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REGULAR ARTICLE

Root respiration and biomass responses to experimental soil warming vary with root diameter and soil depth

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Abstract
Aims Respiration of sugar maple (*Acer saccharum*) surface fine roots has been shown to partially acclimate to experimentally increased soil temperature. In this study, we assessed how larger roots and roots at deeper depths responded to experimental warming.
Methods We quantified specific root respiration and root biomass for three different diameter classes (<1, 1–2, and 2–10 mm) from three soil depths (0–10, 10–30, and 30–50 cm) in a sugar maple forest that had received a factorial combination of increased soil temperature (4 to 5 °C above ambient) and supplemental precipitation for three growing seasons.
Results Partial temperature acclimation occurred for respiration of fine-roots (<1 mm) at 0–10 cm, limiting the increase to 30% above that for roots in the control treatment. In contrast, there was no evidence for acclimation of fine-roots at deeper depths, where soil warming caused respiration to more than double. There was evidence of acclimation for 1–2 mm roots at the 0–10 cm depth (20% reduction in respiration at an 18 °C reference temperature) but not for the larger diameter roots at any of the three soil depths. Root biomass was not altered by soil warming or moisture addition.
Conclusions Despite partial thermal acclimation in surface fine-root respiration, soil warming caused an overall 41% increase in the C flux to the atmosphere from respiration of roots in the upper 30 cm of soil, from 21.3 to 30.1 μmol m^{-2} s^{-1}, potentially reducing C availability for biomass production.

Keywords Specific respiration rate · Temperature acclimation · Soil heating · Soil moisture · *Acer saccharum*

Mickey P. Jarvi and Andrew J. Burton contributed equally to this work.

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Introduction

Global soil respiration has recently been estimated to range from 83 to 108 Pg C yr^{-1} (Hursh et al. 2017), exceeding C emissions from the combustion of fossil fuels (Ballantyne et al. 2015) by more than ten-fold. However, this large C flux is one of the least understood among all terrestrial ecosystem C fluxes (Litton and Giardina 2008; Litton et al. 2007). Although root production can utilize much of the C allocated belowground in forests (Hendrick and Pregitzer 1993; Schimel et al. 1994), a significant proportion also is allocated to root respiration, which can account for anywhere from 10 to 90% of soil respiration, with estimates from 40 to 60% being most common (Hanson et al. 2000).

How root respiration will be affected by global climate change is not well understood. Global land surface temperatures from 2081 to 2100 are expected to be between 1.2 and 4.8 °C greater than they were from 1986 to 2005 (Collins et al. 2013), and those in the Lake States of the US are expected to increase by 1.5 to 5.5 °C (IPCC 2013). This projected warming could have consequences on ecosystem C balance by altering rates of photosynthesis and respiration and potentially changing the strength of an ecosystem as a C sink or source (Canadell et al. 2007; Melillo et al. 1990, 1995; Shaver et al. 2000).

Previously, Jarvi and Burton (2013, 2018) found evidence that surface fine-roots (0–10 cm depth, < 1 mm diameter) of sugar maple in a soil warming experiment exhibited respiratory acclimation to elevated soil temperature representative of a future climate scenario of 4 to 5 °C above current conditions. However, they did not find evidence that surface fine-root biomass had been affected by several years of soil warming. These findings are in contrast to those of Melillo et al. (2011) for the long-term Harvard Forest soil warming experiment in central Massachusetts, which showed no indication of acclimation of specific fine-root respiration to elevated temperatures, but did find reductions in fine-root biomass. These two different response scenarios, however, both result in a reduction in ecosystem fine-root respiration relative to what would be predicted using an exponential response of respiration to temperature, as both a down-regulation in root-respiration rate at a given temperature (acclimation) or a reduction of root biomass (ultimately less tissue to respire) would help constrain the increase in ecosystem level root respiration.

Root functions differ with root diameter or order and with rooting depth (Pregitzer et al. 1998). Finer, lower order roots at shallow soil depth perform the majority of nutrient uptake, fine roots at all depths contribute to water uptake, and coarser roots perform functions such as transport, support, and carbohydrate storage (Xia et al. 2010; McCormack et al. 2015). It has been found that root biomass is often greater when nutrient availability is lower within the same geographical area (Finér et al. 2007; Helmisaari et al. 2007) or water availability is limiting (Meier and Leuschner 2008).

