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SUPPLEMENTARY INFORMATION

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Permafrost collapse after shrub removal shifts tundra ecosystem to a methane source

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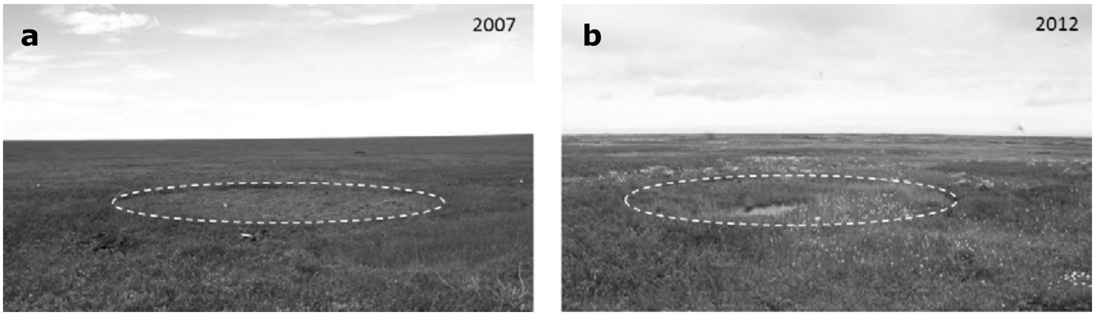
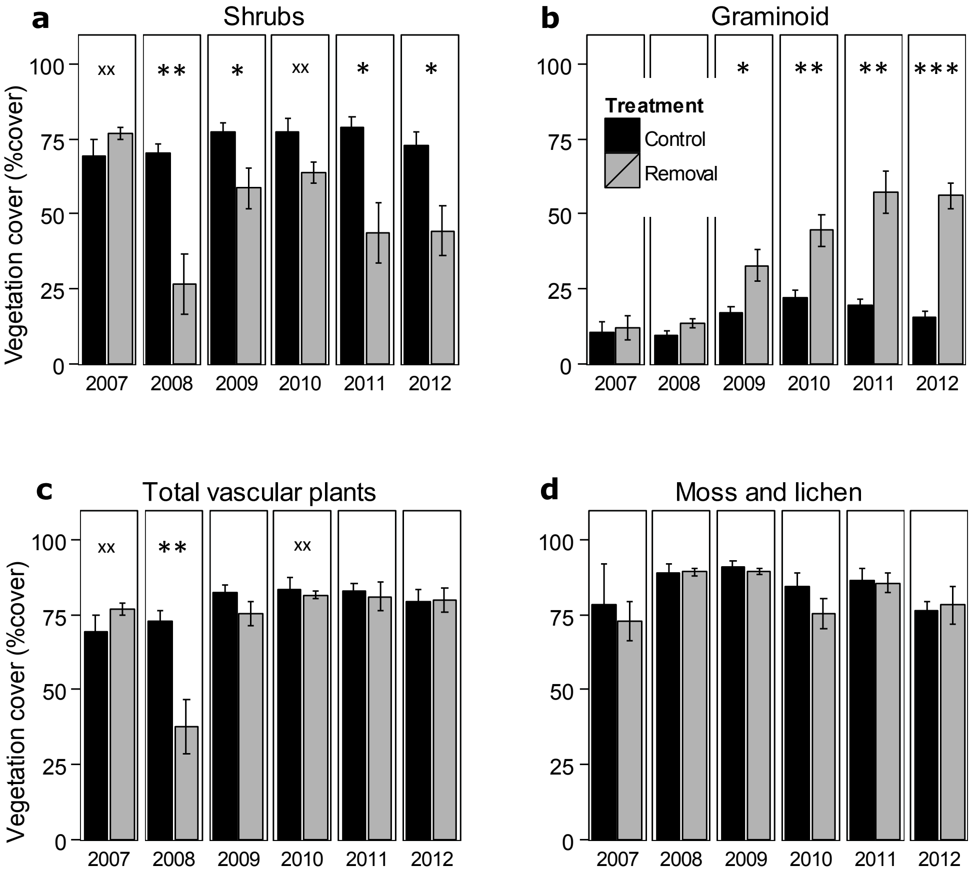


