright microscope until a sum of at least 500 arboreal pollen (AP) grains was reached. In 33 samples, this sum was not met due to the rapid peat accumulation rate, including seven samples in which a 100 AP sum was not achieved. Pollen taxa identification was carried out using atlases by Beug (1961) and Moore et al. (1991), along with reference grain samples from the Institute of Geoecology and Geoinformation, Adam Mickiewicz University, Poznań. In addition to pollen, selected non-pollen palynomorphs (NPPs) such as fungi and algae, as well as microscopic charcoal particles in size fractions of 0.01–0.1 mm and >0.1 mm, were counted. The counting of microscopic charcoal particles continued until their sum, combined with simultaneously counted *Lycopodium* spores, reached 200, following the approach of Finsinger and Tinner (2005) and Tinner and Hu (2003). Finally, palynological markers of human influence were categorized based on the guidelines by Behre (1981) and Gaillard (2013).

4.4. DATA PROCESSING AND ANALYSIS

RADIOCARBON DATING

The oft-used package rbacon (Blaauw et al., 2021) was used to develop an age-depth model and to calculate peat accumulation rates. The Bacon model uses Bayesian statistics (Blaauw and Christen, 2011) for its age-depth curves. To ensure accuracy, the youngest radiocarbon date (-552 +/- 23 cal yr BP) was disregarded due to its significant deviation from the stratigraphic context of other dates. To maintain consistency, an estimated surface sample age of -70 yr BP +/- 5 was introduced.. Calibration was performed using the IntCal20 (Reimer et al., 2020) and NH1 (Hua et al., 2013) calibration curves for pre-bomb and post-

bomb dates respectively. Mean values generated by the age-depth model at 1 cm resolution are referenced in subsequent sections.

BIOMARKER RATIOS AND INDICES

Biomarker data was reported as absolute concentrations in units of microgram per gram dry weight. With these values, a number of ratios and indices developed to better interpret biomarker data were calculated. Table 2 contains those used during the course of this PhD project.

Table 2. Biomarker ratios and proxies.

Compound class	Name	Equation	Indicates	Source
<i>n</i> -alkanes	CPI ¹	$\frac{(\sum_{i=n}^m C_{2i+1} + \sum_{i=n+1}^{m+1} C_{2i+1})}{2(\sum_{i=n+1}^{m+1} C_{2i})}$	Source and degree of degradation	Marzi et al., 1993
	ACL ¹	$\frac{\sum_{i=n}^m (2i + 1) * C_{2i+1}}{\sum_{i=n}^m C_{2i+1}}$	Dominant vegetation type, environmental conditions	Poynter et al., 1989
	P _{aq}	$\frac{C_{23} + C_{25}}{C_{23} + C_{25} + C_{29} + C_{31}}$	Terrestrial plant versus aquatic macrophyte input to lake sediments; water levels in peat bogs	Ficken et al., 2000
	P _{wax}	$\frac{C_{27} + C_{29} + C_{31}}{C_{23} + C_{25} + C_{27} + C_{29} + C_{31}}$	Proportion of waxy hydrocarbons from terrestrial plants to total hydrocarbons;	Zheng et al., 2007

			moisture levels	
	C_{23}/C_{25}	$\frac{C_{23}}{C_{25}}$	Shift in dominant <i>Sphagnum</i> species	McClymont et al., 2008
	$C_{23}/(C_{27}+C_{31})$	$\frac{C_{23}}{C_{27} + C_{31}}$	Proportion of mosses to vascular plants; moisture levels	Andersson et al., 2011
	$C_{23}/(C_{23}+C_{29})$	$\frac{C_{23}}{C_{23} + C_{29}}$	Proportion of mosses to vascular plants; moisture levels	Ronkainen et al., 2013
	$C_{25}/(C_{25}+C_{29})$	$\frac{C_{25}}{C_{25} + C_{29}}$	Proportion of mosses to vascular plants; moisture levels	Ronkainen et al., 2013
	OEP	$\frac{\sum_{i=13}^{16} C_{2i+1}}{\sum_{i=13}^{16} C_{2i}}$	Source and degree of degradation	Hoefs et al., 2002
<i>n</i> -alkanols	CPI ¹	$\frac{(\sum_{i=n}^m C_{2i} + \sum_{i=n+1}^{m+1} C_{2i})}{2(\sum_{i=n+1}^{m+1} C_{2i+1})}$	Source and degree of degradation	Adapted from Marzi et al., 1993
	ACL ¹	$\frac{\sum_{i=n}^m (2i) * C_{2i}}{\sum_{i=n}^m C_{2i}}$	Dominant vegetation type, environmental conditions	Adapted from Poynter et al., 1989

	C_{22}/C_{24}	$\frac{C_{22}}{C_{24}}$	Grass vs forest vegetation	Van Bergen et al. 1997
	$(C_{22}+C_{24})/(C_{26}+C_{28})$	$\frac{C_{22} + C_{24}}{C_{26} + C_{28}}$	Climatic changes in peat bog	Zheng et al., 2011
	C_{24}/C_{28}	$\frac{C_{24}}{C_{28}}$	Potential shifts in vegetation	Thomas et al., 2023
<i>n</i> -fatty acids	CPI^1	$\frac{(\sum_{i=n}^m C_{2i} + \sum_{i=n+1}^{m+1} C_{2i})}{2(\sum_{i=n+1}^{m+1} C_{2i+1})}$	Source and degree of degradation	Adapted from Marzi et al., 1993
	ACL^1	$\frac{\sum_{i=n}^m (2i) * C_{2i}}{\sum_{i=n}^m C_{2i}}$	Dominant vegetation type, environmental conditions	Adapted from Poynter et al., 1989
	$C_{15}/(C_{15}+C_{16})$	$\frac{C_{15}}{C_{15} + C_{16}}$	Warm and wet conditions favoring microbial degradation	Zheng et al., 2007
	C_{24}/C_{28}	$\frac{C_{24}}{C_{28}}$	Abundance of <i>Sphagnum</i> moss	Thomas et al., 2023

1. where C_x is the concentration of each lipid containing x carbon atoms; n and m are the chain lengths of, respectively, the starting and ending lipids divided by 2 (note: both $2n$ and $2m$ should be even numbers). For the n -alkanes, m is 11 and n is 15. For the n -alkanols, m is 10, and n is 14. For the n -fatty acids, m is 10, and n is 16.

CLUSTER ANALYSIS

The biomarker and elemental results of the core were subject to a constrained hierarchical clustering approach (CONISS, Grimm (1987)) to determine dissimilarity between the samples and to identify significant zones that separate the core into characteristic phases.

This approach is most often used on pollen data and determines major changes in vegetation assemblage through time. The analysis was performed with the R packages *vegan* (Oksanen et al., 2020) and *rioja* (Juggins, 2020). For the elemental data, the Euclidean distance was calculated based on the concentrations of C and N, the C/N ratio, and the C stable isotope values using the *dist* function. Conversely, biomarker concentration data was used to calculate Bray-Curtis dissimilarity, which is deemed more suitable for compositional data, computed through the *vegdist* function.. The CONISS analysis was performed on the dissimilarities with the *chclust* function, with clusters constrained by depth. The number of zones was determined by employing the broken-stick model (MacArthur, 1957; Bennett, 1996) with the *bstick* function.

ORDINATION

Because the plant biomarker data does not have a stratigraphic component, it was subject to ordination rather than constrained clustering. A PCA was completed using the R packages *FactoMineR* (Husson et al., 2023) and *factoextra* (Kassambara and Mundt, 2020). The abundances were normalized so that greater abundances were not given extra weight. The peat core data was also subject to ordination to facilitate comparison between the core and the modern plant samples.

4.5. MODELING

VERHIB IN MATLAB

The VERHIB model mentioned in the introduction is fully described in Jansen et al., 2010. It consists of a linear regression model describing the accumulation of plant-derived biomarkers over time in an archive; this forward model is inverted to estimate the plant assemblage that was most likely to have been present. Briefly, the forward model is based on the equation:

$$\begin{bmatrix} b_1(d, t) \\ \vdots \\ b_i(d, t) \end{bmatrix} = \begin{bmatrix} lf_1(d)lc_{1,1} & \cdots & lf_j(d)lc_{1,j} & rf_1(d)rc_{1,1} & \cdots & rf_j(d)rc_{1,j} \\ \vdots & \ddots & \vdots & \vdots & \ddots & \vdots \\ lf_1(d)lc_{i,1} & \cdots & lf_j(d)lc_{i,j} & rf_1(d)rc_{i,1} & \cdots & rf_j(d)rc_{i,j} \end{bmatrix} \begin{bmatrix} lm_1(t) \\ \vdots \\ lm_j(t) \\ rm_1(t) \\ \vdots \\ rm_j(t) \end{bmatrix}$$

in which $b_i(d, t)$ is the mass of biomarker i accumulated at depth d during time t , $lc_{i,j}$ is the concentration of biomarker i in the leaf of plant j , $rc_{i,j}$ is the concentration of biomarker i in the root of plant j , $lf_j(d)$ is the fraction of leaf mass of plant j accumulating at depth d , and $rf_j(d)$ is the same for root mass, $lm_j(t)$ is the leaf mass of plant j during time t and $rm_j(t)$ is the root mass of plant j during time t . The leaf to root mass ratio is a parameter that can be specified for each plant species based off of field data, literature, or estimation. Additionally, each plant's leaf and root biomass must be greater than or equal to zero as a constraint. It is also important to carefully consider which plant species to include in the database for the model as they should be those most likely to occur in the investigated location over the specified time period.

The inverse model is based on the matrix equation:

$$b = Ap$$

Where b is a vector of the biomarker composition in each depth layer, A is a matrix containing known constants, including those related to plant groupings and autocorrelation between depth layers, and p is a matrix of the parameters to be solved for. The equation is solved using a least-squares approach. Additionally, it is possible to specify two additional coefficients to increase the weight of the autocorrelation and the plant groupings to smooth the results.

EVALUATION

We calculated the Pearson correlation coefficient between the predicted core biomarker composition (y) and the actual values (x) using the equation:

$$r = \frac{\sum(x - \bar{x})(y - \bar{y})}{\sqrt{\sum(x - \bar{x})^2 \sum(y - \bar{y})^2}}$$

The Pearson correlation coefficient was used to check for equifinality.

