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Alster, CJ

Allison, SD

Treseder, KK

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Title: Carbon budgets for soil and plants respond to long-term warming in an Alaskan boreal forest

Authors: Charlotte J. Alster¹, Steven D. Allison^{1,2}, Kathleen K. Treseder¹

Affiliations:

¹Ecology and Evolutionary Biology, University of California Irvine, Irvine, CA 92697, USA

²Department of Earth System Science, University of California, Irvine, Irvine, CA 92697, USA

Correspondence:

Charlotte J. Alster

Department of Ecology and Evolutionary Biology

University of California, Irvine

Irvine, CA

+0019498249423

Charlotte.Alster@uci.edu

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25 **Abstract**

26 The potential consequences of global warming for ecosystem carbon stocks
27 are a major concern, particularly in high-latitude regions where soil carbon
28 pools are especially large. Research on soil and plant carbon responses to
29 warming are often based on short-term (<10 year) warming experiments.
30 Furthermore, carbon budgets from boreal forests, which contain at least 10-
31 20% of the global soil carbon pool, have shown mixed responses to warming.
32 In this study, we measured carbon and nitrogen budgets (i.e., soil and
33 understory vegetation carbon and nitrogen stocks) from a 13-year
34 greenhouse warming experiment in an Alaskan boreal forest. Although there
35 were no differences in total aboveground + belowground pools, the carbon in
36 the moss biomass and in the soil organic layer significantly decreased with
37 the warming treatment (-88.3% and -19.1%, respectively). Declines in moss
38 biomass carbon may be a consequence of warming-associated drying, while
39 shifts in the soil microbial community could be responsible for the decrease
40 in carbon in the soil organic layer. Moreover, in response to warming,
41 aboveground plant biomass carbon tended to increase while root biomass
42 carbon tended to decrease, so carbon allocation may shift aboveground with
43 warming. Overall these results suggest that permafrost-free boreal forests
44 are susceptible to soil carbon loss with warming.

45

46 **Introduction**

47   Global warming is expected to alter the amount of carbon stored in
48 soils and plants; however, it is unclear whether warming will lead to a net
49 loss or gain of carbon (C). Soil C stocks are the balance of inputs and outputs
50 (Melillo et al. 2011; Lu et al. 2013). The effect of warming on soil C stocks,
51 especially in high-latitude areas (above 60°N) with large C pools (Dixon et al.
52 1994; Hobbie et al. 2000), depends on the magnitude of change associated
53 with these C fluxes and on their temperature sensitivity (Knorr et al. 2005;
54 Sistla et al. 2013). Soil carbon losses can occur due to increased microbial
55 and enzyme activity (Schimel et al. 2004; Davidson and Janssens 2006). On
56 the other hand, soil C may increase with warming through increased litter
57 inputs and root production (Majdi and Ohrvik 2004; Rinnan et al. 2008).
58 Further complicating predictions of C gain or loss under warming are
59 changes in soil moisture (Lavelle et al. 1993; Davidson et al. 2000; Saleska
60 et al. 2002; Xu et al. 2015). For example, while warming may stimulate
61 decomposition and plant production, warming-associated drying may
62 decrease decomposition and plant growth. This type of interaction
63 complicates predictions of net C gains and losses (van Gestel et al. 2018).

64   Determining how boreal forests will respond to warming, and
65 associated drying, is of particular interest. Boreal forests contain at least 10-
66 20% of global soil C (Jobbagy and Jackson 2000; Allison and Treseder 2011;
67 Pan et al. 2011), and their high latitude distribution makes them especially
68 vulnerable to climate change since warming in these regions is expected to

69 occur faster (Chapin et al. 2000; Shukla et al. 2019). Here, we took
70 advantage of a 13-year greenhouse warming experiment in an Alaskan
71 boreal forest to examine aboveground and belowground C and N budgets.
72 Previous studies at this site have shown that, when compared to control
73 plots, warmed plots have reduced fungal abundance and increased fungal
74 diversity (Allison and Treseder 2008; Treseder et al. 2016). Fungal
75 decomposers tend to dominate under warming and drying because of their
76 drought-resistant growth forms (Barnard et al. 2013; Treseder and Lennon
77 2015) and their ability to decompose recalcitrant compounds (e.g., cellulose
78 and lignin), which may become more abundant with warming due to changes
79 in the plant community (Mcguire et al. 2010; Fontaine et al. 2011; Xiong et
80 al. 2014). Fungi tend to specialize on recalcitrant compounds that may have
81 higher temperature sensitivities for decomposition, thus conferring an
82 advantage over bacteria which tend not to target recalcitrant compounds
83 (Romero-Olivares et al. 2017). Warmed plots at this site were also found to
84 have slower litter decomposition (Romero-Olivares et al. 2017), increased
85 cellulose- and starch-degrading enzyme production (German and Allison
86 2015), and lowered respiration rates (German and Allison 2015). However,
87 no studies have determined whether any of these changes in decomposition
88 dynamics have elicited measurable changes in soil and plant C and N pools.
89 A fuller understanding of the relationship between aboveground and
90 belowground C and N dynamics will provide better insight into whether or

not ecosystem C gains or losses should be expected with warming in this critical ecosystem.