Supplementary Figure 1. Pictures of a removal plot in 2007 and 2012. **a**, shows the *B. nana* removal plot at the start of the experiment in 2007 (after clipping). **b**, shows the appearance of a pool and invasion of flowering graminoids in 2012. The 10-m diameter plot area is delineated by the white dashed line.



Supplementary Figure 2. Vegetation composition in control (black bars) and *B. nana* removal (grey bars) plots from 2007 till 2012. Data are mean values ± standard error, $n = 5$ plots for cover of (**a**) shrubs (deciduous + evergreen shrubs), (**b**) graminoids (grasses + sedges), (**c**) total vascular plants (shrubs + graminoids + forbs), and (**d**) mosses and lichens. Cover data are in percentage of the total number of grid points within the circular 10-m diameter plots. xx indicates years where *B. nana* shrubs have been removed by clipping (note that shrubs have been removed after measuring the vegetation cover). *, ** and *** indicate statistically significant differences between the two treatments ($P < 0.05$, $P < 0.01$, $P < 0.001$, respectively).

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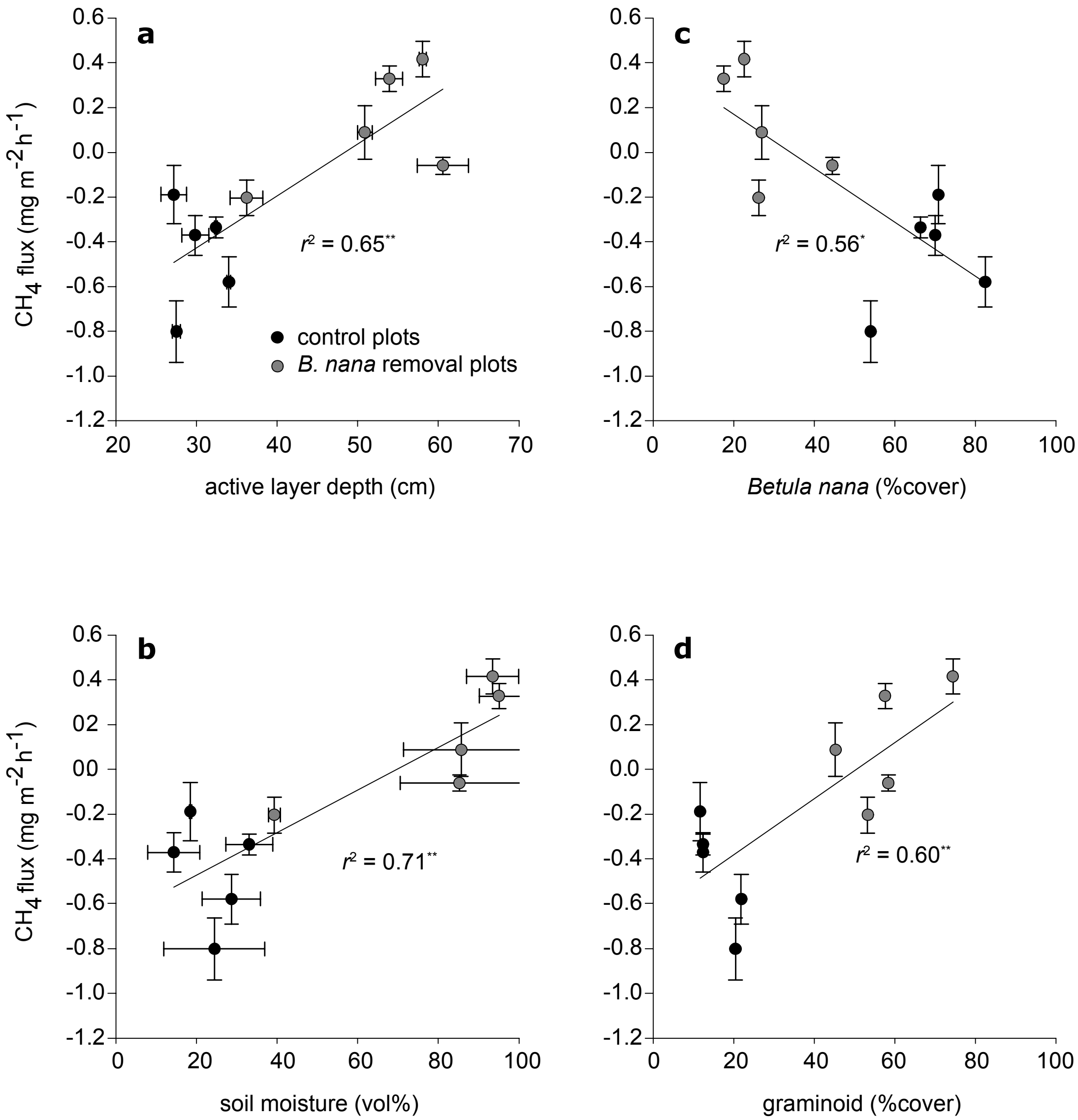

