Soil microbial biomass, activity and community composition along altitudinal gradients in the High Arctic (Billefjorden, Svalbard)

Petr Kotas[1,2], Hana Šantrůčková[1], Josef Elster[3,4], Eva Kaštovská[1]

[1]Department of Ecosystem Biology, Faculty of Science, University of South Bohemia, České Budějovice, 370 05, Czech Republic

[2]Institute of Chemistry and Biochemistry, Faculty of Science, University of South Bohemia, České Budějovice, 370 05, Czech Republic

[3]Centre for Polar Ecology, Faculty of Science, University of South Bohemia, České Budějovice, 370 05, Czech Republic

[4]Centre for Phycology, Institute of Botany, Academy of Sciences of the Czech Republic, Třeboň, 379 82, Czech Republic

Correspondence to: Petr Kotas (kotyno@prf.jcu.cz)

Abstract The unique and fragile High Arctic ecosystems are vulnerable to proceeding global climate warming. Elucidation of factors driving microbial distribution and activity in Arctic soils is essential for comprehensive understanding of the ecosystem functioning and its response to environmental change. The goals of this study were to investigate the microbial biomass, activity, microbial community structure (MCS) and their environmental controls in soils along three elevational transects in coastal mountains of Billefjorden, Central Svalbard. Soils from four different altitudes (25, 275, 525, and 765 m above sea level) were analysed for a suite of characteristics including temperature regimes, organic matter content, base cation availability, moisture, pH, basal respiration, and microbial biomass and community structure using phospholipid fatty acids (PLFA). We observed significant spatial heterogeneity of edaphic properties among transects, resulting in transect-specific effect of altitude on most soil parameters. We did not observed any clear elevation pattern in the microbial biomass and the microbial activity revealed contrasting elevational patterns between transects. We found relatively large horizontal variability in MCS, mainly due to different composition of bacterial PLFAs, but also systematic altitudinal shift in MCS related with different habitat preferences of fungi and bacteria, resulting in high fungi to bacteria ratios at the most elevated sites. Our data further showed that the biological soil crusts on these most elevated, unvegetated sites can host microbial assemblages of the size and activity comparable with the arctic tundra ecosystem. The key environmental factors determining horizontal and vertical changes in soil microbial properties were soil pH, organic carbon content, soil moisture and Mg^{2+} availability.

1 Introduction

Knowledge about the spatial distribution and activity patterns of soil microbial communities is essential to understand ecosystem functioning as the soil microbes play fundamental role in biogeochemical cycling and drive productivity in terrestrial ecosystems (van de Heijden et al., 2008). The soil microbial diversity in the Arctic is comparable to that in other biomes (Chu et al., 2010) and the spatiotemporal variability in microbial community composition is large (Lipson, 2007; Blaud et al., 2015; Ferrari et al., 2016). However, it is still uncertain which environmental factors drive the heterogeneity of soil microbial properties in the Arctic.

Altitudinal transects offer great opportunity to study a distribution of microbial communities adapted to local habitats and explain the patterns by natural gradients of soil conditions, vegetation occurrence and climate regimes over short spatial distances (Ma et al., 2004; Körner et al., 2007). The proceeding climate change will further affect environmental conditions in the Arctic (Collins et al., 2013) including expected upward migration of the vegetation and increasing plant cover (Vuorinen et al., 2017; Yu et al. 2017). Therefore, the knowledge of current microbial distribution and activity patterns along the altitudinal gradients together with identifying their controlling factors can help to predict future development of ecosystems in this region. However, such studies are scarce despite the fact that Arctic tundra comprises 5% of the land on Earth (Nemergut et al., 2005) and most coastal areas in the northern circumpolar region have mountainous character. So far, only few studies assessing altitudinal trends in soil microbial properties were conducted in the Scandinavian Arctic (Löffler et al., 2008; Männistö et al., 2007). The research on spatial variation in microbial community composition and activity in polar regions was conducted mainly at narrow elevation range (Oberbauer et al., 2007; Trevors et al., 2007; Björk et al., 2008; Chu et al., 2010; Van Horn et al., 2013; Blaud et al., 2015; Tytgat et al., 2016) or was focused on initial soil development following glacier retreat (Bekku et al., 2004; Yoshitake et al., 2007; Schűtte et al., 2010). Majority of studies on the elevational patterns in microbial community structure (MCS) and activity has been done in mountain regions of lower latitudes from tropics to temperate zone. The studies commonly show that the microbial activity decreases with increasing elevation (Schinner, 1982; Niklińska and Klimek, 2007; Margesin et al., 2009), while there are no general altitudinal patterns in soil microbial diversity and community structure. For example, the microbial community composition did not change along elevational gradients in Swiss Alps (Lazzaro et al., 2015), while other studies have documented decreasing bacterial (Ma et al., 2004; Lipson, 2007; Shen et al., 2013) and fungal (Schinner and Gstraunthaler, 1981) diversity with an increasing altitude, and several studies reported the mid–altitudinal peak in microbial diversity (Fierer et al., 2011; Singh et al., 2012; Meng et al., 2013). Beside the fungal and bacterial diversity, the relative abundance of these main microbial functional groups is also variable. For example, Djukic et al. (2010), Xu et al. (2014) and Hu et al. (2016) found decreasing fungi to bacteria (F/B) ratio with an increasing elevation, while Margesin et al. (2009) reported opposite trend in Central Alps.

The research focusing on environmental controls over microbial communities in polar and alpine regions recognized many significant factors, including vegetation, litter C : N stoichiometry, organic carbon content, soil pH, nutrient availability, microclimatic conditions, and bedrock chemistry. However, the effect of these variables was site- and scale-specific (Van Horn et al., 2013; Blaud et al., 2015; Ferrari et al., 2016), which highlights the need for further research on environmental controls of microbial community size, activity and structure at local and regional scales. To extend our knowledge about microbial ecology and soil functioning in the arctic alpine ecosystems, we conducted study aiming to assess the activity, biomass and structure of soil microbial communities and to determine their controlling environmental factors along three altitudinal transects located in Central Svalbard. These transects spanned from the vegetated tundra habitats at the narrow areas at the sea level to unvegetated soils at the top of the coastal mountains. The specific objectives of our study were (i) to describe gradients of microclimatic and geochemical soil properties; (ii) to assess microbial activity (soil respiration) and abundance of main microbial groups (fungi, Gram–negative and Gram–positive bacteria, Actinobacteria, phototrophic microorganisms) using phospholipid fatty acid (PLFA) analysis; and (iii) to identify environmental factors explaining the trends in soil microbial parameters along these altitudinal gradients.

2 Materials and methods

2.1 Study area and soil sampling

The Petunia bay (Billefjorden; 78° 40´ N, 16° 35´ E) is located in the center of Svalbard archipelago and represents typical High Arctic ecosystem in the northern circumpolar region. The mean, minimum and maximum air temperatures recorded in the area at 25 m above the sea level (a.s.l.) were –3.7, –28.3 and 17 °C in the period of 2013–2015, respectively, and stayed permanently below 0 °C for eight months a year (Ambrožová and Láska, 2017). The mean annual precipitation in the Central Svalbard area is only 191 mm (Svalbard Airport, Longyearbyen, 1981–2010) and is equally distributed throughout the year (Førland et al., 2010).

In August 2012, we collected soils from three altitudinal transects (Tr1–3) on the east coast of Petunia bay. Each transect was characterized by four sampling sites at altitudes 25, 275, 525 and 765 m a.s.l. (± 5 m). Transects were located on slopes with similar exposition (Tr1 W–E, Tr2 WNW–ESE, Tr3 WSW–ENE; Fig. 1) and lithostratigraphy. Soils at the lowest elevations developed from Holocene slope (Tr1 and Tr3) and marine shore deposits (Tr2), while the bedrock at more elevated sites is formed by dolomite and limestone with units of basal calcareous sandstone (Dallmann et al., 2004). The soils were classified as Leptic Cryosols (Jones et al., 2010) with loamy texture and clay content increasing with altitude (Table 2), and were from 0.15–0.2 m to only few cm deep at 25 and 765 m a.s.l., respectively. The poorly developed organic horizon was present only at the lowest elevation. The sampling locations were selected in geomorphologically stable areas with a similar slope (20±5°). On each sampling site, nine soil cores (4 cm deep, 5.6 cm diameter) were collected and mixed into three representative samples. Each representative sample was mixed from one soil core taken from the edge of the vegetation tussocks (if vegetation was present) and two other cores taken in increasing distance from the vegetation to maintain the consistency with respect to heterogeneity of vegetation cover and soil surface. The triplicates were collected approximately 5 m apart from each other. Immediately after sampling, the soil was sieved (2 mm) to remove larger rocks and roots, sealed in plastic bags and kept frozen at –20 °C till further processing. Soil subsamples for biomarker analysis were as soon as possible freeze–dried and stored at –80 °C until extraction.

Transects represented climosequences from high Arctic tundra to unvegetated bare soil. Vegetation of two lowest sites was dominated by *Dryas octopetala*, with significant contribution of *Saxifraga oppositifolia,* and variable contribution of *Cassiope tetragona, Salix polaris* and grasses (*Carex nardina, C. rupestris, C. misandra*; Prach et al., 2012; personal observations). The vascular plants species formed scattered vegetation patches at the altitude of 525 m a.s.l. with *Salix polaris* and *Saxifraga oppositifolia* being the most abundant species. The soils at the most elevated sites were covered mainly by soil crusts with scarcely occurring *Saxifraga oppositifolia* and *Papaver dahlianum* (personal observations). The percentage cover of main surface types (i.e. stones, bare soil, vegetation, crusts and mosses) was estimated on each sampling site from approximately $1 m^2$ area in a close vicinity of coring sites (Table S1, Fig. S6).

2.2 Monitoring of microclimatic characteristics

To describe the soil microclimatic conditions along the altitudinal transects, we continuously measured soil temperature at –5 cm from 2012–2013 directly at the sampling sites of Tr1 using dataloggers (Minikin Ti Slim, EMS Brno, CZ). The soil water content at the time of sampling was determined in soil subsamples by drying to constant weight at 105 °C. The temperature regimes at particular altitudinal levels were characterized by 10 climatic variables (Table 1). The period of above–zero daily mean ground temperatures is referred to as summer season throughout the text. We also considered number of days with daily mean ground temperatures above 5 °C, characterizing a period with conditions suitable for vascular plant growth (Kleidon and Mooney, 2000). The positive soil surface energy balance was calculated as a sum of daily mean summer temperatures. The records from three years (2011–2013) continuous measurements at two automated weather stations located at 25 and 455 m a.s.l. approximately 3 km apart from the observed transects (hereafter referred as AWS_{25}

and AWS_{455}, respectively; Fig. 1; see Ambrožová and Láska, 2017 for detailed description) were used to evaluate seasonal variation of soil temperature and moisture regimes (Figs. S2, S3, respectively), and coupling of soil and atmospheric temperatures (measured at -5 cm and 2 m above terrain, respectively; Fig. S2). Even though we were not able to continuously measure soil moisture directly at the sampling sites, we regarded data from both AWS locations as representative for the evaluation of seasonal moisture regimes.

