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Substrate potential of Eemian to Holocene permafrost organic matter for future microbial greenhouse gas production

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Abstract. Multiple permafrost cores from Bol´shoy Lyakhovsky Island in NE Siberia comprising deposits from Eemian to modern time are investigated to evaluate the stored potential of the freeze-locked organic matter (OM) to serve as substrate for the production of microbial greenhouse gases from thawing permafrost deposits. Deposits from Late Pleistocene glacial periods (comprising MIS 3 and MIS 4) possess an increased aliphatic character and a higher amount of potential substrates, and therefore higher OM quality in terms of biodegradation compared to interglacial deposits from the Eemian (MIS 5e) as well as from the Holocene (MIS 1). To assess the potential of the individual permafrost deposits to provide substrates for microbially induced greenhouse gas generation, concentrations of free and bound acetate as an excellent substrate for methanogenesis are used. The highest free (in pore water and segregated ice) and bound (bound to the organic matrix) acetate-substrate pools of the permafrost deposits are observed within the interstadial MIS 3 and stadial MIS 4 period deposits. In contrast, deposits from the last interglacial MIS 5e show only poor substrate pools. The Holocene deposits reveal a significant bound-acetate pool, representing at least a future substrate potential upon release during OM degradation. Biomarkers for past microbial communities (branched and isoprenoid GDGTs) show also highest abundance of past microbial communities during the MIS 3 and MIS 4 deposits, which indicates higher OM quality with respect to microbial degradation during time of deposition. On a broader perspective, Arctic warming will increase permafrost thaw and favour substrate availability from freeze-locked older permafrost deposits. Therefore, especially those deposits from MIS 3 and MIS 4 show a high potential for providing substrates relevant for methanogenesis.

1 Introduction

The northern areas of the Eurasian landmass are underlain by permafrost, which is defined as ground that remains under 0 °C for at least 2 consecutive years (Washburn, 1980). These areas are a large reservoir of organic carbon (French, 2007; Zimov et al., 2009) freeze-locked in the permafrost deposits. The formation of permafrost accompanied by low temperature and anoxic soil conditions associated with reduced rates of organic matter (OM) decomposition (Schimel and Schaeffer, 2012) resulted in high OM accumulation (Kuhry et al., 2009; Zimov et al., 2009; Schirrmeister et al., 2011a). Hugelius et al. (2014)

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estimated that about 1300 Pg (1 Pg = 10^{15} = 1 Gt) soil organic carbon is stored in the upper 1 to 3 m in the northern circumpolar permafrost regions, of which 822 Pg is perennially frozen. This freeze-locked soil OM is highly vulnerable to climate warming (Grosse et al., 2011; Mu et al., 2014) and possesses a high potential for global climate feedback (Beer, 2008). Today, Arctic summer temperatures are higher than in the past 400 years (Chapin III et al., 2005). Increases in ground temperature, changes in soil drainage, deepening of the active layer (seasonally thawed surface layer), spatial retreat of permafrost and drastic changes in the ecosystem have already been reported for the Arctic as a consequence of northern hemisphere warming (Davidson and Janssens, 2006; Anisimov, 2007; Mueller et al., 2015; Romanovsky et al., 2010). Permafrost thawing leads to re-mobilization of freeze-locked OM and nutrients (Lawrence and Slater, 2005; Vonk et al., 2015) resulting in increasing decomposition rates (Dutta et al., 2006; Schmidt et al., 2011) and increasing accessibility of the formally persevered OM for microbial degradation. This will enhance microbial production and the release of greenhouse gasses such as carbon dioxide and methane to the atmosphere (Wagner et al., 2003; Schuur et al., 2008; McGuire et al., 2009; Knoblauch et al., 2013). Incubation experiments on permafrost samples of different ages revealed that permafrost thawing is accompanied by outgassing of greenhouse gases (Waldrop et al., 2010; Lee et al., 2012; Lipson et al., 2012; Knoblauch et al., 2013; Schadel et al., 2014; Walz et al., 2017). Former studies on samples from NE Siberian Holocene and Late Pleistocene (LP) Yedoma deposits, an ice-rich paleosol formation that is wide-spread in NE Siberia, have shown that microbial degradability of the freeze-locked OM in permafrost seems to depend on the amount and quality of organic carbon (OC) rather than on the age of the deposits (Knoblauch et al., 2013; Strauss et al., 2015; Stapel et al., 2016).

On Bol´shoy Lyakhovsky Island formation of permafrost started already in the Late Pliocene (Arkhangelov et al., 1996) and provides a unique paleo-environmental archive with stratigraphic patterns of long-lasting accumulation periods of permafrost during glacial periods, as well as permafrost degradation features during interglacial periods (Andreev et al., 2004, 2009; Wetterich et al., 2009, 2011). Here, permafrost deposits were accumulated under continental, cold climate conditions accompanied by syngenetic ice-wedge growth (Wetterich et al., 2011) during glacial periods, e.g. the middle Pleistocene (Saalian) and Late Pleistocene (Weichselian; Yedoma deposits) (Andreev et al., 2004; Schirrmeister et al., 2013). In contrast, in the Eemian and Holocene interglacial environments, extensive thawing of ice wedges and permafrost deposits led to the formation of thermokarst depressions, as well as of thermal erosional valleys and small rivers (Andreev et al., 2004; Ilyashuk et al., 2006; Wetterich et al., 2009). According to pollen and insect data, the Eemian climate resulted in a tundra environment including thermokarst and lake depressions. Open grass and grass-sedge tundra was similar to the modern one (Kienast et al., 2008). In the mid Eemian the environment was characterized by shrub tundra with summer temperatures of 4-5 °C higher than modern and with greater seasonal temperature variations in the Northern Hemisphere (Andreev et al., 2004; Dahl-Jensen et al., 2013). Therefore, Eemian deposits in this study are used as model for an interglacial period containing information on how an ongoing warming climate in the Arctic may affect permafrost OM degradation.

