



Preservation and degradation of ancient organic matter in mid-Miocene Antarctic permafrost

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Abstract. The Antarctic environment is amongst the coldest and driest environments on Earth. The ultraxerous soils in the McMurdo Dry Valleys support exclusively microbial communities, however, 15 million years ago, a tundra ecosystem analogous to present-day southern Greenland occupied this region. The occurrence of ancient soil organic carbon combined with low input rates makes it challenging to differentiate between ancient and modern organic processes. Here, we document the additions of modern organic carbon, and the preservation and degradation of organics and lipid biomarkers, in a 1.4 m mid-Miocene age permafrost soil column from Friis Hills. The total organic carbon is low throughout the soils (< 1%wt). The near-surface (upper 35 cm) dry permafrost has lower C:N ratios, higher $\delta^{13}\text{C}_{\text{org}}$ values, higher proportion of iso-FAs relative to *n*-FAs, lower phytol abundance and higher contributions of low-molecular weight homologues of *n*-alkanes, than the underlying icy permafrost. Conversely, the icy permafrost contains higher molecular weight *n*-alkanes, *n*-fatty acids and *n*-alkanols, along with phytosterols (e.g., sitosterol, stigmasterol) and phytol (and its derivatives pristane and phytane) that are indicative of the contributions and preservation of higher-level plants. This implies that legacy mid-Miocene age carbon in the near-surface soils (c. 35 cm) has been prone to microbial organic matter degradation during times when the permafrost thawed, likely during relatively warm intervals through the late Neogene. Biomolecules found deeper in the permafrost have been preserved for millions of years. These results suggest that ancient organics preserved in permafrost could underpin significant ecological changes in the McMurdo Dry Valleys as Earth's current climate warms in the coming decades and centuries.

1 Introduction

The McMurdo Dry Valleys (MDV) are amongst the coldest and driest environments on Earth (e.g. Horowitz et al., 1972). The hyperarid polar desert can be divided into three microclimate zones (Marchant and Head III, 2007): (i) the coastal thaw (or *subxerous*) zone (<400 m above sea level; a.s.l.) where mean daily summer air temperatures exceed 0°C and soil water can exist seasonally; (ii) the inland mixed (or *xerous*) zone where summer air temperatures may rise above 0°C for short periods and where soil water may be present periodically; and the *ultraxerous* or stable upland zone, where maximum air temperatures



rarely exceed 0°C and snow is scarce (<10 mm water equivalent per year). The MDV lack vascular plants, and the subxerous zone contains edaphic communities with sparse cryptogamic vegetation (mosses and lichens), low diversity of invertebrates, and heterotrophic soil organisms, as well as endolithic communities of phototrophic and heterotrophic organisms in sandstone outcrops and other lithic substrates (e.g. Bargagli et al., 1999; Barrett et al., 2007; Cary et al., 2010; Freckman and Virginia, 1997; Horowitz et al., 1972; Moorhead et al., 2002). By contrast, the ultraxerous zone lacks all types of edaphic vegetation and appears to only support endolithic organisms in sandstone outcrops (Friedmann, 1982). Heterotrophic microbial communities are present in soils but at very low metabolic levels or in a dormant state (Goordial et al., 2016; Tamppari et al., 2012). Similar conclusions were drawn from the Shackleton Glacier region of the Transantarctic Mountains where cultivation-dependent, cultivation-independent and metabolic assays were unable to detect viable microbial life (Dragone et al., 2021). However, in places in University Valley where the ground surface raises above 0°C in summer, the permafrost contains about 5× more organic carbon (C_{org}), and microbial activity has been indirectly inferred from C:N ratios, $\delta^{13}C_{org}$ and $\delta^{13}C_{CaCO_3}$ (Faucher et al., 2017).

The studies about the source and cycling of C_{org} in University Valley were from permafrost cores that represents about 152 ky of sediment accumulation (Lacelle et al., 2013). At Friis Hills, the permafrost consists of early to middle Miocene age sediments (Chorley et al., 2023; Lewis and Ashworth, 2016), and is potentially the oldest on Earth (Verret et al., 2021). During the mid-Miocene, the region harboured a tundra environment (Lewis and Ashworth, 2016; Chorley et al., 2023; Lewis et al., 2008) analogous to that found in southern Greenland today (including vascular plants). Since there have been limited periods of melt since the mid-Miocene (Verret et al., 2023), the site offers the opportunity to analyse the preservation and degradation of organic carbon in permafrost over millions of years. Lipid biomarkers are among the most stable organic molecules and can be preserved over long time periods (e.g. Naeher et al., 2022; Schouten et al., 2013; Castañeda and Schouten, 2011; Duncan et al., 2019). Low-molecular weight organic compounds (e.g. sugars and amino acids) and lipids with double bonds or polar functional groups (e.g. fatty acids and alcohols) are typically susceptible to microbial decomposition. However, other lipid biomarkers, such as apolar, saturated hydrocarbons (e.g. alkanes), isoprenoids (e.g. phytane) and cyclic compounds (i.e. hopanes or steranes), are refractory compounds formed during diagenesis (Peters et al., 2007) and may be preserved for long geological timescales (e.g. Eigenbrode, 2008). As such, their distribution and occurrence in the Friis Hills Miocene age sediments can provide insight about depositional conditions as well as paleoenvironmental changes (e.g. Duncan et al., 2019). In this study, we investigate the source of ancient carbon reservoirs, diagenetic degradation and potential overprint of modern organic carbon sources by combining bulk organic carbon analysis (total C_{org} , total N, $\delta^{13}C_{org}$), with lipid biomarker indicators, ramped pyrolysis-gas chromatography-mass spectrometry (Py-GC-MS), and radiocarbon dating. We also use bacterial branched glycerol dialkyl glycerol tetraether (brGDGT) lipids to infer threshold temperatures required to unlock organic carbon stored within the permafrost. The exposure of ancient organic carbon to modern degradation at Friis Hills provides valuable information on the present-day organic processes, which can be juxtaposed to the distinct organic signature of the tundra environment that dominated the site during sediment deposition. This allows us to assess the degradation of ancient organic carbon pools, soil carbon mineralization and cycling in the context of a warming Antarctic climate.



2 Study Site

The Friis Hills (77°45'S, 161°30'E, 1200-1500 m a.s.l.) are a 12 km wide inselberg situated at the head of Taylor Valley, ca. 50 km from the Ross Sea coast (Fig. 1a). The Friis Hills are situated within the ultraxerous zone (Marchant and Head III, 2007), where modern climatic conditions are extremely cold and arid, and microbial life is limited. The mean annual air temperature is -22°C, with an average reaching -13°C during the summer months (Friis Hills Meteorological Station 2011-2015; Doran and Fountain, 2016). The area receives less than 10 mm snow water equivalent of precipitation per year, most of which is displaced by strong katabatic winds (yearly average wind speed at Friis Hills is 4.7 m s⁻¹; Bliss et al., 2011). The local geology consists primarily of Ordovician and Jurassic age intrusive igneous rocks and Devonian sedimentary rocks of the Beacon Supergroup (Allibone et al., 1993; Lewis and Ashworth, 2016; Cox et al., 2012). A ca. 80 m thick sequence of glacial drifts interbedded with lacustrine sediments record multiple episodes of advance and retreat of local alpine glaciers and intervals of ice sheet over-riding (Chorley et al., 2023; Lewis and Ashworth, 2016).

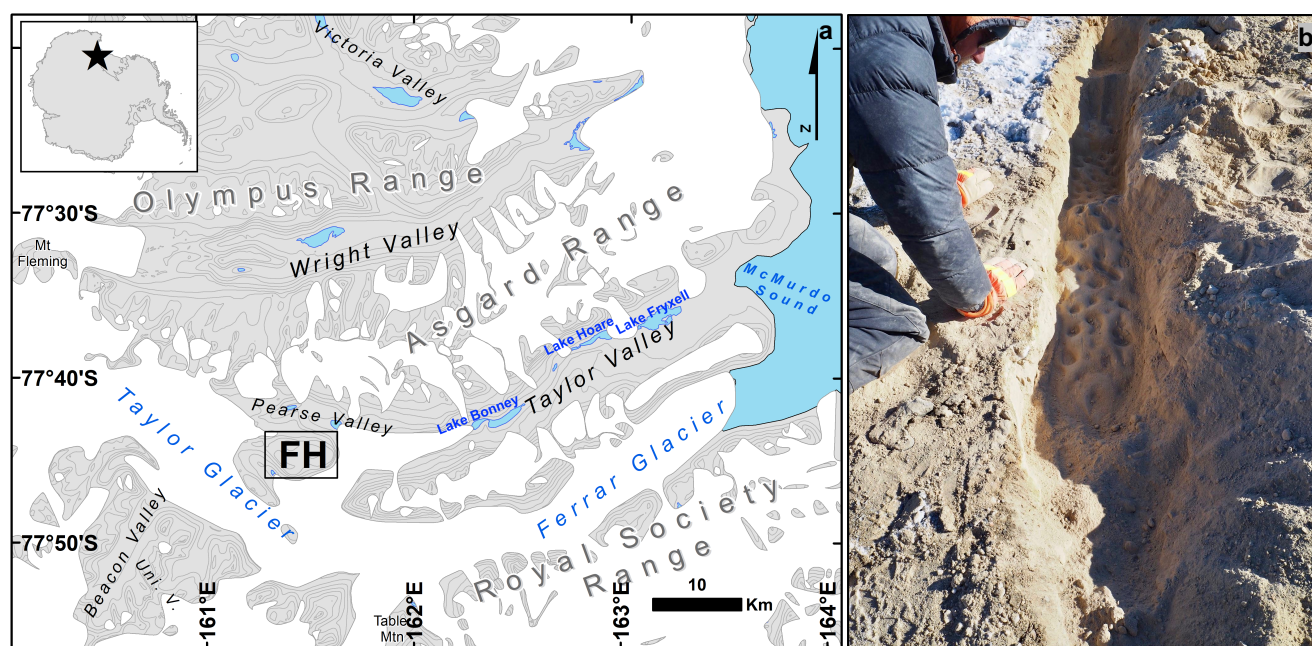


Figure 1 a. Location map of Friis Hills (FH) within the McMurdo Dry Valleys of Antarctica. Contour lines at 200 m intervals. **B.** Uncovered ice table at Friis Hills, overlain by a 35 cm thick layer of dry permafrost (photo credit: H. Chorley).

Major paleoenvironmental changes at Friis Hills can be divided into four distinct periods: (1) the early to mid-Miocene, a period when tundra persisted in the Friis Hills and throughout many locations across the Transantarctic Mountains (~20–14.6



Ma; Lewis et al., 2008; Duncan et al., 2019), (2) a period when tundra vegetation disappeared from high elevation localities in the MDV (~13.8 to 12.5 Ma; Lewis et al., 2008; Lewis and Ashworth, 2016; Chorley et al., 2023); (3) the mid-late Miocene (12.5–6.0 Ma), a period through which high elevations became progressively drier, culminating in hyper-arid conditions (Verret et al., 2023); and (4) the late Neogene and Quaternary (6.0 Ma–present) period, when the high elevations remained frozen and hyper-arid, even during relatively warm interglacial conditions. Low erosion rates (on the order of 0.1 m Myr⁻¹) associated with a relatively cold and dry environment enhanced the preservation of the sediment record through the Neogene, while exposing the mid-Miocene organic carbon to surficial processes (Verret et al., 2023).