5. RESULTS

5.1. TRANSFORMATION OF BIOMARKERS FROM PLANT TO SOIL

From the data gathered during the literature review, two distinct trends were readily identifiable: (1) decrease in total concentration of *n*-alkanes along the plant to soil continuum and (2) preferential degradation of odd chain lengths and shorter chain lengths. Additionally, the overall challenge of performing a systematic review of biomarker data highlighted a need for more uniform reporting standards going forward as well as a need for more research in underrepresented geographic areas. As *n*-alkanes are by far the most widely used and reported plant-derived biomarker, these findings are also applicable to *n*-alkanols and *n*-fatty acids, as well as other classes of compounds such as sterols and isoprenoids.

N-ALKANES FROM PLANT TO SOIL

Within the 37 studies, there were both litterbag experiments and studies on open plant-soil systems, defined as natural systems with no manipulation. Across all studies, there was a nearly ubiquitous trend of decreased concentration of *n*-alkanes with time or with depth. While this was not unexpected as the concentration of *n*-alkanes in plant tissue will be higher than in soil due to the changes in medium as soil is a heterogenous mixture of multiple components and not only organic matter, the extent of the decrease belies the assumption that *n*-alkanes are relatively recalcitrant. Although *n*-alkanes may persist longer than other organic compounds, they are still readily degradable.

RESULTS FROM LITTERBAG EXPERIMENTS

Six litterbag experiments were reviewed, ranging from 300 days to 23 years (Table 1, Manuscript I). These studies explored *n*-alkane degradation in various species and settings. The majority of experiments showed a substantial decline in *n*-alkane concentrations compared to initial biomass, with *Neosinocalanus affinis*, *Zea mays*, and *Pisum sativum* exhibiting the most significant drops (Fig. 1, Manuscript I). However, *Erigeron speciosus* and *Setaria viridis* demonstrated an increase in *n*-alkane concentration (Fig. 1, Manuscript I). The *n*-alkane distribution patterns remained relatively consistent across species (Supplementary Material, Manuscript I).

In four studies, the ACL was assessed, revealing minor changes over time. Notably, *N. affinis* had a significant increase in ACL, while *Calluna vulgaris*'s ACL decreased (Fig. 2a, Manuscript I). CPI data, available from the same studies, showed diverse trends. *N. affinis*'s CPI initially dropped but later increased, while *Fagus sylvatica*'s CPI grew over two years (Fig. 2b, Manuscript I). *C. vulgaris* and *Osmanthus fragrans* experienced slight CPI declines, spanning 23 years and 369 days, respectively. Wang et al.'s (2014) grass experiments displayed varied CPI changes, with *Amaranthus retroflexus* showing a unique pattern. Additionally, Zech et al. (2011) reported OEP, finding changes in *Acer pseudoplatanus*, *Sorbus aucuparia*, and *F. sylvatica* over 27 months (Fig. 2b, Manuscript I).

RESULTS FROM OPEN-PLANT SOIL SYSTEMS

In 21 studies, *n*-alkane compositions in litter, fresh plants, and soil were compared (Fig. 3, Table 2, Manuscript I). These studies spanned various climates and environments, grouped into six biomes: coniferous forest, deciduous forest, mixed forest, grassland or shrubland, peatland, and steppe, based on their locations.

Across most sites, *n*-alkane concentrations decreased consistently from fresh leaves to senescent leaves (if present), litter, organic layers (if present), and topsoil (Fig. 3, Manuscript I). The drop from litter to topsoil averaged 46%, and from fresh plants to topsoil, it was 87%. Notably, coniferous and mixed forest vegetation displayed increased *n*-alkane concentrations when normalized to TOC.

Distribution patterns of *n*-alkanes from plants to soil varied (Supplement, Manuscript I). Some findings were opposing. For instance, Zhang et al. (2017) found more long-chain *n*-alkanes (C₂₇-C₃₃) in peatlands dominated by *Sphagnum*, and fewer mid-chain *n*-alkanes (C₂₁-C₂₅). Conversely, Marseille et al. (1999) identified more mid-chain lengths in deeper litter layers of *F. sylvatica* forest soils. Other studies reported decreased relative concentrations of long-chain *n*-alkanes (Chikaraishi and Naraoka, 2006; Otto and Simpson, 2005; Hirave et al., 2020), while Nguyen Tu et al. (2001) noted a preference for shorter chain lengths in *Ginkgo biloba* leaves.

Figures 4a-c (Manuscript I) illustrate changes in ACL, CPI, and OEP from plants to soil, as reported or calculated. ACL generally rose from fresh plant material to topsoil, except in grassland, shrubland, and mixed forest sites (Fig. 4a, Manuscript I). CPI and OEP both decreased from fresh material to topsoil across all vegetation types, except for coniferous forests (Fig. 4b, 4c, Manuscript I).

RESULTS FROM SOIL PROFILES

Over time, partially decomposed plant material from the litter layer is integrated into the topsoil, aided by soil microbiota and organisms like earthworms. Below lies increased root-derived carbon and reduced litter-derived carbon (Angst et al., 2016). Limited research suggests root biomass could be a significant soil *n*-alkane source, especially in subsoil under specific conditions (Jansen and Wiesenberg, 2017). Numerous studies examined *n*-alkane levels in soil profiles across the six biomes.

Figures 5a and 5b (Manuscript I) display *n*-alkane concentration changes normalized to dry weight for different depths. Figures 6a and 6b (Manuscript I) show TOC-normalized data, available for deciduous forest, grassland, and peat vegetation. Results varied across studies and biomes. Coniferous forests showed mixed A horizon trends, with some increase and mostly decrease (Schäfer et al., 2016) (Fig. 5a, Manuscript I). Deciduous forests had varying results, depending on TOC normalization (Figs. 5a, 5b, 6a, 6b, Manuscript I). Contradictory A horizon trends emerged; Anokhina et al. (2018) reported an increase (Fig. 6b, Manuscript I), while Schäfer et al. (2016) found decreases (Fig. 5a, Manuscript I). Anokhina et al. (2018) found changes below, including an E horizon increase and B horizon decrease (Figs. 6a, 6b, Manuscript I). Others mostly saw decreasing concentrations (e.g., Angst et al., 2016; Bull et al., 2000; Cui et al., 2010; Wu et al., 2019). Grassland sites usually showed *n*-alkane concentration decline from A to B horizons (Fig. 5a, Manuscript I). Two grassland studies explored horizons below A: Celerier et al. (2009) reported a 53.56% A to B decrease, Feng and Simpson (2007) noted mixed A to B and B to C changes (Fig. 5a, Manuscript I). In the steppe biome, alternating *n*-alkane concentration increases and decreases were found down to 97.5 cm (Buggle et al., 2010).

Soil contained a broader *n*-alkane chain length range than plants or litter (e.g., Angst et al., 2016; Anokhina et al., 2018). Soil *n*-alkanes often included more even-chain-length types

than plants or litter, implying diverse sources (Almendros et al., 1996). Dominant chain lengths mostly remained consistent or shifted to longer lengths at lower depths (e.g., Angst et al., 2016; Anokhina et al., 2018; Bull et al., 2000). Most studies found C₂₅ as the dominant chain length in soil, confirming leaf wax as primary *n*-alkane source. Index measurements varied. ACL changed with depth (Figs. 7a, 7b, Manuscript I). CPI generally decreased (e.g., Angst et al., 2016; Celerier et al., 2009; Huang et al., 1996; Wu et al., 2019). OEP mostly decreased, with exceptions (Figs. 9a, 9b, Manuscript I).

5.2. BIOMARKERS AS PALEOVEGETATION PROXIES

BEERBERG CASE STUDY

RADIOCARBON DATES AND AGE-DEPTH MODEL

Using the radiocarbon dates and the Bacon age-depth model, the mean age of the lowest depth (340 cm) calculated was 2528 cal yr BP. The age-depth model (Fig. 2, Manuscript II) also enabled three clear phases of distinct accumulation rates to be seen: 0.66 mm/yr from 2528—1826 cal yr BP (340—293.5 cm), 1.99 mm/yr from 1826—978 cal yr BP (293.5—124.5 cm), and 1.27 mm/yr from 978 cal yr BP—Present (124.5—0 cm).

ELEMENTAL C AND N, STABLE C ISOTOPE

The CONISS analysis based on Euclidean distance separated the elemental data (C%, N%, C/N, $\delta^{13}\text{C}$) into five phases: 2528—2113 cal yr BP (Phase I-G), 2113—1569 cal yr BP (Phase II-G), 1569—1151 cal yr BP (Phase III-G), 1151—809 cal yr BP (Phase IV-G), and 809 cal yr BP—Present (Phase V-G) (Fig. 3, Manuscript II). The transitions between phases appear to be most correlated with changes in the $\delta^{13}\text{C}$ values, which generally increased through Phases I-G through III-G (ranging from -23.5‰— -26.9‰) and then decreased through Phases IV-G and V-G (ranging from -24.0‰— -29.3‰). The C/N ratio also followed a similar pattern to the $\delta^{13}\text{C}$ values. C/N ranged from 19.5-197.7, with the highest values in Phases III-G and IV-G.

BIOMARKER COMPOSITION AND ABUNDANCE

The CONISS analysis of the biomarker abundance data based on the Bray-Curtis dissimilarity index indicated four phases: 2528—1657 cal yr BP (Phase I-B), 1657—809 cal yr BP (Phase II-B), 809—35 cal yr BP (Phase III-B), and 35 cal yr BP—Present (Phase IV-B) (Fig. 6, Manuscript II). These phases are shortly described below:

PHASE I-B (2528—1657 CAL YR BP)

In the first phase, the total lipid extract (TLE) was relatively high but began to decrease around 2272 cal yr BP. The biomarker ratios followed differing patterns: P_{aq} , $C_{23}/(C_{23}+C_{29})$, $C_{25}/(C_{25}+C_{29})$, and $C_{23}/(C_{27}+C_{31})$ generally increased, while C_{23}/C_{25} and P_{wax} generally decreased. Of the *n*-alkanol ratios, $(C_{22}+C_{24})/(C_{26}+C_{28})$ and C_{24}/C_{28} increased throughout the phase while C_{22}/C_{24} decreased. For the *n*-fatty acid ratios, $C_{15}/(C_{15}+C_{16})$ stayed about the same while C_{24}/C_{28} increased.

PHASE II-B (1657—809 CAL YR BP)

In the second phase, the TLE continued to decrease until around 1209 cal yr BP when it began to increase. During this phase, many of the proxies followed a similar curve or its inverse, reaching either a peak or dip near the middle of the phase. These include ACL_{ALK} , $C_{23}/(C_{23}+C_{29})$, $C_{25}/(C_{25}+C_{29})$, $C_{23}/(C_{27}+C_{31})$, P_{aq} , P_{wax} , *n*-alkanol C_{22}/C_{24} , and *n*-fatty acid C_{24}/C_{28} (Fig. 7, Manuscript II).