According to prior studies by Wetterich et al. (2014) and references therein, the cores investigated in this study can be integrated into an already described environmental and climatic history. To access information on quality and biodegradability of the freeze-locked OM, low molecular weight organic acids (LMWOAs) and characteristic OM parameters (amount and quality) are examined in this study. In addition, investigations of microbial biomarkers such as phospholipid fatty acids (PLFAs) and glycerol dialkyl glycerol tetraethers (GDGTs) are used to examine present and past microbial communities in the context of modern and past environmental conditions. Phospholipids are essential membrane components of living cells (Zelles, 1999) and hydrolysed rapidly after cell death (White et al., 1979; Logemann et al., 2011), therefore their fatty acid side chain inventory (PLFA) are used as indicator for viable microorganisms in sediments (Haack et al., 1994). In contrast, GDGTs and archaeol represent membrane lipids of past microbial biomass, since they are already partly degraded as indicated by the loss of their head groups (Pease et al., 1998). While archaeol and GDGTs with isoprenoid tetraether bridges represent archaeal biomass, GDGTs with branched tetraether bridges derive from bacteria (Weijers et al., 2006). However, in this context it has to be mentioned that the bacterial biomarkers only represent part of the bacterial community, while the archaeal markers cover the essential part of the past archaeal community.

The feedback between climate warming and microbial greenhouse gas generation from thawing permafrost is a topic of intensive modern scientific debate (Zimov et al., 2006; Koven et al., 2011; Schuur et al., 2015). However, the feedback effects on permafrost deposits of different ages are not well understood. Therefore, the aim of this study is to compare the stored potential for microbial greenhouse gas production in permafrost deposits from different glacial/ interglacial and stadial/ interstadial periods. Due to the expected similarities permafrost deposits from the Eemian interglacial period in comparison to Holocene permafrost deposits are also in the focus of this paper.

2 Study area and material

Bol´shoy Lyakhovsky Island is located between the Laptev and East Siberian seas as the southernmost part of the New Siberian Archipelago (Fig. 1a). During Pleistocene periods of low sea level the island was part of west Beringia, an unglaciated landmass stretching from NE Siberia to Alaska (Hubberten et al., 2004; Andreev et al., 2009). The area is part of the northern tundra zone with an active layer thickness of 30 to 40 cm and a permafrost thickness of 500 to 600 m (Andreev et al., 2004). The study site is located west of the Zimov´e River mouth on the south coast of Bol´shoy Lyakhovsky Island along the Dmitry Laptev Strait (Fig. 1b). This southern coast is characterized by exposed permafrost deposits while the hinterland is formed by gradually sloping terrain intersected by rivers and valleys developed through thermo-erosion. Based on previous studies (Andreev et al., 2004, 2009; Ilyashuk et al., 2006; Wetterich et al., 2014) in the study area the stratigraphy and regional setting are well known. Therefore, the drill sites (Fig. 1c) were chosen to maximize stratigraphic coverage and age with the aim to obtain a permafrost record from the Holocene (MIS 1) back to the Eemian (MIS 5e).

The field work was conducted in April 2014 as part of the joint Russian-German research project CarboPerm (Schwamborn and Wetterich, 2015). Four cores were drilled using a KMB-3-15M (rotary) drill rig. The drilled core segments were kept frozen and transported in frozen state for further processing to Potsdam, Germany. In the home laboratory sampling was conducted in a climate chamber at -10 °C. 40 inner core samples distributed throughout the cores were taken with exception of intervals, where ice-wedge ice was encountered. Samples were investigated for microbial biomarkers, free (pore-water) and bound acetate concentrations and OM characteristics such as total organic carbon (TOC), total organic carbon to total nitrogen (TOC/TN) ratio, hydrogen index (HI) and compositional OM analysis using open-pyrolysis gas chromatography (Pyr-GC).

2.1 Core descriptions

Cores are described stratigraphically from younger to older deposits. Core L14-05 (Fig. 1c, Table 1) is 7.89 m long and consists of silty fine sediments with scattered organic remains. Overall this core possesses lens-like cryostructures which are distinct between 1.00 to 2.45 m and 6.71 to 7.89 m core depth. According to prior studies by Andreev et al. (2009) and Wetterich et al. (2009), the upper core section approximately down to 5.5 m consists of a Holocene (MIS 1) unit, further below the deposits are of MIS 3 age. According to previous paleo-environmental interpretations the MIS 1 deposits represent Alas deposits, where Early Holocene lake sediments have accumulated on top of a MIS 3 surface. During late Holocene time (<3.7 ka BP) the site drained and froze over.

Core L14-02 (Table 1) is 20.02 m in length. The upper 11.26 m consist of fine-silty sediments with macroscopical organic remains and an alternation of horizontal, vertical and reticulated ice veins, and lens-like cryostructures. Below 11.26 m the core consists of an ice wedge, and no samples were taken from this part. The core material is of Late Pleistocene age and was deposited under subaerial conditions during the last interstadial MIS 3 (Wetterich et al., 2014). The deposits represent the infill of an ice-wedge polygon with a succession of paleosols.

The upper 4.90 m of core L14-03 (15.49 m in length; Table 1) are comparable in their sedimentology and cryostructures to the silty-fine sediments of core L14-02. Below 4.90 m the sediment has sandy portions. While between 4.90 to 8.45 m the deposits still have visible organic remains and similar cryostructures, the sediments below 8.45 m are characterized by only scattered organic remains but similar cryostructures as described above. Below 10.90 m the deposits mainly consist of sand and gravel, and in the lowermost 40 cm of gravel. Cryostructures are partly formed as vertically aligned cm-thick ice veins. In earlier studies on the same study site these deposits are interpreted to represent early Weichselian (MIS 4) floodplain deposits (Andreev et al., 2009).

Core L14-04 (Table 1) is 8.10 m long and consists of silty-fine sediments with visible organic remains and cryostructures comparable to core L14-02. Between 4.24 to 4.89 m the core consists of massive ice. The upper 6 m were probably deposited during the MIS 4 stadial period. The deposits below 6 m were deposited during the Eemian interglacial (MIS 5e) and have been interpreted as thermokarst lake sediments (Andreev et al., 2004).

3 Methods

3.1 Organic matter parameters

After freeze-drying and grounding of the samples total organic carbon (TOC) and total nitrogen (TN) (wt%) were determined by a carbon-nitrogen-sulfur analyser (Vario EL III, Elementar) with a device-specific accuracy of ± 0.1 wt%. For

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further information on characteristic OM parameters the hydrogen index (HI) was determined by Rock-Eval pyrolysis using a Rock-Eval 6 instrument (Behar et al., 2001). 17 freeze-dried and grounded samples of different TOC content covering all time intervals were analysed. Measurements were conducted by Applied Petroleum Technology AS (Kjeller, Norway). To obtain additional information on the macromolecular structure of the OM 10 mg from the selected samples were used for open-system pyrolysis after Horsfield et al. (1989). Measurements were performed on a pyrolysis-gas chromatograph (AGILENT GC 6890A Chromatograph) equipped with a flame ionization detector (Py-GC-FID). For details on the listed methods see Stapel et al. (2016).