2.3 Soil characteristics

The particle size distribution was assessed using aerometric method (Lovelland and Whalley, 2001), the soil type was classified according to U.S. Department of Agriculture. The soil pH was determined in soil–water mixture (1:5, w/v) using glass electrode. The cation exchange capacity (CEC) was considered to be equal to the sum of soil exchangeable base cations Mg^{2+}, Ca^{2+}, Na^{+}, K^{+} extracted with 1M NH_4Cl (Richter et al., 1992). The amount of H^{+} and Al^{3+} ions was neglected due to the high soil pH. Base cations accessible for plant and microbial uptake (Mg^{2+}, Ca^{2+}, Na^{+}, K^{+}) were extracted by the Mehlich 3 reagent (Zbíral and Němec, 2000). Cations were measured by atomic absorption spectroscopy (AA240FS instrument, Agilent Technologies, USA). Total soil organic carbon (TOC) and nitrogen (TN) contents were measured in HCl fumigated samples (Harris et al., 2001) using elemental analyser (vario MICRO cube, Elementar, Germany).

2.4 Microbial respiration

Since we were not able to measure soil respiration on site or immediately after soil collection, we measured the potential respiratory activity (soil CO_2 production) in the laboratory incubation experiment. We stored and transported the soils frozen because it was previously demonstrated that freezing-thawing has a weaker effect on microbial activity than long-term refrigeration (Stenberg et al., 1998) and comparable effect as drying-rewetting (Clein and Schimel, 1994). We then measured microbial respiration in slowly melted field-moist soils twice during the adaptation period (day 4 and 12), which allowed stabilization of the microbial activity after a respiratory flush following freeze-thaw events (Schimel and Clein, 1996), and at day 13, when we expected a stabilized microbial activity. Briefly, soil subsamples (10 g) were incubated in 100 mL flasks at 6 °C, which corresponds to the mean summer soil temperature of all sites along Tr1. At days 4, 12 and 13, a cumulative CO_2 production from the soils was measured using Agilent 6850 GC system (Agilent technologies, CA, USA). The flasks were then thoroughly ventilated and sealed again. Due to high soil pH, the total amount of produced CO_2 was corrected for its dissolution and dissociation in soil solution according to Henderson-Hasselbach equation (Sparling and West, 1990) and expressed as the microbial respiration rate per day. The daily microbial respiration rates measured between days 4-12 and after stabilization (day 13) were not significantly different in any soil samples, therefore, we present only the later one.

2.5 Microbial biomass and community structure

The soil microbial community structure was defined using PLFA analysis according to modified protocol of Frostegård et al. (1993). Briefly, 1-3 g (according to TOC content) of freeze–dried soil samples was extracted twice with a single–phase extraction mixture consisting of chloroform, methanol and citrate buffer. After overnight phase separation achieved by adding more chloroform and buffer, the organic phase was purified on silica columns (SPE–SI Supelclean 250mg/3 mL; Supelco®, PA, USA) using chloroform, acetone and methanol. The polar fraction was trans–esterified to the fatty acid methyl esters (FAME) (Bossio and Scow, 1998). All FAMEs were quantified by an internal standard calibration procedure using methyl-nonadecanoate (19:0) as an internal standard. To identify the FAMEs, retention times and mass spectra were compared with those obtained from standards (Bacterial Acid Methyl Esters standard, the 37–component FAME Mix,

PUFA–2, and PUFA–3; Supelco, USA). The ISQ mass spectrometer (MS) equipped with Focus gas chromatograph (GC) (Thermo Fisher Scientific, USA) was used for chromatographic separation and detection.

Only specific PLFAs were used to assess the microbial community structure: a14:0, i15:0, a15:0, i16:0, i17:0, a17:0 were used as markers of Gram–positive bacteria (G+); 16:1ω9, 16:1ω5, cy17:0, 18:1ω11, 18:1ω7, cy19:0 as markers of Gram–negative bacteria (G–); 10Me16:0 and 10Me18:0 as markers of Actinobacteria (Kroppenstedt, 1985), 18:1ω9, 18:2ω6,9 as fungal markers (Frostegård and Bååth, 1996) and polyunsaturated fatty acids 18:4ω3, 20:5ω3 were used as markers of phototrophic microorganisms (Hardison et al., 2013; Khotimchenko et al., 2002). A sum of Actinobacterial markers, PLFAs specific to G+ and G– bacteria and general bacterial markers 15:0, 17:0 and 18:1ω5 was used to calculate bacterial biomass and fungi to bacteria (F/B) ratio. The sum of all lipid markers mentioned above and nonspecific PLFAs 14:0, 16:0, 18:0 and 16:1ω7 was used as proxy for microbial biomass ($PLFA_{tot}$).

2.6 Sterol analyses

The β–sitosterol and brassicasterol were used as biomarkers of plant (Sinsabaugh et al., 1997) and microalgal (Volkman, 1986; 2003) residues in organic matter (OM), respectively. Sterols were simultaneously determined using microwave assisted extraction adapted from Montgomery et al. (2000) and GC/MS (ISQ MS equipped with Focus GC, Thermo Fisher Scientific, USA) analysis. Briefly, 0.5 g of freeze–dried soil was treated with 6 mL of methanol and 2 mL of 2 M NaOH. Vials were heated twice at the centre of a microwave oven (2450 MHz and 540 W output) for 25 s. After cooling, the contents were neutralized with 1 M HCl, treated with 3 mL of methanol and extracted with hexane (3×4 mL). Extracts were spiked by an internal standard (cholesterol), evaporated and derivatized by adding of pyridine and 1 % BSTFA at 60 °C for 30 min prior analysis. Sterols were quantified by an internal standard calibration procedure.

2.7 Statistical analyses

All data were checked for normality and homoscedasticity, and log–transformed if necessary. The relative PLFA data (mol%) were log-transformed in all statistical tests. The significance of environmental gradients and corresponding shifts in MCS (mol% of summed PLFA specific for fungi, G– and G+ bacteria, Actinobacteria and soil phototrophic microorganisms) in horizontal direction (ie. effect of transect) and vertical direction (ie. effect of altitude) were tested using the partial redundancy analyses (RDA) with covariates. Variation partitioning was subsequently performed to quantify the unique and shared effects of transect and altitude on variability of MCS. Forward selection procedure was used to identify the soil geochemical parameters best explaining the shifts in MCS. During the forward selection procedure, only *P* values adjusted by Holms correction were considered. This procedure is slightly less conservative compared to the often recommended Bonferroni correction, but it is a sequential procedure and takes into account that the candidate predictors with stronger effect were selected first (Holm, 1979). The multivariate tests were performed without standardization by samples, but with centering and standardization by variables (because the variables were not always measured at the same scale, see Šmilauer and Lepš 2014) and Monte Carlo test with 1999 permutations. Only adjusted explained variation is referred throughout the text. Since the samples from each triplicate cannot be considered as independent observations due to relatively low inter-sample distance (otherwise we had 9 independent transects), only the sampling sites were freely permuted while the individual samples were exchangeable only within the sampling sites. The differences in particular soil and microbial parameters between respective transects and altitudes were addressed by ANOVA complemented with Tukey–HSD post hoc test. To find out how tightly were variables related to each other, Pearson correlation coefficient was used. All statistical tests were considered significant at $P < 0.05$. Multivariate statistical analyses were performed with CANOCO for Windows

version 5.0 (Ter Braak and Šmilauer 2012), for ANOVA, Tukey-HSD test and correlations between soil and/or microbial parameters, Statistica 13 was used (StatSoft, USA).

3 Results

3.1 Altitudinal changes in soil microclimate

The soil microclimate at the studied sites was characterized by two distinct periods respecting the air temperature dynamics (compare Fig. S2a with S2b). The winter period lasted typically from the middle of September to early June. The winter soil temperatures were stratified according to the elevation and the temperature means decreased from –4 °C at 25 m a.s.l. to –10 °C at 765 m a.s.l. (Table 1, Fig. S2). In contrast, a short summer period was characterized by a significant diurnal fluctuation of soil temperatures and weak altitudinal temperature stratification (Fig. S2). The length of the summer season more than doubled at the lowest elevations compared to the most elevated study sites, while the period with daily mean soil temperatures above 5 °C shortened almost four times. Correspondingly, the positive surface energy balance gradually decreased with an increasing altitude (Table 1). The maximum daily mean temperatures and diurnal temperature fluctuation were highest at the mid–elevated sites, with the highest mean summer soil temperature reached at 275 m a.s.l. In contrast, the least and most elevated sites experienced lower summer maximum daily means and soil temperature amplitudes (Table 1). The effect of altitude on soil moisture was significant along Tr1 and Tr3 ($P < 0.001$ and 0.01, $F = 22.76$ and 7.39, respectively) with soil moisture content decreasing along with increasing elevation, but nonsignificant along Tr2. Continual volumetric measurements of soil water content at AWS_{25} and AWS_{455} showed that the soil moisture was relatively stable during the summer season and desiccation events did not occur during the summer periods 2011–2013 (for more information, see Fig. S3).

3.2 Gradients of soil geochemical properties and surface vegetation cover

Both factors, transect and altitude, significantly affected soil geochemical properties (partial RDA, pseudo–$F = 8.3$, $P < 0.001$) and explained 61% of the total variation in soil characteristics. The RDA ascribed most of the explained variability (73%) to vertical zonation. Accordingly, the effect of altitude was significantly reflected in all soil parameters (Table 2, 3, Fig. S4), but the significant interactive effect between transect and altitude indicated that the elevational trends were in most cases specific for particular transects (Tables 2, 3). Especially the CEC and availabilities of Ca^{2+}, Mg^{2+}, K^{+} and Na^{+} were spatially variable, reflecting complicated geology of the Petunia bay area. The soils along Tr1 were significantly richer in available Mg^{2+} and K^{+} than soils from other two transects (Table 2). The Mg^{2+} availability also significantly increased with increasing elevation along the Tr1 (Table 2). Other soil properties showed more systematic altitudinal patterns. The mean soil pH ranged from 7.8 to 9.0 and increased with altitude along all transects (Table 2, Fig. S4). Oppositely, the soil TOC and TN contents declined towards higher elevations along all transects; the exception was the lowest site along Tr2 with lower soil OM content compared to the respective sites from Tr1 and Tr3. The OM poorest soil occurred at the highest site of Tr1 (Table 3). The soil C/N ratio, sitosterol content in TOC and the ratio between plant–derived sitosterol and brassicasterol of algal origin were solely affected by the altitude. Their values systematically decreased with an increasing elevation irrespective of the soil OM content (Table 3), indicating an altitudinal shift in the OM quality and origin. The percentage of plant cover also continuously decreased with an increasing elevation along Tr1 and Tr3 but was comparable on the three lower sites along Tr2 (Fig. S5), which significantly resembled the trends in soil OM content ($r = 0.53$; $P = 0.001$). The lichenized soil crusts were predominant type of soil surface cover at all sites, while mosses covered very small proportion of

surface area. The bare surface without any vegetation (bare soil) occurred only at the two most elevated sites (Fig. S5, Table S1).