The Friis Hills Drilling Project (FHDP) recovered a series of sediment cores from three sites (1, 2, and 3) during the 2016-17 Antarctic Field season (Chorley et al., 2023). Three cores (A, B, and C) were drilled at Site 2 (77°45'17.028"S, 161°27'20.632"E, 1244 m a.s.l.) and this study focuses on the uppermost 140 cm of the ~ 5 metre-long core 'C' (FHDP2C; Fig. 2a). The ice table (i.e. the boundary between the dry and the ice-cemented permafrost) occurs at 35 cm below the top of the core (Fig. 1b). Magnetostratigraphy constrained by ⁴⁰Ar/³⁹Ar dating of a tephra at 5.13-5.18 m depth indicate the sedimentary sequence in FHDP2C was deposited between ~14.5 and ~14.3 Ma (magnetozone IN1, C5Acn; 14.070 to 13.739 Ma; Chorley et al., 2023), corresponding to the mid-Miocene climate transition (MMCT; Flower and Kennett, 1994; Shevenell et al., 2004). The core includes two sedimentary cycles comprising two glacial (i.e., matrix-supported diamicts attributed to subglacial traction tills) and two interglacial facies (i.e., muddy-sand units attributed to proglacial ponds). An assemblage of organic macrofragments such as lichens, liverworts, mosses, dicots, grasses and sedges, along with cuticles belonging to the *Nothofagaceae* family were observed within core samples (Chorley et al., 2023). Similar plant assemblages are widespread in the modern southern Greenland or Svalbard in the Arctic (e.g. Berke et al., 2019). Tundra macrofossils are found in all sedimentary facies but are most prevalent in the finer grained units, suggesting that higher plants were relatively abundant across the Friis Hills during the interglacial periods. These data indicate that the mid-Miocene climate at relatively high elevations in the MDV region of the Transantarctic Mountains remained warm and wet enough to support growth of higher plants (Chorley et al., 2023).

3 Methods

3.1 Core description and sampling

Lithological descriptions were undertaken in the field and cross-checked on the frozen core prior to sampling at GNS Science's National Ice Core Facility, Lower Hutt, New Zealand. The core was sub-sampled at ~5 cm intervals using a tile cutter with a 2.2 mm thick blade and samples were placed into polyethylene bags to thaw before being dried at 80°C for 24 hours (plastic contamination was not found in the samples). The surficial dry permafrost layer was sampled in the field at 5 cm intervals. A total of 23 samples from the upper 140 cm of the core were analysed for bulk organic carbon and nitrogen and lipid biomarkers.



3.2 Bulk carbon and nitrogen analysis

The total organic carbon (TOC), total nitrogen (TN) was measured to determine the soil organic carbon density (SOCd), and if the C:N ratios in the bulk sediments follow the Redfield biological stoichiometry ratio (6:61; Redfield, 1934). The TOC and TN was measured using an Elemental VarioEl Cube instrument at the Jan Veizer Laboratory, University of Ottawa, Canada.

120 The samples were first acidified with 10% HCl to remove the inorganic carbon and isolate the organic carbon fraction. 100 mg of sediment was analysed within tin capsules, along with 100 mg of tungstic oxide (WO₃), a combustion catalyst and binder. Calibrated standards of Sulfanilic Acid were prepared in a range of weights and ran cyclically to ensure instrument precision, and approximately 20% of samples were analysed in duplicate with analytical precision of $\pm 0.1\%$.

125 The $\delta^{13}\text{C}_{\text{org}}$ of the bulk samples was measured using a DeltaPlus Advantage instrument coupled with the ConFlo III interface to assess potential source and alteration of organic carbon in the sediments. The results are expressed in δ notation, which represents the parts per thousand difference of $^{13}\text{C}/^{12}\text{C}$ in per mil (‰) with respect to the Vienna Pee-Dee Belemnite (VPDB) standard. Analytical precision was $\pm 0.2\%$.

3.3 Lipid biomarker analyses

Lipid biomarkers were analysed to differentiate between the mid-Miocene tundra carbon and subsequent microbial degradation. Lipid biomarkers were analysed in the Organic Geochemistry Laboratory at GNS Science, Lower Hutt, New Zealand, following Naeher et al. (2012; 2014) with some modifications. Homogenised sediment (~5–15g) was extracted 4× by ultrasonic extraction using dichloromethane (DCM):methanol (MeOH) (3:1, v:v). Total lipid extracts (TLEs) were saponified using 6% KOH in MeOH at 80°C for 3 hours. After the addition of ultrapure H₂O, neutrals were extracted from the aquatic solution using *n*-hexane. Following acidification with 6M HCl to pH <2, fatty acids (FAs) were then recovered using *n*-hexane.

135 The neutral fraction was further separated into apolar and polar fractions by silica gel chromatography using *n*-hexane and DCM:MeOH (1:1, v: v), respectively. Prior to analysis by gas chromatography-mass spectrometry (GC-MS), polar fractions were derivatized with BSTFA in pyridine at 80°C for 1 hour, whereas FA fractions were derivatized with BF₃ MeOH at 100°C for 2 hours to obtain fatty acid methyl esters (FAMES).

140 The resulting lipid fractions were analysed by GC-MS on an Agilent 7890A GC System, equipped with an Agilent J&W DB-5ms capillary column [60 m × 0.25 mm inner diameter (i.d.) × 0.25 μm film thickness (f.t.)], and coupled to an Agilent 5975C inert MSD mass spectrometer. The oven was heated from 70°C (held for 1 min) to 100°C at 20°C min⁻¹, then to 320°C at 4°C min⁻¹ and held at that temperature for 20 minutes. Helium was used as carrier gas with a constant flow of 1.0 ml min⁻¹. Samples (1 μl) were injected splitless at an inlet temperature of 300°C. The MS was operated in electron impact ionisation mode at 70 eV using a source temperature of 230°C.



Lipid biomarkers were quantified relative to an internal standard (50 µL added to the TLEs following solvent extraction; consisting of 110.8 µg ml⁻¹ 5α-cholestane, 118.8 µg ml⁻¹ *n*-nonadecanoic acid and 116.8 µg ml⁻¹ *n*-nonadecanol). Procedural blanks were also analysed to ensure data quality and absence of laboratory contamination.

The polar fractions containing glycerol dialkyl glycerol tetraethers (GDGTs) were dissolved in *n*-hexane/isopropanol (99:1, v/v) and filtered with 0.45 µm PTFE filters prior to analysis by high performance liquid chromatography-atmospheric pressure chemical ionisation-mass spectrometry (HPLC-APCI-MS). These analyses were on an Agilent 1260 Infinity II Prime LC system coupled to an Agilent 6125B single quadrupole MS following the method of Hopmans et al. (2016). In brief, two UHPLC silica columns (Acquity BEH HILIC columns, 2.1 × 150 mm, 1.7 µm; Waters) were used in series, fitted with a 2.1 × 5 mm pre-column of the same material (Waters), and maintained at 30°C. GDGTs were eluted isocratically for 25 minutes with 82% A:18% B, followed by a linear gradient up to 35% B in 25 minutes, then a linear gradient to 100% B in 30 min, where A is *n*-hexane and B is *n*-hexane/isopropanol (9:1, v/v). Flow rate was 0.2 ml/min. The abundances of GDGTs were monitored using selective ion monitoring mode (SIM) with *m/z* 1302, 1300, 1298, 1296, 1294, 1292, 1050, 1048, 1046, 1036, 1034, 1032, 1022, 1020, 1018. Compounds were identified by comparing mass spectra and retention times with those in the literature (e.g. Hopmans et al., 2016). The concentration of biomarkers is expressed in microgram per gram of TOC (µg g⁻¹ TOC).

3.4 Lipid biomarker proxies and indices

The measurements of the lipid biomarkers were used to calculate a series of indices to: i) differentiate between plant- and bacteria-derived organic carbon; and ii) define the level of degradation of the organic carbon.

3.4.1 Carbon preference index

The carbon preference index (CPI) is a parameter that quantifies the ratio of odd to even *n*-alkanes based on its carbon number and is an indicator of the degradability of organic carbon. The CPI value decreases with ongoing alteration of organic carbon (Bray and Evans, 1961):

$$CPI_{25-33} = 0.5 \cdot \left(\frac{\sum \text{odd } C_{25-31}}{\sum \text{even } C_{26-32}} + \frac{\sum \text{odd } C_{27-33}}{\sum \text{even } C_{26-32}} \right) \quad (1)$$

Natural distributions or well-preserved *n*-alkane signatures are expected to show a predominance of odd-numbered carbon chains because of decarboxylation of fatty acids that show characteristic even carbon number predominance. This typically reflects the degree of bacterial alteration during diagenesis, as odd chains get altered into smaller chain lengths, while their odd-over-even predominance decreases (Bray and Evans, 1961; Grimalt et al., 1985). A similar index was also calculated for fatty acids and *n*-alcohols (Meyers and Ishiwatari, 1993):

$$CPI_{22-28} = \frac{\sum \text{even } C_{22-26} + \sum \text{even } C_{24-28}}{2 \cdot \sum \text{odd } C_{23-27}} \quad (2)$$



175 3.4.2 Average chain length and dominant chain length

In general, cuticle waxes of terrestrial plants contain predominantly high molecular weight *n*-alkanes ($>n\text{-C}_{27}$; with leaf waxes containing mainly *n*-C₂₇ and *n*-C₂₉ and grasses containing mainly *n*-C₃₁ and *n*-C₃₃ alkanes) while mid molecular weight (*n*-C₂₁ to *n*-C₂₅) alkanes are indicative of aquatic macrophytes, mosses (e.g. *Sphagnum* in peat) or lichen, and low molecular weight (*n*-C₁₂ to *n*-C₂₀) alkanes are common in algae and bacteria (e.g. Naafs et al., 2019; Killops and Killops, 2013; Zech et al., 180 2010). A similar relation can be extracted from even carbon numbered *n*-alkanols and *n*-fatty acids. The average chain length (ACL) of *n*-alkanes can be used to identify the main source of organic carbon and it can be useful to infer environmental changes in a particular ecosystem (e.g. Killops and Killops, 2013). The ACL was calculated for odd-chain *n*-alkanes with C₂₇ to C₃₃ following this equation (Poynter and Eglinton, 1990):

$$ACL_{C_{27}-C_{33}} = \frac{\sum i \cdot C_i}{\sum C_i} \quad (3)$$

185 where *i* is the carbon number and *C_i* is the concentration of *n*-alkane.

ACL was also calculated for even-chain FAME (C₂₂ to C₃₂) and *n*-alkanols (C₂₂ to C₂₈). We also present the dominant chain length (C_{max}) for each biomarker class.

3.4.3 Ratio of short-chain and long-chain *n*-alkanes

The ratio of short-chain *n*-alkanes (SC, $< C_{23}$) and long-chain *n*-alkanes (LC, $\geq C_{23}$) was calculated using the following 190 equation:

$$SC:LC = \frac{\sum C_{C_{15}-C_{22}}}{\sum C_{C_{23}-C_{33}}} \quad (4)$$

High values of SC:LC are indicative of an environment dominated by bacteria while low values are indicative of an environment dominated by higher plants and/or macrophyte waxes (e.g. Killops and Killops, 2013).

3.4.4 Higher plant fatty acid index

195 The higher plant fatty acid index (HPFA), which can be used as a relative indicator of chemical degradation, was calculated following this equation (Strauss et al., 2015):

$$HPFA = \frac{\sum \text{fatty acids } C_{24}, C_{26}, C_{28}}{\sum \text{fatty acids } C_{24}, C_{26}, C_{28} + \sum \text{n-alkanes } C_{27}, C_{29}, C_{31}} \quad (6)$$

Since *n*-alkanes are typically preserved better than *n*-fatty acids, low HPFA are indicative of high degrees of organic carbon degradation.

200 3.4.5. brGDGT indices

The BIT index, which differentiates between inputs from a terrestrial environment (BIT = 1) and a marine environment (BIT = 0) was calculated as follows (Hopmans et al., 2004):



$$BIT\ Index = \frac{(I+II+III)}{(I+II+III+IV)} \quad (7)$$

where I, II and III denote the relative abundances of brGDGTs and IV reflects the relative abundance of crenarchaeol.