We hypothesized that the greenhouse warming treatment would reduce soil C storage owing to greater activity of recalcitrant C decomposers and decrease above- and below-ground plant biomass owing to water limitation (Hypothesis 1; Figure 1A). Warming-associated drying could also increase soil C storage by inhibiting decomposer activity, while the warming itself could augment above- and below-ground plant biomass by alleviating temperature or nutrient limitation of plants (Hypothesis 2; Figure 1A). Alternatively, if decomposer activity and plant biomass inputs simultaneously increase or decrease, or if previously reported changes at our site are ephemeral, soil C storage would remain the same (Null Hypothesis). To test these hypotheses, we compared changes in understory aboveground and belowground C and N pools from greenhouse warmed and control plots in order to better understand boreal forest ecosystem response to long-term warming treatment.

Methods

Our study site is located in a mature black spruce (*Picea mariana*) boreal forest on the Fort Greely military base near Delta Junction, Alaska, USA (63°55'N, 145°44'W). The understory vegetation is dominated by mosses, lichens, and shrubs (*Vaccinium uliginosum*, *V. vitis-idaea*, *Ledum groenlandicum*, *Empetrum nigrum*, and *Betula glandulosa*) (Treseder et al.

114 2004). The soil is an Inceptisol, with an average organic horizon of 9.8 cm
115 (King et al. 2002) and pH of 4.9 (Hanson et al. 2008).

116 In 2005, a greenhouse warming experiment was established with five
117 pairs of 2.5 x 2.5 m plots within a 1 km² area in the open canopy forest, as
118 described in Allison and Treseder (2008). In each pair, one plot was covered
119 with a wood frame structure covered in greenhouse plastic film in order to
120 warm the plots. Gaps between the frame and the plastic allowed air to
121 circulate, and gutters and tubing allowed water to flow in; the other plot was
122 left unmanipulated as a control (Allison and Treseder 2008). All plots
123 excluded large trees. Air temperature increased by an average of 1.6°C and
124 Onset HOBO data loggers recorded an average of 0.5°C increase in soil
125 temperature at 5 cm depth (Table S1; Allison and Treseder 2008). As a result
126 of the greenhouse warming treatment, soil moisture also decreased by an
127 average of 22% in the warmed plots (Table S1; Allison and Treseder 2008).
128 Passive warming approaches, such as this one, not only warm and dry but
129 can also alter temperature variation, light intensity, CO₂ concentration, wind
130 speed, snow cover, and herbivory (Kennedy 1995; Aronson and McNulty
131 2009; Bokhorst et al. 2011). However, in remote areas where line power is
132 not available, greenhouse warming is a practical and cost-efficient way to
133 elevate temperature. Passive warming treatments are also good at
134 minimizing soil disturbance compared to other approaches like heated cables
135 (Aronson and McNulty 2009). Here, when we refer to the *warming treatment*,

we are referring to the collective change in the microclimate which includes, but is not limited to, warming and drying.

In July 2017, two 0.6 x 0.6 m subsamples of aboveground biomass were collected from each plot. The two subsamples were combined in the field and the contents were sorted into different bags by moss and plant type. Two soil cores (7 cm in diameter x 12 cm in depth) were taken from each plot and divided into three fractions: lichens, O (organic) horizon, and A (mineral) horizon. Depth of the soil organic layer is reported in the supplement (Table S2). We then combined fractions of the same type in the field. Samples were kept cool during transportation and subsequently stored at -20°C until processing at the University of California, Irvine. In the lab, we separated the vegetation samples into herbaceous (leaves and stems) and woody biomass (see Table S3 for list of plant species and for how plant types were categorized). The O and A soil horizons were sieved and separated manually into root biomass, soil, and other organic matter biomass components. We estimated bulk density of the soil horizons by calculating soil volumes and dry weights. We separated the lichen fraction into lichen biomass, litter, and soil components (see Table S4 for approximate percentages). However, since it was difficult to disentangle these components precisely, the lichen fraction was treated as a single unit in subsequent analyses. After final partitioning of each of the plant, moss, and soil samples, all samples were dried at 60°C, weighed for biomass, and subsamples were finely ground using a ball mill. The subsamples were then

159 combusted for C:N using a Thermo Scientific FlashEA 1112 Nitrogen and
160 Carbon analyzer. Pool size for each aboveground and belowground
161 component was estimated using the biomass, bulk density (when
162 applicable), and elemental analysis data. It was not possible to estimate pool
163 size for the mineral horizon because, unlike the organic horizon, we did not
164 sample the entire horizon.