3.3 Soil microbial biomass and activity

The soil PLFA content, used here as a measure of soil microbial biomass, was significantly correlated with soil TOC and TN contents (r = 0.773 and 0.719, respectively; both $P < 0.0001$) and soil moisture (r = 0.772; $P < 0.0001$), and negatively affected by Mg^{2+} availability (r = -0.775; $P < 0.0001$). Despite these relations, the soil PLFA content did not show any altitudinal pattern. The soil PLFA amounts were comparable among differently elevated sites along particular transect (Fig. 2a). Only the most elevated site of Tr1 had significantly lower soil PLFA content than other sites, which corresponded with its very low stock of OM (Table 3). Similarly, neither the flush of microbial respiration measured after soil thawing (day 4 of incubation) nor the respiration measured after stabilization (day 12, not shown, and day13) showed any systematic altitudinal pattern (Fig. 3b, c). Generally, the flush respiration rate was closely related (r = 0.74, P < 0.0001, n = 36) to microbial respiration after stabilization and ca 2.3 ± 0.3 times faster, showing similar freezing-thawing effect on the whole set of samples independently of altitude and transect. Along each transect, the three lower sites (from 25 to 525 m a.s.l.) had after stabilization comparable microbial respiration rates, but the most elevated sites always differed - along Tr1 had the most elevated site significantly lower microbial respiration rate, whilst the most elevated sites along Tr2 and Tr3 produced markedly more CO_2 compared to remaining sites along these transects (Fig. 2b). The respiration rate was related neither to PLFA nor to TOC contents, but significant positive correlation with soil Ca^{2+} availability and F/B ratio, and negative correlation with Mg^{2+} availability (r = 0.489, 0.661and -0.545; P = 0.003, < 0.001 and 0.001, respectively) was observed.

3.4 Microbial community structure

The partial RDA revealed significant interactive effect of altitude and transect on MCS (pseudo–F = 4.8, $P < 0.001$). Both factors explained 51% of the total variation in the MCS, with 66 % of explained variability ascribed to altitude, 26% to transect, and 8% of explained variability shared by both factors. The soil geochemical variables explained 72% of the variation in the MCS (pseudo–F = 7.1; $P < 0.001$) indicating that the interactive effect of altitude and transect on MCS was largely driven by vertical and horizontal variability in soil properties. The forward selection of explanatory variables retained four geochemical parameters: Mg^{2+} availability, pH, moisture and TOC content, all together accounting for 55% of variation in the data (pseudo–F = 11.6, $P < 0.001$). The most pronounced shift in the MCS was given by different altitudinal preferences of bacteria and fungi. The bacteria were consistently more abundant in the soils from lower elevations, having lower pH and higher TOC and moisture contents (Fig. 3). In general, PLFAs specific to G– bacteria were more abundant than PLFAs of G+ bacteria (Fig. 4a; mean G–/G+ ratio ± SD = 1.76 ± 0.17; n = 36). Oppositely, the fungal contribution to microbial community increased with an increasing altitude, at the sites having TOC poorer soils and higher pH (Fig. 3). Therefore, the F/B ratio gradually increased with an increasing altitude along all three transects (Fig. 4b). The significant interactive effect of altitude and transect on MCS was mainly connected with a strong effect of soil Mg^{2+} availability, which was higher along the whole Tr1 and differentiated its microbial communities from sites located along Tr2 and Tr3, where microbial communities of respective sites were more similar. The differences in MCS among the respective sites along Tr1 and other two transects further increased towards higher elevations in coincidence with an increasing soil Mg^{2+} availability along Tr1 (Fig. 3). In result, the TOC poorest and Mg^{2+} richest soil at the highest site on Tr1 had the most distinct MCS from all the sites. Its microbial community was characterized by higher abundance of Actinobacteria and PLFAs of phototrophic microorganisms and much lower contribution of G– bacteria compared to communities of all other sampling sites (Fig. 3, 4a).

4 Discussion

4.1 Climatic and soil edaphic conditions along altitudinal transects

The coastal area of the Petunia Bay in Svalbard is characteristic by ca four months lasting summer, long winter period (Ambrožová and Láska, 2017) and very low precipitations (Førland et al., 2010). Our measurements in this area further showed that soils along an elevation gradient from 25 to 765 m a.s.l. face significantly different microclimatic regimes. During winter, when the air temperatures varied a lot in time but less with elevation (Fig. S2b, data from AWS_{25} and AWS_{455}), the soil temperatures were relatively stable but significantly stratified with altitude (Fig. S1, S2a). The mean winter soil temperatures decreased from –4 to –10 °C along the elevation gradient from 25 to 765 m a.s.l. (Table 1), which can strongly reduce winter soil microbial activity at high altitudes (Drotz et al., 2010; Nikrad et al., 2016). In contrast, the mean summer soil temperatures did not reflect the site elevation (Table 1) and the comparison of temperature fluctuations, mean and maximum daily mean temperatures showed that the lowest and highest sites experienced during summer on average colder, but more stable soil microclimate compared to the mid–elevated sites (Table 1). However, the summer season prolonged with decreasing elevation and the increasing number of days with mean temperature above 5 °C and a rising positive surface energy balance (Table 1) positively affected the occurrence and spreading of vascular plants (Kleidon and Mooney, 2000; Klimeš and Doležal, 2010), which had strong implications for a transition of edaphic conditions along studied elevation transects. Together with increased litter inputs and stocks of soil OM with lower C/N ratio (Table 3) was the plant growth associated with root respiration, cation uptake, and release of H^+ and organic acids from roots, all together accounting for decreased soil pH (van Breemen at al., 1984). The increasing soil OM content was further positively related to soil moisture (Fig. 3). Interestingly, the soils in general did not suffer from desiccation (Fig. S3), commonly identified among the most stressing factors in polar and alpine ecosystems (Ley et al., 2004; Van Horn et al., 2013; Tytgat et al., 2016), probably due to high cloudiness and fog occurrence (Sawaske and Freyberg, 2015) in the maritime climate.

The alkaline bedrock material resulted in high soil pH (7.8–9) and high availabilities of basic cations, which were, however, spatially variable due to diverse geology of the studied area (Dallmann et al., 2004; Table 2). Beside clear altitudinal trends in soil edaphic conditions connected mostly with the soil OM content, the Mg^{2+} availability was recognized as main factor driving differences in soil microbial properties between transects (Fig. 3). In result, the character of the parent substrate mostly controlled soil microbial properties at the most elevated sites, which had generally low OM content and the most divergent MCS compared to lower located sites (Fig. 3). The highest site along Tr1 was the most extreme habitat among all the chosen sites, with the highest proportion of bare unvegetated soil surface (Fig. S5), the lowest OM and moisture contents, highest Mg^{2+} availability and soil pH, and consequently also the most distinct microbial characteristics (Fig. 2, 3). Towards lower elevations, the soil OM content became increasingly important and the microbial characteristics of the sites on different transects were more similar.

4.2 Soil microbial properties along altitudinal transects

The altitudinal shifts in soil edaphic properties were not significantly reflected in the soil microbial biomass and potential microbial respiration. Generally, the soil PLFA contents were comparable between all the sites along particular elevation transects, with the exception of very low soil PLFA concentration on the highest site of the Tr1 (Fig. 2a). There are no other studies from the High Arctic ecosystems reporting about altitude effect on soil microbial biomass. However, other studies conducted on alpine gradients in the temperate and boreal zones documented weak or absent altitudinal trends in the

microbial biomass (Djukic et al. 2010, and Xu et al., 2014 using PLFA; Löffler et al., 2008 using cell counts) but also a negative effect of elevation in the Alps (Margesin et al., 2009) and northewestern Finland (Väre et al., 1997). Importantly, none of the studies considered unvegetated habitats and all of them were conducted in soils with acidic or neutral soil pH.

Microbial respiration also did not change systematically with increasing elevation. The three lowest sites along each transect always had comparable soil microbial respiration rates (Fig. 2b), while soil microbial activities of the highest sites differed. The most elevated site on the Tr1 showed significantly lower respiration rates than the lower sites on this transect, which was in line with the lowest OM content as well as soil PLFA content. However, the soils from the highest sites on both Tr2 and Tr3 respired significantly more than the soils from lower sites on these transects, irrespective of relatively stable microbial biomass. This is in contrast to other studies, which reported decreasing microbial activity with increasing elevation (Schinner, 1982; Väre et al., 1997; Niklińska and Klimek, 2007). However, these studies were conducted in lower latitudes and the studied altitudinal gradients did not include unvegetated habitats. To comment on and justify our results, we are aware that microbial activities were measured in freeze-stored and not fresh samples (see section 2.3 for details) and, therefore, the respiration rates measured after thawing show the potential activity of soil microbial communities in the soils. However, the respiration rates in three subsequent measurements (after flush, during adaptation and after stabilization) were positively correlated ($r = 0.93$ and 0.74, both $P < 0.0001$, $n = 36$), the ratios between the flush and stabilized respiration rates were comparable across all the soils (compare Fig. 2b and c) and the above-described differences in microbial activities among the sites were consistent. Our data are in accord with the study of Larsen et al., (2002), who found comparable response to freeze-thaw events between two different arctic ecosystem types. We thus suggest that the soils responded similarly to the storage treatment independently of site location and that observed differences in soil microbial activities are representative for the studied transects. Therefore, the higher soil microbial respiration at the most elevated sites point to a higher lability of the present OM (Lipson et al., 2000; Uhlířová et al., 2007) and/or to a shift in microbial communities towards groups with higher potential to mineralize the OM (Gavazov, 2010; Djukic et al., 2013). Previous studies, considering either bare soil or vegetated habitats, reported rather increasing complexity of soil OM with elevation (Ley et al., 2004; Xu et al., 2014). However, in this study was majority of OM and microbial biomass at the most elevated sites associated with biological soil crusts with high algal and cyanobacterial abundance (Table S1, Fig. S5), known for their high microbial activity (Pushkareva et al., 2017; Bastida et al., 2014). The high microbial activity in the most elevated sites could be ascribed to prevalence of compounds of algal/cyanobacterial origin with very low portion of complex and slowly decomposable lignin and lignified compounds and protective waxes (like cutin and suberin) mainly derived from vascular plants. In accord, the sitosterol to brassicasterol ratio gradually decreasing with increasing elevation (Table 3) and increasing sitosterol content in the TOC pool at lower elevations pointed to growing importance of microalgal sources of OM in high elevation habitats (Sinsabaugh et al., 1997; Rontani et al., 2012). Even though both sterols can be found in higher plants and microalgae, the changing ratio indicates shift in the origin of OM (reviewed by Volkman, 1986, see also Volkman, 2003). Changes within microbial communities, which can also help to explain higher soil microbial respiration at the most elevated sites are discussed below.