3.2 Low molecular weight organic acids (LMWOAs) analyses

After slow thawing of a subset of the frozen samples at about 4 °C, the pore water within the samples were separated by centrifugation (Sigma, laboratory centrifuge 6K15, 2500 rpm (908 x g), 20 °C, 10 min). Free LMWOAs and anions were measured by ion chromatography with conductivity detection (ICS 3000, Dionex). Furthermore, LMWOAs bound via ester-bonds to the complex OM were analysed by conducting an alkaline ester cleavage approach developed by Glombitza et al. (2009a) on pre-extracted sediment samples. Details are described in Stapel et al. (2016).

3.3 Microbial lipid biomarker analysis

Approximately 30 - 50 g of the freeze-dried and grounded samples were extracted using a flow blending system modified after Bligh and Dyer (1959) as described in Stapel et al. (2016). Subsequently, the obtained sediment extract was separated into four different fractions of polarity (low polar lipids, free fatty acids, glycolipids, and polar lipids) following a method described by Zink and Mangelsdorf (2004). Finally, all four fractions were evaporated to dryness and stored at -20 °C until analysis. After a fatty acid cleavage procedure described in Müller et al. (1998), the phospholipid fatty acids (PLFA) within the PL fraction were measured by gas chromatography-mass spectrometry (GC-MS). Details on instrument settings are described in Stapel et al. (2016).

After asphaltene precipitation the low polar lipid fraction was separated into an aliphatic, aromatic and hetero-compound (containing nitrogen, oxygen and sulphur-components, NSO) fraction using a medium-pressure liquid chromatography (MPLC) (Radke et al., 1980). An aliquot of the NSO fraction was investigated for tetraether lipids (glycerol dialkyl glycerol tetraether, GDGT) and archaeol using a Shimadzu LC20AD HPLC instrument coupled to a Finnigan TSQ 7000 triple quadrupole MS with an atmospheric pressure chemical ionization (APCI) interface. Details on instrument settings are described in Stapel et al. (2016).

4 Results

Characteristic OM parameters (TOC and TOC/TN), biomarkers for living microbial communities (PLFA), past bacterial (br-GDGTs) and past archaeal communities (iso-GDGTs + archaeol) as well as the concentration of free and bound acetate are

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presented in figure 2 for all four cores from Bol´shoy Lyakhovsky Island. Every single core consists of at least one sample (core L14-03 consist of two samples) representing the overlaying soil as part of the active layer above the permafrost deposits. Additionally, the results of 17 selected samples for open-system pyrolysis are shown in figure 3.

4.1 Characteristic OM parameters

Active layers:

In the active layers TOC values are slightly above 2.3 wt%, except in core L14-05 with 1.5 wt% (Fig. 2a). The TOC/TN values range between 8.2 and 11.1 (Fig. 2b). The representative active layer sample for Rock-Eval analysis revealed a hydrogen index (HI) of 236 mg HC/ g TOC (Fig. 2c). Overall, the samples reveal strongly increased PLFA concentrations (84.1, 149.3, 86.3 and 37.8 µg/g sediment, respectively) compared to the permafrost deposits below (Fig. 2d). The concentrations of past bacterial marker (brGDGTs) vary between 849.1, 53.2, 27.2 and 196.0 ng/g sediment, while the concentrations of the past archaeal markers are much lower with 22.0, 5.2, 6.5 and 7.4 ng/g sediment in each core, respectively (Fig. 2e,f). Free acetate concentration (Fig. 2g) is, compared to the rest of the cores, rather low (0.9 to 1.5 mg/l). In contrast, the bound acetate concentrations are comparatively high with 44.5 to 64.2 mg/l (Fig. 2h).

Marine Isotope Stage 1 (MIS 1):

In unit MIS 1 from core L14-05, the TOC, TOC/TN and HI-profiles (Fig. 2.a,b,c) correlate (TOC:TOC/TN, R^2=0.9; p=0.047 and TOC:HI, R^2=0.79; p=0.015). TOC values vary between 1.5 to 1.8 wt%, TOC/TN values between 7.1 to 8.2 and HI data between 71 to 194 mg HC/ g TOC (H1 to H5; Fig. 2c). Overall, the concentration of PLFAs is lower than in the active layer ranging between 11 and 27 µg/g sediment (Fig. 2d). The PLFA data resemble the TOC profile. Concentrations for past bacterial markers vary between 22.5 and 370.0 ng/g sediment and for the past archaeal marker between 10.5 and 65.1 ng/g sediment (Fig. 2e,f), whereas both marker profiles correlate (R^2=0.9; p=0.015). Free acetate values (Fig. 2g) of about 2.2 mg/l were detected between 0.4 to 2.7 m, followed by an increase to 106 mg/l at 3.3 m depth. Bound acetate concentrations (Fig. 2h) correlate with TOC (R^2= 0.8; p=0.046) and are comparatively low (10.7 mg/l) at 0.4 m depth, but is significantly higher with values between 29.7 to 48.1 mg/l for the rest of the unit.

Marine Isotope Stage 3 (MIS 3):

Unit MIS 3 comprises the core segments MIS 3-1 of core L14-05, MIS 3-2 and MIS 3-3 of core L14-02 (Fig. 2). Overall, TOC and TOC/TN (Fig 2a,b) correlate (R^2=0.8; p=0.003). The core segment MIS 3-1 shows increased TOC (4.8 wt%) and TOC/TN (12.8) values, while core segment MIS 3-2 is characterized by TOC values of 1.4 to 4.7 wt% with a maximum at 1.6 m and TOC/TN values between 6.2 to 11.8. Within core segment MIS 3-3 TOC varies from 0.9 to 3.4 wt% and TOC/TN values range between 6.2 and 11.9. HI values of 316 (LP1), 322 (LP2) and 126 mg HC/ g TOC (LP3) are indicated for unit MIS 3 (Fig. 2c). Figure 2d shows PLFA concentrations of 33 µg/g sediment in core segment MIS 3-1, a decreasing trend from 35.6 to 11.1 µg/g sediment in core segment MIS 3-2, and values of 12.9 to 22.9 µg/g sediment in core segment MIS 3-