165 Data were tested for normality and log-transformed if needed. Two-
166 tailed, paired *t*-tests at $\alpha < 0.05$ were conducted to test for differences
167 between the paired control and warmed plots. We also used a generalized
168 linear model, weighted by mass, to check for differences in plant community
169 composition in the plots and treatments in R version 3.4.1 (R Core Team
170 2017). Correlations were tested (also in R) to identify relationships between
171 the aboveground and belowground variables measured. Because our sample
172 size was relatively small (five pairs of plots), we conducted a power analysis
173 in G*Power (Erdfelder et al. 2009) to determine the sample size needed to
174 achieve a power of 0.80.

175

176 **Results**

177 After 13 years of the greenhouse warming treatment, soil C in the
178 organic layer decreased by 19.1% in the warmed treatment (Table 1; *P* =
179 0.048), while understory aboveground biomass trended towards C stocks
180 increasing by 1.5- to 4-fold (except for moss). Total aboveground biomass C
181 was higher under the warmed plots, but the difference was not statistically

significant (Table 1; $P = 0.236$). However, separating the data by vegetation type, we observed that moss decreased by 88.3% with greenhouse warming (Table 1; $P = 0.034$). In contrast, the C biomass of herbaceous vegetation, woody vegetation, and lichen increased with the warming treatment, although these trends are only marginally significant at most ($P = 0.166$, $P = 0.069$, and $P = 0.285$, respectively). Based on the power analysis, we suspect that we may have detected significance with a larger sample size (Table S5). In terms of aboveground biomass of individual plant species, there were no significant differences between the treatments (Table S6). In addition, soil organic horizon depth did not change with the warming treatment ($P = 0.922$).

The greenhouse warming treatment increased allocation of aboveground vegetative C by 13%. Root biomass decreased in the warming treatment by nearly a third, albeit non-significantly ($P = 0.595$), while total aboveground vegetation increased (Table 1). There was also a positive relationship between root biomass C and moss biomass C ($R = 0.698$; $P = 0.025$; Table S7). However, no other significant relationships between aboveground and belowground C pools were evident (Table S7).

Percent C was higher for herbaceous and woody vegetation in the greenhouse warmed plots, but this trend was not statistically significant (Table 2; $P = 0.270$ and $P = 0.108$, respectively). Percent N was significantly lower in warmed plots for both herbaceous and woody vegetation (Table 2; $P = 0.044$ and $P = 0.028$, respectively), resulting in higher C:N ratios in the

205 warmed plots (Table 2; herbaceous $P = 0.055$; woody $P = 0.118$). Percent C
206 and %N in the soil layers did not change significantly with the warming
207 treatment (Table 2).

208

209 **Discussion**

210 To our knowledge, this work is the first to directly measure how C and
211 N pools respond to long-term warming in an Alaskan, permafrost-free boreal
212 forest. We found that the long-term greenhouse warming treatment
213 significantly reduced soil C in the organic layer and moss biomass C (Figure
214 1B). At the same time, aboveground plant biomass C tended to increase
215 while root C tended to decrease. Altogether, the distribution of C stocks
216 within this ecosystem tended to shift from belowground to aboveground in
217 response to the warming treatment.

218 These results are important because permafrost-free boreal forests are
219 understudied, yet represent approximately 45-60% of all boreal forests
220 (Allison and Treseder 2011). Forests cover over 30% of Earth's land surface,
221 with more than a third of that coming from boreal forests (Bonan 2008).
222 Understanding the uncertainties associated with these systems' responses
223 provides greater clarity for biogeochemical model parameterization. These
224 findings also corroborate other permafrost-free boreal forest studies (Niinisto
225 et al. 2004; Bronson et al. 2008), which find that CO₂ fluxes from boreal
226 forest soils increase with warming. Altogether, these results suggest that
227 warming can alter C pools in boreal forests lacking permafrost.