Although the soil PLFA content did not change along the studied elevation transects, we have found a systematic altitudinal shift in the PLFA composition, resulting in significantly increasing F/B ratio towards higher elevations. This shift was best explained by a decreasing soil OM content and soil moisture and increasing pH (Fig. 3). Reports about soil F/B ratios and their altitudinal changes from the High Arctic are missing, but studies from lower latitudes showed either a similar trend of increasing F/B ratio with an altitude in the Alps (Margesin et al., 2009) or the opposite altitudinal effect in the Alps (Djukic et al., 2010) and Himalayas (Xu et al., 2014; Hu et al., 2016). Such divergent results indicate that altitude alone is not the key driving factor of the soil F/B ratio. In contrast to our observation, these studies reported very low soil F/B ratios of 0.05-0.2, which may indicate important role of fungi in functioning of the Arctic habitats. Soil pH was previously

identified as the main driver of fungal-bacterial dominance in the soil (Baath and Anderson, 2003; Högberg et al., 2007; Rousk et al., 2009; Siles and Margesin, 2016). Fungi have been found more acid tolerant than bacteria, leading to higher F/B ratio in acidic soils (Högberg et al., 2007; Rousk et al., 2009; reviewed by Strickland and Rousk, 2010). However, here we report high F/B ratios in the alkaline soils (pH 7.8-9.0) and increasing F/B ratios with an increasing soil pH. Similar trend was reported also by Hu et al., (2016), but the authors found F/B ratios one order of magnitude lower compared to our study. The possible explanation of generally high fungal abundance and increasing F/B ratio at more elevated sites, which are typical by unfavourable edaphic conditions and severe winter microclimate, could be higher competitiveness of fungi compared to bacteria in suboptimal conditions due to their wider pH (Wheeler et al., 1991) and lower temperature (Margesin et al., 2003) growth optima. We further found that the increasing F/B ratio was significantly coupled with an increasing soil respiration ($r = 0.649$; $P < 0.001$). Indeed, such relationship can be related to higher fungal ability either to prosper in the soil conditions at the most elevated sites, or to utilize more efficiently available C sources (Ley et al., 2004; Bardgett et al., 2005; Nemergut et al., 2005; van der Heijden et al., 2008). In turn, the higher bacterial contribution at lower elevations may be associated with more benign soil conditions and bacterial preference for utilization of labile root exudates released by vascular plants (Lipson et al., 1999; Lipson et al., 2002). Since the projected warming in the Arctic (Collins et al., 2013) will likely cause an upward migration of the vegetation and increasing plant cover in detriment of lichens and biological soil crusts (Vuorinen et al., 2017; Yu et al. 2017; de Mesquita et al., 2017), the soil microbial communities will likely respond by decreasing F/B ratios at higher elevations.

Apart from the systematic altitudinal shift in the F/B ratio, we observed a strong shift in the bacterial composition, which differentiated the altitudinal trends in the soil MCS along Tr1 from trends along Tr2 and Tr3. This difference between transects increased towards higher elevations and was best explained by Mg^{2+} availability (Fig. 3). The soils from Tr1, except the lowest site, had a lower G– to G+ bacterial ratios within microbial communities than soils from other two transects. Further, the microbial community of the most elevated site along Tr1 was significantly more contributed by actinobacteria and phototrophic microorganisms compared to all other sites (Fig. 3, 4a). It is known that the high Mg^{2+} availability inhibits growth of many soil bacterial species. The observed inhibitive Mg^{2+} levels were 5 and 50 p.p.m for G– and G+ bacteria, respectively (Webb 1949), indicating that these bacterial groups significantly differ in their tolerance for enhanced Mg^{2+} levels. Considering half of available Mg^{2+} in soil solution and average soil moisture content 20%, the Mg^{2+} concentrations ranged approximately from 16-140 p.p.m,, which could explain decreased abundance of G- bacteria in sites with high Mg^{2+} availability. This inhibitive Mg^{2+} effect further corresponds with the negative correlations between Mg^{2+} availability and soil microbial biomass and respiration found in our study, and could explain the lower microbial biomass and respiration in the soils from Tr1. Our data thus indicate that beside the traditionally identified drivers of microbial activity and MCS such as soil OM content, moisture and pH, Mg^{2+}availability is in important factor shaping the microbial environment along the arctic altitudinal transects on dolomitic parent materials.

5 Conslusions

The results obtained in this study have shown significant altitudinal zonation of most edaphic properties, but also significant spatial heterogeneity in horizontal direction, resulting in transect-specific effect of altitude on abiotic soil properties. Our data demonstrated that soils on the most elevated, unvegetated sites around the Petunia Bay can host microbial assemblages comparable in size and activity with the tundra ecosystem. The high microbial biomass and activity at the most elevated sites were almost exclusively associated with biological soil crusts, largely contributed by fungi. However, their development was retarded on some sites by high pH, low moisture and high Mg availability, resulting in pronouncedly low OM content, microbial biomass and distinct MCS. Despite the ubiquitous occurrence of soil crusts, the gradually increasing plant productivity and litter inputs down along transects were associated with decreasing soil pH, increasing OM content and soil

moisture. Concurrently, the soil edaphic and microbial properties become more uniform. As the rise in temperatures and humidity predicted by climatic models will likely cause an upward migration of the vegetation and increasing plant cover, the higher plant litter inputs will overreach the influence of parent material and entail an increasing abundance of bacteria and decreasing F/B ratio in the summer microbial assemblages.

Author contribution

P. Kotas and E. Kaštovská analysed the data and wrote the manuscript with assistance of all coauthors. P. Kotas and J. Elster designed the study and performed sampling. The microbial community structure and environmental parameters were assessed by P. Kotas, E. Kaštovská and H. Šantrůčková.

Acknowledgements

The field work on Svalbard was funded by the Ministry of Education, Youth and Sports of the Czech Republic through CzechPolar – Czech polar stations, construction and logistic expenses (LM2010009 and RVO67985939). The lab work was supported by the SoWa Research Infrastructure funded by the Ministry of Education, Youth and Sports of the Czech Republic, programme "Projects of Large Infrastructure for Research, Development, and Innovations" (grant LM2015075). We thank also to Jan Kavan for help with logistics during the field work.

Competing interests

The authors declare that they have no conflict of interest.

References

Ambožová, K. and Láska, K.: Air temperature variability in the vertical profile over the coastal area of Petuniabukta, central Spitsbergen, Pol. Polar Res., 38, 41–60, 2017.

Bååth, E. and Anderson, T.–H.: Comparison of soil fungal/bacterial ratios in a pH gradient using physiological and PLFA based techniques. Soil Biol. Biochem., 35, 955–63, 2003.

Bardgett, R. D., Bowman, W. D., Kaufmann, R., and Schmidt, S.K.: A temporal approach to linking aboveground and belowground ecology, Trends Ecol. Evol., 20, 634–641, 2005.

Bastida, F., Jehmlich, N., Ondoño, S., von Bergen, M., García, C., and Moreno, J. L.: Characterization of the microbial community in biological soil crusts dominated by Fulgensia desertorum (Tomin) Poelt and Squamarina cartilaginea (With.) P. James and in the underlying soil, Soil Biol. Biochem., 76, 70–79, 2014.

Bekku, Y. S., Kume, A., Masuzawa, T., Kanda, H., Nakatsubo, T., and Koizumi, H.: Soil respiration in a high arctic glacier foreland in Ny-Ålesund, Svalbard, Polar Bioscience, 17, 36–46, 2004.

Björk, R. G., Björkman, M. P., Andersson M. X., and Klemedtsson L.: Temporal variation in soil microbial communities in Alpine tundra, Soil Biol. Biochem., 40, 266–268, 2008.

Blaud, A., Lerch, T. Z., Phoenix, G. K., and Osborn M. A.: Arctic soil microbial diversity in a changing world, Res. Microbiol., 166, 796–813, 2015.

Bossio, D.A. and Scow, K.M.: Impacts of carbon and flooding on soil microbial communities: phospholipid fatty acid profiles and substrate utilization patterns, Microb. Ecol., 35, 265–278, 1998.

Clein, J. S. and Schimel, J. P.: Reduction in microbial activity of litter due to drying and rewetting events, Soil Biol. Biochem., 26, 403–406, 1994.

Collins, M., Knutti, R., Arblaster, J., Dufresne, J. L., Fichefet, T., Friedlingstein, P., Gao, X., Gutowski, W. J., Johns, T., Krinner, G., Shongwe, M., Tebaldi, C., Weaver, A. J., and Wehner, M.: Long-term climate change. Projections, commitments and irreversibility. In: Stocker, T. F., Qin, D., Plattner, G. K., Tignor, M. M. B., Allen, S. K., Boschung, J., Nauels, A., Xia, Y., Bex, V., and Midgley, P. M. (eds.).: Climate Change 2013. The physical science basis. Contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change. Cambridge University Press. Cambridge. p. 1029-1136, ISBN: 9781107057991, 2013.

Chu, H., Fierer, N., Lauber, Ch. L., Caporaso, J. G., Knight, R., and Grogan, P.: Soil bacterial diversity in the Arctic is not fundamentally different from that found in other biomes, Environ. Microb., 12, 2998–3006, 2010.