This study was conducted at the Sugar Maple Altered Rainfall and Temperature (SMART) experiment in August 2013, during the third full growing season after the initiation of the soil warming treatments. The objectives were to determine if thermal acclimation of specific root respiration still existed for fine-roots (< 1 mm) from the shallow soil depth (0–10 cm), in agreement with the previous findings of Jarvi and Burton (2013, 2018), and to conduct a novel investigation for the presence or absence of respiratory acclimation to ongoing thermal treatments for roots from multiple diameter classes (<1, 1–2, and 2–10 mm) and soil depths (0–10, 10–30, and 30–50 cm). Additionally, root biomass (g m^{-2}) was quantified to determine if soil warming had altered biomass for any root size class and soil depth.

We hypothesized that fine-root respiration (<1 mm) for the 0–10 cm soil depth would still be experiencing acclimation on the heated treatments, based on previous findings for the experiment (Jarvi and Burton 2013, 2018) and that acclimation would also occur for deeper and larger diameter roots. We additionally hypothesized that root biomass would remain unchanged across all root diameters, depths, and soil treatments.

Materials and methods

Study site location

The SMART experiment was located in a natural second-growth sugar maple (*Acer saccharum* Marsh.) forest at Michigan Technological University's Ford Center and Forest (46°38.41' N latitude, 88°29.07' W longitude). Sugar maple dominated the site with 89.3% of the 21.7 m^2 ha^{-1} of basal area. Other less dominant species included: American elm (*Ulmus americana* L.), eastern hemlock (*Tsuga canadensis* (L.) Carr.), eastern hophornbeam (*Ostrya virginiana* (Mill.) K. Koch), and yellow birch (*Betula alleghaniensis* Britton). The soil is classified as a sandy loam with 62% sand, 29% silt, and 9% clay for the 0–10 cm depth and 59% sand, 29% silt, and 12% clay for the 10–20 cm depth. A fragipan occurs at approximately 40 cm below the soil surface.

The full factorial heating and water addition experiment warmed the soil with infrared lamps beginning in September 2010 (Jarvi and Burton 2013). The treatment plots were 10 m by 10 m, with four treatments replicated three times: control, heat (4 to 5 °C above ambient soil temperature), water (addition of 30% of average growing season ambient precipitation), and heat + water.

Soil warming was achieved using sixteen infrared heating lamps (model MRM1215 heaters, Kalglo Electronic Co., Bethlehem, PA, USA) per plot, situated

1.5 m above the soil surface, with four rows of four lamps spaced 2.5 m apart. The lamp's heat output was adjusted manually as needed to achieve the desired increase in soil temperature. The treatments were applied during the snow-free season with a general ramping up of soil temperature for the heated plots during the spring (April), and ramping down of temperature on the heated plots during late fall (mid-October to November). The infrared lamps were shut off but left in place during the snow season (approximately mid-November to early April).

Water was applied via sprinkler heads positioned on the four corners of the 10 m × 10 m plots. Each covered a 90° field of application, with slight overlap at plot center to ensure even and complete distribution of water throughout the plot. Water additions were made weekly, in concert with natural rain events when possible, to help maintain natural wetting and drying cycles. A total of 16 cm of precipitation equivalent was added annually between mid-May and mid-October.

Treatment effectiveness was monitored with temperature and moisture probes placed at 2, 5, and 10 cm soil depths beneath heater rows, and at 2 and 5 cm depths midway between heater rows (Em50 data loggers with 5TM temperature/moisture probes, Decagon Devices Inc., Pullman, Washington, USA). Additional monitoring of soil temperature and moisture at 20 and 30 cm depths was conducted in 2014 to ensure that the temperature differential extended to deeper soil depths. Over the course of the experiment, the targeted degree of soil warming in heated treatments has occurred at all soil depths, as well as directly beneath and midway between rows of heat lamps (Jarvi and Burton 2018). Collins et al. (2018) measured air temperatures and vapor pressure deficits at 3, 8 and 12 m heights on all four treatments at the SMART site and found no significant differences among the heights and treatments, which was expected at these locations above the infrared lamps. Water additions were intended to offset the drying effects of the soil warming treatment, allowing the heat + water treatment to have soil moisture availability similar to the control. Since the initiation of treatments in 2010, the water additions have kept the volumetric soil moisture on the heat + water treatment similar to the control treatment (Jarvi and Burton 2013). In late August 2013, when sampling occurred, volumetric soil moisture contents were 0.24, 0.22, 0.27 and 0.25 cm^3 cm^{-3} for the control, heat, water and heat + water treatments, respectively.