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3. In unit MIS 3 no correlation between PLFA concentrations and TOC is observable. The profile of past bacterial markers (Fig. 2e) in unit MIS 3 correlates with TOC (R^2=0.9; p=0.016), but only partly with the past archaeal profile (Fig. 2f). In MIS 3-1 both markers are strongly increased (2070.4 ng/g sediment for bacterial and 224.5 ng/g sediment for archaeal). MIS 3-2 is characterized by concentrations of 34.4 to 591.1 ng/g sediment for bacterial and of 6.3 to 37.7 ng/g sediment for archaeal markers. Much lower concentrations of 3.5 to 147.7 ng/g sediment in the bacterial profile and concentrations of 1.6 to 14.0 ng/g sediment in the archaeal profile are observed for MIS 3-3. Free acetate concentration (Fig. 2g) of 51.1 mg/l are indicated for MIS 3-1, while in MIS 3-2 the free acetate concentrations rises to 412.0 mg/l at 1.6 m. In MIS 3-3 the highest free acetate concentrations of 757.5 mg/l in 4.2 m are measured, followed by decreasing values of 13.8 to 160.0 mg/l for the rest of the core segment. The bound acetate concentrations (Fig. 2h) correlate with TOC (R^2= 0.8; p=0.049) and show a concentration of 59.2 mg/l in MIS 3-1. Both, core segments MIS 3-2 and 3-3 reveal average concentrations of 11.7 to 49.1 mg/l with a maximum of 93.7 mg/l at 1.6 m and a maximum of 86.8 mg/l at 4.2 m.

Marine Isotope Stage 4 (MIS 4):

Unit MIS 4 comprises the core segments MIS 4-1 of core L14-03 and MIS 4-2 of core L14-04. TOC and TOC/TN values (Fig. 2a,b) correlate within unit MIS 4 (R^2=0.8; p=0.009). In core segment MIS 4-1 TOC values range between 2.7 to 1.9 wt% and the TOC/TN ratio between 8.6 and 9.7 in the upper 2.5 m, below TOC values are ≤ 1.5 wt% and TOC/TN values are < 6. Core segment MIS 4-2 is characterized by a decreasing TOC trend revealing values between 0.5 and 2.4 wt% and TOC/TN values between 4.5 and 9. Four HI values were measured in MIS 4-1 with values of 388 (LP4), 226 (LP5), 80 (LP6) and 256 mg HC/ g TOC (LP7) and one HI value of 276 mg HC/ g TOC (LP8) in MIS 4-2 (Fig. 2c). The PLFA concentrations (Fig. 2d) resemble TOC (R^2= 0.7; p=0.002) with values between 19.4 to 35.5 µg/g sediment in the upper 5 m of MIS 4-1 and lower concentrations of 5.2 to 10.0 µg/g sediment below 6 m. MIS 4-2 shows low PLFA concentrations of 8.1 to 19.2 µg/g sediment. In MIS 4-1 both past microbial biomarker profiles (Fig. 2e,f) correlate (R^2= 0.8; p=0.032) and resemble the TOC profile (R^2=0.7; p=0.047). Here, the concentrations for bacterial markers range between 0.02 to 48.9 ng/g sediment with an increase from 149.1 to 208.0 ng/g sediment between 1.3 and 2.0 m, and at 4.7 m to 128.0 g/g sediment. For archaeal markers concentrations are between 0.01 to 3.9 ng/g sediment with a rise in concentrations to 42.3 g/g sediment between 1.3 and 2.0 m and at 4.7 m to 13.1 g/g sediment. In MIS 4-2 the bacterial concentrations are decreasing from 294.0 ng/g sediment to 34.2 ng/g sediment and correlate with TOC (R^2=0.9; p=0.018). The archaeal concentrations range between 7.4 and 19.1 ng/g sediment forming maxima at 5.3 and 6.3 m. The data does not correlate with the profile of past bacterial markers or with TOC. Within core segment MIS 4-1 usually free acetate concentrations (Fig. 2g) were below 100.1 mg/l. However, extreme maxima occurred at 2.5 m (193.2 mg/l), and 6.4 m depth (628.5 mg/l). In core segment MIS 4-2 the free acetate concentrations range from 31.2 to 70.0 mg/l. The bound acetate concentrations (Fig. 2h) of unit MIS 4 resemble TOC (R^2=0.7; p=0.018) and usually are characterized by values of 6.7 to 23.7 mg/l with maxima between 2.0 to 3.4 m (40.5 to 61.8 mg/l) and at 5.7 m (44.6 mg/l) in MIS 4-1, and decreasing values from 41.7 to 12.1 mg/l in MIS 4-2.

Marine Isotope Stage 5e (MIS 5e):

In unit MIS 5e (Eemian) of core L14-04, TOC and TOC/TN correlate (R^2=0.99; p=0.018) and show values of about 0.6 wt% TOC and TOC/TN values from 3.7 to 4.9 (Fig. 2a,b). Samples from MIS 5e show HI values (Fig. 2c) of 61 (E1), 81 (E2) and 78 mg HC/ g TOC (E3). The PLFA concentrations (Fig. 2d) are with values between 4.5 to 5.2 µg/g sediment quite low compared to the other intervals and resemble the TOC profile. Both past microbial markers (Fig. 2e,f) correlate (R^2=0.9; p=0.036). The concentrations of past bacterial markers vary between 21.7 to 27.1 ng/g sediment, while the concentrations of the archaeal markers range from 1.8 to 3.9 ng/g sediment. The free acetate concentration (Fig. 2g) increases from 12.5 to 89.5 mg/l with depth, while the bound acetate concentration (Fig. 2h) scatter between of 0.5 to 19.6 mg/l.

4.2 Open system-pyrolysis GC

Results provided by open system-pyrolysis experiments on 17 representative samples (high/low TOC) enable a deeper insight into the OM characteristics:

Figure 3a (after Eglinton et al., 1990) classifies the deposited OM into aliphatic-, aromatic- or sulphur-rich OM. All samples from the Holocene (H1, H2, H3, H4, H5) and Eemian (E1, E2, E3) unit and two samples from the Late Pleistocene unit (LP3, LP6) fall within the range of kerogen type III (terrestrial OM Type). Late Pleistocene samples (LP4, LP5, LP7, LP8) corresponding to higher HI values indicate a mixture of kerogen III and II (increased aliphatic character). Additionally, two Late Pleistocene sample (LP1, LP2) and the AL sample (AL), all hold the highest HI values, also indicate a mixture of kerogen type III and II but with a stronger input of type II compared to all other samples. All samples, especially the Eemian samples, show only a very low abundance of sulphur compounds generated by pyrolysis indicating sulphur lean OM (2,3-DMThio).