PHASE III-B (809—35 CAL YR BP)

In Phase III-B, the TLE continued to increase, though with a slight dip from 466 cal yr BP to 259 cal yr BP. During this phase, many of the proxies reached a local maximum or minimum around 345 cal yr BP (Figs. 6 and 7, Manuscript II).

PHASE IV-B (35—PRESENT CAL YR BP)

In the fourth, most recent phase, there are only four samples, so patterns are difficult to identify. The TLE initially dipped from its values in Phase III-B but was relatively high in the uppermost sample.

MACROFOSSIL COMPOSITION

The CONISS analysis of the macrofossil composition, utilizing Bray-Curtis dissimilarity, identified four significant phases (Fig. 4, Manuscript II). Phase I-M (2528–2251 cal yr BP) was characterized by a substantial presence of *E. vaginatum* and a relatively high abundance of macrocharcoal, particularly at a depth of 328.5 cm (2388 cal yr BP). In Phase II-M (2251–1671 cal yr BP), *S. fuscum* became dominant. Moving into Phase III-M (1671–64 cal yr BP), *S. fuscum* maintained its dominance, but there was also a consistent, albeit minor, presence of *E. vaginatum* and Ericaceae rootlets. Lastly, Phase IV-M (64 cal yr BP–Present) saw the replacement of *S. fuscum* by *S. medium/divinum*, accompanied by an increase in Ericaceae rootlets and *E. vaginatum*.

POLLEN COMPOSITION

The CONISS analysis of the pollen assemblage led to the identification of four distinct phases, each indicating a unique regional vegetation composition (Fig. 5, Manuscript II).

PHASE I-P (2528–1816 CAL YR BP)

In the initial stage of Phase I-P (340–292.5 cm), the dominant tree species in the forests was *Fagus sylvatica*, accounting for pollen percentages ranging from 24% to 44.5% (Fig. 5, Manuscript II). Pollen from *P. sylvestris*, *Betula undiff.*, *Alnus undiff.*, *Abies alba*, and *P. abies* also contributed significantly to the arboreal pollen composition. Towards 1810 cal yr BP, there was a notable increase in the percentages of *Abies alba* and *P. abies*, coinciding with a period of heightened fire activity, as indicated by elevated CHAR_{micro} values (ranging from 6035 to 38,139 particles cm⁻² yr⁻¹) and the presence of *Neurospora* and *Gelasinospora* ascospores between 2500–2300 cal yr BP (Stivrins et al., 2019). Towards the end of this phase, there was a rise in *Sphagnum*, signaling a transition to moss-dominated peat.

PHASE II-P (1816–1092 CAL YR BP)

In the second phase, *F. sylvatica* continued to dominate the forests, with percentages ranging from 18% to 54.5% (Fig. 5, Manuscript II). However, a noticeable decline in *F. sylvatica* occurred between 1280–1210 cal yr BP. Human impact, based on indicator pollen counts, was least significant during this phase across the entire paleorecord. Towards the end, there was evidence of crop introduction in the region, reflected in an increased share of *Cerealia* pollen. The peatland maintained stable conditions during this time, dominated by *Sphagnum*, *C. vulgaris*, and other Ericaceae species.

PHASE III-P (1092–366 CAL YR BP)

Arboreal pollen decreased from 97.5% to 77.5% between 1090–570 cal yr BP (Fig. 5, Manuscript II). Late successional species like *F. sylvatica* and *C. betulus* experienced a significant decline, particularly towards the end of this phase. By contrast, pioneer trees such as *P. sylvestris*, *Betula*, and *Corylus avellana* increased in proportion. Cultivated indicators, mainly *Cerealia undiff.* and *Secale cereale*, maintained a consistent share of the pollen assemblage, especially from 740 cal yr BP. Simultaneously, there was a sharp increase in CHAR_{micro} and coprophilous fungi taxa. *Sphagnum* proportions decreased sharply from 740 to 570 cal yr BP, rebounding at the phase's conclusion.

PHASE IV-P (366 CAL YR BP – PRESENT)

Deciduous trees that were previously dominant gradually receded from the site, reflected in decreasing percentages of *F. sylvatica* (from 18% to 4.4%), *Quercus*, and *Corylus avellana* (Fig. 5, Manuscript II). *P. abies* and *P. sylvestris* reached their highest proportions in the forest (13.5%–58% and 21%–32.5%, respectively), while *A. alba* appeared to decline completely.

5.3. TOWARDS IMPROVED QUANTITATIVE ASSESSMENT OF BIOMARKER DATA

Following the evaluation of the Beerberg peat biomarkers, the dataset was used to test the VERHIB model. First, the modern plant samples collected at the Beerberg site were analyzed for their biomarker composition to serve as input for the model.

BEERBERG PLANT BIOMARKER SIGNATURES

Across all species and plant parts, some general trends could be identified. Generally, the *n*-fatty acids had the highest absolute concentration, greater than the *n*-alkane and *n*-alkanol absolute concentrations by an order of magnitude. Between the *n*-alkanes and *n*-alkanols, *n*-alkanes were typically more abundant in leaves/non-woody aboveground biomass. In contrast, *n*-alkanols were primarily the more abundant compound class in stem and root samples. Of the *n*-alkanes, C₃₁ was most often the most abundant homologue (C_{max}), followed by C₂₉. For *n*-alkanols, C₂₈ was the most frequent C_{max}, closely followed by C₂₄. For *n*-fatty acids, C₂₄ was the most frequent C_{max}, followed by C₂₀ and C₂₈.

When a PCA was performed using all of the identified biomarkers and calculated ratios (see Methods section), PC1 was found to explain 25.1% of the variance and PC2 21.6% (Figure 2a, Manuscript III). The variables contributing the most to the variance were P_{aq}, P_{wax}, C₂₅ *n*-alkane, C₂₆ and C₂₈ *n*-alkanols, and the *n*-alkane ratio C₂₃/(C₂₃+C₂₉). The leaves, moss and root samples all had distinct clusters, while the stem samples were spread out.

The same PCA was also performed on the peat core samples. PC1 explained 51.6% of the variance while PC2 explained 12.8% (Figure 3a, Manuscript III). The top variables contributing to the variance were C₂₉ and C₃₁ *n*-alkanes, C₂₂ and C₂₈ *n*-alkanols, P_{wax}, and the C₂₅/(C₂₅+C₂₉) ratio.

VERHIB MATLAB WITH BEERBERG

Four groups of scenarios were run in the VERHIB model using the Beerberg dataset. For all of these scenarios, the plants were grouped as follows: Group 1: *Sphagnum angustifolium*, *S. magellanicum*, *S. capillifolium*, *S. fuscum*, Group 2: *C. vulgaris*, *Vaccinium uliginosum*, *V. myrtillus*, *V. vitis-idaea*, *Oxycoccus palustris*, *Empetrum nigrum*, Group 3: *Eriophorum vaginatum*, Group 4: *Polytrichum strictum*, Group 5: *Pinus sylvestris*, Group 6: *Picea abies*, and Group 7: *Betula pendula* and *B. pubescens*. The two regularization parameters were kept at 0.1, and the maximum iterations were set at 2000. In each group, the first run included all three compound classes, the second *n*-alkanes and *n*-alkanols, the third included only *n*-alkanes, and the fourth *n*-alkanes and *n*-fatty acids.

The first group included all of the measured chain lengths, not only the plant-derived, and root data was not included. When compared to the Beerberg plant macrofossil analysis, if *n*-fatty acid data was excluded from the run, the contribution of tree species was overestimated. All of the runs underestimated the contribution of *E. vaginatum*, especially in the bottom part of the core, which also coincided with the highest residual values for the predicted versus actual core biomarker composition.

The second group included all of the chain lengths and the root biomarker data, with a leaf:root mass ratio of 5:1. The results of this group were similar to that of the first in that *E. vaginatum* was underestimated in all of the scenarios. If the *n*-fatty acid data was excluded, the contribution of *S. fuscum* was underestimated.

For the third group of scenarios only plant-derived chain lengths were included and the root biomarker data was excluded. Again, all of the runs underestimated the proportion of *E. vaginatum*. In the runs including *n*-fatty acids, the contribution of *S. magellanicum* was overestimated. In runs excluding *n*-fatty acids, the contribution of *P. sylvestris* and *P. abies* was overestimated.

For the last group of scenarios, only the plant-derived chain lengths were included as well as both leaf and root biomarker data, with a leaf:root mass ratio of 5:1. The primary difference between the reconstructions of this group (Fig. 10, Manuscript III) with the last

group (plant-derived, no root data) (Fig. 8, Manuscript III) is that the estimated contribution of *P. strictum* increased in all the scenarios.

6. DISCUSSION

In this chapter, the main findings of the thesis are synthesized by addressing the overarching objectives and research questions presented in Chapter 1. Additionally, general conclusions and a perspective on future research are presented.

6.1. MAIN FINDINGS

Objective I: Identify processes and parameters that should be considered in the improved model

RQ 1: Can changes in biomarker composition along the plant to soil continuum be readily quantified into a widely applicable parameter?

In Manuscript I, a systematic literature review was performed with the goal of developing a quantitative, mechanistic understanding of how *n*-alkane composition transforms from its origin in fresh plant tissue to its final destination incorporated into soil or peat. The results would be used to determine what processes should be considered for an updated version of the VERHIB model, as well as to calculate a potential degradation parameter that could be used in the model to apply globally, as previously degradation rates and coefficients have only been calculated at specific sites. *n*-Alkanes were chosen as the representative biomarker for the review because they are by far the most commonly studied (Jansen and Wiesenberg, 2017).

Using as systematic of an approach as possible, data were extracted from 37 studies available via Web of Science out of 9297 results from the initial search. A meta-analysis was performed on the extracted data by grouping the studies into six biome types based on dominant vegetation: coniferous forest, deciduous forest, mixed forest, grassland or shrubland, peatland, and steppe. The analysis examined how *n*-alkane concentrations, normalized to either the samples dry weight or total organic carbon, changed with time and with depth, as well as how the most common biomarker proxy measures (CPI, OEP, ACL) varied with time and depth.

Ultimately, only two clear trends could be identified in the analysis. The first was that relative to the dry weight of biomass, the concentration of *n*-alkanes decreased from fresh plant tissue to soil, though if normalized to total organic carbon, there were instances in which the *n*-alkane concentration increased. This was not unexpected as the concentration in plant tissue will be higher as soil is a heterogenous mixture of many more components than organic material. However, the considerable extent of the decrease in most studies shows that *n*-alkanes are readily degradable, though potentially less so than other sources of soil carbon (Schmidt et al., 2011).