205 Past summer soil temperatures (Raberg et al., 2024) were estimated using the MBT'_{5ME} index (De Jonge et al., 2014) and the soil-specific, Bayesian temperature calibration BayMBT₀ of Crampton-Flood et al. (2020). The MBT'_{5ME} index was calculated as:

$$MBT'_{5ME} = \frac{I_a + I_b + I_c}{III_a + II_a + II_b + II_c + I_a + I_b + I_c} \quad (8)$$

210 3.5 Py-GC-MS analysis

One sample in the dry permafrost (sample 2-C0-3) was analysed by full, rapid-ramp pyrolysis and then incremental, partitioned ramped Py-GC-MS analysis to characterize the composition and sources of the organic carbon at different temperatures (Ginnane et al., 2024). For this, 37–50 mg of the sample was pyrolyzed in deactivated stainless-steel cups. For full, rapid-ramp pyrolysis, samples were pyrolyzed with a ramp of 100°C min⁻¹ from 100°C to the Py-GC-MS instrumental maximum of 650°C.

215 For incremental, partitioned ramped-Py-GC-MS, sample splits were obtained similar to RPO analysis by utilizing thermal desorption mode with a ramp of 10°C min⁻¹ run from 100°C to 259°C as the first split, then the subsequent splits collected to their respective maximum temperatures (395, 449, 535 and 650°C). After each split collection, the sample was removed from the pyrolyser furnace and GC-MS analysis of this step was completed, before reinserting the same sample cup and heating to the next split maximum temperature. The front inlet of the GC over was set to 5:1 split and the GC oven programme started at
220 40°C (held 5 min isothermal), ramped to 300°C at 5 °C min⁻¹, and then held isothermal for 5 min. All other GC-MS settings were the same as described in Section 3.3.

Compounds were identified from the Py-GC-MS and grouped into nine compound classes following existing literature interpretations of dominant OM sources (i.e. bacteria or plant). The sources of *n*-alkanes were distinguished based on molecular weight (as detailed in section 3.4.2.). Pyrroles, furans and phenols were attributed to a plant-derived OM as they are derived
225 from pigments such as chlorophylls and polysaccharides (e.g. Keely, 2006). Polycyclic aromatic hydrocarbons, thiophenes, alkylbenzenes and other aromatics are undifferentiable compounds.

3.6 Radiocarbon Analysis

To determine if biogeochemical processes are relict or currently active at Friis Hills, one sample of dry sediment (2-C0-3; 10 cm depth) was measured for radiocarbon activity at the Rafter Radiocarbon Laboratory, GNS Science. The bulk sediment
230 sample (< 300 µm) was first acid-treated to remove carbonates and then combusted at 900°C for 4 hours in an evacuated, sealed quartz tube with cupric oxide and silver wire. The resulting CO₂ was graphitized by reduction with hydrogen over iron catalyst and measured by accelerator mass spectrometry (Turnbull et al., 2015; Zondervan et al., 2015). Radiocarbon activity is presented as F¹⁴C (fraction modern carbon; Donahue et al., 1990; Reimer et al., 2004) and conventional radiocarbon age (as described in Stuiver and Polach, 1977).



235 3.7 Statistical analysis

To simplify the data, we carried out hierarchical clustering of all 25 variables in R Studio 1.3.1056 using the *chclust* function in the package *rioja* with the method *coniss* (Juggins, 2020).

4 Results

The hierarchical clustering analysis of all samples in the 1.4 m sediment core and their 25 variables, which included TOC, C:N ratio, $\delta^{13}\text{C}_{\text{org}}$, lipid abundance and main biomarker indices, grouped the samples by depth. Group 1 consists of the dry permafrost from 0 to 35 cm depth; group 2 consists of the icy permafrost from 38 to 71 cm; and group 3 was the icy permafrost from 93 to 140 cm. An outlier sample was identified at 79 cm depth and not classified in any group. An ANOVA within subjects and post hoc Tukey HSD tests were then conducted on the results to determine whether the means of each variable in the three units were statistically different. The results first describe the general trends found throughout the core, followed by variables and indices that are statistically different between the units.

4.1 General biomarker trends

Lipid biomarkers that show little variations throughout the 1.4 m core include: *n*-alkanes (*n*-C₁₅ to *n*-C₃₄), isoprenoids, hopanoids, *n*-fatty acids (*n*-C₁₂ to *n*-C₃₀), branched fatty acids with iso- and anteiso- (*n*-C₁₃ to *n*-C₁₇) configurations, *n*-alkanols (*n*-C₁₂ to *n*-C₂₈), ketones, and sterols. The ACL₂₇₋₃₃ of *n*-alkanes, ACL₂₂₋₃₂ of *n*-fatty acids and ACL₂₂₋₂₈ of *n*-alkanols averaged 28.7 ± 0.5, 24.0 ± 0.3 and 23.3 ± 0.4 respectively, and showed little variation in the core (Figs 3d, 4d & 5d). The cholesterol to plant sterol ratio remained low throughout the core (avg. 2.4; Fig. 5c), which indicates a terrestrial setting. The GDGT analysis showed that the distribution of all samples was dominated by brGDGTs. All samples had a BIT index of 1.0, and MBT'5_{ME} index ranged between 0.24 and 0.48 (Supplementary Data). In general, the homogenous distribution of these biomarkers in the core are indicative of a terrestrial environment with higher plant components such as high molecular weight *n*-alkanes, *n*-fatty acids and *n*-alkanols and plant sterols. Overall, these biomarkers reflect the background composition of the tundra-dominated ecosystem of the site during the mid-Miocene as inferred from the macrofossils.

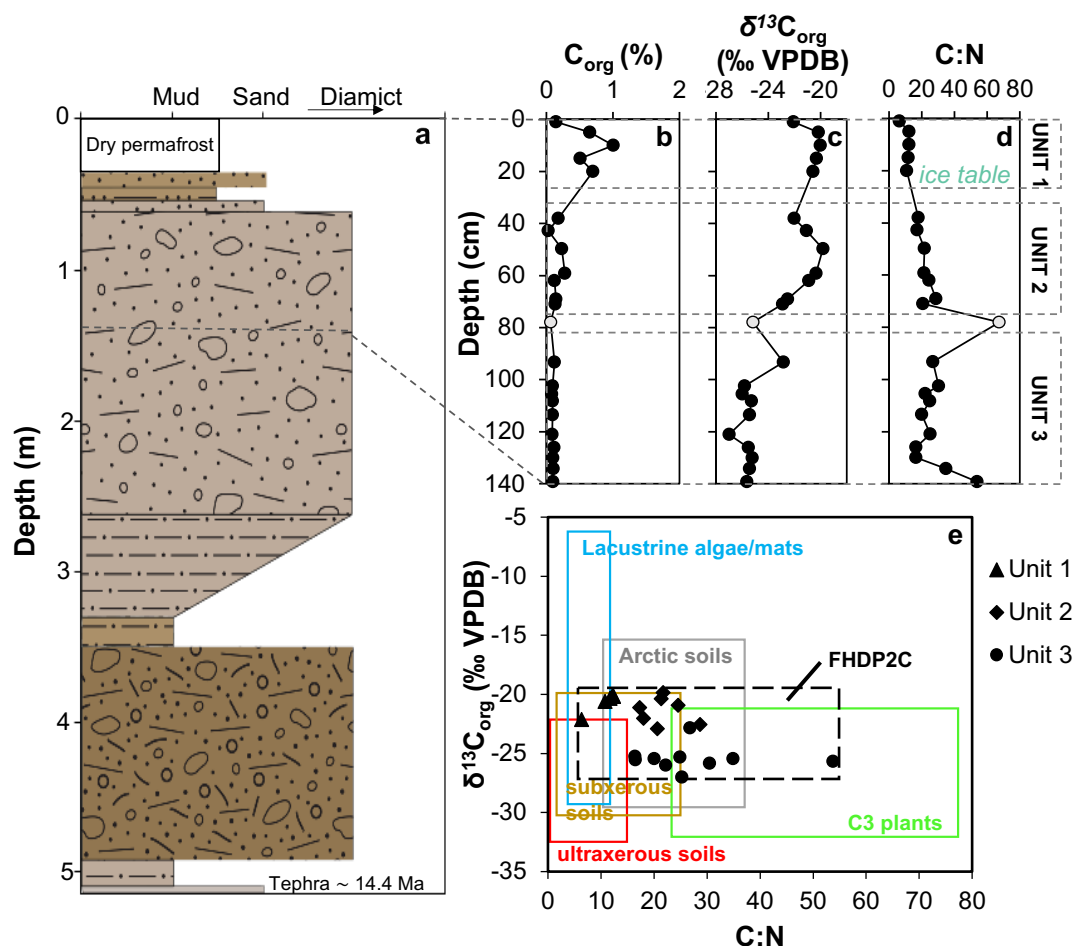


Figure 2 Carbon and nitrogen in the upper section of core FHDP2C. a. sedimentary log of core FHDP2C from Friis Hills with the upper 140 cm studied in detail in this paper, where colours reflect Munsell colours of each unit. b. organic carbon concentration, expressed in wt%, c. $\delta^{13}\text{C}_{\text{org}}$, d. C:N ratio and e. $\delta^{13}\text{C}_{\text{org}}$ and C:N biplot with bulk sediment measurements from three different units in FHDP2C compared to ultraxerous environments; University Valley and Mackay Glacier (Faucher et al., 2017; Van Goethem et al., 2020), subxerous environments; Miers, Garwood, Taylor and Victoria Valleys (Barrett et al., 2007; Hopkins et al., 2009), Antarctic lake/stream mats (Hopkins et al., 2009; Lawson et al., 2004), Arctic soils (Haugk et al., 2022; Osburn et al., 2019) and signature for C3 plants (Meyers, 1994). Unit separation was established by hierarchical clustering, by which sample in grey is an outlier.

4.2 Group 1: dry permafrost (0-35 cm)

The dry permafrost has the highest TOC and TN concentrations, averaging 6.0 ± 0.3 and $0.53 \pm 0.22 \text{ mg g}^{-1}$ dry soil, respectively (Fig. 2b). The C:N ratio averaged of 10.6 ± 2.5 , which is near the Redfield ratio and suggests balanced microbial growth (Fig. 2d & SFig. 1). The $\delta^{13}\text{C}_{\text{org}}$ values were highest in the core, ranging from -22.1 to -20.0 ‰VPDB (Fig. 2c). The sum of odd *n*-alkane concentrations (avg. $628.2 \mu\text{g g}^{-1}$ TOC; Fig. 3a), even *n*-fatty acids concentrations (avg. 1.67 mg g^{-1}



TOC; Fig. 4a) and even *n*-alkanols concentrations (avg. 432.1 $\mu\text{g g}^{-1}$ TOC; Fig. 5a) were all lowest in Unit 1. Within Unit 1, the CPI₂₅₋₃₃ of *n*-alkanes ranged from 3.7 to 6.9 (avg. 5.5 ± 1.2 ; Fig. 3b) and the SC:LC ratio from 1.6 to 7.7 (Fig. 3c); both of which are the highest values found in the core. The highest CPI₂₂₋₂₈ of *n*-fatty acids were also found in Unit 1 (Fig. 4b), along with the ratio of $100 \times \text{C}_{15-17} \text{ iso-FA} + \text{anteiso-FA} / \text{n-FA}$ (up to 12.6; Fig. 4c). The dominant *n*-alkane chain length was C₁₈ (Fig. 3e). The HPFA remained <1 throughout the whole core, ranging from 0.3 to 1.0 (Fig. 4f), but was generally lower in Unit 1. Low HPFA are typically indicative of high degrees of organic carbon degradation. Hopanoids, which were detected in all measured fractions (as hopanes, hopanols and hopanoic acids), were highest in Unit 1 (avg. $84.6 \pm 65.3 \mu\text{g g}^{-1}$ TOC; Fig. 6a). Phytosterols such as stigmasterol and sitosterol and their derivatives were identified in highest concentrations in Unit 1, with the exception of the surface sample (Fig. 6b). Pr/Ph ratios were also highest in Unit 1 (avg. 6.2 ± 1.4 ; Fig. 6d).