Figure 3b (after Horsfield et al., 1989) suggests different aliphatic characters for the selected samples indicating an increasing aliphatic character with higher HI and TOC. Samples from the Eemian unit (E1, E2, E3) and the Holocene sample H1 as well as two samples from the Late Pleistocene unit (LP3, LP6) with low HI (< 130) and low TOC (< 1) reveal the smallest aliphatic character. In comparisons, the samples from the Holocene unit (H2, H3, H4, H5) with intermediate HI (140-200) and TOC values > 1 show a slightly increased aliphatic character. All these samples are characterized by kerogen type III (Fig. 3a). Most of the samples from the Late Pleistocene unit (LP1, LP2, PL4, PL5, PL6, PL7) and the active layer (AL) sample reveal the highest aliphatic character corresponding to HI > 200 and to TOC >1 and to a mixture of kerogen type III and II (Fig. 3a).

5 Discussion

5.1 Organic matter characterization

The biodegradability of the OM in permafrost soils is generally assigned to its amount, composition and quality (Schirrmeister et al., 2011b; Strauss et al., 2015; Stapel et al., 2016), and strongly depends on the OM source and its degree of degradation (White, 2013). The permafrost deposits on Bol´shoy Lyakhovsky Island are dominated by terrestrial OM as indicated by the results of the open system-pyrolysis (Fig. 3a). Samples from the active layer and the Late Pleistocene (LP) glacial period (comprising MIS 3 and MIS 4, Table 1) reveal a terrestrial OM source mixed with aquatic OM (e.g. algae material) reflecting intervals of increased soil-moisture during deposition. Especially the interstadial deposits of MIS 3 (core sections 3-1 and 3-2) show the highest accumulation of OM as characteristic for Yedoma deposits (Schirrmeister et al., 2013). The measured TOC/TN values of 5 to 12 are within the range of terrestrial permafrost deposits reported for the NE Siberian Arctic (Wetterich et al., 2009; Schirrmeister et al., 2011a; Strauss et al., 2015). Usually, the TOC/TN ratio describes the amount of sedimentary OM that originates from aquatic vs. terrestrial sources and is commonly used to characterize the dominant origin of the OM (Meyers and Teranes, 2002). However, the TOC/TN signal can be overprinted by different processes during OM decomposition such as microbial consumption and pedogenic processes resulting in lower TOC/TN values for stronger decomposed OM (Carter and Gregorich, 2008). Furthermore, the HI is used as indicator for OM quality in terms of microbial degradability (Talbot and Livingstone, 1989; Stapel et al., 2016) since OM with higher HI is considered to contain a higher proportion of aliphatic molecular structures, while OM with low HI contain a higher proportion of aromatic structures. Thus, low TOC and TOC/TN values (< 5) together with a low HI during the Eemian (MIS 5e, Table 1) might point to an increased degree of OM decomposition and therefore to a reduced OM quality. The Eemian interglacial period represents a warm and dry climate interval in NE Siberia (Andreev et al., 2004; Wetterich et al., 2014; Wetterich et al., 2016) and was characterized by warmer summer temperatures compared to the LP glacial period (Bond et al., 2001; Shackleton et al., 2003; Kienast et al., 2008, 2011) accompanied by permafrost thawing, thermokarst formation and thermal erosion (Table 2).

The same conclusions can be drawn from the component-specific analysis of the OM by open system-pyrolysis GC (Fig. 3). Here, the Eemian deposits (E1, E2, E3) show a more pronounced kerogen type III character due to higher content of the aromatic components (as indicated by ortho-xylene content) and minor content of the aliphatic components (as indicated by n-nonen (n-C9:1) content) compared to the Holocene (MIS 1) deposits (except of sample H1; Fig. 3a). Also by a closer look on their aliphatic compositions (Fig. 3b), the differences between the Eemian and Holocene interglacial deposits are expressed by a higher aliphatic character of the Holocene deposits accompanied by higher HI values indicating presumably less decomposed OM and higher OM quality in comparison to the Eemian deposits.

The LP glacial period have slowed down decomposition of soil OM (Dutta et al., 2006) and led to the accumulation of higher amounts of organic carbon under anaerobic conditions with higher aliphatic character to the soils (Andreev et al., 2011; Schirrmeister et al., 2011a; Wetterich et al., 2014). The Yedoma samples (comprising MIS 3 and partly MIS 4) with a higher proportion of the aliphatic component n-nonen (Fig. 3a) and a stronger aliphatic character (Fig. 3b) indicate a moisture increased depositional settings with higher input of aquatic OM (e.g. algae material), especially during the MIS 3 (LP1, LP2) accompanied by increased accumulation of TOC and higher values of HI (LP1, LP2, LP4, LP5, LP7, LP8; Fig. 2). Compared to that, LP glacial period samples with lower TOC and minor HI values (LP3, LP6; Fig. 2) reveal a minor aliphatic character similar to the Holocene deposits reflecting changes in the depositional environments with probably less moist conditions.

Overall, the results from the open system-pyrolysis identify a terrestrial OM source for all investigated samples affected by environmental shifts in moisture as seen in the MIS 3 Yedoma deposits, and higher rates of decomposed OM as observed in the Eemian deposits. Additionally, higher aliphatic character and, therefore, higher quality in terms of biodegradability seems generally assigned to increased HI values (Stapel et al., 2016).

5.2 Signals of present and past microbial communities in permafrost deposits

PLFA life markers indicate the occurrence of a living bacterial community in the investigated deposits from Bol´shoy Lyakhovsky Island. While the signals are low in the permafrost deposits, all active layers contain higher amounts of PLFAs indicating a larger bacterial community in these surface layers. Although the permafrost surface layers contain not only old OM but also fresh organic material (Fontaine et al., 2007), the increase of PLFA in the different active layers with different old OM from different depositional times, might at least provide a first hint that older OM has the potential to stimulate microbial life when reaching the thawing front of the active layer. The PLFA profiles also suggest higher bacterial activity in the active layer compared to the deeper permafrost deposits.