228   We found that the soil organic layer experienced a 19.1% reduction in
229 C with the warming treatment, supporting Hypothesis 1. Declines in soil C
230 are consistent with results of a prior study at this site predicting soil C loss
231 with warming due to shifts in the fungal community, which could improve
232 breakdown of recalcitrant C (Treseder et al. 2016). In contrast, other studies
233 from this site found that warming and drying during the growing season
234 suppressed microbial activity and decomposition (Allison and Treseder 2008;
235 Romero-Olivares et al. 2017), suggesting a delay in soil C loss. However, in
236 the context of our results, this suppression of microbial activity may be
237 reflective of only the short-term or seasonal response (Schmidt et al. 2007).
238 This could be due to the relatively quick successional changes of the
239 microbial community (Schmidt et al. 2007; Voriskova and Baldrian 2013) or
240 depletion of the labile organic matter (Knorr et al. 2005). Microbial activity
241 may return to pre-disturbance levels or increase once the community is
242 adapted to the new environmental conditions (Allison et al. 2010; Karhu et
243 al. 2014). Since aboveground C biomass increased with the warming
244 treatment, it is unlikely that lower aboveground litter production was
245 responsible for the decline in soil C.

246   Declines in soil C in the organic layer with long-term warming are
247 common (Kane and Vogel 2009; DeAngelis et al. 2015). However, a recent
248 study from a boreal forest in Eastern Canada contrasts our results. They
249 found no change in soil C stocks with 9 years of warming treatment (Marty et
250 al. 2019). This discrepancy could be due to differences in topography, C

251 quality, soil depth, or experimental design between the Eastern Canadian
252 study and ours.

253   Also in support of Hypothesis 1, moss biomass C decreased with
254 warming by 88.3%. This considerable decline in moss could potentially be
255 attributed to greater susceptibility to water loss with warming (Charron and
256 Quatrano 2009) since moisture declined in our warmed plots by 22% (Allison
257 and Treseder 2008). However, in support of Hypothesis 2, the herbaceous
258 and woody vegetation increased with the warming treatment (Figure 1B).
259 This increase in aboveground vegetation could be due to removal of
260 temperature limitations (Hobbie et al. 1999) or the ability to produce deeper
261 roots to acquire water (Comas et al. 2013; Lindh et al. 2014). These
262 increases in aboveground vegetation could also be an unintended
263 consequence of the greenhouse warming treatment, such as decreased
264 herbivory (Aronson and McNulty 2009). In contrast, the greenhouse warming
265 treatment reduced photosynthetic active radiation by 30-40% (Allison and
266 Treseder 2008), yet we found an increase in photosynthetic biomass.
267 Perhaps with a different warming technique, the aboveground C response to
268 warming would be more pronounced. The decrease in moss and increase in
269 herbaceous and woody vegetation may neutralize the effect of the warming
270 treatment on total aboveground C pools.

271   In addition to these changes in aboveground biomass pools, root
272 biomass C declined with the warming treatment, although not significantly
273 (Table 1). Additionally, root and moss biomass C positively correlated (Table

274 S7). These results are surprising given our predictions about moisture
275 structuring the vegetation response. However, since aboveground vegetation
276 %N decreased with the warming treatment (increasing aboveground
277 vegetation C:N), perhaps N limitation plays a role. While warming is typically
278 thought to decrease N limitation owing to increased N mineralization (Rustad
279 et al. 2001), low soil moisture can slow N mineralization (Beier et al. 2008).
280 This mechanism could elicit lower root growth with moisture loss.
281 Alternatively, perhaps warming and drying resulted in a deeper rooting
282 system to acquire water and nitrogen that was not captured in our
283 experiment. Inclusion of deeper soil horizons, and their roots, may provide
284 additional clarity, especially since C and N stocks in boreal forests can be
285 substantial in these deeper layers (Kane et al. 2005; Vogel et al. 2005; Marty
286 et al. 2015, 2017). Additionally, we only measured understory aboveground
287 biomass. Inclusion of trees may further explain the relationship between
288 aboveground and belowground pools in this critical ecosystem.

289   In conclusion, we did not find that warming led to a net loss or gain of
290 C. However, our results suggest that permafrost-free boreal forests are
291 susceptible to C loss from soil and moss with warming. Carbon losses from
292 soil and moss represent approximately 9% and 4% of the total C stock
293 calculated in our experiment, respectively. These changes could contribute
294 to climate change if increases in herbaceous and woody biomass no longer
295 compensate for these losses. Furthermore, if vegetative C continues to
296 transfer from belowground to aboveground, we might expect habitat shifts

297 and future changes to C stocks. Aboveground pools may be less stable than
298 belowground pools for storing C (Zhou et al. 2006), so an increase in the
299 proportion of aboveground C could cause additional C losses with
300 disturbance. Knowledge of individual warming responses from different C
301 pools improves mechanistic understanding of ecosystem responses to
302 climate change, which is especially important for this vulnerable forest
303 system.