A sample taken at 10 cm below the surface was ¹⁴C dated and analyzed to determine the thermally partitioned splits. The sample taken at 10 cm below the surface yielded a bulk radiocarbon age of $41,749 \pm 1,736$ ¹⁴C yrs BP (F¹⁴C of 0.0055 ± 0.0012 ; Table 1). The lowest temperature split (105 to 259°C) is composed primarily of small, mostly undiagnostic molecules that are mainly degradation products from larger (macro-)molecules (e.g. alkylbenzenes and other aromatics; Ginnane et al., 2024). In contrast, furans, which are found mostly in the 259 to 395°C partition (split 2) and 395 to 449°C partition (split 3), are diagnostic source indicators which can originate from carbohydrates, such as cellulose associated with the decay of terrestrial plants (e.g. Kaal, 2019). Pyrroles that are most abundant in split 2 are typical products of labile tetrapyrrole pigments (e.g. chlorophylls and its transformation products), whereas those found in split 4 are rather derived from the decomposition of plant lignin (Keely, 2006). The highest two temperature splits (splits 4 and 5 collected from 449 to 535°C and 535 to 750°C respectively) are comprised of compounds that are mostly derived from alteration of more complex macromolecules commonly associated with more relict carbon or from secondary thermal reactions during pyrolysis at higher temperatures (Ginnane et al., 2024). The resulting compounds derived in these fractions commonly consists of more stable, older material including kerogen, that is less accessible to bacterial degradation. Overall, the TOC-TN and biomarker indices suggest a higher level of organic carbon degradation by microbes relative to the underlying icy permafrost.

4.3 Groups 2 and 3: icy permafrost (38-140cm)

The TOC and TN in the icy permafrost, averaging 2.0 ± 0.1 and 0.11 ± 0.09 mg g⁻¹ dry soil, respectively, were statistically lower than in the dry permafrost. The C:N ratios (16.4 to 53.7) were well above the Redfield ratio (Fig. 2d). The large range of C:N values in the icy permafrost may also partly be attributed to the low concentrations of N nearing the limit of analytical detection. The $\delta^{13}\text{C}_{\text{org}}$ values showed a general decrease with depth: -21.4 ± 1.1 ‰ VPDB in unit 2, and -25.4 ± 1.0 ‰ VPDB in unit 3; (Fig. 2c). The sum of odd *n*-alkane concentrations (Fig. 3a), even *n*-fatty acids concentrations (Fig. 4a) and even *n*-alkanols concentrations (Fig. 5a) increased down core and were highest in Unit 3. The CPI₂₅₋₃₃ of *n*-alkanes ranged from 0.4 to 7.8 (avg. 3.9 ± 1.7 ; Fig. 3b) and the SC:LC ratio, from 0.2 to 3.3 (Fig. 3c) within the icy permafrost, with little difference between unit 2 and 3. The dominant *n*-alkane was C₁₈ in Unit 2 (except for a dominance of C₂₇ at the ice table), whereas Unit 3 was dominated by C₂₅ (Fig. 3e). The sum of the concentrations of hopanoids decreased within the icy permafrost, averaging



59.5 ± 58.9 µg g⁻¹ TOC in Unit 2 and 25.9 ± 18.0 µg g⁻¹ TOC in Unit 3 (Fig. 6a). Phytane (Pr), pristane (Ph) and phytol were also detected in highest concentrations in Unit 3 (Fig. 6c). Pr/Ph ratios ranged from 0.2 to 2.2 in the icy permafrost and decreased with depth (Fig. 6d). Overall, these indices display a gradient of organic carbon degradation that decreases downcore through the icy permafrost. The hierarchical clustering analysis suggest higher level of organic carbon degradation in the dry permafrost, intermediate level in unit 2 and lower level in unit 3.

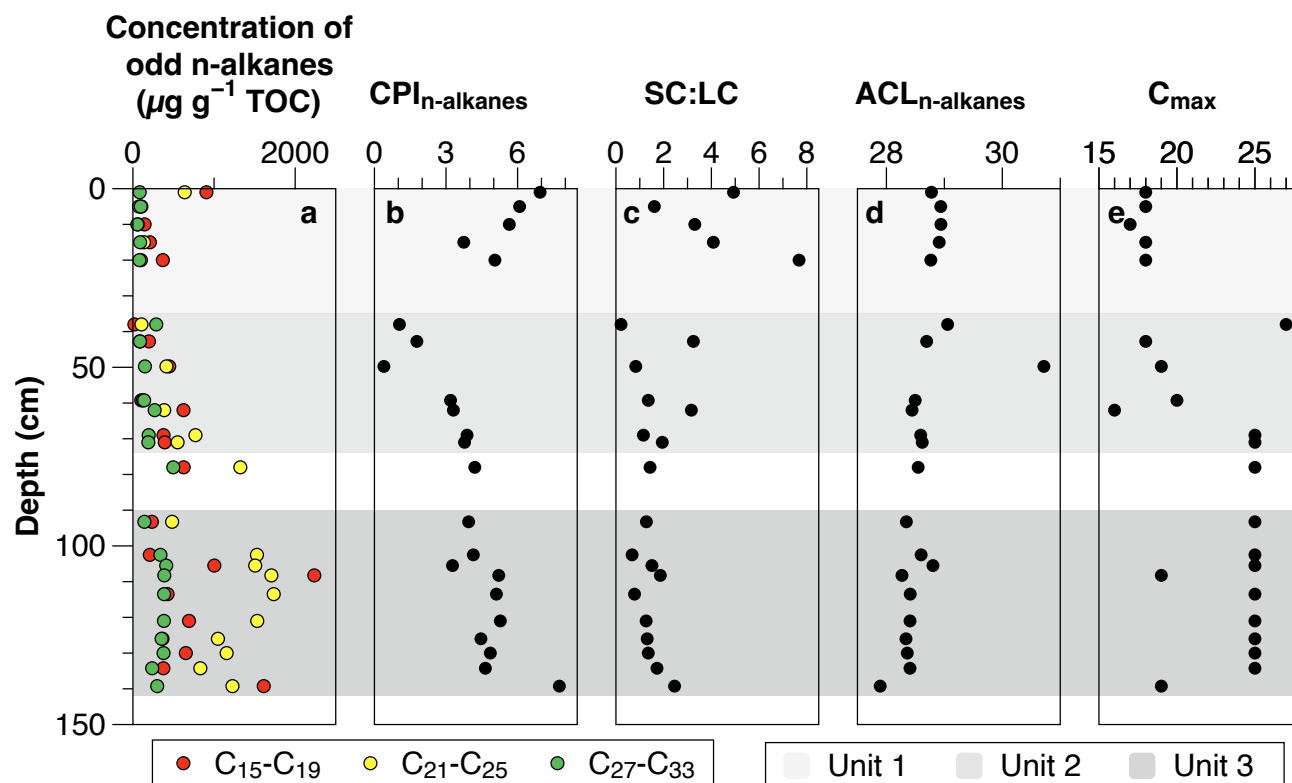


Figure 3 *n*-Alkane parameters of the Friis Hills drill core. a. concentration per gram organic carbon of low molecular weight odd *n*-alkanes (*n*-C₁₅ to *n*-C₁₉), mid molecular weight odd *n*-alkanes (*n*-C₂₁ to *n*-C₂₅) and high molecular weight odd *n*-alkanes (*n*-C₂₇ to *n*-C₃₃), b. carbon preference index (CPI; *n*-C₂₅ to *n*-C₃₃), c. SC/LC (short-chain/long-chain) ratio, d. average chain length (ACL) for high molecular weight *n*-alkanes (*n*-C₂₇ to *n*-C₃₃), e. dominant *n*-alkane chain length.