Dallmann, W. K., Piepjohn, K., and Blomeier, D.: Geological map of Billefjorden, Central Spitsbergen, Svalbard, Temakart Nr. 36, Norsk Polarinstitutt, 2004.

de Mesquita, C. P. B., Knelman, J. E., King, A. J., Farrer, E. C., Porazinska, D. L., Schmidt, S. K., and Suding, K. N.: Plant colonization of moss-dominated soils in the alpine: Microbial and biogeochemical implications, Soil Biol. Biochem., 111, 135–142, 2017.

Djukic, I., Zehetner, F., Mentler, A., and Gerzabek, M. H.: Microbial community composition and activity in different Alpine vegetation zones, Soil Biol. Biochem., 42, 155–161, 2010.

Djukic, I., Zehetner, F., Watzinger, A., Horacek, M., and Gerzabek, M. H.: In situ carbon turnover dynamics and the role of soil microorganisms therein: a climate warming study in an Alpine ecosystem, FEMS Microbiol. Ecol., 83, 112–124, 2013.

Drotz, S. H., Sparrman, T., Nilsson, M. B., Schleucher, J., and Öquist, M. G.: Both catabolic and anabolic heterotrophic microbial activity proceed in frozen soils, PNAS, 107, 21046–21051, 2010.

Ferrari, B. C., Bissett, A., Snape, I., van Dorst, J., Palmer, A. S., Ji, M., Siciliano, S. D., Stark, J. S., Winsley, T., and Brown, M. V.: Geological connectivity drives microbial community structure and connectivity in polar, terrestrial ecosystems, Environ. Microb., 18, 1834–1849, 2016.

Fierer, N., McCain C. M., Meir, P., Zimmermann, M., Rapp, J. M., Silman, M. R., and Knight, R.: Microbes do not follow the elevational diversity patterns of plants and animals, Ecology, 92, 797–804, 2011.

Førland, E.J., Benestad, R., Hanssen-Bauer, I., Haugen, J.E., and Skaugen, T.: Temperature and Precipitation Development at Svalbard 1900-2100, Advances in Meterology, doi:10.1155/2011/893790, 2010.

Frostegård, A. and Bååth, E.: The use of phospholipid fatty acid analysis to estimate bacterial and fungal biomass in soil, Biol. Fert. Soils, 22, 59–65, 1996.

Frostegård, Å., Bååth, E., and Tunlid, A.: Shifts in the structure of soil microbial communities in limed forests as revealed by phospholipid fatty acid analysis, Soil Biol. Biochem., 25, 723–730, 1993.

Gavazov, K. S.: Dynamics of alpine plant litter decomposition in a changing climate, Plant Soil, 337, 19–32, 2010.

Hardison, A. K., Canuel, E. A., Anderson, I. C., Tobias, C. R., Veuger, B., and Waters, M. N.: Microphytobenthos and benthic macroalgae determine sediment organic matter composition in shallow photic sediments, Biogeosciences, 10, 5571–5588, 2013.

Harris, D., Horwáth, W. R., and van Kessel, Ch.: Acid fumigation of soils to remove carbonates prior to total organic carbon or carbon-13 isotopic analysis, Soil Sci. Soc. Am. J., 65, 1853–1856, 2001.

Holm, S.: A simple sequentially rejective multiple test procedure. Scand. J. Stat., 6, 65–70, 1979.

Högberg, M. N., Högberg, P., and Myrold, D. D.: Is microbial community composition in boreal forest soils determined by pH, C-to-N ratio, the trees, or all three? Oecologia, 150, 590–601, 2007.

Hu, L., Xiang, Z., Wang, G., Rafique, R., Liu W., and Wang, C.: Changes in soil physicochemical and microbial properties along elevation gradients in two forest soils, Scandinavian Journal of Forest Research, 31, 242–253, 2016.

Jones, A, Stolbovoy, V., Tarnocai, C., Broll, G., Spaargaren, O., and Montanarella, L. (eds.): Soil Atlas of the Northern Circumpolar Region. European Commission, Publications Office of the European Union, Luxembourg, 144pp., 2010.

Khotimchenko, S. V., Vaskovsky, V. E., and Titlyanova, T. V.: Fatty acids of marine algae from the pacific coast of North California, Bot. Mar., 45, 17–22, 2002.

Kleidon, A. and Mooney, H. A.: A global distribution of biodiversity inferred from climatic constraints: results from a process-based modelling study, Global Change Biol., 6, 507–523, 2000.

Klimeš, L. and Doležal J.: An experimental assessment of the upper elevational limit of flowering plants in the western Himalayas, Ecography, 33, 590–596, 2010.

Körner, Ch.: The use of 'altitude' in ecological research, Trends in Ecol. And Evol., 22, 569–574, 2007.

Kroppenstedt, R. M.: Fatty acid and menaquinone analysis of actinomycetes and related organisms. In: Goodfellow, M., Minikin, D. E., Eds. Chemical methods in bacterial systematics. Academic Press, London, pp. 173–194, 1985.

Larsen, K. S., Jonasson, S., and Michelsešn, A.: Repeated freeze–thaw cycles and their effects on biological processes in two arctic ecosystem types, Appl. Soil Ecol., 21, 187–195, 2002.

Lazzaro, A., Hilfiker, D., and Zeyer, J.: Structure of microbial communities in alpine soils: seasonal and elevational effects, Frontiers in Microbiology, doi: 10.3389/fmicb.2015.01330, 2015.

Ley, R. E., Williams, M. W., and Schmidt, S. K.: Microbial population dynamics in an extreme environment: controlling factors in talus soils at 3750m in the Colorado Rocky Mountains, Biogeochemistry, 68, 313–335, 2004.

Lipson, D. A.: Relationships between temperature responses and bacterial community structure along seasonal and altitudinal gradients, FEMS Microbiol. Ecol., 59, 418–427, 2007.

Lipson, D. A., Schmidt, S. K., and Monson, R. K.: Links between microbial population dynamics and nitrogen availability in an alpine ecosystem, Ecology, 80, 1623–1631, 1999.

Lipson, D. A., Schmidt, S. K., and Monson, R. K.: Carbon availability and temperature control the post-snowmelt decline in alpine soil microbial biomass, Soil Biol. Biochem., 32, 441–448, 2000.

Lipson, D. A., Schadt, C. W., and Schmidt, S. K.: Changes in soil microbial community structure and function in an alpine dry meadow following spring snow melt, Microb. Ecol., 43, 307–314, 2002.

Lovelland P. J. and Whalley W. R.: Particle size analysis. In: Soil and Environmental Analysis Physical Method (Eds Smith, K.A. and Mullins, Ch.E.), Dekker Press, New York, USA, 2001.

Löffler, U. C. M., Cypionka, H., and Löffler, J.: Soil microbial activity along an arctic-alpine altitudinal gradient from a seasonal perspective, Eur. J. Soil Sci., 59, 842–854, 2008.

Ma, X., Chen, T., Zhang, G., and Wang R.: Microbial community structure along an altitude gradient in three different localities, Folia Microbiol., 49, 105–111, 2004.

Männistö, M., Tiirola, M., and Häggblom, M. M.: Bacterial communities in Arctic fjelds of Finnish Lapland are stable but highly pH-dependent, FEMS Microbiol. Ecol., 59, 452–465, 2007.

Margesin, R., Gander, S., Zacke, G., Gounot, A. M., and Schinner, F.: Hydrocarbon degradation and enzyme activities of cold-adapted bacteria and yeasts, Extremophiles, 7, 451–458, 2003.

Margesin, R., Jud, M., Tscherko, D., and Schinner, F.: Microbial communities and activities in alpine and subalpine soils, FEMS Microbiol. Ecol., 67, 208–218, 2009.

Meng, H., Li, K., Nie, M.,Wan, J.-R., Quan, Z.-X., Fang, C.-M., Chen, J.-K., Gu, J.-D., and Li, B.: Responses of bacterial and fungal communities to an elevation gradient in a subtropical montane forest of China, Appl. Microbiol. Biot., 97, 2219–2230, 2013.

Montgomery, H. J., Monreal, C. M., Young, J. C., and Seifert, K. A.: 2000. Determination of soil fungal biomass from soil ergosterol analyses, Soil Biol. Biochem., 32, 1207–1217, 2000.

Nemergut, D. R., Costello, E. K., Meyer, A. F., Pescador, M. Y., Weintraub, M. N., and Schmidt, S. K.: Structure and function of alpine and arctic soil microbial communities, Res. Microbiol., 156, 775–784, 2005.

Niklińska, M. and Klimek, B.: Effect of temperature on the respiration rate of forest soil organic layer along an elevation gradient in the Polish Carpathians, Biol. Fert. Soils, 43, 511–518, 2007.

Nikrad, M. P., Kerkhof, L. J., and Häggblom, M. M.: The subzero microbiome: microbial activity in frozen and thawing soils, FEMS Microbiol. Ecol., 92, doi:10.1093/femsec/fiw08, 2016.

Oberbauer, S. F., Tweedie, C. E., Welker, J. M., Fahnestock, J. T., Henry, G. H. R., Webber, P. J., Hollister, R. D., Walker, M. D., Kuchy, A., Elmore, E., and Starr, G.: Tundra CO2 fluxes in response to experimental warming across latitudinal and moisture gradients. Ecol. Monogr., 77, 221–238, 2007.

Prach, K., Klimešová, J., Košnar, J., Redčenko O., and Hais M.: Variability of contemporary vegetation around Petuniabukta, central Spitsbergen, Pol. Polar Res., 33, 383–394, 2012.

Pushkareva, E., Kvíderová, J., Šimek, M., and Elster, J.: Nitrogen fixation and diurnal changes of photosynthetic activity in Arctic soil crusts at different development stage, Europ Journal of Soil Biology, 79, 21–30, 2017.

Richter, D. D., Johnson, D. W., and Dai, K. H.: Cation exchange reactions in acid forested soils: effects of atmospheric pollutant deposition. In: Johnson, D.W., Lindberg, S.E. (eds.), Atmospheric Deposition and Nutrient Cycling in Forest Ecosystems, Spring-Verlag, New York, pp 339–358, 1992.

Rontani, J.–F., Charriere, B., Petit, M., Vaultier, F., Heipieper, H. J., Link, H., Chaillou, G., and Sempéré, R.: Degradation state of organic matter in surface sediments from the Southern Beaufort Sea: a lipid approach, Biogeosciences, 9, 3513–3530, 2012.

Rousk, J., Brookes, P. C., and Bååth, E.: Contrasting soil pH effects on fungal and bacterial growth suggest functional redundancy in carbon mineralization, Appl. Environ. Microb., 75, 1589–1596, 2009.

Sawaske, S. R. and Freyberg, D. L.: Fog, fog drip, and streamflow in the Santa Cruz Mountains of the California Coast Range, Ecohydrology, 8, 695–713, 2015.