304

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312

313 **References**

- 314 Allison SD, Treseder KK (2011) Climate change feedbacks to microbial
315 decomposition in boreal soils. *Fungal Ecol* 4:362–374. doi:
316 10.1016/j.funeco.2011.01.003
- 317 Allison SD, Treseder KK (2008) Warming and drying suppress microbial
318 activity and carbon cycling in boreal forest soils. *Glob Chang Biol*
319 14:2898–2909. doi: 10.1111/j.1365-2486.2008.01716.x

320 Allison SD, Wallenstein MD, Bradford MA (2010) Soil-carbon response to
 321 warming dependent on microbial physiology. *Nat Geosci* 3:336–340
 322 Aronson EL, McNulty SG (2009) Appropriate experimental ecosystem
 323 warming methods by ecosystem, objective, and practicality. *Agric For*
 324 *Meteorol* 149:1791–1799. doi: 10.1016/j.agrformet.2009.06.007
 325 Barnard RL, Osborne CA, Firestone MK (2013) Responses of soil bacterial and
 326 fungal communities to extreme desiccation and rewetting. *ISME J*
 327 7:2229–2241. doi: 10.1038/ismej.2013.104
 328 Beier C, Emmett BA, Peñuelas J, et al (2008) Carbon and nitrogen cycles in
 329 European ecosystems respond differently to global warming. *Sci Total*
 330 *Environ* 407:692–697. doi: 10.1016/j.scitotenv.2008.10.001
 331 Bokhorst S, Huiskes A, Convey P, et al (2011) Microclimate impacts of
 332 passive warming methods in Antarctica: Implications for climate change
 333 studies. *Polar Biol* 34:1421–1435. doi: 10.1007/s00300-011-0997-y
 334 Bonan GB (2008) Forests and climate change: forcings, feedbacks, and the
 335 climate benefits of forests. *Science* (80-) 320:1444–1450
 336 Bronson DR, Gower ST, Tanner M, et al (2008) Response of soil surface CO₂
 337 flux in a boreal forest to ecosystem warming. *Glob Chang Biol* 14:856–
 338 867. doi: 10.1111/j.1365-2486.2007.01508.x
 339 Chapin FS, Mcguire AD, Randerson J, et al (2000) Arctic and boreal
 340 ecosystems of western North America as components of the climate
 341 system. *Glob Chang Biol* 6:211–223. doi: 10.1046/j.1365-
 342 2486.2000.06022.x

343 Charron AJ, Quatrano RS (2009) Between a rock and a dry place: The water-
 344 stressed moss. *Mol Plant* 2:478–486. doi: 10.1093/mp/ssp018
 345 Comas LH, Becker SR, Cruz VM V., et al (2013) Root traits contributing to
 346 plant productivity under drought. *Front Plant Sci* 4:1–16. doi:
 347 10.3389/fpls.2013.00442
 348 Davidson EA, Janssens IA (2006) Temperature sensitivity of soil carbon
 349 decomposition and feedbacks to climate change. *Nature* 440:165–173
 350 Davidson EA, Trumbore SE, Amundson R (2000) Biogeochemistry: soil
 351 warming and organic carbon content. *Nature* 408:789–790
 352 DeAngelis KM, Pold G, Topcuoglu BD, et al (2015) Long-term forest soil
 353 warming alters microbial communities in temperate forest soils. *Front*
 354 *Microbiol* 6:. doi: 10.3389/fmicb.2015.00104
 355 Dixon RK, Brown S, Houghton RA, et al (1994) Carbon pools and flux of global
 356 forest ecosystems. *Science* (80-) 263:185–190. doi:
 357 10.1126/science.263.5144.185
 358 Erdfelder E, FAul F, Buchner A, Lang AG (2009) Statistical power analyses
 359 using G*Power 3.1: Tests for correlation and regression analyses. *Behav*
 360 *Res Methods* 41:1149–1160. doi: 10.3758/BRM.41.4.1149
 361 Fontaine S, Henault C, Aamor A, et al (2011) Fungi mediate long term
 362 sequestration of carbon and nitrogen in soil through their priming effect.
 363 *Soil Biol Biochem* 43:86–96. doi: 10.1016/j.soilbio.2010.09.017
 364 German DP, Allison SD (2015) Drying and substrate concentrations interact
 365 to inhibit decomposition of carbon substrates added to combusted