Glycerol dialkyl glycerol tetraethers (GDGTs) and archaeol represent past microbial biomass. GDGTs are the cores of former membrane lipids which are already partly degraded as indicated by the loss of their head group moieties. However, the core lipids are very stable over geological time scales (Pease et al., 1998; Schouten et al., 2013) and can be found in many different habitats (Bischoff et al., 2013; Schouten et al., 2013). Past bacterial (branched GDGTs (Weijers et al., 2006)) and archaeal (isoprenoid-GDGTs (Koga et al., 1993; Pancost et al., 2001)) markers provide information on the abundance and most likely also on the activity of microbial communities in the geological past. Periods of increased OM accumulation and increased past microbial activity can be observed in the Yedoma deposits (MIS 3 and 4). Especially, the Yedoma core sections MIS 3-1, 3-2 and the upper part of 4-1 and 4-2 are characterized by increased OM accumulation with increased aliphatic character (HI values; LP1, LP2, LP4 and LP8) and increase bacterial and archaeal GDGT concentrations (Fig. 2e,f). These data reflect a depositional environment of increased soil moisture and warmer temperature resulting in excellent living conditions for anaerobic bacteria (Weijers et al., 2006) and archaea (Wagner et al., 2007) during parts of MIS 3 and 4. According to Wetterich et al. (2014), the MIS 3 interstadial optimum occurred between 48 to 38 ka BP on Bol´shoy Lyakhovsky Island and is characterized by warmer climatic conditions compared to LP stadial periods (Andreev et al., 2011).

5.3 Microbial substrate potential for greenhouse gas generation

The potential for providing substrates for microbial induced greenhouse gas generation depends on the amount and quality of the OM (Stapel et al., 2016). To assess the potential for future substrate provision, acetate as an excellent substrate for microbial turnover is used (Ivarson and Stevenson, 1964; Sørensen and Paul, 1971; Sansone and Martens, 1981; Balba and

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Nedwell, 1982). Acetate is the terminal electron acceptor for methanogens in cold-temperate environments (Chin and Conrad, 1995; Wagner and Pfeiffer, 1997), especially for acetoclastic methanogens (Thauer, 1998) and methanogenic archaea which are ubiquitous in anoxic environments and in permafrost sediments (Kobabe et al., 2004).

In this study two acetate pools are investigated: The free-acetate pool within the pore water representing an easily accessible substrate source for microbial metabolism, and the bound-acetate fraction still linked to the OM forming a future substrate source (Glombitza et al., 2009b). Overall, the concentrations of the bound acetate (Fig. 2h) correlate well with the amount and quality of OM indicating larger reservoir pools for future microbial turnover in depths with increased TOC and HI (Fig. 2, Table 2). Therefore, deposits from the MIS 1, 3 and 4 possess increased bound acetate mean values indicating the largest future substrate reservoir within MIS 3 (~ 48.9 mg/l), followed by MIS 4 (~ 33.26 mg/l) and MIS 1 (~30.05 mg/l) (Table 2). In contrast, the bound-acetate concentration in the Eemian (MIS 5e) interglacial deposits suggests a depleted and maybe already altered bound-substrate pool since the concentration is considerably lower (~ 9.98 mg/l).

The concentration of free acetate in permafrost pore waters is not only the result of acetate released from the OM but is also influenced by lateral and vertical diffusion supported by capillary pressure (Parlange, 1971) as well as thawing and freezing processes and microbial generation and consumption. Overall, the availability of substrates is influenced by a variety of factors, e.g. soil biogeochemistry composition, grain and molecular size, and environmental factors such as temperature and nutrient concentrations (Deshpande et al., 2016). The very low concentrations of free acetate in all active layers is due to higher microbial acetate consumption by an active microbial community indicated by the high PLFA life marker signal. All active layer samples reveal increased PLFA concentrations compared to the underlying permafrost deposits indicating good living conditions for microorganisms, mainly due to the fresh OM input during the thawing period (Fontaine et al., 2007; Liebner et al., 2008). Additionally, old freeze-look permafrost carbon, currently incorporated into the active layers from the underlying permafrost, is particularly sensitive to temperature-induced microbial decomposition (Knorr et al., 2005; Davidson and Janssens, 2006) and, therefore, is considered as an important substrate source. The highest PLFA concentration is detected in the active layer on top of the MIS 3 deposits. This might reflect a high potential of the MIS 3 Yedoma deposits to serve as substrate provider for a living microbial community when reaching the active layer. In contrast, local environmental differences also effect on the PLFA concentration. In this case, the core containing MIS 3 deposits was drilled in a stable tundra environment, while the other cores were either drilled in an instable thermos terrace influenced by continuously sediment and water input or on the Holocene thermokarst basin characterized by a moisture-reduced environment with shallower active layers (Schwamborn and Wetterich, 2015). However, as already mentioned above the increased PLFA concentrations in all active layers indicate that to a certain extent the permafrost deposits from MIS 3, 4 and 1 serve as good substrate provider when thawed.

In the permafrost deposits similarities between the concentration of the free-substrate pool and the TOC content is more defined within the deposits from the LP glacial period (comparing MIS 3 and 4). Here, the mean concentration (~ 93.6 and 82.1 mg/l) of free acetate is at least two to three times higher than in the interglacial periods MIS 1 (24.1 mg/l) or MIS 5e (46.1 mg/l). The minor free-acetate pool in the Holocene deposits might either be a result of OM composition deposited during that period or as also proposed for the modern active layer the result of intense microbial consumption during time of deposition. The latter could be favoured by stronger pronounced active layers with increased active microbial acetate consumption (Xue et al., 2016) due to the onset of the early Holocene which caused warmer but unstable environmental conditions especially during the Holocene Optimum (10.3 to 9.3 ka BP) and the late Holocene (Andreev et al., 2004; Wagner et al., 2007; Wetterich et al., 2008). The same might be the reasons for the low free-acetate concentration in the Eemian deposits; whereas in addition to microbial consumption the warmer environmental conditions during the Eemian interstadial have probably also altered the concentrations of free substrates in the sediments due to lateral transportation of water and/or sediment as a result of permafrost thaw. Thus, based on the results obtained in this study it can be noted that the interglacial periods contained reduced amounts of free-acetate in comparison to the LP glacial period investigated.