Schimel, J. P. and Clein, J. S.: Microbial response to freeze-thaw cycles in tundra and taiga soils, Soil Biol. Biochem., 28, 1061–1066, 1996.

Schinner, F.: Soil microbial activities and litter decomposition related to altitude, Plant Soil, 65, 87–94, 1982.

Schinner, F. and Gstraunthaler, G.: Adaptation of microbial activities to the environmental conditions in alpine soils, Oecologia, 50, 113–116, 1981.

Schűtte, U. M. E., Abdo, Z., Foster, J., Ravel, J., Bunge, J., Solheim, B., and Forney, L. J.: Bacterial diversity in a glacier foreland of the high Arctic, Mol. Ecol., 19, 54–56, 2010.

Shen, C., Xiong, J., Zhang, H., Feng, Y., Lin, X., Li, X., Liang, W., and Chu, H.: Soil pH drives the spatial distribution of bacterial communities along elevation on Changbai Mountain, Soil Biol. Biochem., 57, 204–211, 2013.

Siles, J. S. and Margesin, R.: Abundance and diversity of bacterial, archaeal, and fungal communities along an altitudinal gradient in alpine forest soils: What are the driving factors? Soil Microbiology, 72, 207–220, 2016.

Singh, D., Takahashi, K., Kim, M., Chun, J., and Adams, J. M.: A hump-backed trend in bacterial diversity with elevation on Mount Fuji, Japan, Microb. Ecol. 63, 429–437, 2012.

Sinsabaugh, R. L., Antibus, R. K., Jackson, C. R., Karpanty, S., Robinson, M., Liptak, M., and Franchini, P.: A β-sitosterol assay for fine-root mass in soil, Soil Biol. Biochem., 29, 39–44, 1997.

Sparling, G. P. and West, A. W.: A comparison of gas chromatography and differential respirometer methods to measure soil respiration and to estimate the soil microbial biomass, Pedobiologia, 34, 103–112, 1990.

Stenberg, B., Johanssson, M., Pell, M., Sjödahl-Svensson, K., Stenström, J., and Torstensson, L.: Microbial biomass and activities in soil as affected by frozen and cold storage, Soil Biol. Biochem., 30, 393–402, 1998.

Strickland, M. S. and Rousk, J.: Considering fungal:bacterial dominance in soils – Methods, controls, and ecosystem implications, Soil Biol. Biochem., 42, 1385–1395, 2010.

Šmilauer, P. and Lepš, J.: Multivariate analysis of ecological data using CANOCO5. Cambridge University Press, Cambridge, 2014.

Ter Braak, C. J. F. and Šmilauer, P.: Canoco reference manual and user's guide: software for ordination, version 5.0. Microcomputer Power, Ithaca, USA, 496 pp, 2012.

Trevors, J. T., Kevan, P. G., and Tam, L.: Microbial diversity across a Canadian sub-Arctic, isostatically rebounding, soil transect. Polar Science, 4, 81–91, 2010.

Tytgat, B., Verleyen, E., Sweetlove, M., D'hondt, S., Clercx, P., Van Ranst, E., Peeters, K., Roberts, S., Namsaraev, Z., Wilmotte, A., Vyverman, W., and Willems, A.: Bacterial community composition in relation to bedrock type and macrobiota in soils from the Sør Rondane Mountains, East Antarctica, FEMS Microbiol. Ecol., 92, doi: 10.1093/femsec/fiw126, 2016.

Uhlířová, E., Šantrůčková, H., and Davidov, S. P.: Quality and potential biodegradability of soil organic matter preserved in permafrost of Siberian tussock tundra, Soil Biol. Biochem., 39, 1978–1989, 2007.

van Breemen, N., Driscoll, C. T., and Mulder, J.: Acidic deposition and internal proton sources in acidification of soils and waters, Nature, 307, 599–604, 1984.

van de Heijden, M. G. A., Bardgett, R. D., and van Straalen, N. M.: The unseen majority: soil microbes as drivers of plant diversity and productivity in terrestrial ecosystems, Ecol. Lett., 11, 296–310, 2008.

Van Horn, D. J., Van Horn, M. L., Barrett, J. E., Gooseff, M. N., Altrichter, A. E., Geyer, K. M., Zeglin, L. H., and Takacs-Vesbach, K.: Factors controlling soil microbial biomass and bacterial diversity and community composition in a cold desert ecosystem: Role of geographic scale, PLoS One, 8, doi: 10.1371/journal.pone.0066103, 2013.

Väre, H., Vestberg, M., and Ohtonen, R.: Shifts in mycorrhiza and microbial activity along an oroarctic altitudinal gradient in northern Fennoscandia, Arctic Alpine Res., 29, 93–104, 1997.

Volkman, J. K.: Sterols in microorganisms, Appl. Microbiol. Biot., 60, 495–506, 2003.

Volkman, J. K.: A review of sterol markers for marine and terrigenous organic matter, Org. Geochem., 9, 83–99, 1986.

Vuorinen, K. E. M., Oksanen, L., Oksanen, T., Pyykönen, A., Olofsson, J., and Virtanen, R.: Open tundra persist, but arctic features decline–Vegetation changes in the warming Fennoscandian tundra, Global Change Biol., 23, 3794–3807, 2017.

Webb, M.: The influence of magnesium on cell division. 2. The effect of magnesium on the growth and cell division of various bacterial species in complex media, Microbiology, 3, 410–417, 1949a.

Wheeler, K. A., Hurdman, B. F., and Pitt, J. I.: Influence of pH on the growth of some toxigenic species of *Aspergillus*, *Penicillium* and *Fusarium*, Int. J. Food Microbiol., 12, 141–150, 1991.

Xu, M., Li, X., Cai, X., Gai, J., Li, X., Christie, P., and Zhang, J.: Soil microbial community structure and activity along a montane elevational gradient on the Tibetan Plateau, Eur. J Soil Biol., 64, 6–14, 2014.

Yoshitake, S., Uchida, M., Koizumi, H., and Nakatsubo, T.: Carbon and nitrogen limitation of soil microbial respiration in a High Arctic successional glacier foreland near Ny-Ålesund, Svalbard, Pol. Res., 26, 22–30, 2007.

Yu, Q., Epstein, H., Engstrom, R., and Walker, D.: Circumpolar arctic tundra biomass and productivity dynamics in response to projected climate change and herbivory, Global Change Biol., 23, 3895–3907, 2017.

Zbíral, J. and Němec, P.: Data presentation, interpretation, and communication: Integrating of Mehlich 3 extractant into the Czech soil testing scheme, Communications in Soil Science and Plant Analysis, 31, 2171–2182, 2000.

Tables

Table 1. Climatic variables; temperatures given in °C

Sites [m a.s.l.]	Means Summer	Means Winter	Means Year	Min daily means Winter	Max daily means Summer	Mean daily amplitude Summer	Max daily amplitude Summer	Number of days with daily mean > 0 °C	Number of days with daily mean > 5 °C	Positive soil surface energy balance
25	5.8	-3.6	-0.8	-7.0	11.2	5.2	10.9	110	62	615
280	7.1	-5.7	-2.7	-10.3	14.5	8.5	18.2	96	54	571
520	5.8	-8.9	-4.9	-15.8	14.7	8.1	17.7	91	40	480
765	5.3	-9.5	-6.6	-17.1	11.6	5.5	14.0	51	11	290

Table 2. Geochemical characteristics of soils along the studied altitudinal transects (Tr1-Tr3). Means ± SD (n = 3) are given in the upper part of the table. Results of two–way ANOVAs (F–values) of the effects of transect (Tr), altitude (Alt) and their interaction (Tr x Alt) are presented in the lower part of the table.

transect	altitude [m a.s.l]	soil type	soil moisture [%]	pH	CEC [meq/100g^{-1}]	Ca^{2+} [mg g^{-1}]	Mg^{2+} [mg g^{-1}]	K^{+} [µg g^{-1}]	Na^{+} [µg g^{-1}]
Tr1	25	sandy loam	**a** 28.4 ± 2.5	**b** 7.8 ± 0.1	**a** 35.8 ± 0.4	**b** 4.9 ± 0.2	**c** 0.50 ± 0.03	**b** 104 ± 2.3	**a** 16.0 ± 1.4
	275	sandy loam-loam	**b** 18.0 ± 0.5	**b** 7.9 ± 0.2	**b** 27.4 ± 2.3	**b** 5.2 ± 0.6	**c** 0.55 ± 0.08	**b** 81 ± 8.8	**bc** 8.4 ± 1.3
	525	loam	**b** 18.6 ± 2.5	**b** 8.1 ± 0.1	**b** 30.3 ± 0.7	**b** 4.3 ± 0.4	**b** 0.85 ± 0.04	**a** 160 ± 18.1	**b** 11.3 ± 1.1
	765	clay-loam	**c** 12.1 ± 1.8	**a** 9 ± 0.0	**b** 26.8 ± 2.3	**a** 19.8 ± 1.0	**a** 1.25 ± 0.06	**c** 11 ± 2.7	**c** 7.3 ± 0.0
Tr2	25	sandy loam	**a** 21.1 ± 2.4	**c** 7.8 ± 0.1	**b** 25.6 ± 2.7	**b** 14.7 ± 2.6	**c** 0.19 ± 0.01	**ab** 52 ± 4.0	**a** 13.2 ± 1.7
	275	sandy loam-loam	**a** 21.1 ± 2.4	**c** 7.9 ± 0.1	**b** 30.3 ± 1.7	**ab** 16.5 ± 1.1	**b** 0.26 ± 0.01	**a** 59 ± 4.3	**ab** 10.1 ± 1.7
	525	sandy loam-loam	**a** 21.7 ± 5.3	**b** 8.4 ± 0.1	**b** 30.8 ± 1.1	**c** 7.8 ± 1.6	**a** 0.34 ± 0.01	**a** 69 ± 3.3	**ab** 9.6 ± 1.8
	765	loam	**a** 22.5 ± 1.7	**a** 8.8 ± 0.1	**a** 45.1 ± 0.5	**a** 27.9 ± 9.3	**b** 0.25 ± 0.01	**b** 41 ± 8.8	**b** 8.1 ± 1.4
Tr3	25	sandy loam	**a** 39.5 ± 1.4	**b** 8.1 ± 0.1	**a** 49.4 ± 2.1	**c** 7.7 ± 0.3	**a** 0.20 ± 0.03	**b** 52 ± 5.3	**a** 17.1 ± 1.1
	275	sandy loam-loam	**ab** 31.9 ± 2.9	**b** 8.1 ± 0.1	**b** 39.2 ± 5.4	**b** 10.8 ± 0.6	**a** 0.21 ± 0.01	**ab** 59 ± 1.9	**a** 18.5 ± 0.5
	525	loam	**ab** 28.2 ± 6.5	**b** 8 ± 0.1	**b** 34.9 ± 3.0	**ab** 13.0 ± 4.6	**a** 0.22 ± 0.00	**a** 66 ± 6.6	**a** 18.4 ± 3.1
	765	loam	**b** 22.5 ± 1.7	**a** 8.8 ± 0.1	**b** 30.6 ± 3.9	**a** 14.2 ± 0.1	**b** 0.16 ± 0.00	**b** 52 ± 1.6	**b** 9.9 ± 0.2
		d.f.							
Tr		2	**31.4 *****	0.10	**22.1 *****	**6.43 ****	**634 *****	**51.7 *****	**36.2 *****
Alt		3	**11.1 *****	**98 *****	**4.61 ***	**14.1 *****	**66.9 *****	**74.9 *****	**18.7 *****
Tr x Alt		6	**5.07 ****	**5.6 *****	**20.5 *****	0.83	**60.6 *****	**31.6 *****	**3.94 ****

Different letters indicate significant differences between sampling sites along particular transects ($P < 0.05$; upper part of the table). Statistically significant differences are indicated by: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ (lower part of the table).