366 Inceptisols from a boreal forest. *Biol Fertil soils* 51:525–533. doi:
 367 10.1007/s00374-015-0998-z
 368 Hanson CA, Allison SD, Bradford MA, et al (2008) Fungal taxa target different
 369 carbon sources in forest soil. *Ecosystems* 11:1157–1167. doi:
 370 10.1007/s10021-008-9186-4
 371 Hobbie SE, Schimel JP, Trumbore SE, Randerson JR (2000) Controls over
 372 carbon storage and turnover in high-latitude soils. *Glob Chang Biol*
 373 6:196–210. doi: 10.1046/j.1365-2486.2000.06021.x
 374 Hobbie SE, Shevtsova A, Chapin FS (1999) Plant responses to species
 375 removal and experimental warming in Alaskan tussock tundra. *Oikos*
 376 84:417–434
 377 Jobbagy EG, Jackson RB (2000) The vertical distribution of soil organic carbon
 378 and its relation to climate and vegetation. *Belowgr Process Glob Chang*
 379 10:423–436
 380 Kane ES, Valentine DW, Schuur EAG, Dutta K (2005) Soil carbon stabilization
 381 along climate and stand productivity gradients in black spruce forests of
 382 interior Alaska. *Can J For Res* 35:2118–2129. doi: 10.1139/x05-093
 383 Kane ES, Vogel JG (2009) Patterns of total ecosystem carbon storage with
 384 changes in soil temperature in boreal black spruce forests. *Ecosystems*
 385 12:322–335. doi: 10.1007/s10021-008-9225-1
 386 Karhu K, Auffret MD, Dungait JA, et al (2014) Temperature sensitivity of soil
 387 respiration rates enhanced by microbial community response. *Nature*
 388 513:81–84. doi: 10.1038/nature13604

389 Kennedy AD (1995) Simulated climate change: are passive greenhouses a
 390 valid microcosm for testing the biological effects of environmental
 391 perturbations? *Glob Chang Biol* 1:29–42. doi: 10.1111/j.1365-
 392 2486.1995.tb00004.x

393 King S, Harden J, Manies KL, et al (2002) Fate of carbon in Alaskan
 394 Landscapes Project: database for soils from eddy covariance tower sites,
 395 Delta Junction, AK. Open-File Rep 20

396 Knorr W, Prentice IC, House JI, Holland EA (2005) Long-term sensitivity of soil
 397 carbon turnover to warming. *Nature* 433:298–301

398 Lavelle P, Blanchart E, Martin A, et al (1993) A hierarchical model for
 399 decomposition in terrestrial ecosystems: application to soils of the humid
 400 tropics. *Biotropica* 25:130–150

401 Lindh M, Zhang L, Falster D, et al (2014) Plant diversity and drought: The role
 402 of deep roots. *Ecol Modell* 290:85–93. doi:
 403 10.1016/j.ecolmodel.2014.05.008

404 Lu M, Zhou X, Yang Q, et al (2013) Responses of ecosystem carbon cycle to
 405 experimental warming : a meta-analysis. *Ecology* 94:726–738

406 Majdi H, Ohrvik J (2004) Interactive effects of soil warming and fertilization
 407 on root production, mortality, and longevity in a Norway spruce stand in
 408 Northern Sweden. *Glob Chang Biol* 10:182–188. doi: 10.1111/j.1529-
 409 8817.2003.00733.x

410 Marty C, Houle D, Gagnon C (2015) Variation in stocks and distribution of
 411 organic C in soils across 21 eastern Canadian temperate and boreal

412 forests. *For Ecol Manage* 345:29–38. doi: 10.1016/j.foreco.2015.02.024

413 Marty C, Houle D, Gagnon C, Courchesne F (2017) The relationships of soil
 414 total nitrogen concentrations, pools and C:N ratios with climate,
 415 vegetation types and nitrate deposition in temperate and boreal forests
 416 of eastern Canada. *Catena* 152:163–172. doi:
 417 10.1016/j.catena.2017.01.014

418 Marty C, Piquette J, Morin H, et al (2019) Nine years of in situ soil warming
 419 and topography impact the temperature sensitivity and basal respiration
 420 rate of the forest floor in a Canadian boreal forest. *PLoS One* 14:1–22.
 421 doi: 10.1371/journal.pone.0226909

422 McGuire KL, Bent E, Borneman J, et al (2010) Functional diversity in resource
 423 use by fungi. *Ecology* 91:2324–2332

424 Melillo JM, Butler S, Johnson J, et al (2011) Soil warming, carbon–nitrogen
 425 interactions, and forest carbon budgets. *PNAS* 108:9508–9512. doi:
 426 10.1073/pnas.1018189108

427 Niinisto SM, Silvola J, Kellomaki S (2004) Soil CO₂ efflux in a boreal pine
 428 forest under atmospheric CO₂ enrichment and air warming. *Glob Chang*
 429 *Biol* 10:1363–1376. doi: 10.1111/j.1365-2486.2004.00799.x

430 Pan Y, Birdsey RA, Fang J, et al (2011) A large and persistent carbon sink in
 431 the world’s forests. *Science* (80-) 333:988–993