Supplementary Figure 3. Methane (CH_4) fluxes plotted against (**a**) active layer thickness, (**b**) soil moisture, (**c**) *Betula nana* cover, and (**d**) graminoid cover in control (black symbols) and *B. nana* removal (grey symbols) plots. Data are average plot CH_4 fluxes ± standard error (n = 3 measurement dates), measured on 3, 7 and 12 August 2012. No standard error bars are shown for plant cover data, as these were measured only once during the peak of the growing season in mid-July 2012. Linear regression lines with coefficient of determination (r^2) are indicated in each graph. * and ** indicate statistically significant linear relationships ($P < 0.05$, $P < 0.01$, respectively).

Supplementary Table 1. Soil and vegetation measurements at the start of the experiment (July 2007) in control and *B. nana* removal plots. Data are mean values and standard error between brackets, $n = 5$ plots. P-values above 0.05 indicate that there was no statistically significant difference between the two treatments.

Variable	Control		Removal		P-value
Soil measurements (14-18 July 2007)					
Active layer thickness (cm)	22.1	(1.1)	19.8	(1.1)	0.187
Soil moisture (vol. %)	25.7	(2.8)	30.8	(2.8)	0.224
Soil temperature 5 cm depth (°C)	5.7	(0.4)	5.0	(0.4)	0.347
Soil temperature 10 cm depth (°C)	4.4	(0.4)	3.5	(0.3)	0.105
Soil temperature 20 cm depth (°C)	2.4	(0.4)	1.9	(0.3)	0.347
Plant measurements (July 2007)					
Total aboveground biomass (g m^{-2})	414	(45)	430	(88)	0.871
Total leaf biomass (g m^{-2})	80	(8)	85	(14)	0.743
B. nana aboveground biomass (g m^{-2})	379	(42)	374	(79)	0.957
B. nana leaf biomass (g m^{-2})	62	(5)	66	(13)	0.776
B. nana leaf N concentration (mg N g^{-1})	24.1	(1.0)	23.6	(0.0)	0.710
B. nana leaf P concentration (mg P g^{-1})	2.0	(0.0)	2.1	(0.2)	0.856
B. nana leaf K concentration (mg K g^{-1})	6.0	(0.5)	6.7	(0.5)	0.349

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Supplementary Table 2. Temperature and precipitation for the years 2007-2012, observed at the weather station near Chokurdakh (WMO station 21649).