Additionally, there was preferential degradation of odd chain lengths and shorter chain lengths. This was clear through the decreases in CPI and OEP and increase in ACL. While this was generally known, as CPI and OEP can be used to determine if organic matter has been degraded, the finding underscores the importance of taking care when comparing the biomarker signatures of fresh plant material to that of soil or peat samples, particularly if only concentration is being measured. The preferential degradation of certain chain lengths means that the distribution pattern of *n*-alkanes changes and could result in misattribution of source. These results indicate that the degradation process should be considered in future iterations of the VERHIB model and others aiming to use biomarker concentration data to increase accuracy.

Disappointingly, the review also showed that there is a lack of uniform reporting standards for biomarker study results as well as a lack of publicly accessible data. As systematic reviews and meta-analyses have been used increasingly in fields outside of medicine and as the movement for open science expands, aggregating studies and data to determine global trends is much more efficient than endless iterations of smaller more limited studies (Gurevitch et al., 2018). Therefore, as part of Manuscript I, guidelines for future reporting of biomarker data were recommended, adapted from previously published guidelines for soil incubation datasets (Schädel et al., 2020). These guidelines could enable future systematic reviews to be more comprehensive and enable the calculation of widely applicable degradation parameters.

Objective II: Paleoecological reconstruction of the Beerberg peatland using plant-derived biomarkers and traditional paleobotanical proxies

RQ 1: How did the climate and vegetation develop over time at the Beerberg peatland?

The radiocarbon dates and age-depth model indicated that the Beerberg core spanned 2600 years. Although the CONISS analysis of the different proxies identified different significant phases, as macrofossils are seen as the proxy most reliably reflecting in situ vegetation (Birks and Birks, 2000), the development of the peatland is described using the phases from the macrofossil analysis.

In the first phase, from 2528–2251 cal yr BP, the low C/N ratio and negative $\delta^{13}\text{C}$ values (Fig. 3, Manuscript II), along with the main vegetation being *E. vaginatum* (Fig. 4, Manuscript II), are typical indicators for fen or transitional peat underlying bog (Kuhry et al., 1992; Jones et al., 2010). There was also evidence of high fire activity during this phase, which could have been the spark for peatland development, as fen peat can form on wet ground following fire, then develop into bog peat (e.g., Tuittila et al., 2007; Gałka et al., 2019). Towards the end of the phase, there is a shift towards more ombrotrophic conditions, indicated by the increasing C/N ratio and $\delta^{13}\text{C}$ values (Fig. 3, Manuscript II) (Wang et al., 2015) and the first incidences of *Sphagnum* (Figs. 4 and 5, Manuscript II). The biomarker measurements also support this interpretation with the relative abundance of C_{23} and C_{25} *n*-alkanes increasing throughout the phase, indicating an increase in *Sphagnum* mosses and the beginning of bog peat development (Baas et al., 2000; Pancost et al., 2002; Bingham et al., 2010).

Following the transition to more ombrotrophic conditions, *S. fuscum* was dominant and the main peat-forming plant in the second phase, 2251—1671 cal yr BP (Fig. 4, Manuscript II). The pollen data also showed a rapid increase in the proportion of *Sphagnum* spores (Fig. 5, Manuscript II) and the *n*-alkane ratios continued to indicate an increase in the proportion of C_{23} and C_{25} (Fig. 7, Manuscript II). Rapid peat growth and low decomposition was also echoed in the increase of the C/N ratio and $\delta^{13}\text{C}$ values (Loisel et al., 2010; Kuhry and Vitt, 1996), as well as an increased accumulation rate (Fig. 2, Manuscript II).

Although *S. fuscum* remained dominant in the third phase, 1671—64 cal yr BP, there was slightly more variation in the plant macrofossils in the latter half of the phase, with *E. vaginatum*, *S. medium/divinum*, and *Polytrichum* returning (Fig. 4, Manuscript II). The biomarker records were not as constant as the macrofossils. The TLE and individual concentrations were low but began to increase around 809 cal yr BP, followed by a dip from around 466 cal yr BP to 259 cal yr BP (Fig. 6, Manuscript II). Most of the biomarker ratios also followed this pattern, with a maximum or minimum point around 1200 cal yr BP (Fig. 7, Manuscript II) and a local maximum or minimum around 345 cal yr BP. This was echoed in the $\delta^{13}\text{C}$ values and corresponded to a color change in the peat itself (Fig. 3, Manuscript II).

Considering these proxies together, a decrease in water level and moss abundance occurs around 809 cal yr BP, followed by an increase from 500 cal yr BP to 345 cal yr BP with a subsequent decrease. These conditions are likely related to the regional climate patterns in Europe during this time period. The first decrease around 809 cal yr BP, with lower water table levels and drier conditions, is likely caused by the Medieval Climate Anomaly (MCA) (Luterbacher et al., 2016). Following this period, there is likely colder, wetter conditions causing an increase in water table levels or surface moisture around 500 cal yr BP caused by the Little Ice Age (LIA). Similar changes have been noted in other peat archives (Barber et al., 2004; Marcisz et al., 2020).

In the fourth phase, 64 cal yr BP—Present, the dominant *Sphagnum* species shifted to *S. medium/divinum* and the proportion of *E. vaginatum* increased (Fig. 4, Manuscript II). This shift in *Sphagnum* species has been identified as being related to pollution and increased dust deposition in other studies (Gałka et al., 2019, 2022a, b) and the increase in fen vegetation could indicate further dry conditions, consistent with recent drying of peatlands across Europe (Swindles et al., 2019), related to drainage as well as climate change.

While the plant macrofossils alone provide an adequate reconstruction of the Beerberg peatland vegetation, the addition of the biomarker analysis in tandem with the geochemical data enabled deeper insight into the local response to regional climate shifts. As the timing of the MCA and LIA varies across European paleoclimate records (Wanner et al., 2022) and

such records are sparse for the Thuringian Forest, this is an important insight into the development of the Beerberg peatland.

RQ 2: What biomarker ratios are most indicative for paleo transitions at the Beerberg peatland?

As paleoreconstructions including *n*-alkanes, *n*-alkanols, and *n*-fatty acids are rare, the Beerberg study provided an opportunity to evaluate known biomarker ratios as well as investigate potential new ones. Biomarker ratios are important for the interpretation of biomarker data because single compounds are not source-specific, but the relationship between multiple compounds can be. There are many established ratios for interpreting *n*-alkane data (see Methods section), but far fewer for *n*-alkanol and *n*-fatty acid data.

In this study, the included biomarker ratios allowed for the identification of a local response to the MCA and LIA, which was seen in the shifts of *Sphagnum* abundance in the pollen data, but not in the more locally representative plant macrofossils. The P_{aq} and related *n*-alkane ratios (P_{wax} , $C_{23}/(C_{27}+C_{31})$, $C_{23}/(C_{23}+C_{29})$, $C_{25}/(C_{25}+C_{29})$) generally correlated well with the *Sphagnum* trends seen in the macrofossil and pollen data, and in the case of the Beerberg site, all of these ratios provided similar results. The *n*-alkanol ratio C_{22}/C_{24} and the *n*-fatty acid ratio C_{24}/C_{28} also roughly followed these trends. While they have not been previously linked to *Sphagnum* mosses as indicative ratios, future peat biomarker studies could investigate their potential further, enabling better interpretation of *n*-alkanol and *n*-fatty acid datasets. Although the use of biomarker ratios allows for improved qualitative interpretation of biomarker data, there is still room for quantitative interpretation to be better integrated into such paleovegetation and paleoclimate reconstruction studies.

Objective III: Evaluate VERHIB (MATLAB) performance using Beerberg peatland biomarker dataset

RQ 1: How does the biomarker composition of modern vegetation samples vary across plant organs and groups?

For the VERHIB model, the biomarker compositions of likely vegetation species are used as input. Due to the various climate and environmental factors influencing the composition, it has been the common practice to sample and measure biomarkers in modern vegetation

at the study site, even though data has been previously published for some species. Previously published data however does not typically include *n*-alkanols and *n*-fatty acids and is often limited to only leaves. Overall, the results from the Beerberg study, particularly the *n*-alkane measurements of leaves, generally confirm what has been found in other northern and central European peatland settings with some differences in dominant chain lengths and absolute concentrations. These discrepancies can be explained either through the different climate and ontological factors that can influence plant-derived biomarker composition, and differences in absolute concentrations can also arise due to methodological differences in sample preparation.

The results also show overlap between plant groups (Moss, Sedge, Shrub, Tree) and plant parts (Aboveground (non-woody), Leaves, Moss, Needles, Roots, Stems), making source apportionment based on biomarker compositions more challenging. This was most apparent in the *n*-alkane measurements, where *Sphagnum* mosses had similar results to tree stems or tree roots, depending on the considered ratio (Figs. 1 and 2, Manuscript III). These results illustrate the need of having a localized library of plant biomarker data to compare to archives, as well as the need to consider multiple indicative biomarker ratios in case the local signatures of the plants cause the ratios to be less effective.

RQ2: What combination of parameters provides the best model fit for the Beerberg sequence?

The combination of parameters that provided the most comparable results to the plant macrofossil records were those that included all of the measured chain lengths for *n*-alkanes, *n*-alkanols, and *n*-fatty acids, not only plant-derived chain lengths (Run 1, Run 4, Run 5, Run 8) (Figs. 4a, 4d, 6a, 6d; Manuscript III). In these runs, *S. fuscum* was correctly identified as the dominant moss species through the majority of the core. These reconstructions also correctly estimated a higher proportion of Ericaceae shrubs at the bottom and top of the core. However, *E. vaginatum* was underestimated, particularly at the base, showing that the model struggled to recognize the transition from poor fen to peat bog. If only plant-derived chain lengths were included in the model, *S. fuscum* was not the dominant moss. Additionally, the proportion of tree species was overestimated in the runs in which *n*-fatty acid data was excluded. In the Beerberg sequence, the addition of root data

did not change the results significantly, likely due to the domination of *Sphagnum* moss and relatively little root input from other plants.

*RQ 3: What does the inclusion of *n*-alkanols and *n*-fatty acids add to the analysis?*

Most previous studies of biomarkers focus solely on *n*-alkane measurements. During the investigation of the Beerberg sequence, both the qualitative comparison of biomarker measurements to plant macrofossil and pollen records, the addition of *n*-alkanols and *n*-fatty acids allowed for improved interpretation due to the overlap in *n*-alkane composition between plant species and plant parts. This was the first attempt at using the VERHIB model with *n*-fatty acid data included and the results showed that all three compound classes were necessary to produce the most comparable reconstruction. Additionally, the Pearson correlation coefficients indicated that without the *n*-fatty acid data, the model runs were overparameterized and could be solved with a non-unique solution. While root data did not affect the outcome significantly using the Beerberg sequence, the inclusion of *n*-alkanols and *n*-fatty acids will enable the VERHIB model to better differentiate aboveground versus belowground matter.