Sample collection

Samples for root respiration measurement and biomass determination were collected in late August 2013. Three 10 cm diameter soil cores were taken per plot using a steel corer designed for use in rocky forest soils (Jurgensen et al. 1977). Cores were separated by depth increment (0–10 cm, 10–30 cm, and 30–50 cm), creating a total of 9 samples per plot and 108 samples for the entire study site. All of these were used for root biomass determination, and a subset of roots for respiration measurements were immediately extracted from one of the three soil samples per plot being excavated, with the residual soil and roots stored on ice, then returned to the laboratory where they were stored frozen until further analysis. All sampling and root respiration measurements were completed over a four-day period.

Root respiration

Root respiration measurements were conducted following the procedure outlined by Jarvi and Burton (2013, 2018) and Burton et al. (2012), with the use of two open-system infrared gas analyzers (IRGA; CIRAS-1 and CIRAS-2, PP Systems, Amesbury MA). Custom aluminum cuvettes that could be placed in water baths to control temperature were connected to the IRGAs. Root samples were separated from the soil and sorted to obtain approximately 2 g fresh weight of roots for each of three diameter classes (<1, 1–2, 2–10 mm). The <1 mm class consisted of primarily 1st and 2nd order roots, and is equivalent to the absorptive fine-root class defined by McCormack et al. (2015). The larger roots would be classified as higher order transport roots (McCormack et al. 2015), with the 1–2 mm class including primarily 3rd, 4th and 5th order roots. Root samples were hand cleansed of soil and organic debris before measurement of root respiration commenced, typically within 15 min of sample collection. Respiration rates (nmol CO_2 s^{-1}) were recorded after allowing 15 min for root samples to equilibrate to the cuvette temperature and the measurement atmosphere of 1000 μl l^{-1} CO_2, representative of upper soil atmospheric conditions (Burton et al. 1998).

It has been shown that respiration measurements at a common reference temperature are useful for assessing differences in metabolic capacity that might be indicative of acclimation (Atkin et al. 2000). Thus, root respiration rates for all samples were measured at ambient

soil temperature and at a common reference temperature of 18 °C. The order of these two temperatures was alternated each sample to reduce bias. We interpret significantly lower respiration rates at the reference temperature for roots from heated soil than for roots from non-heated soil as evidence of acclimation. As a second indicator, we also view increases in respiration with heating that are less than one would expect based on typical Q_{10} values for root respiration as additional support for the existence of acclimation.

For all samples, measurements at both temperatures were completed within 45 min after roots were removed from the soil and no more than one hour after the soil cores were extracted. Previous work with sugar maple fine roots has indicated that respiration rates declined very little during the first four hours after removal from the soil (Burton et al. 2002). We performed repeated measurements of a subset of samples during the early years of the SMART experiment, and found only a slight decline in respiration rates over time (< 10% over 45 min). After respiration measurements, roots were placed on ice, transported back to the laboratory, and stored frozen at −20 °C until further analysis. Later, root respiration samples were cleansed with deionized water to remove any residual adhering soil not removed during field cleaning, dried at 65 °C for 48 h, and weighed to enable calculation of specific root respiration rates (nmol CO_2 g^{-1} s^{-1}).

Root biomass

Root biomass samples were obtained from the same soil samples used to obtain root respiration samples. Soil was passed through a 6.2 mm sieve to separate rocks and larger roots and then passed through a 2 mm sieve. Any remaining pebbles, debris, and non-tree roots were discarded. Only live tree roots (pliable and brown to white in color) were collected for further analysis, while dead roots (brittle and brown to black in color) were discarded. Root samples were then cleansed in deionized water, sorted into the same diameter classes as the root respiration samples (<1, 1–2, 2–10 mm) and dried at 65 °C for 48 h. Dry weights obtained from the respiration samples were added to those sorted from the root biomass soil samples to determine total root biomass by depth increment and diameter class for each core.

Ecosystem root respiration

Ecosystem level root-respiration was calculated as the product of specific root respiration (nmol CO_2 g^{-1} s^{-1}) and root biomass (g m^{-2}) and then divided by 1000 for a final unit of μmol CO_2 g^{-1} s^{-1} for each combination of root depth and diameter class. These values were then summed to estimate total ecosystem root respiration to a depth of 30 cm. The samples from 30 to 50 cm were excluded from the sum due to the inability of our sample design to adequately estimate root biomass at this deeper soil depth, especially for larger diameter roots, which had very high spatial variability, with many soil cores having no coarse roots and a few having a large biomass.