Table 3. Total soil carbon (TOC) and nitrogen (TN) contents, their molar ratios, contents of sitosterol in TOC and sitosterol / brassicasterol ratios and soil PLFA contents in soils along the altitudinal transects (Tr1-Tr3). Means ± SD (n = 3) are given in the upper part of the table. Results of two–way ANOVAs (F–values) of the effects of transect (Tr), altitude (Alt) and their interaction (Tr x Alt) are presented in the lower part of the table.

transect	altitude	TOC	TN	TOC/TN	Sitosterol	Sitosterol / Brassicasterol
	[m a.s.l.]	[mg g^{-1}]	[mg g^{-1}]		[µg g^{-1} TOC]	
Tr1	25	**c** 70.6 ± 13.4	**b** 5.0 ± 1.01	**b** 12.1 ± 0.2	**c** 534 ± 62.8	**b** 5.5 ± 0.4
	275	**b** 21.1 ± 1.9	**a** 2.0 ± 0.29	**ab** 9.0 ± 0.7	**bc** 521 ± 140	**b** 5.3 ± 0.8
	525	**b** 18.5 ± 4.2	**a** 1.8 ± 0.31	**ab** 8.8 ± 0.7	**ab** 293 ± 66.5	**b** 4.7 ± 1.0
	765	**a** 4.4 ± 1.5	**a** 0.5 ± 0.07	**a** 7.9 ± 2.6	**a** 81.1 ± 2.7	**a** 2.3 ± 0.4
Tr2	25	**ab** 30.6 ± 4.8	**a** 1.9 ± 0.40	**c** 13.7 ± 0.9	**bc** 515 ± 44.9	**b** 6.7 ± 0.7
	275	**b** 37.2 ± 5.0	**a** 3.0 ± 0.26	**b** 10.7 ± 0.7	**c** 616 ± 143	**b** 5.6 ± 1.2
	525	**a** 24.4 ± 7.8	**a** 1.9 ± 0.64	**b** 9.8 ± 1.2	**ab** 299 ± 73.3	**a** 2.9 ± 0.4
	765	**a** 21.6 ± 3.6	**a** 2.8 ± 0.20	**a** 6.7 ± 0.6	**a** 161 ± 36.9	**a** 2.7 ± 0.7
Tr3	25	**c** 81.1 ± 8.7	**b** 6.1 ± 0.38	**b** 11.5 ± 0.7	**b** 587 ± 144	**b** 6.4 ± 2.1
	275	**b** 62.2 ± 9.1	**ab** 4.8 ± 0.32	**b** 11 ± 0.7	**ab** 370 ± 42.9	**a** 4.2 ± 0.7
	525	**ab** 39.6 ± 11.4	**a** 4.8 ± 0.32	**b** 10.6 ± 0.6	**a** 270 ± 112	**a** 3.3 ± 1.0
	765	**a** 23.1 ± 3.9	**a** 2.5 ± 0.37	**a** 7.9 ± 0.2	**a** 151 ± 37.8	**a** 3.1 ± 0.9
	d.f.					
Tr	2	**27.8 *****	**31.5 *****	1.57	0.79	1.04
Alt	3	**42.4 *****	**26.4 *****	**23.6 *****	**28.4 *****	**14.4 *****
Tr x Alt	6	**8.33 *****	**11.3 *****	1.96	1.34	2.17

Different letters indicate significant differences between sampling sites along particular transects ($P < 0.05$; upper part of the table). Statistically significant differences are indicated by: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ (lower part of the table).

Figures

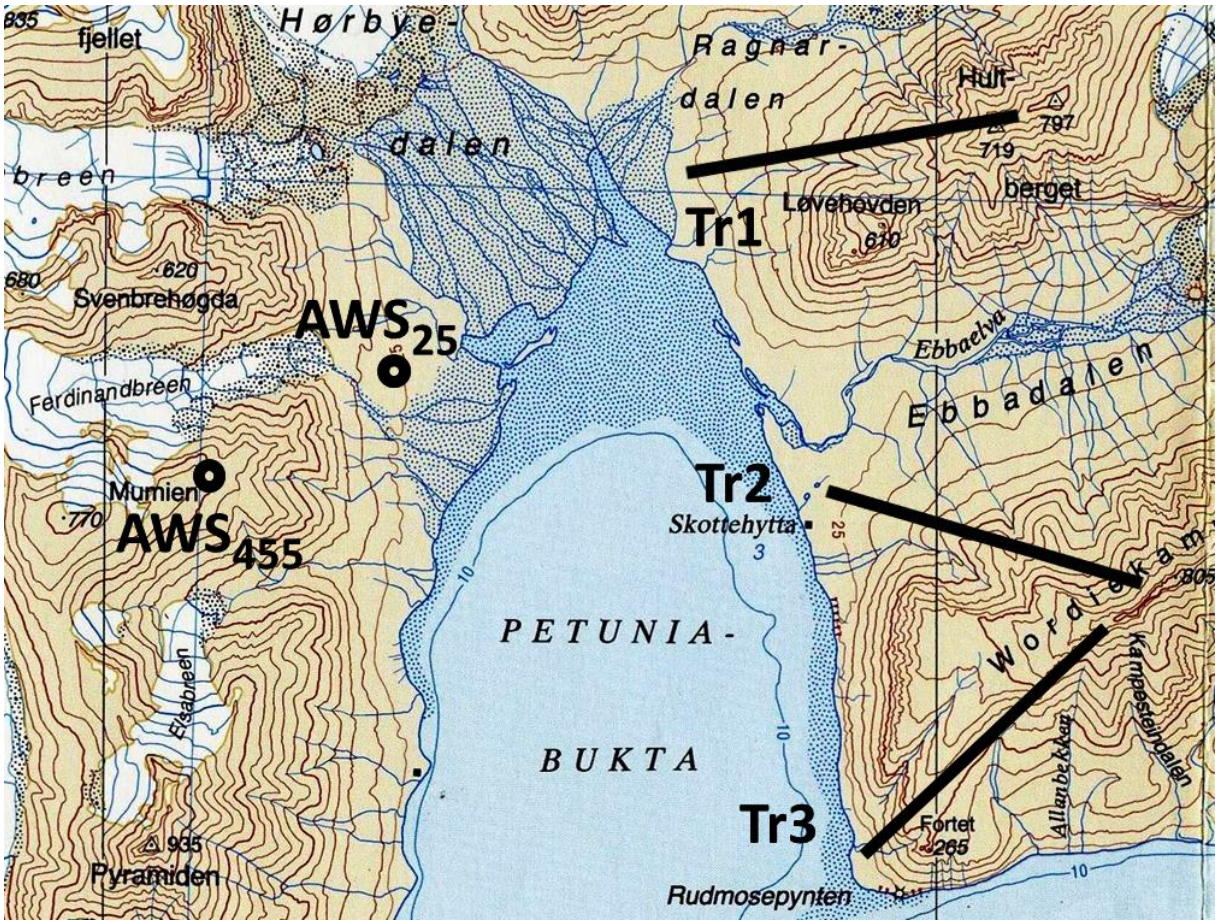

Figure 1. Location of the three investigated transects Tr1–Tr3 and automated weather stations (AWS) in Petunia bay, Billefjorden, Central Spitsbergen. Map source: map sheet C7, Svalbard 1:100 000, Norwegian Polar Institute 2008.