7. CONCLUSIONS AND PERSPECTIVE

MAIN CONTRIBUTIONS AND INSIGHTS

This dissertation explored the possibilities for more quantitative application of plant-derived biomarkers in determining source apportionment of organic matter in terrestrial archives. The included work demonstrated not only the usefulness of biomarkers in multi-proxy studies, but the usefulness of including multiple lipid classes in biomarker investigations and not only relying on *n*-alkanes as well as guidance for the next steps in developing a standard quantitative approach to vegetation reconstruction and source apportionment using the VERHIB model.

During the project, the following insights were gained. The literature review performed in Manuscript I confirmed that, despite degradation being rarely considered in biomarker reconstructions, *n*-alkanes readily degrade with time and/or depth along the trajectory from plant source to depositional archive. Additionally, there is preferential degradation of odd

chain lengths and shorter chain lengths. These results are not unexpected but show that degradation does need to be considered to accurately interpret biomarker data. The review also illustrated a need for better data transparency and standardization across biomarker studies and enabled the development of guidelines for future data reporting.

The paleovegetation reconstruction study performed of the Beerberg peatland in Manuscript II not only provided a high-resolution interpretation of peatland development in an understudied area but the biomarker data enabled the identification of the local timing of the regional climate shifts, the MCA and LIA. Furthermore, the inclusion of *n*-alkanes, *n*-alkanols, and *n*-fatty acids allowed for the identification of two new potential indicative biomarker ratios for *n*-alkanols and *n*-fatty acids related to the abundance of *Sphagnum* mosses. The biomarker dataset gathered during the Beerberg investigation could also serve as a test dataset to evaluate the VERHIB model.

Previously, the model had not been tested using *n*-fatty acid data, only *n*-alkane and *n*-alkanol. Manuscript III aimed to investigate the best combination of parameters for reconstructing the development of the Beerberg peatland. In addition, biomarker measurements were gathered for local vegetation at a high level of detail; some of the included species had never had previous measurements published and some had only had *n*-alkane data previously published. Ultimately, the testing showed that including all of the available compound data from *n*-alkanes, *n*-alkanols, and *n*-fatty acids was essential for producing the reconstruction most comparable to available plant macrofossil records from the Beerberg sequence. It is therefore recommended that future studies include all of the straight-chain lipid biomarkers in analyses rather than just *n*-alkanes as is still most common.

LIMITATIONS AND OPPORTUNITIES OF BIOMARKER APPLICATION FOR PALEOECOLOGICAL RECONSTRUCTIONS CONCENTRATIONS

One important point illustrated by the work included in this dissertation is that there are many limitations on the use of only biomarker concentrations and distribution patterns. The lack of chemotaxonomic specificity and consistency makes their use for vegetation reconstruction purposes complicated as well as potentially misleading. Biomarker concentrations can be used for identifying general sources of organic matter as well as

evaluating how degraded organic matter is. However, for more precision, biomarker concentrations should be measured as part of a multi-proxy study including compound-specific isotope analysis or paleobotanical proxies such as pollen or plant macrofossils.

In spite of these limitations, this work has shown that biomarkers can be a valuable contribution to paleoecological studies as they can confirm or provide deeper insight into paleobotanical proxies. The limitations of chemotaxonomic specificity can be reduced when multiple component classes, i.e., *n*-alkanes, *n*-alkanols, and *n*-fatty acids) are used. Biomarkers can also enable a better understanding of aboveground vs belowground organic matter input due to differences in composition between plant parts. Systematic reconstruction models such as VERHIB can unravel large suites of biomarker composition data to provide a quantitative assessment of source apportionment in a depositional archive.

RECOMMENDATIONS FOR IMPROVEMENT OF MODELING

Historically, soil science has included a lot of qualitative research, including source attribution using biomarker concentrations. Models, such as VERHIB, increase the quantitative rigor and reproducibility of biomarker reconstructions. In its current form, the VERHIB model does not explicitly consider any potential degradation or transportation of biomarkers. Although there is a lot of data on *n*-alkane degradation in different ecosystems, further experimental research is needed to quantify degradation of *n*-alkanols and *n*-fatty acids in comparison.

It also only considers the sources that the user inputs. The robustness of the VERHIB model could be increased if pollen or other proxy data could also be considered to guide the reconstruction of the biomarkers more precisely. However, in order to develop such a module, more complete datasets such as the Beerberg sequence with high-resolution biomarker and other proxy data available are necessary for testing. Finally, the VERHIB model itself should be reprogrammed into R as R is more widely used than MATLAB in environmental research and it is open-source, enabling wider adoption.

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To improve the potential of quantifying degradation coefficients and understanding how biomarkers change through the plant-to-soil continuum in varying environments, the

adoption of uniform reporting standards, as well as increasing access to data from published studies, is essential. Potential guidelines for future data reporting can be found in Table 3 of Manuscript I. These guidelines were adapted from those for a soil incubation database (Schädel et al., 2020) with the rationale that having a more comprehensive overview of the characteristics of the depositional archive will improve future possibilities for systematic reviews like the one attempted in Manuscript I. Additionally, increased open access to data by publication in databases such as Pangaea, will make it easier for the community to identify knowledge gaps as well as to increase the opportunities for using training and testing data to validate models such as VERHIB.

8. REFERENCES

- Almendros, G., Sanz, J., & Velasco, F. (1996). Signatures of lipid assemblages in soils under continental Mediterranean forests. *European Journal of Soil Science*, 47(2), 183–196. <https://doi.org/10.1111/j.1365-2389.1996.tb01389.x>
- Andersson, R. A., Kuhry, P., Meyers, P., Zebühr, Y., Crill, P., & Mörth, M. (2011). Impacts of paleohydrological changes on *n*-alkane biomarker compositions of a Holocene peat sequence in the eastern European Russian Arctic. *Organic Geochemistry*, 42(9), 1065–1075. <https://doi.org/10.1016/j.orggeochem.2011.06.020>
- Angst, G., John, S., Mueller, C. W., Kögel-Knabner, I., & Rethemeyer, J. (2016). Tracing the sources and spatial distribution of organic carbon in subsoils using a multi-biomarker approach. *Scientific Reports*, 6(1), 1–12. <https://doi.org/10.1038/srep29478>
- Anokhina, N. A., Demin, V. V., & Zavgorodnyaya, Yu. A. (2018). Compositions of *n*-alkanes and *n*-methyl ketones in soils of the forest-park zone of Moscow. *Eurasian Soil Science*, 51(6), 637–646. <https://doi.org/10.1134/S1064229318060030>
- Baas, M., Pancost, R., Van Geel, B., & Sinninghe Damsté, J. S. (2000). A comparative study of lipids in *Sphagnum* species. *Organic Geochemistry*, 31(6), 535–541. [https://doi.org/10.1016/S0146-6380\(00\)00037-1](https://doi.org/10.1016/S0146-6380(00)00037-1)
- Barber, K., Chambers, F., & Maddy, D. (2004). Late Holocene climatic history of northern Germany and Denmark: Peat macrofossil investigations at Dosenmoor, Schleswig-Holstein, and Svanemose, Jutland. *Boreas*, 33(2), 132–144. <https://doi.org/10.1080/03009480410001082>
- Barber, K. E., Chambers, F. M., Maddy, D., Stoneman, R., & Brew, J. S. (1994). A sensitive high-resolution record of late Holocene climatic change from a raised bog in northern England. *The Holocene*, 4(2), 198–205. <https://doi.org/10.1177/095968369400400209>
- Behre, K. E. (1981). Interpretation of anthropogenic indicators in pollen diagrams, *Pollen et Spores*, 23, 225–245.
- Bennett, K. D. (1996). Determination of the number of zones in a biostratigraphical sequence, *New Phytologist*, 132, 155–170, <https://doi.org/10.1111/j.1469-8137.1996.tb04521.x>

- Berglund, B. E. & Ralska-Jasiewiczowa, M. (1986). Pollen analysis and pollen diagrams. In Berglund, B. E. (Ed.), *Handbook of Holocene Palaeoecology and Palaeohydrology*, pp. 455–484, John Wiley & Sons: Chichester.
- Beug, H. J. (1961). *Leitfaden der Pollenbestimmung für Mitteleuropa und angrenzende Gebiete*, G. Fischer.
- Bingham, E. M., McClymont, E. L., Väiliranta, M., Mauquoy, D., Roberts, Z., Chambers, F. M., Pancost, R. D., & Evershed, R. P. (2010). Conservative composition of *n*-alkane biomarkers in *Sphagnum* species: Implications for palaeoclimate reconstruction in ombrotrophic peat bogs. *Organic Geochemistry*, *41*(2), 214–220. <https://doi.org/10.1016/j.orggeochem.2009.06.010>
- Birks, H. H., & Birks, H. J. B. (2000). Future uses of pollen analysis must include plant macrofossils. *Journal of Biogeography*, *27*(1), 31–35. <http://www.jstor.org/stable/2655981>
- Blaauw, M. & Christen, J. A. (2011). Flexible paleoclimate age-depth models using an autoregressive gamma process, *Bayesian Analysis*, *6*, 457–474, <https://doi.org/10.1214/11-BA618>
- Blaauw, M., Christen, J. A., & Aquino López, M. A. (2021). *rbacon: Age-Depth Modelling using Bayesian Statistics* [Computer software], <https://CRAN.R-project.org/package=rbacon>, r package version 2.5.2.
- Buggle, B., Wiesenberg, G. L. B., & Glaser, B. (2010). Is there a possibility to correct fossil *n*-alkane data for postsedimentary alteration effects? *Applied Geochemistry*, *25*(7), 947–957. <https://doi.org/10.1016/j.apgeochem.2010.04.003>
- Bull, I. D., van Bergen, P. F., Nott, C. J., Poulton, P. R., & Evershed, R. P. (2000). Organic geochemical studies of soils from the Rothamsted classical experiments—V. The fate of lipids in different long-term experiments. *Organic Geochemistry*, *31*(5), 389–408. [https://doi.org/10.1016/S0146-6380\(00\)00008-5](https://doi.org/10.1016/S0146-6380(00)00008-5)
- Canadell, J. G., Monteiro, P. M. S., Costa, M. H., Cotrim da Cunha, L., Cox, P. M., Eliseev, A. V., Henson, S., Ishii, M., Jaccard, S., Koven, C., Lohila, A., Patra, P. K., Piao, S., Rogelj, J., Syampungani, S., Zaehle, S., & Zickfeld, K. (2021). Global Carbon and other Biogeochemical Cycles and Feedbacks. In *Climate Change 2021: The Physical Science Basis. Contribution of Working Group I to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change* (pp. 673–816). Cambridge University Press.
- Celerier, J., Rodier, C., Favetta, P., Lemee, L., & Ambles, A. (2009). Depth-related variations in organic matter at the molecular level in a loamy soil: Reference data for a long-term experiment devoted to the carbon sequestration research field. *European Journal of Soil Science*, *60*(1), 33–43. <https://doi.org/10.1111/j.1365-2389.2008.01085.x>
- Chambers, F. M., Booth, R. K., De Vleeschouwer, F., Lamentowicz, M., Le Roux, G., Mauquoy, D., Nichols, J. E., & van Geel, B. (2012). Development and refinement of proxy-climate indicators from peats. *Quaternary International*, *268*, 21–33. <https://doi.org/10.1016/j.quaint.2011.04.039>
- Chibnall, A. C., Piper, S. H., Pollard, A., Williams, E. F., & Sahai, P. N. (1934). The constitution of the primary alcohols, fatty acids and paraffins present in plant and insect waxes. *Biochemical Journal*, *28*(6), 2189–2208. <https://doi.org/10.1042/bj0282189>