	Annual air temperature (^{0}C)	January air temperature (^{0}C)	July air temperature (^{0}C)	Annual precipitation (mm)	July precipitation (mm)
2007	-10.8	-33.4	11.6	149	9
2008	-12.2	-34.3	9.4	145	21
2009	-12.6	-31.4	8.5	191	25
2010	-11.6	-32.7	15.5	237	8
2011	-12.0	-30.9	12.9	427	94
2012	-12.0	-30.7	9.3	236	17
Mean over 2007-2012	-11.8	-32.2	11.2	231	29

Supplementary Table 3. Plant species cover in control and removal plots for the years 2007-2012. Cover is expressed as percentage of number of hits divided by total number of grid points within the circular 10-m diameter plot. Data are mean values and s.e. between brackets, n = 5 plots.

	2007		2008		2009		2010		2011		2012	
	Control	Removal	Control	Removal	Control	Removal	Control	Removal	Control	Removal	Control	Removal
Deciduous shrub	61.8 (3.5)	66.4 (5.6)	66.6 (2.4)	16.5 (3.1)	73.4 (2.7)	47.9 (5.2)	74.2 (4.5)	55.9 (3.4)	75.0 (4.1)	32.3 (3.7)	70.2 (4.7)	34.5 (6.8)
Betula nana	*59.4 (3.6)*	*60.1 (5.1)*	*65.1 (2.2)*	*12.8 (3.7)*	*72.3 (2.5)*	*42.3 (2.9)*	*72.0 (3.9)*	*50.4 (2.1)*	*85.0 (5.1)*	*26.3 (3.9)*	*68.8 (4.6)*	*27.6 (4.6)*
Salix spp.	*2.3 (1.5)*	*6.0 (2.0)*	*2.3 (0.9)*	*2.9 (1.3)*	*3.2 (1.3)*	*5.8 (3.0)*	*3.6 (1.9)*	*9.5 (3.2)*	*3.2 (1.9)*	*9.1 (3.0)*	*2.6 (1.2)*	*8.2 (3.6)*
Vaccinium uliginosum	*0.0 (0.0)*	*0.3 (0.3)*	*0.0 (0.0)*	*0.7 (0.6)*	*0.0 (0.0)*	*0.7 (0.7)*	*0.0 (0.0)*	*0.4 (0.4)*	*0.0 (0.0)*	*0.6 (0.3)*	*0.0 (0.0)*	*0.3 (0.3)*
Evergreen shrub	7.4 (2.7)	10.2 (6.9)	8.0 (2.6)	12.4 (9.4)	8.5 (2.2)	15.3 (10.3)	9.1 (3.0)	12.4 (7.8)	11.1 (3.1)	15.9 (10.4)	9.2 (2.7)	13.4 (9.4)
Ledum palustre	*0.6 (0.3)*	*0.3 (0.2)*	*0.6 (0.3)*	*0.3 (0.3)*	*0.4 (0.3)*	*0.1 (0.1)*	*0.4 (0.4)*	*0.0 (0.0)*	*0.6 (0.4)*	*0.1 (0.1)*	*0.3 (0.2)*	*0.3 (0.3)*