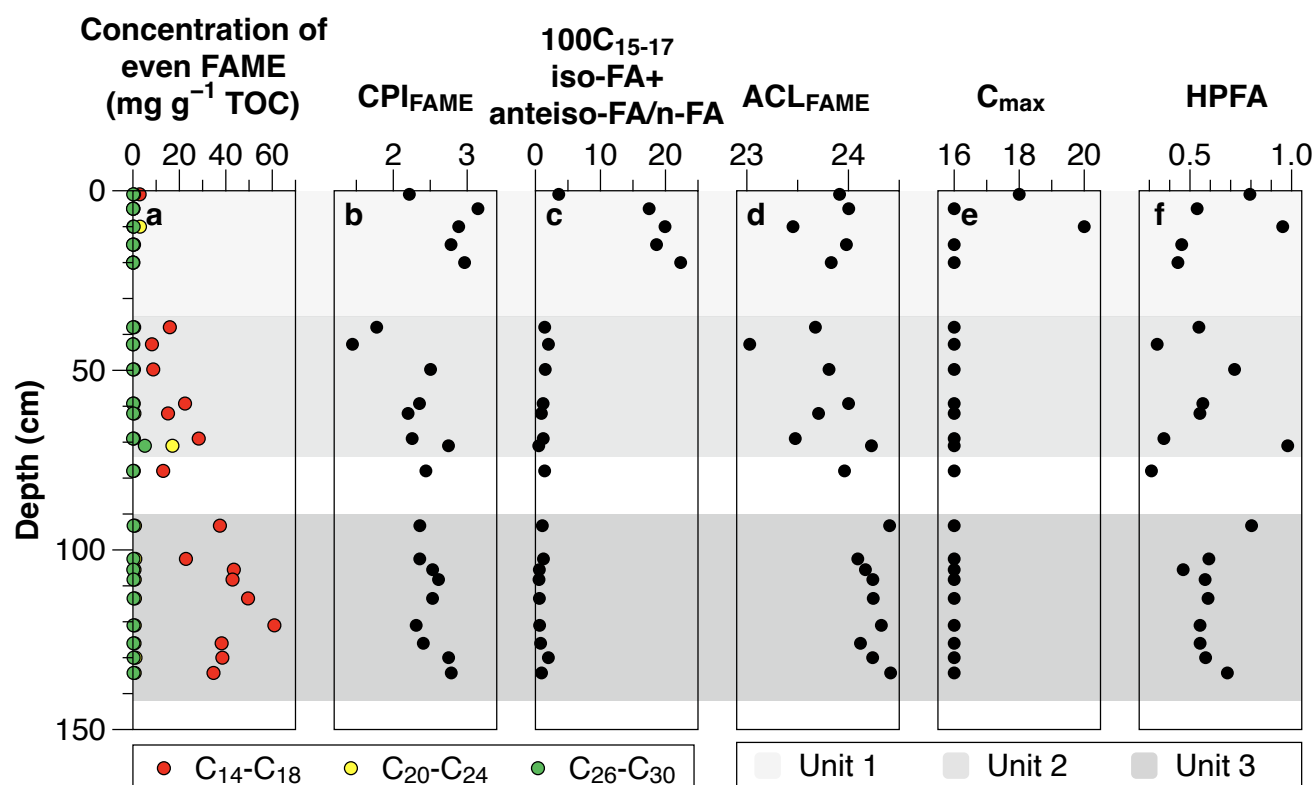
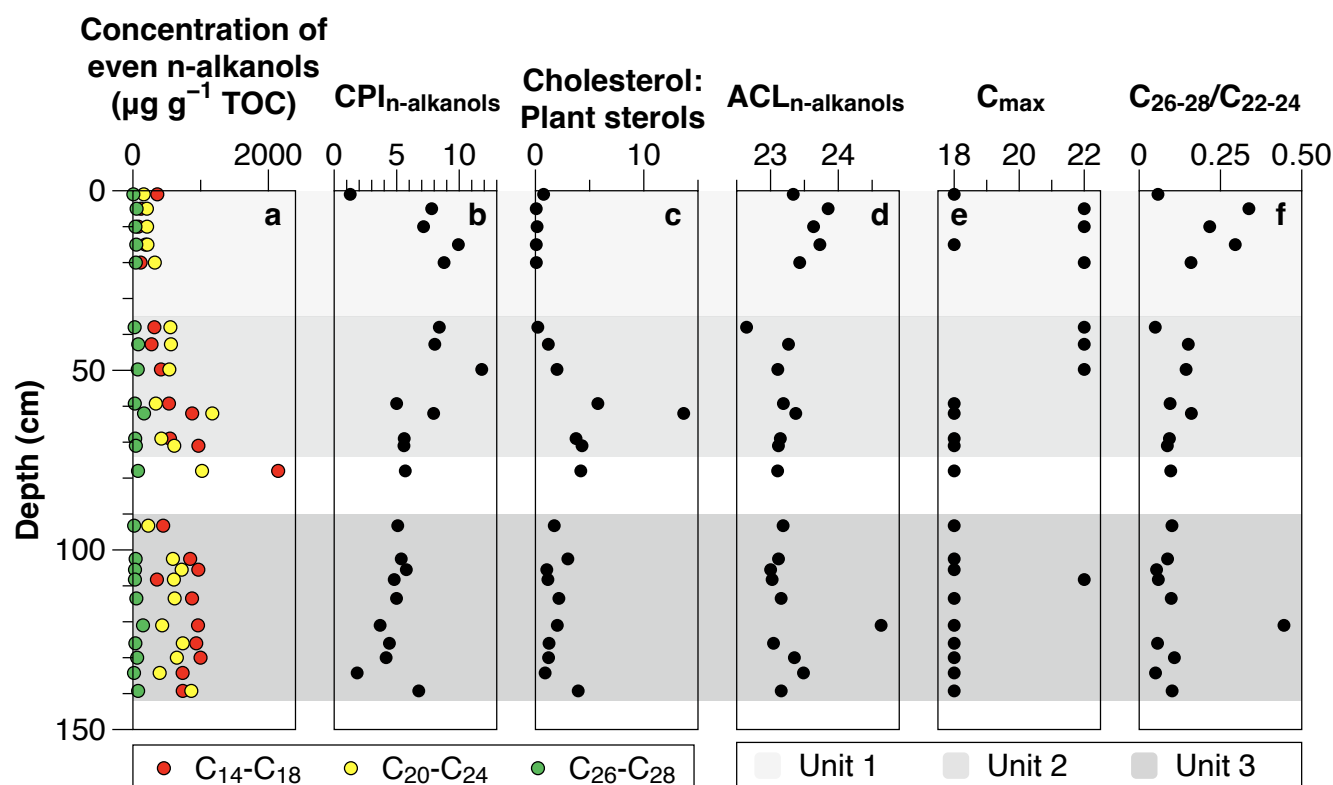
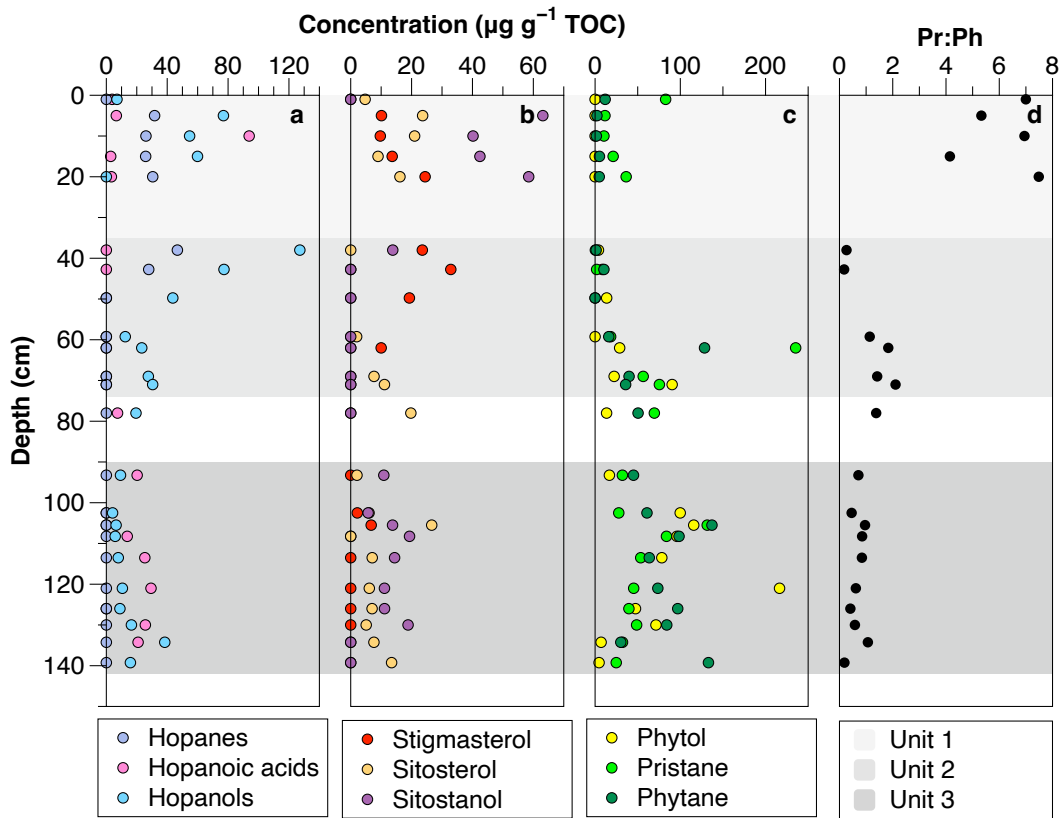


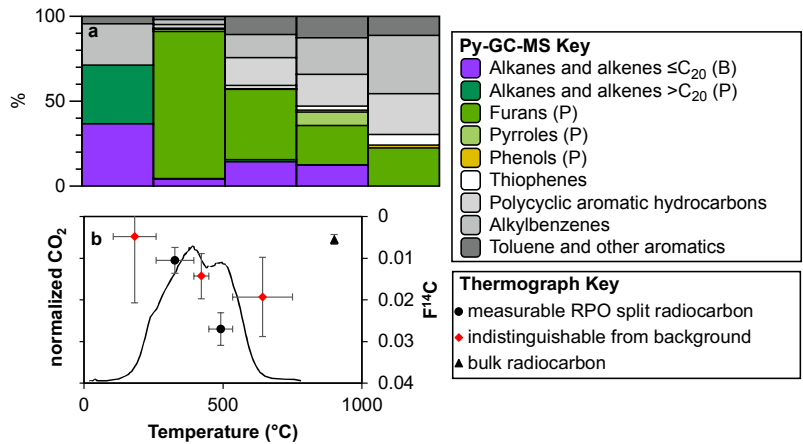
Figure 4 Fatty-acids parameters of the Friis Hills drill core. a. concentration per gram organic carbon of even low molecular weight FAME (n -C₁₄ to n -C₁₈), even mid molecular weight FAME (n -C₂₀ to n -C₂₄) and even high molecular weight FAME (n -C₂₆ to n -C₃₀), b. carbon preference index (CPI) for FAME (n -C₂₂ to n -C₂₈), c. ratio of iso-+anteiso- to straight chain FAME (n -C₁₅ to n -C₁₇), d. average chain length (ACL) for FAME (n -C₂₂ to n -C₃₂), e. dominant FAME chain length and f. higher plant fatty acid index (HPFA).



325 **Figure 5** *n*-Alkanol parameters of the Friis Hills drill core. a. concentration per gram organic carbon of even low molecular weight *n*-alkanols (*n*-C₁₂ to *n*-C₁₈), even mid-molecular weight *n*-alkanols *n*-C₂₀ to *n*-C₂₄, and even high molecular weight *n*-alkanols (*n*-C₂₆ to *n*-C₂₈), b. carbon preference index (CPI) for *n*-alkanols (*n*-C₂₂ to *n*-C₂₈), c. ratio of cholesterol to plant sterols, d. average chain length (ACL) for *n*-alkanols (*n*-C₂₂ to *n*-C₂₈), e. dominant *n*-alkanol chain length and f. Ratio of C₂₆₋₂₈ to C₂₂₋₂₄ *n*-alkanols.



330 **Figure 6** Concentrations of various biomarker compounds. a. hopanes, hopanoic acids and hopanols, b. plant sterols, c. phytol, pristane and phytane and d. pristane/phytane ratio (Pr:Ph ratio).



335 **Figure 7** Py-GC MS split composition for sample 2-C0-3, situated at 10 cm depth in the dry permafrost layer. a. compounds are classified approximately on bacterial- (B) or plant- (P) derived sources based on carbon number of alkanes and alkenes detected. b. RPO thermograph of CO₂ evolution and RPO and bulk radiocarbon measurements. Note: data points in red are indistinguishable from background measurements.



5 Discussion

5.1 Content and source of organic carbon: the legacy of the mid-Miocene tundra ecosystem

340 Soil organic carbon density in the dry and icy permafrost 140 cm core from Friis hills is about 4 times higher than those found in other ultraxerous soils like University Valley (SFig. 2; Faucher et al., 2017) and soils in the Mackay Glacier region (Fig. 10; Van Goethem et al., 2020), but multiple orders of magnitude less than carbon stocks in mineral soils of maritime Antarctica (e.g. Alekseev and Abakumov, 2024; Simas et al., 2007). Currently, no vascular plants grow in the MDV (e.g. Virginia and Wall, 1999). As such, the low SOCd in most Quaternary-age soils in the MDV is attributed to the soil organic carbon being
345 sourced either from endolithic micro-organisms (Faucher et al., 2017), glacially eroded material from older Cenozoic sediment and/or the Beacon Sandstone (e.g. Matsumoto et al., 1990a, Matsumoto et al., 2010), or Holocene age legacy carbon from aquatic systems (e.g. Lancaster, 2002, Barrett et al., 2006).

Conversely, the higher SOCd at Friis Hills is attributed to the tundra ecosystem that occupied the site during the mid-Miocene. The tundra macrofossil assemblage from the site included lichens, liverworts, mosses, dicots, grasses and sedges, along with
350 cuticles belonging to the *Nothofagaceae* family (Chorley et al., 2023). The lipid biomarkers that had a homogenous distribution in the 140 cm core are also consistent with the macrofossils assemblage. The BIT index of 1.0 derived from the brGDGTs samples suggests a fully terrestrial environment. The presence of high molecular weight *n*-alkanes, *n*-fatty acids and *n*-alkanols, along with phytosterols (i.e. sitosterol, stigmasterol and their respective stanols) and phytol (and pristane/phytane) are all indicative of a terrestrial environment dominated by higher order plants (Kögel-Knabner and Amelung, 2014; and references
355 therein). The presence of mid-molecular weight (C_{21} - C_{25}) *n*-alkanes, *n*-fatty acids and *n*-alkanols indicate contributions of mosses and lichens. Thus, the Friis Hills sediments harbour a tundra-type biomarker signature and explains why Friis Hills has higher SOCd than other sites in the MDV (Fig. S2).