- Chikaraishi, Y., & Naraoka, H. (2006). Carbon and hydrogen isotope variation of plant biomarkers in a plant–soil system. *Chemical Geology*, 231(3), 190–202. <https://doi.org/10.1016/j.chemgeo.2006.01.026>
- Cranwell, P. A. (1981). Diagenesis of free and bound lipids in terrestrial detritus deposited in a lacustrine sediment. *Organic Geochemistry*, 3(3), 79–89. [https://doi.org/10.1016/0146-6380\(81\)90002-4](https://doi.org/10.1016/0146-6380(81)90002-4)
- Crausbay, S., Genderjahn, S., Hotchkiss, S., Sachse, D., Kahmen, A., & Arndt, S. K. (2014). Vegetation dynamics at the upper reaches of a tropical montane forest are driven by disturbance over the past 7300 years. *Arctic, Antarctic, and Alpine Research*, 46(4), 787–799. <https://doi.org/10.1657/1938-4246-46.4.787>
- Cui, J., Huang, J., Meyers, P. A., Huang, X., Li, J., & Liu, W. (2010). Variation in solvent-extractable lipids and *n*-alkane compound-specific carbon isotopic compositions with depth in a southern China karst area soil. *Journal of Earth Science*, 21(4), 382–391. <https://doi.org/10.1007/s12583-010-0101-5>
- Eglinton, G., & Hamilton, R. J. (1967). Leaf Epicuticular Waxes. *Science*, 156, 1322–1335. <https://doi.org/10.1126/science.156.3780.1322>
- Eglinton, T. I., & Eglinton, G. (2008). Molecular proxies for paleoclimatology. *Earth and Planetary Science Letters*, 275(1–2), 1–16. <https://doi.org/10.1016/j.epsl.2008.07.012>
- Erb, K.-H., Kastner, T., Plutzer, C., Bais, A. L. S., Carvalhais, N., Fetzel, T., Gingrich, S., Haberl, H., Lauk, C., Niedertscheider, M., Pongratz, J., Thurner, M., & Luysaert, S. (2018). Unexpectedly large impact of forest management and grazing on global vegetation biomass. *Nature*, 553(7686), 73–76. <https://doi.org/10.1038/nature25138>
- Farrimond, P., & Flanagan, R. L. (1996). Lipid stratigraphy of a Flandrian peat bed (Northumberland, UK): Comparison with the pollen record. *The Holocene*, 6(1), 69–74. <https://doi.org/10.1177/095968369600600108>
- Feng, X., & Simpson, M. J. (2007). The distribution and degradation of biomarkers in Alberta grassland soil profiles. *Organic Geochemistry*, 38(9), 1558–1570. <https://doi.org/10.1016/j.orggeochem.2007.05.001>
- Ficken, K. J., Li, B., Swain, D. L., & Eglinton, G. (2000). An *n*-alkane proxy for the sedimentary input of submerged/floating freshwater aquatic macrophytes. *Organic Geochemistry*, 31(7), 745–749. [https://doi.org/10.1016/S0146-6380\(00\)00081-4](https://doi.org/10.1016/S0146-6380(00)00081-4)
- Finsinger, W. & Tinner, W. (2005). Minimum count sums for charcoal concentration estimates in pollen slides: Accuracy and potential errors. *The Holocene*, 15, 293–297. <https://doi.org/10.1191/0959683605hl808rr>
- Gaillard, M.-J. (2013). Archaeological applications. In, *The Encyclopedia of Quaternary Science*, pp. 880–904, Elsevier.
- Gałka, M., Diaconu, A.-C., Feurdean, A., Loisel, J., Teickner, H., Broder, T., & Knorr, K.-H. (2022a). Relations of fire, palaeohydrology, vegetation succession, and carbon accumulation, as reconstructed from a mountain bog in the Harz Mountains (Germany) during the last 6200 years. *Geoderma*, 424, 115991. <https://doi.org/10.1016/j.geoderma.2022.115991>
- Gałka, M., Hölzer, A., Feurdean, A., Loisel, J., Teickner, H., Diaconu, A.-C., Szal, M., Broder, T., & Knorr, K.-H. (2022b). Insight into the factors of mountain bog and forest development in the Schwarzwald

- Mts.: Implications for ecological restoration. *Ecological Indicators*, *140*, 109039. <https://doi.org/10.1016/j.ecolind.2022.109039>
- Gałka, M., Szal, M., Broder, T., Loisel, J., & Knorr, K.-H. (2019). Peatbog resilience to pollution and climate change over the past 2700 years in the Harz Mountains, Germany. *Ecological Indicators*, *97*, 183–193. <https://doi.org/10.1016/j.ecolind.2018.10.015>
- Gocke, M., Kuzyakov, Y., & Wiesenberg, G. L. B. (2010). Rhizoliths in loess – evidence for post-sedimentary incorporation of root-derived organic matter in terrestrial sediments as assessed from molecular proxies. *Organic Geochemistry*, *41*(11), 1198–1206. <https://doi.org/10.1016/j.orggeochem.2010.08.001>
- Görner, M., Haupt, R., Hiekel, W., Niemann, E., & Westhus, W. (1984). Die Naturschutzgebiete der Bezirke Erfurt, Suhl und Gera. In Weinitschke, H. (Ed.), *Handbuch der Naturschutzgebiete der Deutschen Demokratischen Republik, Bd. 4* (pp. 99-101), Urania-Verlag, Leipzig, Jena, Berlin.
- Grimm, E. C. (1987). CONISS: a FORTRAN 77 program for stratigraphically constrained cluster analysis by the method of incremental sum of squares, *Computers & Geosciences*, *13*, 13–35. [https://doi.org/10.1016/0098-3004\(87\)90022-7](https://doi.org/10.1016/0098-3004(87)90022-7)
- Gurevitch, J., Koricheva, J., Nakagawa, S., & Stewart, G. (2018). Meta-analysis and the science of research synthesis. *Nature*, *555*(7695). <https://doi.org/10.1038/nature25753>
- Hirave, P., Wiesenberg, G. L. B., Birkholz, A., & Alewell, C. (2020). Understanding the effects of early degradation on isotopic tracers: Implications for sediment source attribution using compound-specific isotope analysis (CSIA). *Biogeosciences*, *17*(8), 2169–2180. <https://doi.org/10.5194/bg-17-2169-2020>
- Hoefs, M. J. L., Rijpstra, W. I. C., & Sinninghe Damsté, J. S. (2002). The influence of oxic degradation on the sedimentary biomarker record I: evidence from Madeira Abyssal Plain turbidites, *Geochimica et Cosmochimica Acta*, *66*, 2719-2735, [https://doi.org/10.1016/S0016-7037\(02\)00864-5](https://doi.org/10.1016/S0016-7037(02)00864-5)
- Hua, Q., Barbetti, M., & Rakowski, A. Z. (2013). Atmospheric radiocarbon for the period 1950–2010, *Radiocarbon*, *55*, 2059–2072. https://doi.org/10.2458/azu_js_rc.v55i2.16177
- Huang, Y., Bol, R., Harkness, D. D., Ineson, P., & Eglinton, G. (1996). Post-glacial variations in distributions, ¹³C and ¹⁴C contents of aliphatic hydrocarbons and bulk organic matter in three types of British acid upland soils. *Organic Geochemistry*, *24*(3), 273–287. [https://doi.org/10.1016/0146-6380\(96\)00039-3](https://doi.org/10.1016/0146-6380(96)00039-3)
- Husson, F., Josse, J., Le, S., & Mazet, J. (2023). *FactoMineR: Multivariate Exploratory Data Analysis and Data Mining* (2.8) [Computer software]. <https://cran.r-project.org/web/packages/FactoMineR/index.html>
- Jackson, R. B., Lajtha, K., Crow, S. E., Hugelius, G., Kramer, M. G., & Piñeiro, G. (2017). The ecology of soil carbon: Pools, vulnerabilities, and biotic and abiotic controls. *Annual Review of Ecology, Evolution, and Systematics*, *48*(1), 419–445. <https://doi.org/10.1146/annurev-ecolsys-112414-054234>
- Jansen, B., Nierop, K. G. J., Hageman, J. A., Cleef, A. M., & Verstraten, J. M. (2006). The straight-chain lipid biomarker composition of plant species responsible for the dominant biomass production along two