432 R Core Team (2017) R: A language and environment for statistical computing
 433 [Internet]. Vienna, Austria; 2014

434 Rinnan R, Michelsen A, Jonasson S (2008) Effects of litter addition and

435 warming on soil carbon, nutrient pools and microbial communities in a
 436 subarctic heath ecosystem. *Appl Soil Ecol* 39:271–281. doi:
 437 10.1016/j.apsoil.2007.12.014
 438 Romero-Olivares AL, Allison SD, Treseder KK (2017) Decomposition of
 439 recalcitrant carbon under experimental warming in boreal forest. *PLoS*
 440 *One* 12:e0179674
 441 Rustad LE, Campbell JL, Marion GM, et al (2001) A meta-analysis of the
 442 response of soil respiration, net nitrogen mineralization, and
 443 aboveground plant growth to experimental ecosystem warming.
 444 *Oecologia* 126:543–562. doi: 10.1007/s004420000544
 445 Saleska SR, Shaw MR, Fischer ML, et al (2002) Plant community composition
 446 mediates both large transient decline and predicted long-term recovery
 447 of soil carbon under climate warming. *Global Biogeochem Cycles* 16:3-1-
 448 3-18. doi: 10.1029/2001GB001573
 449 Schimel JP, Bilbrough C, Welker JM (2004) Increased snow depth affects
 450 microbial activity and nitrogen mineralization in two Arctic tundra
 451 communities. *Soil Biol Biochem* 36:217–227. doi:
 452 10.1016/j.soilbio.2003.09.008
 453 Schmidt SK, Costello EK, Nemergut DR, et al (2007) Biogeochemical
 454 consequences of rapid microbial turnover and seasonal succession in
 455 soil. *Ecology* 88:1379–1385
 456 Shukla PR, Skea J, Calvo Buendia E, et al (2019) IPCC, 2019: Climate Change
 457 and Land: an IPCC special report on climate change, desertification, land

458 degradation, sustainable land management, food security, and
 459 greenhouse gas fluxes in terrestrial ecosystems. Intergovernmental
 460 Panel on Climate Change (IPCC)
 461 Sistla S a, Moore JC, Simpson RT, et al (2013) Long-term warming
 462 restructures Arctic tundra without changing net soil carbon storage.
 463 Nature 497:615–8. doi: 10.1038/nature12129
 464 Treseder KK, Lennon JT (2015) Fungal traits that drive ecosystem dynamics
 465 on land. Microbiol Mol Biol Rev 79:243–262. doi: 10.1128/MMBR.00001-
 466 15
 467 Treseder KK, Mack MC, Cross A, et al (2004) Relationships among fires, fungi,
 468 and soil dynamics in Alaskan boreal forests. Ecol Appl 14:1826–1838
 469 Treseder KK, Marusenko Y, Romero-Olivares AL, Maltz MR (2016)
 470 Experimental warming alters potential function of the fungal community
 471 in boreal forest. Glob Chang Biol
 472 van Gestel N, Shi Z, van Groenigen KJ, et al (2018) Predicting soil carbon loss
 473 with warming. Nature 554:E4–E5. doi: 10.1038/nature25745
 474 Vogel JG, Valentine DW, Ruess RW (2005) Soil and root respiration in mature
 475 Alaskan black spruce forests that vary in soil organic matter
 476 decomposition rates. Can J For Res 35:161–174. doi: 10.1139/x04-159
 477 Voriskova J, Baldrian P (2013) Fungal community on decomposing leaf litter
 478 undergoes rapid successional changes. ISME J 7:477–486. doi:
 479 10.1038/ismej.2012.116
 480 Xiong J, Peng F, Sun H, et al (2014) Divergent responses of soil fungi

481 functional groups to short-term warming. *Microb Ecol* 68:708–715. doi:
482 10.1007/s00248-014-0385-6

483 Xu X, Shi Z, Li D, et al (2015) Plant community structure regulates responses
484 of prairie soil respiration to decadal experimental warming. *Glob Chang*
485 *Biol* 21:3846–3853. doi: 10.1111/gcb.12940

486 Zhou G, Zhou C, Liu S, et al (2006) Belowground carbon balance and carbon
487 accumulation rate in the successional series of monsoon evergreen
488 broad-leaved forest. *Sci China, Ser D Earth Sci* 49:311–321. doi: 10.1007/
489 s11430-006-0311-y

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Table 1. Effects of greenhouse warming on boreal forest carbon and nitrogen pools after 13 y of treatment $\pm$ 1 standard error. Bold indicates significance ($P \leq 0.05$).