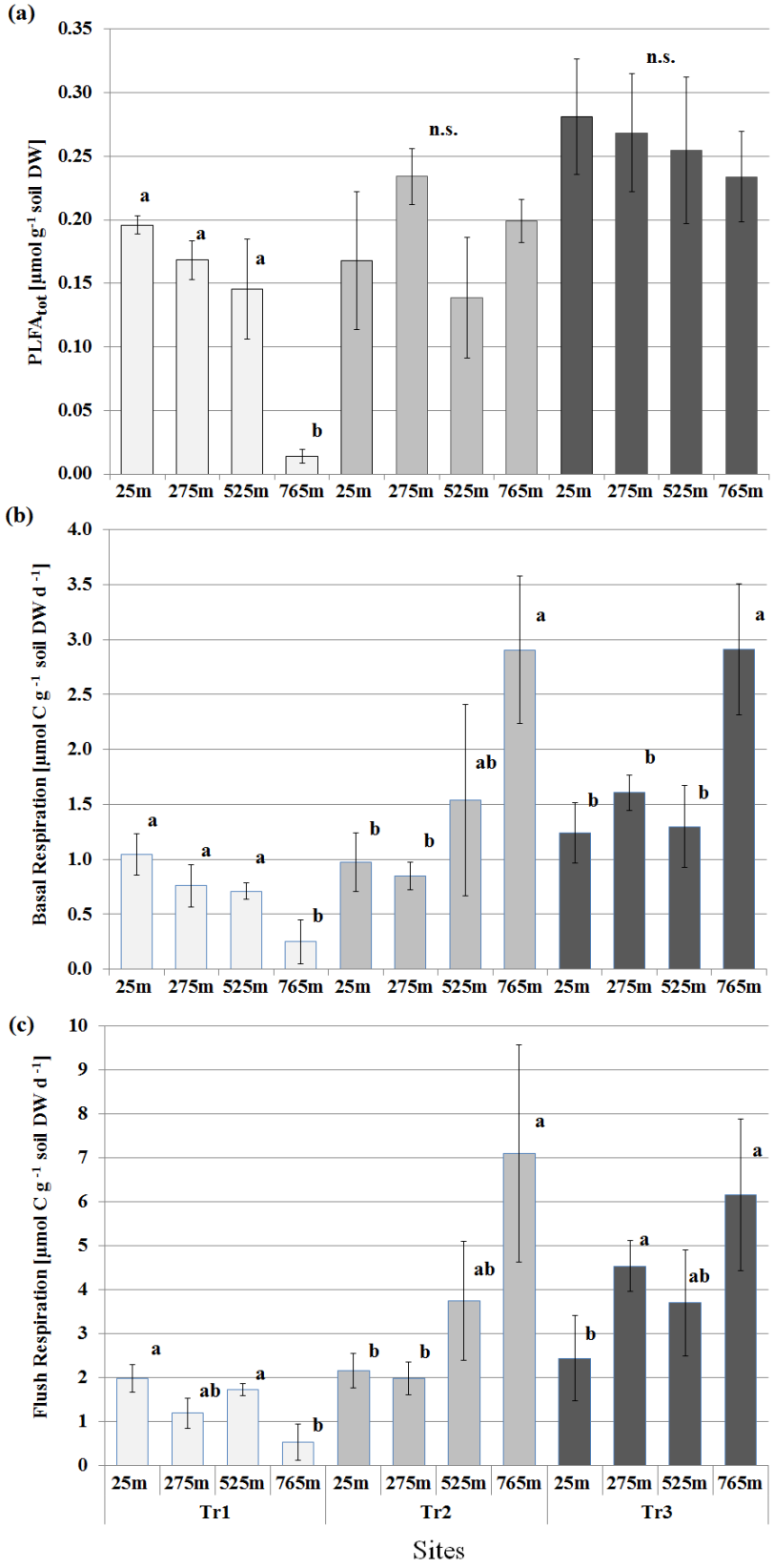

Figure 2. The soil PLFA contents (a), the potential respiration rates (b) and the flush respiration rates (c) in the soils along altitudinal transects (Tr1-Tr3). Error bars indicate mean ± SD (n = 3). Small case letters denote significant differences among altitudes within particular transects ($P < 0.05$; One–way ANOVA combined with Tukey post hoc test).

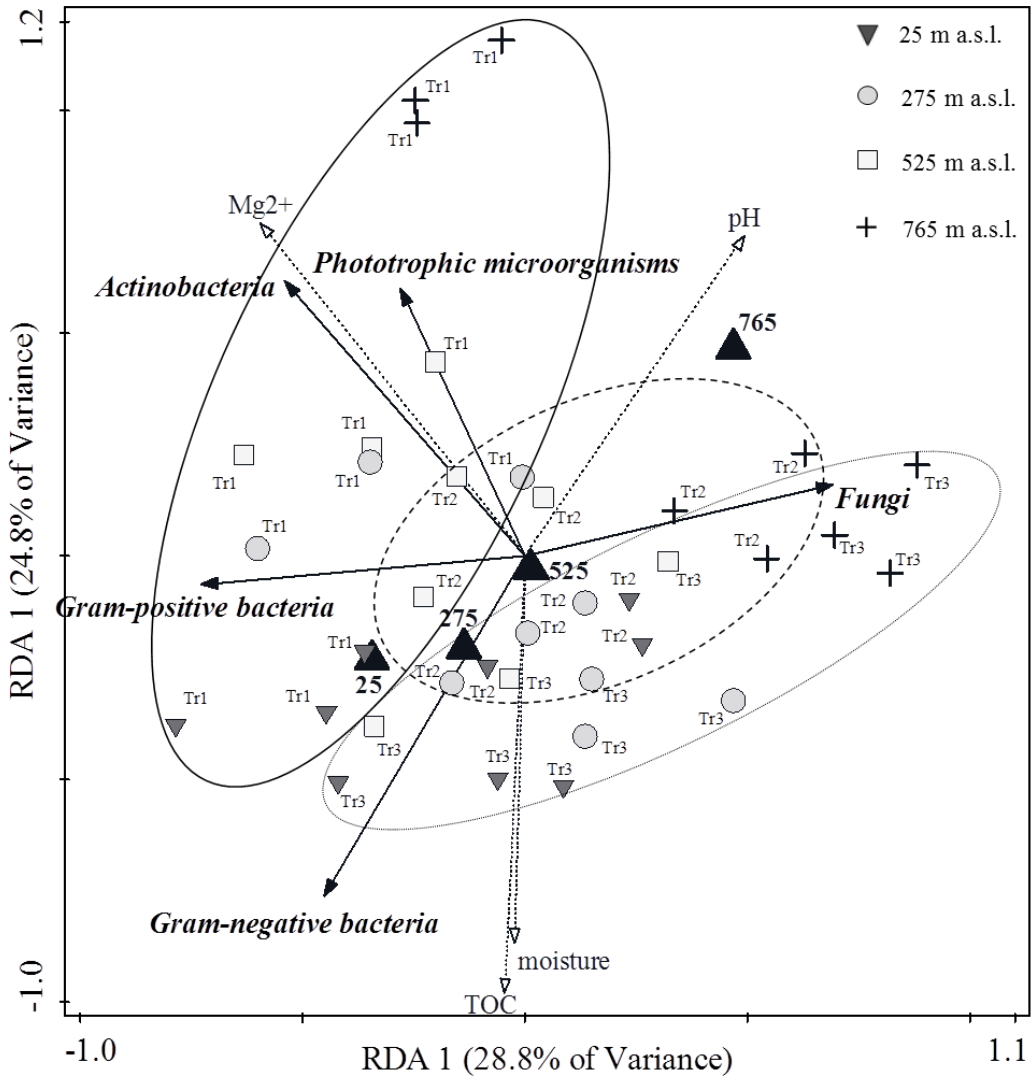

Figure 3. The correlation between abundance of main microbial groups (bold italic) and soil geochemical parameters retained by forward selection of explanatory variables. Results of RDA. Altitude of sampling sites was used as supplementary variable. *Arrows* indicate the direction in which the respective parameter value increases, *solid* lines indicate microbial groups, *dotted* lines indicate selected environmental variables. *Up triangles* are centroids of sites with corresponding elevation (n = 9), numbers indicate elevation (m a.s.l.). The thin solid line encases sites along the Transect 1 (Tr1), the dashed line encases sites along the Transect 2 (Tr2), and the dotted line encases sites along the Transect 3 (Tr3). The numbers in parentheses are the portions of the variation explained by each axis.

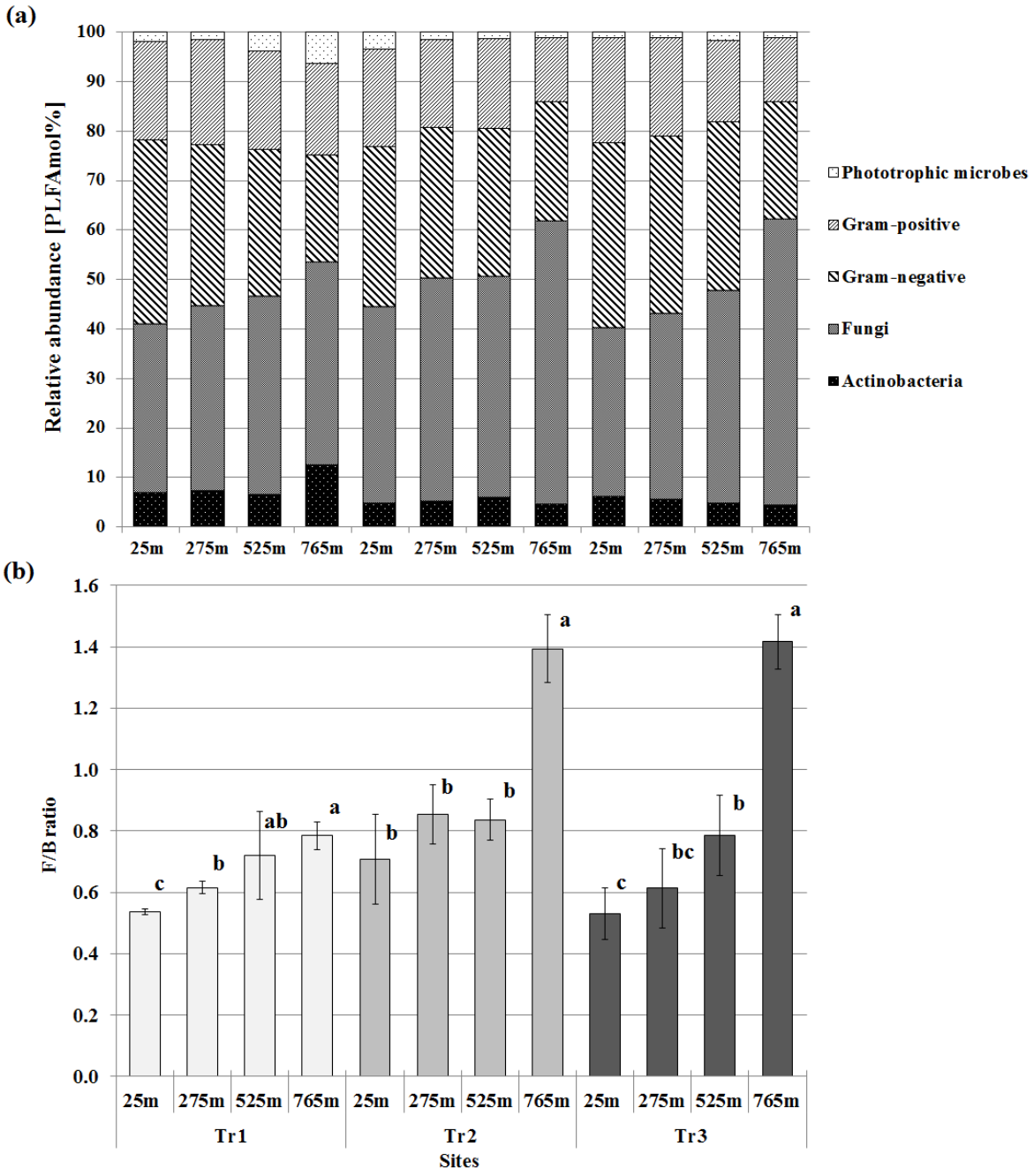

Figure 4. Relative abundance of specific PLFAs within the microbial community (a), and fungi to bacteria (F/B) ratios (b) along altitudinal transects (Tr1-Tr3). Error bars indicate mean ± SD (n = 3). Small case letters denote significant differences between altitudes within particular transects (*P* < 0.05; One–way ANOVA combined with Tukey post hoc test).