Vaccinium vitis-idaea	*6.9 (2.6)*	*9.9 (6.8)*	*7.4 (2.6)*	*12.3 (9.3)*	*8.3 (2.2)*	*13.0 (10.8)*	*8.9 (3.1)*	*12.4 (7.8)*	*10.8 (3.1)*	*15.8 (10.3*	*8.9 (2.7)*	*13.3 (9.3)*
Graminoid	10.7 (3.3)	12.0 (4.2)	9.5 (1.3)	13.6 (1.4)	17.1 (1.8)	32.7 (5.3)	22.2 (2.2)	44.5 (5.2)	19.6 (2.2)	57.1 (7.1)	15.5 (2.2)	56.1 (4.3)
Arctagrostis latifolia	*9.1 (3.8)*	*11.7 (4.3)*	*9.3 (1.2)*	*13.4 (1.4)*	*16.9 (1.9)*	*32.3 (5.4)*	*21.9 (2.3)*	*42.9 (4.8)*	*19.0 (2.0)*	*54.0 (5.9)*	*14.3 (2.1)*	*49.6 (3.7)*
Eriophorum vaginatum	*1.5 (1.5)*	*0.1 (0.1)*	*0.1 (0.1)*	*0.1 (0.1)*	*0.1 (0.1)*	*0.4 (0.3)*	*0.0 (0.0)*	*0.3 (0.2)*	*0.3 (0.2)*	*0.4 (0.3)*	*0.7 (0.6)*	*2.3 (0.8)*
Eriophorum angustifolium	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*0.0 (0.0)*	*3.8 (2.2)*	*0.0 (0.0)*	*5.7 (3.2)*
Carex spp.	*0.1 (0.1)*	*0.1 (0.1)*	*0.0 (0.0)*	*0.0 (0.0)*	*1.8 (1.8)*	*0.7 (0.7)*	*0.3 (0.2)*	*1.9 (0.9)*	*0.3 (0.3)*	*0.0 (0.0)*	*0.7 (0.5)*	*0.1 (0.1)*
Moss	57.1 (7.2)	57.2 (6.2)	80.7 (3.1)	82.6 (2.3)	83.6 (2.6)	83.6 (2.9)	74.2 (3.1)	67.9 (5.0)	80.9 (3.4)	79.6 (4.6)	66.3 (4.2)	73.9 (5.0)
Lichen	21.5 (7.3)	15.8 (2.9)	29.3 (5.9)	23.1 (3.8)	26.7 (6.6)	21.5 (3.7)	25.8 (7.3)	15.6 (4.6)	18.2 (7.3)	10.2 (3.7)	16.9 (5.3)	7.2 (3.6)
Forbs	0.1 (0.1)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.1 (0.1)	1.3 (1.0)	0.1 (0.1)	0.3 (0.2)	0.0 (0.0)	0.1 (0.1)	0.0 (0.0)	0.0 (0.0)
Total litter	46.4 (4.6)	52.0 (5.1)	61.2 (1.9)	51.7 (4.0)	69.8 (3.1)	45.7 (2.3)	59.0 (3.3)	47.3 (4.8)	50.8 (5.8)	29.6 (5.1)	41.3 (3.2)	12.6 (2.0)
Standing dead	23.8 (8.4)	21.3 (7.5)	48.8 (3.7)	55.9 (5.4)	51.5 (0.8)	50.9 (4.4)	55.0 (2.9)	60.6 (6.0)	5.3 (2.0)	17.5 (5.5)	17.7 (2.7)	44.4 (3.1)
Bare soil	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.1 (0.1)	0.1 (0.1)	0.1 (0.1)	0.0 (0.0)	0.0 (0.0)
Water	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	2.2 (0.0)	17.8 (6.8)	0.0 (0.0)	5.0 (5.0)