5.2 Degradation of organic carbon in the dry permafrost

360 The hierarchical clustering analysis of the samples in the 1.4 m core produced three groups and the variables and indices that were statistically different between the units. The dry permafrost (0-35cm) experienced a high degree of organic carbon degradation by microbes, whereas the underlying icy permafrost (38-71cm) experienced intermediate level of degradation. The dry permafrost had C:N ratios near the Redfield ratio (Fig. 2d), higher $\delta^{13}C_{org}$ (Fig. 2c), a higher proportion of iso- and anteiso-FAs relative to *n*-FAs (Fig. 4c), low phytol (Fig. 6c) and a higher contributions of low-molecular weight homologues
365 of *n*-alkanes (Fig. 3c). All of these indicators can be attributed to a balanced microbial activity that degraded the organic carbon in Unit 1. The relative higher concentration of phytosterols, pristane, phytane and phytol in the dry permafrost, the dominance of hopanoids (Fig. 6a) and the absence of steranes, compounds that possess similar long-term preservation potential (Love and Zumberge, 2021), can all be attributed to the higher degradation of the plant material (Figs 5c & 6c). Moreover, the Py-GC-



MS measurements in the thermally partitioned splits for a sample in the dry permafrost indicate a high level of refractory organic matter and degraded organic carbon.

Active microbial growth in dry permafrost has also been reported in other ultraxerous soils. For example, in University Valley, the soils that experience temperature above 0°C and where the icy permafrost is recharged by snowmelt, the C:N ratios were also distributed along the Redfield ratio (Fig. S1; Faucher et al., 2017). Similar ratios were found in the Mackay Glacier region of East Antarctica, supporting active nutrient cycling, although low respiration rates were associated with dormancy (Van Goethem et al., 2020). At Friis Hills, the ice table and underlying icy permafrost is being recharged by evaporated snowmelt (Verret et al., 2022). As such the dry permafrost transiently receives input of liquid water that could support microbial activity, similar to the endoliths growing in bedrocks (e.g. Friedmann, 1982), and to the water tracks that form in spring and summer (Chan-Yam et al., 2019). $\delta^{13}\text{C}$ of bulk sample indicate active microbial activity is small (<1%), but yet it is producing degradation of organic carbon over large time-scales (as shown in the biomarker results). As such, microbial activity is occurring at very low rates, only when enough moisture is present in the dry permafrost layer. The rest of the time, the soil ecosystem remains mostly dormant (e.g. Van Goethem et al., 2020).

5.3 Degradation of organic carbon in the icy permafrost

The degradation of organic carbon is not limited to the dry permafrost and it can also be observed in the underlying icy permafrost (unit 2: 38-71cm). Units 1 and 2 both display a higher concentration of hopanoids and higher SC:LC ratios than Unit 3 (Figs 8d). Additionally, $\delta^{13}\text{C}_{\text{org}}$ values show a sharp shift below the ice table, and between Unit 2 and 3, where the average $\delta^{13}\text{C}_{\text{org}}$ changes from $-25.2 \pm 0.7\text{‰}$ to $-21.4 \pm 1.4\text{‰}$ VPDB (Fig. 2c). This shift in $\delta^{13}\text{C}_{\text{org}}$ values suggests preferential loss of ^{12}C in CO_2 from respiration and organic matter degradation, which leads to sediment enriched in $\delta^{13}\text{C}_{\text{org}}$. The samples just below the ice table also have the lowest $\text{CPI}_{\text{n-alkane}}$ and CPI_{FAME} (Figs 2c & 3c), showing a higher degree of degradation.

These findings imply that bacterial activity extended below the current ice table after the mid-Miocene. Based on a study on $^{10}\text{Be}_{\text{met}}$ concentrations in the upper section of the FHDP2C core, the onset of hyper-arid conditions occurred around 6.0 Ma (Verret et al., 2023); although it is uncertain if wet conditions prevailed throughout the late Miocene period and ended at 6.0 Ma or if a single warm period punctuated the late Miocene cooling trend at 6.0 Ma. Either way, before 6.0 Ma, an active layer that seasonally thawed was present in at the Friis Hills, which could explain the higher degradability of Unit 2 compared to Unit 3. The transition from Unit 2 to 3 at ~80 cm depth could therefore represent the position of a paleo layer of increased biological activity within the active layer. Similar trends have been observed in the Arctic, where paleo-active layers are identified biogeochemically by a higher degree of degradation. (e.g. Lacelle et al., 2019). Furthermore, permafrost carbon reservoirs have also been destabilized at a large scale in the Arctic during past warm periods of the Pleistocene-Holocene (e.g. Tesi et al., 2016). We show here that ancient permafrost carbon stocks in Antarctica, although marginal, could have also been destabilized during past warm periods in the near surface.

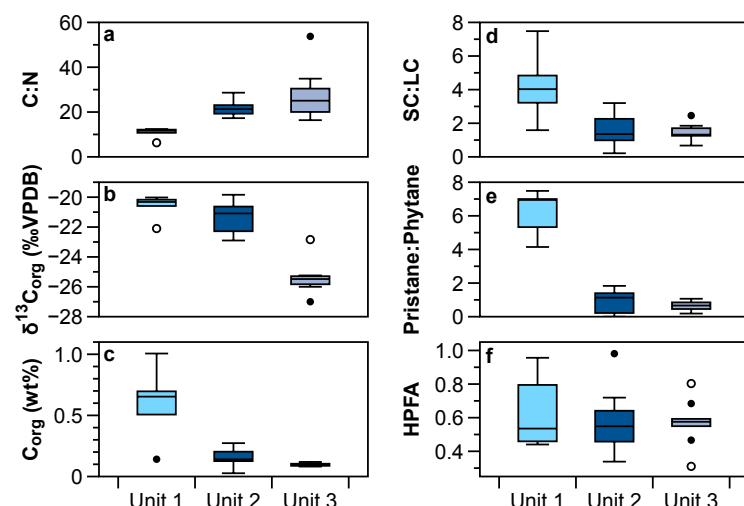


Figure 8 Boxplot of a. C:N ratio, b. $\delta^{13}\text{C}$, c. organic carbon, expressed in wt%, d. short-chain/long-chain *n*-alkane ratio, e. pristane/phytane ratio and f. HPFA index. Units are based on hierarchical clustering.

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5.4 Temperature thresholds to reactivate biological activity in the dry permafrost and paleo-active layer

brGDGTs have been increasingly used to reconstruct past temperatures in Arctic permafrost because the methylation and cyclisation of brGDGTs can be correlated to ground surface temperature (e.g. Raberg et al., 2024). Previous studies in permafrost regions have made the assumption that microbial communities stored in permafrost reflect the environmental conditions at time of enclosure, not the current conditions (closed-system assumption; Kusch et al., 2019). Moreover, recent studies have shown that in cold regions, the brGDGT calibrations best represents the summer ground temperature, and not the mean air temperatures for which brGDGT results are typically calibrated (e.g. Raberg et al., 2024). However, in the ultraxerous zone of the MDV, the mean annual air temperatures approximates those at the ground surface temperatures since there is no vegetation, minimal snow cover and little organic material (Lacelle et al., 2016).

While soil calibrations for Antarctic sites are lacking, sites from Arctic permafrost in Svalbard, Greenland and Alaska are found in the global soil sample database of De Jonge et al. (2014) and these regions show the same dominance of pentamethylated and hexamethylated brGDGTs (Kusch et al., 2019). The BayMBT₀ calibration (Crampton-Flood et al., 2020) also offers a separate calibration that assumes brGDGT distributions only reflect months with mean air temperatures above freezing and was applied to the Antarctic Peninsula (Tibbett et al., 2022). We therefore used these two calibrations to reconstruct mean summer ground temperatures to estimate the threshold temperature required to activate bacteria activity at different depths in core FHDP2C (November to February in the MDV; Obryk et al., 2020). The distribution of all samples in core FHDP2C was dominated by brGDGTs. All samples had a BIT index of 1.0. MBT'5_{ME} index ranged between 0.24 and 0.48 (Supplementary Data). We obtained temperatures varying between 2.3 and 7.1°C using the BayMBT₀ calibration (Crampton-Flood et al., 2020) and between -1.1 and 6.4°C using the De Jonge et al. (2014) calibration. The BayMBT₀

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calibration yields slightly warmer ($\sim 1^{\circ}\text{C}$ overall) temperature reconstruction than the DeJonge calibration, but overall they show similar trends (Fig. 9). The dry permafrost layer would require a mean summer soil temperature of $\sim 2\text{--}4^{\circ}\text{C}$ to reactivate biological processes. The temperature required to unlock the OM stored below the ice table seems a bit more uncertain but lies between 0°C near the ice table up to 7°C down at 140 cm. The latter is in line with the mean summer air temperatures required to thaw the maximum active layer depth (based on meteoric Beryllium-10 concentrations) during the late Miocene (i.e. $7\text{--}10^{\circ}\text{C}$ to thaw a maximum active layer depth of 2.74 m; Verret et al., 2023). In general, the temperature required to reactivate biological activity increases with depth since the thickness of the active layer is largely controlled by summer air temperature. The increase in threshold temperature to $\sim 0^{\circ}\text{C}$ near the ice table based on the De Jonge et al. (2014) calibration could be explained by the soil fauna having a high response rate to increased soil moisture (thawing ground ice) in the MDV (Niederberger et al., 2019; Andriuzzi et al., 2018). These temperature thresholds are in line with current conditions in the lower elevations of Taylor Valley at a site near Lake Fryxell ($77^{\circ}36'06.1''\text{S}$, $163^{\circ}08'19.6''\text{E}$, 21 m a.s.l.; Bakermans et al., 2014), where the mean summer ground temperature is $+2^{\circ}\text{C}$ and psychrophilic species have the ability to function. In short, the OM in unit 3 has a low probability of being reactivated, while the section closest to the ice table, unit 2, has a much higher probability and hence is showing higher levels of degradation.

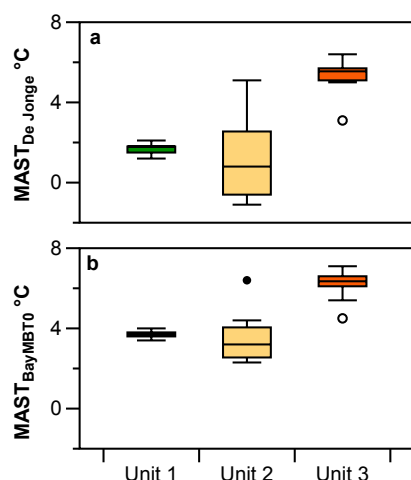


Figure 9 brGDGT ground temperature reconstructions for different units in core FHDP2C. These represent threshold temperatures for unlocking OM at different depths in the core: Unit 1 (0-35 cm depth), Unit 2 (38 -71 cm depth) and Unit 3 (93-140 cm depth).

6 Conclusions

Although our study suggests that Holocene organic carbon is being introduced at high elevation sites such as the Friis Hills, modern C_{org} contributions remain very low ($<1\%$). Beyond the dry permafrost, C_{org} is dominantly ancient and highly degraded. Based on the ramped pyrolysis approach to characterise and date different carbon pools, it seems like soil ecosystems in the high elevations (>1000 m a.s.l.) of the MDV rely mostly on exogenous sources of C_{org} , in this case legacy carbon from the Miocene tundra environment, but could have also marginally been able fix carbon *in situ* through the Holocene. More detailed



radiocarbon studies should be conducted to prove the latter. Moreover, based on our biomarker findings suggesting a gradient of C_{org} degradation through the soil profile, we conclude that legacy carbon *locked-in* at depth in the permafrost has been bioavailable under past warmer climate post-deposition. Seasonal thawing during warm periods is at the origin of C_{org} degradation at depth. Such periods seem to be marginal over the last 14 Myrs. Carbon in the dry upper 35 cm of the core could be bioavailable at a mean ground summer temperature of $+2^{\circ}\text{C}$, conditions similar to those found in the lower elevations of Taylor Valley. Carbon is a key physicochemical factor in the development of soil microbial communities (e.g. Cary et al., 2010). Therefore, future climate warming could lead to unlocking legacy sources of carbon which would cause considerable impacts on the structure and function of ecosystems in the MDV. Overall, the organic matter in the core appears to be compatible with a highly degraded signature of the mid-Miocene paleoenvironment, but also displays a gradual environmental over-print attributable to post-depositional conditions that is more important near the surface. This finding suggests that the OM enclosed within the permafrost at Friis Hills is stable and bacterial alteration is mostly inhibited. However, bacterial degradation has occurred during warmer periods: episodically down to 80 cm and potentially beyond 140 cm during the late Miocene and down to 35 cm through to Holocene.