- altitudinal transects in the Ecuadorian Andes. *Organic Geochemistry*, 37(11), 1514–1536. <https://doi.org/10.1016/j.orggeochem.2006.06.018>
- Jansen, B., Van Loon, E. E., Hooghiemstra, H., & Verstraten, J. M. (2010). Improved reconstruction of palaeoenvironments through unravelling of preserved vegetation biomarker patterns. *Palaeogeography, Palaeoclimatology, Palaeoecology*, 285(1–2), 119–130. <https://doi.org/10.1016/j.palaeo.2009.10.029>
- Jansen, B., & Wiesenberg, G. L. B. (2017). Opportunities and limitations related to the application of plant-derived lipid molecular proxies in soil science. *SOIL*, 3(4), 211–234. <https://doi.org/10.5194/soil-3-211-2017>
- Jeschke, L. & Paulson, C. (2000). Pflege-und Entwicklungspläne für die Hochmoore in den Kammlagen des Thüringer Waldes, Beerbergmoor, Saukopfmoor, Schneekopfmoore und Schützenbergmoor, Unter Mitarbeit von Ch. Paulson und der Geocad-Ingenieurgesellschaft mbH. Unveröffentlichtes Gutachten im Auftrag des Staatlichen Umweltamtes Erfurt.
- Jobbágy, E. G., & Jackson, R. B. (2000). The vertical distribution of soil organic carbon and its relation to climate and vegetation. *Ecological Applications*, 10(2), 423–436. [http://doi.wiley.com/10.1890/10510761\(2000\)010\[0423: TVDOSO\]2.0.CO;2](http://doi.wiley.com/10.1890/10510761(2000)010[0423: TVDOSO]2.0.CO;2)
- Jones, M. C., Poteet, D. M., & Sambrotto, R. (2010). Late-glacial and Holocene $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ variation from a Kenai Peninsula, Alaska peatland, *Palaeogeography, Palaeoclimatology, Palaeoecology*, 293, 132–143. <https://doi.org/10.1016/j.palaeo.2010.05.007>
- Juggins, S. (2020). *rioja: Analysis of Quaternary Science Data* [Computer software]. <https://cran.r-project.org/package=rioja>, r package version 0.9-26
- Kassambara, A., & Mundt, F. (2020). *factoextra: Extract and Visualize the Results of Multivariate Data Analyses* (1.0.7) [Computer software]. <https://cran.r-project.org/web/packages/factoextra/index.html>
- Koch, K., & Ensikat, H.-J. (2008). The hydrophobic coatings of plant surfaces: Epicuticular wax crystals and their morphologies, crystallinity and molecular self-assembly. *Micron*, 39(7), 759–772. <https://doi.org/10.1016/j.micron.2007.11.010>
- Kögel-Knabner, I. (2002). The macromolecular organic composition of plant and microbial residues as inputs to soil organic matter. *Soil Biology and Biochemistry*, 34(2), 139–162. [https://doi.org/10.1016/S0038-0717\(01\)00158-4](https://doi.org/10.1016/S0038-0717(01)00158-4)
- Kuhry, P., Halsey, L. A., Bayley, S. E., & Vitt, D. H. (1992). Peatland development in relation to Holocene climatic change in Manitoba and Saskatchewan (Canada). *Canadian Journal of Earth Sciences*, 29(5), 1070–1090. <https://doi.org/10.1139/e92-086>
- Kuhry, P., & Vitt, D. H. (1996). Fossil carbon/nitrogen ratios as a measure of peat decomposition. *Ecology*, 77(1), 271–275. <https://doi.org/10.2307/2265676>
- Lavorel, S., Díaz, S., Cornelissen, J. H. C., Garnier, E., Harrison, S. P., McIntyre, S., Pausas, J. G., Pérez-Harguindeguy, N., Roumet, C., & Urcelay, C. (2007). Plant Functional Types: Are We Getting Any Closer to the Holy Grail? In J. G. Canadell, D. E. Pataki, & L. F. Pitelka (Eds.), *Terrestrial Ecosystems in a Changing World* (pp. 149–164). Springer Berlin Heidelberg. https://doi.org/10.1007/978-3-540-32730-1_13

- Loisel, J., Garneau, M., & Hélie, J.-F. (2010). Sphagnum $\delta^{13}\text{C}$ values as indicators of palaeohydrological changes in a peat bog. *The Holocene*, 20(2), 285–291. <https://doi.org/10.1177/0959683609350389>
- Luterbacher, J., Werner, J. P., Smerdon, J. E., Fernández-Donado, L., González-Rouco, F. J., Barriopedro, D., Ljungqvist, F. C., Büntgen, U., Zorita, E., Wagner, S., Esper, J., McCarroll, D., Toreti, A., Frank, D., Jungclauss, J. H., Barriendos, M., Bertolin, C., Bothe, O., Brázdil, R., Camuffo, D., Dobrovolný, P., Gagen, M., García-Bustamante, E., Ge, Q., Gómez-Navarro, J. J., Guiot, J., Hao, Z., Hegerl, G. C., Holmgren, K., Klimenko, V. V., Martín-Chivelet, J., Pfister, C., Roberts, N., Schindler, A., Schurer, A., Solomina, O., von Gunten, L., Wahl, E., Wanner, H., Wetter, O., Xoplaki, E., Yuan, N., Zanchettin, D., Zhang, H., & Zerefos, C. (2016). European summer temperatures since Roman times. *Environmental Research Letters*, 11(2), 024001. <https://doi.org/10.1088/1748-9326/11/2/024001>
- MacArthur, R. H. (1957). On the relative abundance of bird species, *Proceedings of the National Academy of Sciences*, 43, 293–295, <https://doi.org/10.1073/pnas.43.3.293>
- Marcisz, K., Kołaczek, P., Gałka, M., Diaconu, A.-C., & Lamentowicz, M. (2020). Exceptional hydrological stability of a *Sphagnum*-dominated peatland over the late Holocene. *Quaternary Science Reviews*, 231, 106180. <https://doi.org/10.1016/j.quascirev.2020.106180>
- Marseille, F., Disnar, J. R., Guillet, B., & Noack, Y. (1999). *n*-Alkanes and free fatty acids in humus and A1 horizons of soils under beech, spruce and grass in the Massif-Central (Mont-Lozère), France. *European Journal of Soil Science*, 50(3), 433–441. <https://doi.org/10.1046/j.1365-2389.1999.00243.x>
- Marzi, R., Torkelson, B. E., & Olson, R. K. (1993). A revised carbon preference index. *Organic Geochemistry*, 20(8), 1303–1306. [https://doi.org/10.1016/0146-6380\(93\)90016-5](https://doi.org/10.1016/0146-6380(93)90016-5)
- Mauquoy, D. & Van Geel, B.: Plant macrofossil methods and studies: mire and peat macros, in: *Encyclopedia of Quaternary Science*, pp. 2315–2336, Elsevier Science, 2007.
- Mauquoy, D., Yeloff, D., Van Geel, B., Charman, D. J., & Blundell, A. (2008). Two decadal resolved records from north-west European peat bogs show rapid climate changes associated with solar variability during the mid-late Holocene. *Journal of Quaternary Science*, 23(8), 745–763. <https://doi.org/10.1002/jqs.1158>
- McClymont, E. L., Mauquoy, D., Yeloff, D., Broekens, P., van Geel, B., Charman, D. J., Pancost, R. D., Chambers, F. M., & Evershed, R. P. (2008). The disappearance of *Sphagnum imbricatum* from Butterburn Flow, UK. *The Holocene*, 18(6), 991–1002. <https://doi.org/10.1177/0959683608093537>
- McClymont, E. L., Mackay, H., Stevenson, M. A., Damm-Johnsen, T., Honan, E. M., Penny, C. E., & Cole, Y. A. (2023). Biomarker proxies for reconstructing Quaternary climate and environmental change. *Journal of Quaternary Science*, 38(7), 991–1024. <https://doi.org/10.1002/jqs.3559>
- McGovern, P. E., & Hall, G. R. (2016). Charting a future course for organic residue analysis in archaeology. *Journal of Archaeological Method and Theory*, 23(2), 592–622. <https://doi.org/10.1007/s10816-015-9253-z>
- Moore, P. D., Webb, J. A., & Collison, M. E. (1991). *Pollen Analysis*, Blackwell Scientific Publications.
- Naafs, B. D. A., Inglis, G. N., Blewett, J., McClymont, E. L., Lauretano, V., Xie, S., Evershed, R. P., & Pancost, R. D. (2019). The potential of biomarker proxies to trace climate, vegetation, and

- biogeochemical processes in peat: A review. *Global and Planetary Change*, 179, 57–79. <https://doi.org/10.1016/j.gloplacha.2019.05.006>
- Nelson, D. B., Ladd, S. N., Schubert, C. J., & Kahmen, A. (2018). Rapid atmospheric transport and large-scale deposition of recently synthesized plant waxes. *Geochimica et Cosmochimica Acta*, 222, 599–617. <https://doi.org/10.1016/j.gca.2017.11.018>
- Nguyen Tu, T. T., Derenne, S., Largeau, C., Mariotti, A., & Bocherens, H. (2001). Evolution of the chemical composition of *Ginkgo biloba* external and internal leaf lipids through senescence and litter formation. *Organic Geochemistry*, 32(1), 45–55. [https://doi.org/10.1016/S0146-6380\(00\)00152-2](https://doi.org/10.1016/S0146-6380(00)00152-2)
- Oades, J. M. (1988). The retention of organic matter in soils. *Biogeochemistry*, 5(1), 35–70. <https://doi.org/10.1007/BF02180317>
- Oksanen, J., Blanchet, F. G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P. R., O’Hara, R. B., Simpson, G. L., Solymos, P., Stevens, M. H. H., Szoecs, E., & Wagner, H. (2020). *vegan: Community Ecology Package* [Computer software], <https://CRAN.R-project.org/package=vegan>, r package version 2.5-7
- Otto, A., & Simpson, M. J. (2005). Degradation and preservation of vascular plant-derived biomarkers in grassland and forest soils from western Canada. *Biogeochemistry*, 74(3), 377–409. <https://doi.org/10.1007/s10533-004-5834-8>
- Pancost, R. D., Baas, M., van Geel, B., & Sinninghe Damsté, J. S. (2002). Biomarkers as proxies for plant inputs to peats: An example from a sub-boreal ombrotrophic bog. *Organic Geochemistry*, 33(7), 675–690. [https://doi.org/10.1016/S0146-6380\(02\)00048-7](https://doi.org/10.1016/S0146-6380(02)00048-7)
- Peuple, M. D., Tierney, J. E., McGee, D., Lowenstein, T. K., Bhattacharya, T., & Feakins, S. J. (2021). Identifying plant wax inputs in lake sediments using machine learning. *Organic Geochemistry*, 156, 104222. <https://doi.org/10.1016/j.orggeochem.2021.104222>
- Peters, K. E., Walters, C. C., & Moldowan, J. M. (2005). *The Biomarker Guide*. Cambridge University Press.
- Poynter, J. G., Farrimond, P., Robinson, N., & Eglinton, G. (1989). Aeolian-Derived Higher Plant Lipids in the Marine Sedimentary Record: Links with Palaeoclimate. In M. Leinen & M. Sarnthein (Eds.), *Paleoclimatology and Paleometeorology: Modern and Past Patterns of Global Atmospheric Transport* (pp. 435–462). Springer Netherlands. https://doi.org/10.1007/978-94-009-0995-3_18
- Reimer, P. J., Austin, W. E. N., Bard, E., Bayliss, A., Blackwell, P. G., Bronk Ramsey, C., Butzin, M., Cheng, H., Edwards, R. L., Friedrich, M., Grootes, P. M., Guilderson, T. P., Hajdas, I., Heaton, T. J., Hogg, A. G., Hughen, K. A., Kromer, B., Manning, S. W., Muscheler, R., Palmer, J. G., Pearson, C., van der Plicht, J., Reimer, R. W., Richards, D. A., Scott, E. M., Southon, J. R., Turney, C. S. M., Wacker, L., Adolphi, F., Büntgen, U., Capano, M., Fahrni, S. M., Fogtmann-Schulz, A., Friedrich, R., Köhler, P., Kudsk, S., Miyake, F., Olsen, J., Reinig, F., Sakamoto, M., Sookdeo, A., & Talamo, S. (2020). The IntCal20 Northern Hemisphere radiocarbon age calibration curve (0–55 cal kBP), *Radiocarbon*, 62, 725–757, <https://doi.org/10.1017/RDC.2020.41>.
- Ronkainen, T., McClymont, E. L., Väiliranta, M., & Tuittila, E.-S. (2013). The *n*-alkane and sterol composition of living fen plants as a potential tool for palaeoecological studies. *Organic Geochemistry*, 59, 1–9. <https://doi.org/10.1016/j.orggeochem.2013.03.005>