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SUPPLEMENTARY METHODS

Site description

The study was conducted at the Chokurdakh Scientific Tundra Station (70°49'N, 147°29'E, 10 m a.s.l.) located in the Kytalyk (Siberian crane) Wildlife Reserve in the Indigirka lowlands in Northeastern Yakutia, Russian Federation. The site is 28 km northwest of the town Chokurdakh and 150 km south of the East Siberian Sea. The research site is in the Low Arctic climate zone (bioclimatic zone E) and experiences a more continental climate than most other tundra research sites[31]. Chokurdakh mean annual air temperature is −13.4 °C (1981-2010) with −34.0 °C mean January temperature and 10.3 °C mean July temperature. Mean annual precipitation is 196 mm (1981-2010), of which 76 mm (39 %) falls in the summer months June to August. Chokurdakh temperature and precipitation data for the study period are presented in Supplementary Table 1. Climate data were from the GHCN monthly climate dataset (http://www.ncdc.noaa.gov/ghcnm) obtained through the KNMI climate explorer tool (http://climexp.knmi.nl)[32].

The study area is underlain by thick continuous permafrost with a high ice content (> 20 % by volume, Circum-Arctic Map of Permafrost and Ground-Ice Conditions,)[20] which reaches more than 300 m depth. At locations close to our study site, ice contents of 40-95 % by volume have been measured[33]. The lowland mostly consists of floodplains, drained thaw lake basins and higher Pleistocene remnant surfaces. The Circumpolar Arctic Vegetation Map[34] classifies the vegetation of the research area as G4, moist tussock-sedge dwarf-shrub moss tundra, which is a widely occurring tundra type. The field experiment was set up in a former thaw lake that was drained by intersection of the Berelekh River. The microtopography within the drained thaw lake basin consists mostly of elevated shrub patches dominated by *Betula nana* L. (dwarf birch) shrubs within a diffuse drainage network of wet depressions dominated by the graminoid species *Eriophorum augustifolium* Honck. Soils are Gelisol and consist of an organic top layer overlying silty clay parent deposits. The surface organic layer varies in thickness from few cm up to 25 cm, which has together with the overlying moss layer an insulating function, particularly after removal of the shrub layer.

Measurements

At the start of the experiment (July 2007), several vegetation and soil variables were measured in all 10 plots. Destructive harvests in two 50 x 50 cm subplots just outside each of the 10-m diameter plots were made to assess leaf and branch biomass per plant species. Dry weight was determined after oven drying for at least 48 hours at 70 °C. For *B. nana* leaf nitrogen (N), phosphorous (P) and potassium (K) concentrations, each dried sample was grinded and digested with sulphuric acid, salicylic acid, selenium and hydrogen peroxide. Subsequently, the N and P concentrations were measured colorimetrically using a continuous flow analyser (SKALAR SAN plus systems, Breda, The Netherlands). K concentrations were measured by atomic emission flame spectroscopy (Varian AAS, Palo Alto, USA). Soil moisture was measured at nine points in each plot on July 15, 2007 using a handheld ML2x ThetaProbe (Delta-T Devices Ltd., Cambridge, United Kingdom). Soil temperature was measured at nine points in each plot on July 16, 2007 using a K-thermocouple thermometer (Eijkelkamp Agrisearch Equipment, Giesbeek, The Netherlands).

Permafrost thaw depth was assessed by measuring the active layer thickness at nine points in each plot using a bluntly tipped metal probe[10]. For comparison among the years (2007-2012) the active layer thickness measurements between 24th and 28th of July were used.

Surface elevation was measured along two transects in each plot in July 2011 and 2012. The two transects were oriented perpendicular to each other, crossing each other in the plot centre and extending 2-13 meter beyond the plot border until the closest wet depression. Along the transect we measured the surface elevation at 1 meter intervals. The surface elevation was measured using an optical levelling instrument (Kompensator-Nivellier NI 025, Jena, Germany). For a single plot, elevation measurements were obtained with an accuracy of 5 mm or less. For each plot, the elevation measurements from beyond the plot border were averaged and used as a local reference for the elevation measurements within the plot. Although we do not have elevation data from the start of the experiment, we can assume that control and removal plots were very similar prior to the experimental manipulation given their similarities in measured vegetation composition, aboveground biomass, leaf nutrient concentration, active layer thickness, soil moisture and soil temperature during the start of the experiment (Fig. 1, Supplementary Fig. 2, Supplementary Table 1). These data show that there is no reason to assume that there was any difference in methane emission, snow depth or

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other variable among the plots. Assuming equal relative surface elevation at the start, we calculated soil subsidence due to the removal treatment as the average difference in relative surface elevation between the control and removal plots.