Data availability

The data is uploaded as supplementary material.

Author Contributions

M.V., S.N., D.L., W.D. and K.N. designed this project and contributed to data analysis/interpretation and writing the manuscript. C.G. and J.T. ran the ramped pyrolysis experiment. R.L. developed the Friis Hills Drilling Project. All authors edited the whole manuscript.

Competing interests

One of the co-authors of this manuscript is a member of the editorial board of Biogeosciences.

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485 References

- Alekseev, I. and Abakumov, E.: Soil organic matter and biogenic-abiogenic interactions in soils of Larsemann Hills and Bunger Hills, East Antarctica, *Polar Science*, 40, 101040, 2024.
- Allibone, A. H., Cox, S. C., Graham, I. J., Smellie, R. W., Johnstone, R. D., Ellery, S. G., and Palmer, K.: Granitoids of the Dry Valleys area, southern Victoria Land, Antarctica: plutons, field relationships, and isotopic dating, *New Zealand Journal of Geology and Geophysics*, 36, 281-297, 1993.
- 490 Andriuzzi, W., Adams, B., Barrett, J., Virginia, R., and Wall, D.: Observed trends of soil fauna in the Antarctic Dry Valleys: early signs of shifts predicted under climate change, *Ecology*, 99, 312-321, 2018.
- Bakermans, C., Skidmore, M. L., Douglas, S., and McKay, C. P.: Molecular characterization of bacteria from permafrost of the Taylor Valley, Antarctica, *FEMS Microbiology Ecology*, 89, 331-346, 2014.
- 495 Bargagli, R., Sanchez-Hernandez, J., and Monaci, F.: Baseline concentrations of elements in the Antarctic macrolichen *Umbilicaria decussata*, *Chemosphere*, 38, 475-487, 1999.
- Barrett, J. E., Virginia, R. A., Lyons, W. B., McKnight, D. M., Priscu, J. C., Doran, P. T., Fountain, A. G., Wall, D. H., and Moorhead, D.: Biogeochemical stoichiometry of Antarctic dry valley ecosystems, *Journal of Geophysical Research: Biogeosciences*, 112, 2007.
- 500 Berke, M. A., Sierra, A. C., Bush, R., Cheah, D., and O'Connor, K.: Controls on leaf wax fractionation and $\delta^2\text{H}$ values in tundra vascular plants from western Greenland, *Geochimica et Cosmochimica Acta*, 244, 565-583, 2019.
- Bliss, A. K., Cuffey, K. M., and Kavanaugh, J. L.: Sublimation and surface energy budget of Taylor Glacier, Antarctica, *Journal of Glaciology*, 57, 684-696, 2011.
- Bray, E. and Evans, E.: Distribution of n-paraffins as a clue to recognition of source beds, *Geochimica et Cosmochimica Acta*, 22, 2-15, 1961.
- 505 Cary, S. C., McDonald, I. R., Barrett, J. E., and Cowan, D. A.: On the rocks: the microbiology of Antarctic Dry Valley soils, *Nature Reviews Microbiology*, 8, 129-138, 2010.
- Castañeda, I. S. and Schouten, S.: A review of molecular organic proxies for examining modern and ancient lacustrine environments, *Quaternary Science Reviews*, 30, 2851-2891, 2011.
- 510 Chan-Yam, K., Goordial, J., Greer, C., Davila, A., McKay, C. P., and Whyte, L. G.: Microbial activity and habitability of an Antarctic dry valley water track, *Astrobiology*, 19, 757-770, 2019.
- Chorley, H., Levy, R., Naish, T., Lewis, A., Cox, S., Hemming, S., Ohneiser, C., Gorman, A., Harper, M., Homes, A., Hopkins, J., Prebble, J., Verret, M., Dickinson, W., Florindo, F., Golledge, N., Halberstadt, A. R., Kowalewski, D., McKay, R., Meyers, S., Anderson, J., Dagg, B., and Lurcock, P.: East Antarctic Ice Sheet variability during the middle Miocene Climate Transition captured in drill cores from the Friis Hills, Transantarctic Mountains, *GSA Bulletin*, 135, 1503-1529, 2023.
- 515 Cleveland, C. C. and Liptzin, D.: C: N: P stoichiometry in soil: is there a "Redfield ratio" for the microbial biomass?, *Biogeochemistry*, 85, 235-252, 2007.
- Cox, S., Turnbull, I., Isaac, M., Townsend, D., and Lyttle, B.: Geology of southern Victoria Land, Antarctica, *Institute of Geological & Nuclear Sciences 1: 25 0 000 geological map 22. 1 sheet+ 135 pp*, GNS Science, Lower Hutt, New Zealand, 2012.
- 520 Crampton-Flood, E. D., Tierney, J. E., Peterse, F., Kirkels, F. M., and Damsté, J. S. S.: BayMBT: A Bayesian calibration model for branched glycerol dialkyl glycerol tetraethers in soils and peats, *Geochimica et Cosmochimica Acta*, 268, 142-159, 2020.
- De Jonge, C., Hopmans, E. C., Zell, C. I., Kim, J.-H., Schouten, S., and Damsté, J. S. S.: Occurrence and abundance of 6-methyl branched glycerol dialkyl glycerol tetraethers in soils: Implications for palaeoclimate reconstruction, *Geochimica et Cosmochimica Acta*, 141, 97-112, 2014.
- 525



- Donahue, D. J., Linick, T. W., and Jull, A. T.: Isotope-ratio and background corrections for accelerator mass spectrometry radiocarbon measurements, *Radiocarbon*, 32, 135-142, 1990.
- Doran, P. and Fountain, A.: *McMurdo Dry Valleys Friis Hills Meteorological Station Monthly Averages*, 2016.
- 530 Dragone, N. B., Diaz, M. A., Hogg, I. D., Lyons, W. B., Jackson, W. A., Wall, D. H., Adams, B. J., and Fierer, N.: Exploring the boundaries of microbial habitability in soil, *Journal of Geophysical Research: Biogeosciences*, 126, e2020JG006052, 2021.
- Duncan, B., McKay, R., Bendle, J., Naish, T., Inglis, G. N., Moossen, H., Levy, R., Ventura, G. T., Lewis, A., and Chamberlain, B.: Lipid biomarker distributions in Oligocene and Miocene sediments from the Ross Sea region, Antarctica: Implications for use of biomarker proxies in glacially-influenced settings, *Palaeogeography, Palaeoclimatology, Palaeoecology*, 516, 71-89, 535 2019.
- Eigenbrode, J. L.: Fossil lipids for life-detection: a case study from the early Earth record, *Space Science Reviews*, 135, 161-185, 2008.
- Faucher, B., Lacelle, D., Davila, A., Pollard, W., Fisher, D., and McKay, C. P.: Physicochemical and biological controls on carbon and nitrogen in permafrost from an ultraxerous environment, *McMurdo Dry Valleys of Antarctica*, *Journal of Geophysical Research: Biogeosciences*, 122, 2593-2604, 2017.
- 540 Flower, B. P. and Kennett, J. P.: The middle Miocene climatic transition: East Antarctic ice sheet development, deep ocean circulation and global carbon cycling, *Palaeogeography, palaeoclimatology, palaeoecology*, 108, 537-555, 1994.
- Freckman, D. W. and Virginia, R. A.: Low-diversity Antarctic soil nematode communities: distribution and response to disturbance, *Ecology*, 78, 363-369, 1997.
- 545 Friedmann, E. I.: Endolithic microorganisms in the Antarctic cold desert, *Science*, 215, 1045-1053, 1982.
- Ginnane, C. E., Turnbull, J. C., Naeher, S., Rosenheim, B. E., Venturelli, R. A., Phillips, A. M., Reeve, S., Parry-Thompson, J., Zondervan, A., and Levy, R. H.: Advancing Antarctic sediment chronology through combined ramped pyrolysis oxidation and pyrolysis GC-MS, *Radiocarbon*, 1-20, 2024.
- Goordial, J., Davila, A., Lacelle, D., Pollard, W., Marinova, M. M., Greer, C. W., DiRuggiero, J., McKay, C. P., and Whyte, 550 L. G.: Nearing the cold-arid limits of microbial life in permafrost of an upper dry valley, Antarctica, *The ISME journal*, 10, 1613-1624, 2016.
- Grimalt, J., Albaiges, J., Al-Saad, H., and Douabul, A.: n-Alkane distributions in surface sediments from the Arabian Gulf, *Naturwissenschaften*, 72, 35-37, 1985.
- Haugk, C., Jongejans, L. L., Mangelsdorf, K., Fuchs, M., Ogneva, O., Palmtag, J., Mollenhauer, G., Mann, P. J., Overduin, P. 555 P., and Grosse, G.: Organic matter characteristics of a rapidly eroding permafrost cliff in NE Siberia (Lena Delta, Laptev Sea region), *Biogeosciences*, 19, 2079-2094, 2022.
- Hopkins, D., Sparrow, A., Gregorich, E., Elberling, B., Novis, P., Fraser, F., Scrimgeour, C., Dennis, P., Meier-Augenstein, W., and Greenfield, L.: Isotopic evidence for the provenance and turnover of organic carbon by soil microorganisms in the Antarctic dry valleys, *Environmental microbiology*, 11, 597-608, 2009.
- 560 Hopmans, E. C., Schouten, S., and Damsté, J. S. S.: The effect of improved chromatography on GDGT-based palaeoproxies, *Organic Geochemistry*, 93, 1-6, 2016.
- Hopmans, E. C., Weijers, J. W., Schefuß, E., Herfort, L., Damsté, J. S. S., and Schouten, S.: A novel proxy for terrestrial organic matter in sediments based on branched and isoprenoid tetraether lipids, *Earth and Planetary Science Letters*, 224, 107-116, 2004.
- 565 Horowitz, N. H., Cameron, R. E., and Hubbard, J. S.: Microbiology of the Dry Valleys of Antarctica: Studies in the world's coldest and driest desert have implications for the Mars biological program, *Science*, 176, 242-245, 1972.
- Juggins, S.: Package 'rioja', An R Package for the Analysis of Quaternary Science Data., 0.9, 26, 2020.
- Kaal, J.: Analytical pyrolysis in marine environments revisited, *Analytical Pyrolysis Letters*, 6, 1-16, 2019.
- Keely, B. J.: Geochemistry of chlorophylls, in: *Chlorophylls and bacteriochlorophylls: biochemistry, biophysics, functions and applications*, Springer, 535-561, 2006.
- 570 Killops, S. and Killops, V.: 5.1. 2 General differences between major groups of organisms, *Introduction to Organic Geochemistry*, (Somerset: Wiley), 167, 2013.
- Kögel-Knabner, I. and Amelung, W.: 12.7 - Dynamics, Chemistry, and Preservation of Organic Matter in Soils, in: *Treatise on Geochemistry (Second Edition)*, edited by: Holland, H. D., and Turekian, K. K., Elsevier, 157-215, 2014.