The alteration of the free and bound acetate reservoir is implied by either local microbial consumption or by lateral and vertical transportation probably also resulting in microbial acetate consumption on a regional scale. The MIS 1 and MIS 5e deposits, however, possess a similar depleted free-acetate pool, whereas, the bound-acetate concentration in the MIS 1 implies a considerable future substrate reservoir compared to the MIS 5e deposits (Table 2). On the other hand, the investigated interstadial and glacial period deposits provide both acetate-substrates pools in higher concentrations (Table 2). Here, especially the deposits from the interstadial Yedoma period (MIS 3) possess a larger substrate reservoir linked to moister depositional conditions and increased amount and quality of the OM as already been observed in a study of Buor Khaya Peninsula permafrost, 350 km SW from Bol'shoy Lyakhovsky Island (Stapel et al., 2016). Considering the thickness of the LP Yedoma deposit of 20 m on Bol'shoy Lyakhovsky Island (Schennen et al., 2016) and 10-60 m across Russia (Dutta et al., 2006), and a Yedoma extension of about 106 km² (Romanovsky, 1993) a significant substrate pool for future microbial greenhouse gas generation within permafrost thawing is assessable. On a broader perspective, an ongoing Arctic warming will increase active layer thickness and thus favour substrate availability from deeper and older OM for microbial decomposition which enhances the production and release of greenhouse gases (Schuur et al., 2008). Although the complexity of potential positive (e.g. Schuur et al. (2008)) and negative (e.g. Flanagan and Syed (2011)) feedback loops between climate warming and the carbon cycle in the Arctic leads to considerable uncertainties in how far the substrate potential in permafrost deposits plays a key role for future greenhouse gas production concerning shifts in the microbial community composition, vegetation, hydrogeology and soil thermal regime. This study shows that OM especially deposited during the interstadial and glacial periods appear to contain a larger substrate potential than the interglacial deposits.

6 Conclusions

The quality of OM in terms of providing organic substrates for microbial induced greenhouse gas production varies within the investigated permafrost deposits from Eemian to present time and is controlled by depositional, environmental and climatic conditions. The strongest present and future substrate potential appear to be stored within the Yedoma deposits from the last interstadial (MIS 3) and stadial (MIS 4) period. Thus, this material might have strong impact on the greenhouse-gas driven climate-feedback cycle, when reaching the thaw front of the active layer. In contrast, the interglacial periods (Holocene and especially Eemian) show lower substrate potentials. The Eemian deposits reveal both low present and future substrate pools. However, the Holocene deposits at least contain a significant future-substrate pool, which might become available when recycled in the active layer.

Data availability

https://www.pangaea.de/ (follows after acceptance and includes all shown datasets)

Author contributions

J. G. Stapel performed the cores sub-sampling, the laboratory analyses and data interpretations guide by K. Mangelsdorf and B. Horsfield. G. Schwarmborn and L. Schirrmeister planned and coordinated the field work, collected the cores and opened the cores. J. G. Stapel wrote the manuscript that all co-authors commented on.

Competing interests

The authors declare that they have no conflict of interest.

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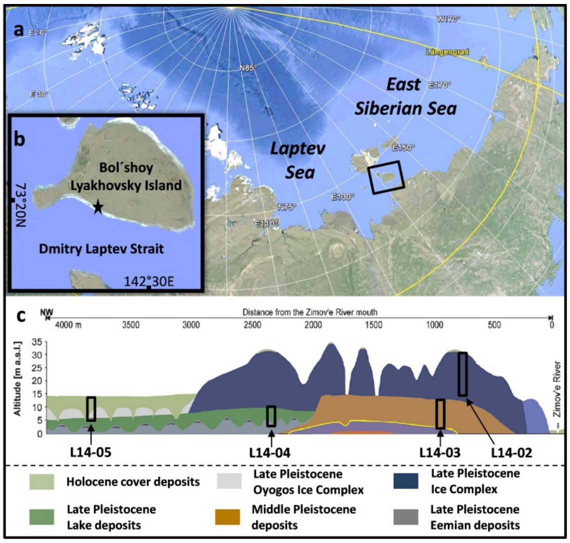

Figure 1: (a) Position of Bol´shoy Lyakhovsky Island in the Siberian Arctic. (b) Study site on Bol´shoy Lyakhovsky Island, indicated by a black star, (c) and location of the drilled cores comprising different age intervals (L14-05, L14-02, L14-03 and L14-04) modified after Wetterich et al. (2014) and Schwamborn and Wetterich (2015).

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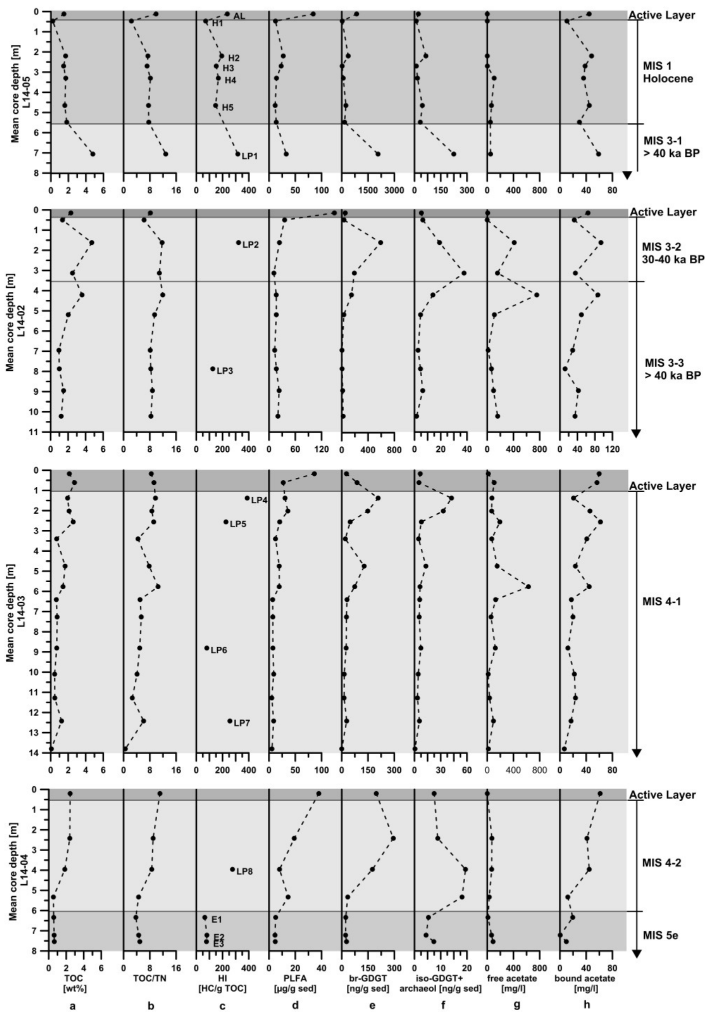