Water table depth was measured 13 times at regular intervals during the growing season in 2011 and 2012 using piezometers. Piezometers were installed until the base of the active layer at the beginning of August in 2010. The piezometers were located in the visually deepest location within the plot. The water table was measured relative to the moss/soil surface at the piezometer, since we were interested in the local water table as that determines growing conditions for vegetation. Note that in case no water level was present, the value of active layer thickness was used instead. Snow depth was measured in May 2008 and 2009 at five randomly chosen points in each plot using a bluntly tipped metal probe. In April 2012 snow depth was measured at nine points in each plot following the same scheme as used for the measurements of active layer thickness.

Vegetation cover and species composition were assessed annually in July by taking point intercept measurements on a grid of 13 by 13 points spaced 75 cm apart[10]. Species cover was calculated as the number of hits divided by the 137 grid points within the circular 10-m diameter plot.

Methane (CH_4) flux measurements were made using the closed chamber method in all 10 plots on 3, 7 and 12 August 2012 using an INNOVA 1312 photoacoustic field gas monitor (LumaSense Technologies A/S, Ballerup, Denmark). A clear Plexiglas cylindrical flux chamber measuring 29 cm diameter and 28 cm tall was attached to flux chamber collar rings (30 cm diameter, 10 cm tall) installed to a depth of approximately 5 cm into the soil of each plot. The flux chamber was covered with a dark cloth during flux measurements. During each flux measurement run, the flux chamber was attached to the collar ring for approximately 25 minutes, during which time 20 readings of atmospheric CH_4 and water vapour (H_2O) concentration were made. Logging started approximately 90 seconds after placing of the chamber to prevent potential disturbance-related release of gases affecting fluxes. The air volume inside the chamber was circulated using a battery-powered fan to ensure homogenous air mixing. Concentrations of H_2O inside the chamber were controlled by passing the sampled air through a tube containing dry silica gel before entering the gas monitor and prevent cross-interference at high concentrations with CH_4 [34,35]. Soil volumetric water content was measured after each flux measurement at 3 points inside the collar ring at 10 cm depth using a

Decagon 5TE soil moisture sensor (Decagon devices, Pullman WA, USA). Air temperature and inner collar rim height were measured after each measurement run to adjust for gas volume size inside the chamber.

Data analyses

The effect of the *B. nana* removal treatment on active layer thickness, relative surface elevation, water level, snow depth and vegetation cover, was assessed using a repeated measures analysis of variance (RM-ANOVA) with treatment (control, removal) as between-subject factor and year as within-subject factor. Subsequently, differences between control and removal treatment in specific years were analysed using one-way ANOVA. As plot pair generally had no effect on the measured variables, it was not included in the ANOVA. All data were tested for normal distribution and equality of variance. These statistical analyses were performed using SPSS 19.

Methane fluxes were calculated by fitting 2nd-order polynomial equations to each flux measurement run and using the slope at t = 1 s (at t = 0 logging of chamber CH_4 concentrations started) to calculate a flux. A quality control check for stability of atmospheric H_2O concentrations was performed for each run, whereby data points fluctuating more than 10% from the previous measured value were discarded. This resulted in the exclusion of 7.2% of all gas concentration readings, with CH_4 fluxes being calculated from the part of the measurement run when H_2O was stable.

The effect of *B. nana* removal on the CH_4 fluxes was assessed using the PROC MIXED model procedure in SAS. Pair was included as a random factor, taking into account the pairwise selection of the plots during the establishment of the experiment in 2007. Day of measurement was included as repeated factor in order to avoid temporal pseudo-replication, with autoregressive (AR1) as covariance structure to take into account potential autocorrelation of CH_4 fluxes within plot among measurement days. Fixed effects were estimated using the Restricted Maximum Likelihood (REML) method, with degrees of freedom estimated by the Kenward and Rogers method. Pearson's correlation coefficients *r* were calculated between 3-day average CH_4 fluxes and active layer thickness, soil moisture, plot fractional cover of *B. nana*, and plot fractional cover of graminoid species. These statistical analyses were performed using SAS 9.4.

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