- 575 Kusch, S., Winterfeld, M., Mollenhauer, G., Höfle, S. T., Schirrmeister, L., Schwamborn, G., and Rethemeyer, J.: Glycerol dialkyl glycerol tetraethers (GDGTs) in high latitude Siberian permafrost: Diversity, environmental controls, and implications for proxy applications, *Organic Geochemistry*, 136, 103888, 2019.
- Lacelle, D., Fontaine, M., Pellerin, A., Kokelj, S. V., and Clark, I. D.: Legacy of holocene landscape changes on soil biogeochemistry: a perspective from paleo-active layers in Northwestern Canada, *Journal of Geophysical Research: Biogeosciences*, 124, 2662-2679, 2019.
- 580 Lacelle, D., Lapalme, C., Davila, A. F., Pollard, W., Marinova, M., Heldmann, J., and McKay, C. P.: Solar radiation and air and ground temperature relations in the cold and hyper-arid Quartermain Mountains, McMurdo Dry Valleys of Antarctica, *Permafrost and Periglacial Processes*, 27, 163-176, 2016.
- Lacelle, D., Davila, A. F., Fisher, D., Pollard, W. H., DeWitt, R., Heldmann, J., Marinova, M. M., and McKay, C. P.: Excess ground ice of condensation–diffusion origin in University Valley, Dry Valleys of Antarctica: Evidence from isotope geochemistry and numerical modeling, *Geochimica et Cosmochimica Acta*, 120, 280-297, 2013.
- 585 Lawson, J., Doran, P. T., Kenig, F., Des Marais, D. J., and Priscu, J. C.: Stable carbon and nitrogen isotopic, *Aquatic Geochemistry*, 10, 269-301, 2004.
- Lewis, A. R. and Ashworth, A. C.: An early to middle Miocene record of ice-sheet and landscape evolution from the Friis Hills, Antarctica, *Bulletin*, 128, 719-738, 2016.
- 590 Lewis, A. R., Marchant, D. R., Ashworth, A. C., Hedenäs, L., Hemming, S. R., Johnson, J. V., Leng, M. J., Machlus, M. L., Newton, A. E., and Raine, J. I.: Mid-Miocene cooling and the extinction of tundra in continental Antarctica, *Proceedings of the National Academy of Sciences*, 105, 10676-10680, 2008.
- Love, G. D. and Zumbege, J. A.: *Emerging patterns in proterozoic lipid biomarker records*, Cambridge University Press 2021.
- 595 Marchant, D. R. and Head III, J. W.: Antarctic dry valleys: Microclimate zonation, variable geomorphic processes, and implications for assessing climate change on Mars, *Icarus*, 192, 187-222, 2007.
- Meyers, P. A.: Preservation of elemental and isotopic source identification of sedimentary organic matter, *Chemical geology*, 114, 289-302, 1994.
- Meyers, P. A. and Ishiwatari, R.: Lacustrine organic geochemistry—an overview of indicators of organic matter sources and diagenesis in lake sediments, *Organic geochemistry*, 20, 867-900, 1993.
- 600 Moorhead, D. L., Wall, D. H., Virginia, R. A., and Parsons, A. N.: Distribution and life-cycle of *Scottinema lindsayae* (Nematoda) in Antarctic soils: a modeling analysis of temperature responses, *Polar Biology*, 25, 118-125, 2002.
- Naafs, B., Inglis, G., Blewett, J., McClymont, E. L., Lauretano, V., Xie, S., Evershed, R., and Pancost, R.: The potential of biomarker proxies to trace climate, vegetation, and biogeochemical processes in peat: A review, *Global and Planetary Change*, 179, 57-79, 2019.
- 605 Naeher, S., Cui, X., and Summons, R. E.: Biomarkers: molecular tools to study life, environment, and climate, *Elements: An International Magazine of Mineralogy, Geochemistry, and Petrology*, 18, 79-85, 2022.
- Naeher, S., Niemann, H., Peterse, F., Smittenberg, R. H., Zigah, P. K., and Schubert, C. J.: Tracing the methane cycle with lipid biomarkers in Lake Rotsee (Switzerland), *Organic Geochemistry*, 66, 174-181, 2014.
- 610 Naeher, S., Smittenberg, R. H., Gilli, A., Kirilova, E. P., Lotter, A. F., and Schubert, C. J.: Impact of recent lake eutrophication on microbial community changes as revealed by high resolution lipid biomarkers in Rotsee (Switzerland), *Organic Geochemistry*, 49, 86-95, 2012.
- Niederberger, T. D., Bottos, E. M., Sohm, J. A., Gunderson, T., Parker, A., Coyne, K. J., Capone, D. G., Carpenter, E. J., and Cary, S. C.: Rapid microbial dynamics in response to an induced wetting event in Antarctic Dry Valley soils, *Frontiers in Microbiology*, 10, 621, 2019.
- 615 Obryk, M. K., Doran, P. T., Fountain, A. G., Myers, M., and McKay, C. P.: Climate from the McMurdo Dry Valleys, Antarctica, 1986–2017: surface air temperature trends and redefined summer season, *Journal of Geophysical Research: Atmospheres*, 125, e2019JD032180, 2020.
- Osburn, C. L., Anderson, N. J., Leng, M. J., Barry, C. D., and Whiteford, E. J.: Stable isotopes reveal independent carbon pools across an Arctic hydro-climatic gradient: Implications for the fate of carbon in warmer and drier conditions, *Limnology and Oceanography Letters*, 4, 205-213, 2019.
- 620 Peters, K. E., Walters, C. C., and Moldowan, J. M.: *The biomarker guide: Volume 2, Biomarkers and isotopes in petroleum systems and earth history*, Cambridge University Press 2007.



- Poynter, J. and Eglinton, G.: 14. Molecular composition of three sediments from hole 717c: The Bengal fan, Proceedings of the Ocean Drilling Program: Scientific results, 155-161,
- 625 Raberg, J. H., Crump, S. E., de Wet, G., Harning, D. J., Miller, G. H., Geirsdóttir, Á., and Sepúlveda, J.: BrGDGT lipids in cold regions reflect summer soil temperature and seasonal soil water chemistry, *Geochimica et Cosmochimica Acta*, 2024.
- Redfield, A. C.: On the proportions of organic derivatives in sea water and their relation to the composition of plankton, university press of liverpool Liverpool1934.
- 630 Reimer, P. J., Brown, T. A., and Reimer, R. W.: Discussion: reporting and calibration of post-bomb ^{14}C data, *Radiocarbon*, 46, 1299-1304, 2004.
- Schouten, S., Hopmans, E. C., and Damsté, J. S. S.: The organic geochemistry of glycerol dialkyl glycerol tetraether lipids: A review, *Organic geochemistry*, 54, 19-61, 2013.
- 635 Shevenell, A. E., Kennett, J. P., and Lea, D. W.: Middle Miocene southern ocean cooling and Antarctic cryosphere expansion, *Science*, 305, 1766-1770, 2004.
- Simas, F., Schaefer, C., Mendonça, E., Silva, I., Santana, R., and Ribeiro, A.: Organic carbon stocks in permafrost-affected soils from Admiralty Bay, Antarctica, US Geological Survey and The National Academies, USGS OF-2007-1047, Short Research Paper, 76, 2007.
- 640 Strauss, J., Schirrmeister, L., Mangelsdorf, K., Eichhorn, L., Wetterich, S., and Herzschuh, U.: Organic-matter quality of deep permafrost carbon—a study from Arctic Siberia, *Biogeosciences*, 12, 2227-2245, 2015.
- Stuiver, M. and Polach, H. A.: Discussion reporting of ^{14}C data, *Radiocarbon*, 19, 355-363, 1977.
- Tamppari, L., Anderson, R., Archer, P., Douglas, S., Kounaves, S., McKay, C., Ming, D., Moore, Q., Quinn, J., and Smith, P.: Effects of extreme cold and aridity on soils and habitability: McMurdo Dry Valleys as an analogue for the Mars Phoenix landing site, *Antarctic Science*, 24, 211-228, 2012.
- 645 Tesi, T., Muschitiello, F., Smittenberg, R. H., Jakobsson, M., Vonk, J., Hill, P., Andersson, A., Kirchner, N., Noormets, R., and Dudarev, O.: Massive remobilization of permafrost carbon during post-glacial warming, *Nature Communications*, 7, 13653, 2016.
- Tibbitt, E. J., Warny, S., Tierney, J. E., Wellner, J. S., and Feakins, S. J.: Cenozoic Antarctic Peninsula Temperatures and Glacial Erosion Signals From a Multi-Proxy Biomarker Study, *Paleoceanography and Paleoclimatology*, 37, e2022PA004430, 2022.
- 650 Turnbull, J. C., Zondervan, A., Kaiser, J., Norris, M., Dahl, J., Baisden, T., and Lehman, S.: High-precision atmospheric $^{14}\text{CO}_2$ measurement at the Rafter Radiocarbon Laboratory, *Radiocarbon*, 57, 377-388, 2015.
- Van Goethem, M. W., Vikram, S., Hopkins, D. W., Hall, G., Woodborne, S., Aspray, T. J., Hogg, I. D., Cowan, D. A., and Makhalanyane, T. P.: Nutrient parsimony shapes diversity and functionality in hyper-oligotrophic Antarctic soils, *bioRxiv*, 2020.
- 655 Verret, M., Dickinson, W., Lacelle, D., Fisher, D., Norton, K., Chorley, H., Levy, R., and Naish, T.: Cryostratigraphy of mid-Miocene permafrost at Friis Hills, McMurdo Dry Valleys of Antarctica, *Antarctic Science*, 33, 174-188, 2021.
- Verret, M., Trinh-Le, C., Dickinson, W., Norton, K., Lacelle, D., Christl, M., Levy, R., and Naish, T.: Late Miocene onset of hyper-aridity in East Antarctica indicated by meteoric beryllium-10 in permafrost, *Nature Geoscience*, 1-7, 2023.
- 660 Virginia, R. A. and Wall, D. H.: How soils structure communities in the Antarctic Dry Valleys, *Bioscience*, 49, 973-983, 1999.
- Zech, M., Buggle, B., Leiber, K., Marković, S., Glaser, B., Hambach, U., Huwe, B., Stevens, T., Sümegi, P., and Wiesenberger, G.: Reconstructing Quaternary vegetation history in the Carpathian Basin, SE-Europe, using n-alkane biomarkers as molecular fossils: problems and possible solutions, potential and limitations, *E&G Quaternary Science Journal*, 58, 148-155, 2010.
- 665 Zondervan, A., Hauser, T., Kaiser, J., Kitchen, R., Turnbull, J., and West, J.: XCAMS: The compact ^{14}C accelerator mass spectrometer extended for ^{10}Be and ^{26}Al at GNS Science, New Zealand, *Nuclear Instruments and Methods in Physics Research Section B: Beam Interactions with Materials and Atoms*, 361, 25-33, 2015.



Table 1: Summary of RPO-AMS results

NZA #	RP Split	RP split min temp. (°C)	RP split max temp. (°C)	Split size (mg C)	CRA (yrs BP)	CRA Error	F ¹⁴ C	F ¹⁴ C Error	Comments
70400	Bulk			1.07	41,749	1,736	0.0055	0.0012	
75354	1	105	259	0.08	42,935	26,742	0.0048	0.0159	This sample is indistinguishable from the background measurements
75365	2	259	395	0.45	36,576	2,379	0.0105	0.0031	
75361	3	395	449	0.22	34,098	3,018	0.0143	0.0054	This sample is indistinguishable from the background measurements
75364	4	449	535	0.32	29,004	1,158	0.0270	0.0039	
75359	5	535	750	0.17	31,716	3,971	0.0193	0.0095	This sample is indistinguishable from the background measurements