Figure 2: Bio- and geochemical parameters of permafrost cores L14-05, L14-02, L14-03 and L14-04 from Bol´shoy Lyakhovsky Island, northern Siberia, presented with respect to core depth (left axis) and stratigraphic and age units (right column). The vertical profiles show (note partly different axis): a) the total organic carbon (TOC) content in wt%, b) the ratio of total organic carbon and total nitrogen (TOC/TN), c) the hydrogen index (HI) in mg HC/ g TOC, d) concentration of phospholipid fatty acids (PLFAs) in µg/g sediment, e) the concentration of branched glycerol dialkyl glycerol tetraethers (br-GDGTs) in ng/g sediment, f) the concentration of isoprenoid glycerol dialkyl glycerol tetraethers (iso-GDGTs) + archaeol in ng/g sediment, g) the concentration of free acetate in mg/l, and h) concentration of bound acetate in mg/l. Active layer samples are dyed in dark grey, interglacial periods (MIS 1 and MIS 5e) are dyed in grey and the last glacial period (MIS 3 and MIS 4) is dyed in light grey. According to age, stratigraphy and core segments, the MIS 3 unit is subdivided into the core segments MIS 3-1, 3-2 and 3-3, and the MIS 4 unit is subdivided into the core segment MIS 4-1 and 4-2. Sample labels within the HI profile correspond to core samples of different ages (H: Holocene; LP: Late Pleistocene glacial period; E: Eemian).

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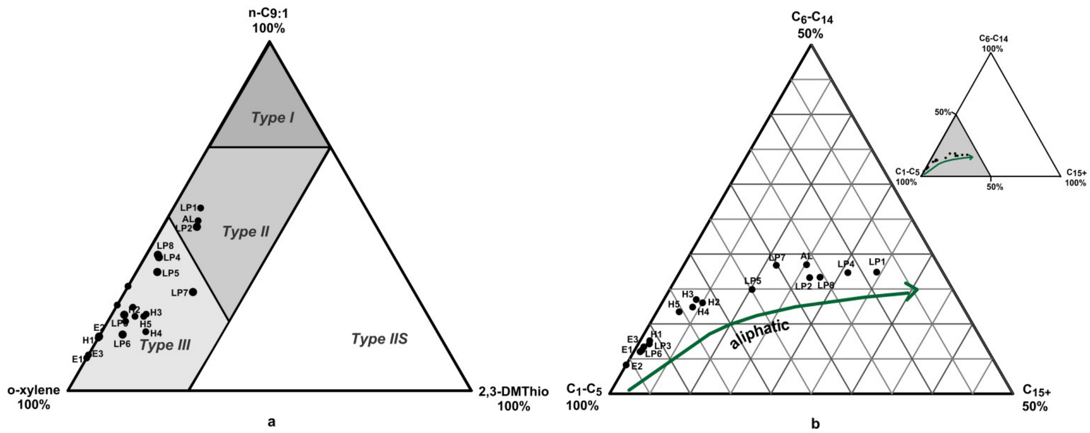

Figure 3: Triangular plots derived from organic matter pyrolysis. (a) Eglington-diagram: Classification of the kerogen type (type I/II: aquatic and marine; type III: terrestrial; type IIS: enriched sulphur content) due to the relative abundance of 1,2 dimethylbenzene (ortho-xylene), n-nonen (n-C9:1) and 2,3-dimethylthiophen (2,3DMThio) in the OM after Eglinton et al. (1990). (b) Horsfield-diagram: Composition of the OM according to the chain length distribution of short (C1-C5), intermediate (C6-C14) and long (C15+) n-alk-1-enes after Horsfield et al. (1989). The arrow indicates an increasing aliphatic proportion in the OM of the investigated samples. Sample labels correspond to core samples of different ages (H: Holocene; LP: Late Pleistocene glacial period; E: Eemian) with different total organic carbon (TOC) and hydrogen index (HI) values (Fig. 2a,c).

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Table 1: Schematic summary of core materials investigated including age periods, marine isotope stage (MIS) after Andreev et al. (2004) and (2009), Wetterich et al. (2004) and (2014), core numbers and coordinates of drill sites.

Age		Cores	Drilling site
Epoch	MIS		
Holocene (interglacial)	1	L14-05	73.34994° N 141.24156° E
Late Pleistocene Glacial Period (glacial) – interstadial	3	L14-02	73.33623° N 141.32761° E
Late Pleistocene Glacial Period (glacial) – stadial	4	L14-03	73.33464° N 141.32822° E
Eemian (interglacial)	5e	L14-04	73.34100° N 141.28587° E

Table 2: Schematic summary (from left to right) including age (epoch and marine isotope stage (MIS) classification) after Andreev et al. (2004) and (2009), Wetterich et al. (2004) and (2014), paleo-environment (1Schirrmeister et al. (2002), 2Andreev et al. (2009), 3Grosse et al. (2007), 4Sher et al. (2005), 5Wetterich et al. (2014)), organic matter (OM) quality, substrate potential (present (free acetate) and future (bound acetate)) and core numbers (related to the age classification). To visualize the OM quality and the substrate potential a relative scaling is used: very good (++), good (+), poor (-) and very poor (--).

Age			Palaeoenvironment	OM quality	Substrate potential		Cores
Epoch		MIS			present	future	
Holocene (interglacial)		1	- climatic warming[1] - moisture increased thawing, thermokarst[2] - unstable environmental conditions[2] - dissected landscape influences by local hydrology[3]	-	- -	+	L14-05
Late Pleistocene Glacial Period (glacial)	interstadial	3	- increased temperature and soil moisture[4] - warm/moderate and dry climate[5] -optimum: warm and dry (48 to 38 ka BP)[5] - warmer summers, open vegetation[2]	+ +	-	+ +	
				+ +	+ +	+ +	L14-02
				-	+	+	
	stadial	4	- cold and dry climate[5] - harsh climate conditions[2] - thin snow cover, low precipitation[2]	+	+	+	L14-03
				-			
				+			L14-04
Eemian (interglacial)		5e	- warmer climate, open-grass tundra similar to modern[2] - permafrost thawing[2] - optimum: 4-5 °C higher summer temperatures than modern, shrub tundra[2]	- -	-	- -	