Soil respiration was measured approximately monthly during the growing season of all years of the study using a dynamic chamber, infrared gas analyzer system (LI-8100 survey system; Li-Cor Biosciences, Lincoln, NE, USA). On each plot, measurements were made at nine soil respiration collars, located in the center 5 m × 5 m portion of each plot, utilizing a 3 × 3 grid of collars, with 2.5 m spacing between collars. The respiration collars (10.2 cm diameter; schedule 40 PVC) were inserted 2.5 cm into the forest floor two weeks before the beginning of the field season and were left in place for the remainder of the year. In this paper we report soil respiration data collected on the date closest to the root respiration measurement period, in order to provide context for our ecosystem root respiration estimates. The data is from 8 September 2013, ten to thirteen days after root respiration measurements were made. Soil temperatures on the soil respiration date were approximately 3 °C cooler than those during the root respiration measurement period.

Statistical analysis

All statistical analyses were conducted in the R environment (R Development Core Team 2008). Two-way nested analysis of variance (ANOVA) was used to test for differences in specific root respiration and root biomass among treatments (heat, water) with nested root size class and soil depth effects also examined. Two-way ANOVA was used to test for differences among treatments (heat, water) in ecosystem root respiration and soil respiration.

Results

Root respiration

The average temperatures across all three soil depths (0–10, 10–30, 30–50 cm) during the root sampling period were 16.4 °C and 21.8 °C for non-heated and heated treatments, respectively. The propagation of the heat treatments to significant depths indicated by these one-time values recorded by a hand probe was consistent with continuous data logger measurements made to depths of 30 cm in August of 2014 (Fig. 1).

Specific root respiration rates measured at treatment ambient temperature were greater for the heated treatments than the non-heated treatments ($P < 0.001$, Table 1, Fig. 2). Effects were more pronounced for the <1 mm roots and 2–10 mm roots than for the 1–2 mm roots ($P = 0.008$, significant heat x diameter interaction, Table 1). In addition, regardless of treatment, specific respiration rates were greater for smaller diameter roots and were lower for all root size classes in deeper soil (Table 1, Fig. 2), with differences between root diameter classes being less pronounced in deeper soil ($P < 0.001$, significant root diameter x soil depth interaction in Table 1).

At the reference temperature of 18 °C, root respiration rates were slightly less for the heated versus the non-heated treatments ($P = 0.082$, Table 1, Fig. 3). This effect is driven largely by lower respiration rates for the heated treatments in surface soil roots, with reduced effects in deeper soil ($P = 0.082$ for heat x depth interaction, Table 1). Relative to the non-heated treatments, there was a 30% increase in respiration of surface fine-roots from warmed soil when measured at treatment ambient temperature, but a 25% reduction of respiration for these same roots, relative to those from unheated soil, when compared at the 18 °C reference temperature (Figs. 2 & 3). This is in contrast to much greater increases for heated versus non-heated treatments in respiration measured at ambient temperature for fine roots from the 10–30 cm and 30–50 cm soil depths (111% and 197%, respectively, Fig. 2). Additionally, there were 3% and 26% increases, rather than decreases, for heated versus non-heated treatments in respiration rates at the reference temperature of 18 °C for fine roots from the same two depths (Fig. 3). Similar to trends observed at ambient temperature, root respiration at the 18 °C reference temperature was much lower for larger root diameters and samples from deeper soil depths ($P < 0.001$, Table 1, Fig. 3), with differences between diameter classes less pronounced in deeper soil (root diameter x soil depth interaction, Table 1).

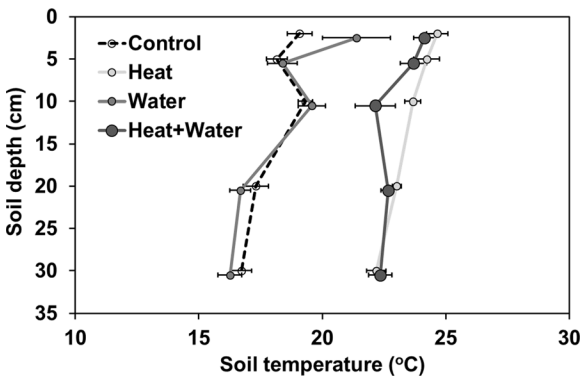

Fig. 1 Temperature differences with depth for heated and non-heated treatments in August 2014. Error bars are one standard error of the mean

Root biomass

Root biomass was not altered by soil warming or water additions (Tables 2 and 3, Fig. 4). There was a significant difference in root biomass across root diameters, with greater biomass in <1 mm and 2–10 mm roots than in 1–2 mm roots, and an indication that fine-root biomass declined with soil depth ($P < 0.001$, Tables 2 and 3). A significant soil depth x root diameter interaction resulted from the tendency for root biomass to be greater for <1 mm diameter roots in surface soil and greater for larger diameter roots at depths beyond 10 cm ($P = 0.012$, Tables 2 and 3, Fig. 4).