- Ronkainen, T., Väiliranta, M., McClymont, E. L., Biasi, C., Salonen, S., Fontana, S., & Tuittila, E.-S. (2015). A combined biogeochemical and palaeobotanical approach to study permafrost environments and past dynamics. *Journal of Quaternary Science*, *30*, 189–200. <https://doi.org/10.1002/jqs.2763>
- Schädel, C., Beem-Miller, J., Aziz Rad, M., Crow, S. E., Hicks Pries, C. E., Ernakovich, J., Hoyt, A. M., Plante, A., Stoner, S., Treat, C. C., & Sierra, C. A. (2020). Decomposability of soil organic matter over time: The Soil Incubation Database (SIDb, version 1.0) and guidance for incubation procedures. *Earth System Science Data*, *12*(3), 1511–1524. <https://doi.org/10.5194/essd-12-1511-2020>
- Schäfer, I. K., Lanny, V., Franke, J., Eglinton, T. I., Zech, M., Vysloužilová, B., & Zech, R. (2016). Leaf waxes in litter and topsoils along a European transect. *SOIL*, *2*(4), 551–564. <https://doi.org/10.5194/soil-2-551-2016>
- Schmidt, M. W. I., Torn, M. S., Abiven, S., Dittmar, T., Guggenberger, G., Janssens, I. A., Kleber, M., Kögel-Knabner, I., Lehmann, J., Manning, D. A. C., Nannipieri, P., Rasse, D. P., Weiner, S., & Trumbore, S. E. (2011). Persistence of soil organic matter as an ecosystem property. *Nature*, *478*(7367), 49–56. <https://doi.org/10.1038/nature10386>
- Shepherd, T., & Wynne Griffiths, D. (2006). The effects of stress on plant cuticular waxes. *New Phytologist*, *171*(3), 469–499. <https://doi.org/10.1111/j.1469-8137.2006.01826.x>
- Smith, A. J. E. (2004). *The moss flora of Britain and Ireland*, Cambridge University Press.
- Speranza, A., Hanke, J., Geel, B., & Fanta, J. (2000). Late-Holocene human impact and peat development in the Černá Hora bog, Krkonoše Mountains, Czech Republic. *Holocene*, *10*, 575–585. <https://doi.org/10.1191/095968300668946885>
- Srivastava, K., & Wiesenberg, G. L. B. (2018). Severe drought-influenced composition and $\delta^{13}\text{C}$ of plant and soil *n*-alkanes in model temperate grassland and heathland ecosystems. *Organic Geochemistry*, *116*, 77–89. <https://doi.org/10.1016/j.orggeochem.2017.11.002>
- Stivrins, N., Aakala, T., Ilvonen, L., Pasanen, L., Kuuluvainen, T., Vasander, H., Gałka, M., Disbrey, H. R., Liepins, J., Holmström, L., & Sepp, H. (2019). Integrating fire-scar, charcoal and fungal spore data to study fire events in the boreal forest of northern Europe. *The Holocene*, *29*, 1480–1490. <https://doi.org/10.1177/0959683619854524>
- Stockmarr, J. (1971). Tablets with spores used in absolute pollen analysis. *Pollen et Spores*, *13*, 615–621.
- Swindles, G. T., Morris, P. J., Mullan, D. J., Payne, R. J., Roland, T. P., Amesbury, M. J., Lamentowicz, M., Turner, T. E., Gallego-Sala, A., Sim, T., Barr, I. D., Blaauw, M., Blundell, A., Chambers, F. M., Charman, D. J., Feurdean, A., Galloway, J. M., Gałka, M., Green, S. M., Kajukała, K., Karofeld, E., Korhola, A., Lamentowicz, Ł., Langdon, P., Marcisz, K., Mauquoy, D., Mazei, Y. A., McKeown, M. M., Mitchell, E. A. D., Novenko, E., Plunkett, G., Roe, H. M., Schoning, K., Sillasoo, Ü, Tsyganov, A. N., van der Linden, M., Väiliranta, M., & Warner, B. (2019). Widespread drying of European peatlands in recent centuries. *Nature Geoscience*, *12*(11), 922–928. <https://doi.org/10.1038/s41561-019-0462-z>
- Thomas, C. L., Jansen, B., Czerwiński, S., Gałka, M., Knorr, K.-H., Van Loon, E. E., Egli, M., & Wiesenberg, G. L. B. (2023). Comparison of paleobotanical and biomarker records of mountain peatland and forest

- ecosystem dynamics over the last 2600 years in central Germany. *Biogeosciences*, *20*, 4893–4914. <https://doi.org/10.5194/bg-20-4893-2023>
- Thomas, C. L., Jansen, B., Van Loon, E. E., & Wiesenberg, G. L. B. (2021). Transformation of *n*-alkanes from plant to soil: A review. *SOIL*, *7*(2), 785–809. <https://doi.org/10.5194/soil-7-785-2021>
- Tinner, W. & Hu, F. S. (2003). Size parameters, size-class distribution and area-number relationship of microscopic charcoal: relevance for fire reconstruction. *The Holocene*, *13*, 499–505. <https://doi.org/10.1191/0959683603hl615rp>
- Tuittila, E.-S., Väiliranta, M., Laine, J., & Korhola, A. (2007). Quantifying patterns and controls of mire vegetation succession in a southern boreal bog in Finland using partial ordinations, *Journal of Vegetation Science*, *18*, 891–902. <https://www.jstor.org/stable/4499301>
- Van Bergen, P. F., Bull, I. D., Poulton, P. R., & Evershed, R. P. (1997). Organic geochemical studies of soils from the Rothamsted Classical Experiments—I. Total lipid extracts, solvent insoluble residues and humic acids from Broadbalk Wilderness. *Organic Geochemistry*, *26*(1-2), 117-135.
- Wang, G., Zhang, L., Zhang, X., Wang, Y., & Xu, Y. (2014). Chemical and carbon isotopic dynamics of grass organic matter during litter decompositions: A litterbag experiment. *Organic Geochemistry*, *69*, 106–113. <https://doi.org/10.1016/j.orggeochem.2014.02.012>
- Wang, M., Moore, T. R., Talbot, J., & Riley, J. L. (2015). The stoichiometry of carbon and nutrients in peat formation: C and nutrients in peat. *Global Biogeochemical Cycles*, *29*(2), 113–121. <https://doi.org/10.1002/2014GB005000>
- Wanner, H., Pfister, C., & Neukom, R. (2022). The variable European Little Ice Age. *Quaternary Science Reviews*, *287*, 107531. <https://doi.org/10.1016/j.quascirev.2022.107531>
- Wiesenberg, G. L. B., & Gocke, M. I. (2015). Analysis of Lipids and Polycyclic Aromatic Hydrocarbons as Indicators of Past and Present (Micro)Biological Activity. In T. J. McGenity, K. N. Timmis, & B. Nogales (Eds.), *Hydrocarbon and Lipid Microbiology Protocols* (pp. 61–91). Springer Berlin Heidelberg. https://doi.org/10.1007/8623_2015_157
- Wu, M. S., West, A. J., & Feakins, S. J. (2019). Tropical soil profiles reveal the fate of plant wax biomarkers during soil storage. *Organic Geochemistry*, *128*, 1–15. <https://doi.org/10.1016/j.orggeochem.2018.12.011>
- Xu, J., Morris, P. J., Liu, J., & Holden, J. (2018). PEATMAP: Refining estimates of global peatland distribution based on a meta-analysis. *CATENA*, *160*, 134–140. <https://doi.org/10.1016/j.catena.2017.09.010>
- Yang, D., & Bowen, G. J. (2022). Integrating plant wax abundance and isotopes for paleo-vegetation and paleoclimate reconstructions: A multi-source mixing model using a Bayesian framework. *Climate of the Past*, *18*(10), 2181–2210. <https://doi.org/10.5194/cp-18-2181-2022>
- Yu, Z., Loisel, J., Brosseau, D. P., Beilman, D. W., & Hunt, S. J. (2010). Global peatland dynamics since the Last Glacial Maximum. *Geophysical Research Letters*, *37*(13). <https://doi.org/10.1029/2010GL043584>

- Zech, M., Pedentchouk, N., Buggle, B., Leiber, K., Kalbitz, K., Marković, S. B., & Glaser, B. (2011). Effect of leaf litter degradation and seasonality on D/H isotope ratios of *n*-alkane biomarkers. *Geochimica et Cosmochimica Acta*, 75(17), 4917–4928. <https://doi.org/10.1016/j.gca.2011.06.006>
- Zhang, Y., Zheng, M., Meyers, P. A., & Huang, X. (2017). Impact of early diagenesis on distributions of *Sphagnum n*-alkanes in peatlands of the monsoon region of China. *Organic Geochemistry*, 105, 13–19. <https://doi.org/10.1016/j.orggeochem.2016.12.007>
- Zheng, Y., Zhou, W., Liu, Z., & Liu, X. (2011). The *n*-alkanol paleoclimate records in two peat deposits: A comparative study of the northeastern margin of the Tibetan Plateau and Northeast China. *Environmental Earth Sciences*, 63(1), 135–143. <https://doi.org/10.1007/s12665-010-0676-2>
- Zheng, Y., Zhou, W., Meyers, P. A., & Xie, S. (2007). Lipid biomarkers in the Zoigê-Hongyuan peat deposit: Indicators of Holocene climate changes in West China. *Organic Geochemistry*, 38(11), 1927–1940. <https://doi.org/10.1016/j.orggeochem.2007.06.012>