	C (g m ⁻²)			N (g m ⁻²)		
			<i>P</i>	Greenhou		<i>P</i>
	Control	Greenhouse		Control	se	
Herbaceous		77.32 $\pm$	0.16	0.50 $\pm$	1.34 $\pm$	0.25
vegetation	19.96 $\pm$ 6.83 33.63 $\pm$	38.75 97.05 $\pm$	6 0.06	0.19 0.37 $\pm$	0.59 0.53 $\pm$	9 0.51
Woody vegetation	20.00 66.50 $\pm$	32.77	9 0.0	0.23 1.23 $\pm$	0.18 0.34 $\pm$	1 0.14
Moss	25.01 193.64 $\pm$	7.78 $\pm$ 6.07 301.83 $\pm$	34 0.28	0.45 2.09 $\pm$	0.30 2.55 $\pm$	2 0.68
Lichen fraction	74.77	67.57	5	0.74	0.48	7
Sum	313.73 $\pm$	483.97 $\pm$	0.23	4.19 $\pm$	4.76 $\pm$	0.77
aboveground	59.94	122.25	6	1.25	1.15	3
Organic soil						
horizon						
	407.85 $\pm$	290.10 $\pm$	0.59	6.13 $\pm$	6.00 $\pm$	0.95
<i>Roots</i>	132.86 679.02 $\pm$	97.8 549.47 $\pm$	5 0.0	2.40 25.25 $\pm$	1.45 20.83 $\pm$	7 0.28
<i>Soil</i>	84.03 15.76 $\pm$	36.95 46.92 $\pm$	48 0.27	3.74 0.17 $\pm$	1.54 0.57 $\pm$	7 0.30
<i>Other</i>	13.11 1102.63 $\pm$	31.50 886.50 $\pm$	6 0.11	0.14 31.55 $\pm$	0.34 27.40 $\pm$	7 0.35
Sum belowground	179.03	148.75	5	4.83	2.44	0
Total above and	1416.36 $\pm$	1370.47 $\pm$	0.81	35.74 $\pm$	32.17 $\pm$	0.37
belowground	181.49	255.38	0	4.56	2.99	6

Table 2. Effects of greenhouse warming on boreal forest percent carbon, percent nitrogen, and C:N ratios after 13 y of treatment $\pm$ 1 standard error. Bold indicates significance ($P \leq 0.05$).

	%C			%N			C:N		
	Greenhou		<i>P</i>	Greenho		<i>P</i>	Greenhous		<i>P</i>
	Control	se		Control	use		Control	e	
Herbaceous	49.76 ±	50.28 ±	0.27		0.92 ±	0.0	44.08 ±	55.48 ±	
vegetation	0.42	0.42	0	1.15 ± 0.07	0.07	44	2.77	3.91	0.055
	49.35 ±	50.73 ±	0.10		0.36 ±	0.0	83.63 ±	253.57 ±	
Woody vegetation	0.43	0.36	8	0.61 ± 0.06	0.13	28	8.29	91.68	0.118
	44.22 ±	44.29 ±	0.95		1.05 ±	0.52	52.86 ±	52.21 ±	
Moss	0.67	5.61	8	0.87 ± 0.07	0.25	3	5.56	14.89	0.896
	48.93 ±	47.49 ±	0.95		0.42 ±	0.41	106.44 ±	118.95 ±	
Lichen fraction	2.39	3.56	9	0.67 ± 0.30	0.05	0	23.03	14.82	0.275
Organic soil									
horizon									
	48.97 ±	48.21 ±	0.86		1.27 ±	0.30	74.28 ±	50.47 ±	
Roots	1.55	3.31	8	0.71 ± 0.10	0.39	5	9.37	11.61	0.292
	24.09 ±	24.71 ±	0.89		0.92 ±	0.70	27.46 ±	26.58 ±	
Soil	3.89	3.57	9	0.86 ± 0.11	0.09	5	1.35	1.40	0.663
Mineral soil									
horizon									
	43.48 ±	47.05 ±	0.40		1.03 ±	0.67	44.64 ±	52.13 ±	
Roots	3.49	2.33	6	1.18 ± 0.19	0.19	7	13.21	10.23	0.730
	9.93 ±	8.93 ±	0.60		0.35 ±	0.65	28.01 ±	27.85 ±	
Soil	1.85	1.81	2	0.37 ± 0.09	0.10	5	2.79	2.51	0.908

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Figure 1. A) Hypothesized positive and negative effects of the greenhouse warming treatment on aboveground and belowground C pools in a permafrost-free boreal forest. In this experiment, we measured aboveground and belowground carbon pools (black, solid-lined boxes). Grey, dashed boxes indicate mechanisms that were not measured in this experiment but were observed in prior experiments at our study site (Allison and Treseder 2008; German and Allison 2015; Treseder et al. 2016; Romero-Olivares et al. 2017). Depending on which mechanisms dominate, we predicted different responses for aboveground and belowground C pools. B) Inlay of changes to aboveground and belowground C pools found in our experiment. Positive and negative signs indicate increases and decreases in pool size. Bolded text (soil, moss, and the relationship between roots and moss) specifies significance ($P \leq 0.05$).