Ecosystem root respiration

For the 0–10 cm depth, soil warming caused no change ($P = 0.31$) in ecosystem root respiration (μmol CO_2 m^{-2} s^{-1}). However, at the 10–30 cm depth there was 107 and 40% more ecosystem root respiration for the heat and heat + water treatments, respectively, than for the control (Table 4, significant heat effect $P = 0.05$). As a result, overall ecosystem root respiration to a depth of 30 cm for the heated (heat, heat + water) treatments of 2.81 μmol CO_2 m^{-2} s^{-1} was 41% more ($P = 0.10$) than that for non-heated treatments (control, water; 1.99 μmol CO_2 m^{-2} s^{-1}). Soil respiration on the date closest to the root respiration sampling period was not affected by the heat or water addition treatments ($P \geq 0.70$, Table 4), and was, on average, 1.52 times our estimated ecosystem root respiration from the top 30 cm of soil.

Table 1 Analysis of variance (ANOVA) table for effects of soil warming and moisture addition on specific root respiration rates by root diameter class and soil depth increment at ambient soil temperature and at an 18 °C reference temperature

	Ambient temperature root respiration				
	d.f	Sum Sq	Mean Sq	F Value	*P* value
Heat	1	17.4	17.14	34.876	**<0.001**
Water	1	0.73	0.73	1.479	0.228
Root diameter	2	79.01	39.50	80.396	**<0.001**
Soil Depth	2	33.34	16.67	33.922	**<0.001**
Heat x Water	1	0.39	0.39	0.789	0.378
Heat x Root diameter	2	5.78	2.89	5.880	**0.008**
Water x Root diameter	2	0.93	0.47	0.947	0.393
Heat x Soil depth	2	0.60	0.30	0.610	0.546
Water x Soil depth	2	1.50	0.75	1.525	0.226
Root diameter x Soil depth	3	18.19	4.55	9.256	**<0.001**
Heat x Water x Root diameter	2	0.22	0.11	0.225	0.779
Heat x Water x Soil depth	2	0.40	0.20	0.406	0.668
Heat x Root diameter x Soil depth	4	0.61	0.15	0.313	0.868
Water x Root diameter x Soil depth	4	1.07	0.27	0.545	0.704
Heat x Water x Root diameter x Soil depth	3	1.21	0.40	0.821	0.487
Residuals	*63*	*30.96*	*0.49*		
	Reference temperature root respiration				
	d.f.	Sum Sq	Mean Sq	F value	*P* value
Heat	1	1.36	1.36	3.118	0.089
Water	1	1.04	1.04	2.372	0.129
Root diameter	2	62.74	31.37	71.771	**<0.001**
Soil Depth	2	34.75	17.37	39.750	**<0.001**
Heat x Water	1	0.00	0.00	0.001	0.973
Heat x Root diameter	2	0.56	0.28	0.635	0.533
Water x Root diameter	2	0.51	0.26	0.583	0.561
Heat x Soil depth	2	2.28	1.14	2.608	0.140
Water x Soil depth	2	1.49	0.74	1.701	0.191
Root diameter x Soil depth	4	22.09	5.52	12.635	**<0.001**
Heat x Water x Root diameter	2	0.44	0.22	0.507	0.605
Heat x Water x Soil depth	2	1.12	0.56	1.278	0.286
Heat x Root diameter x Soil depth	4	2.21	0.55	1.267	0.293
Water x Root diameter x Soil depth	4	1.77	0.44	1.010	0.409
Heat x Water x Root diameter x Soil depth	3	0.39	0.13	0.299	0.826
Residuals	*63*	*27.54*	*0.44*		

Discussion

Fine-root (<1 mm) biomass was unchanged in the 0–10 cm depth in response to experimental warming at the SMART experiment in 2010 and 2011 (Jarvi and Burton 2013), and we similarly found no evidence that root biomass in any size class or depth increment had been altered by warming for the more comprehensive sampling conducted in 2013 (Fig. 4, Table 3). Lower root respiration rates at the reference temperature for samples from warmed soil than from non-heated soil provides evidence that

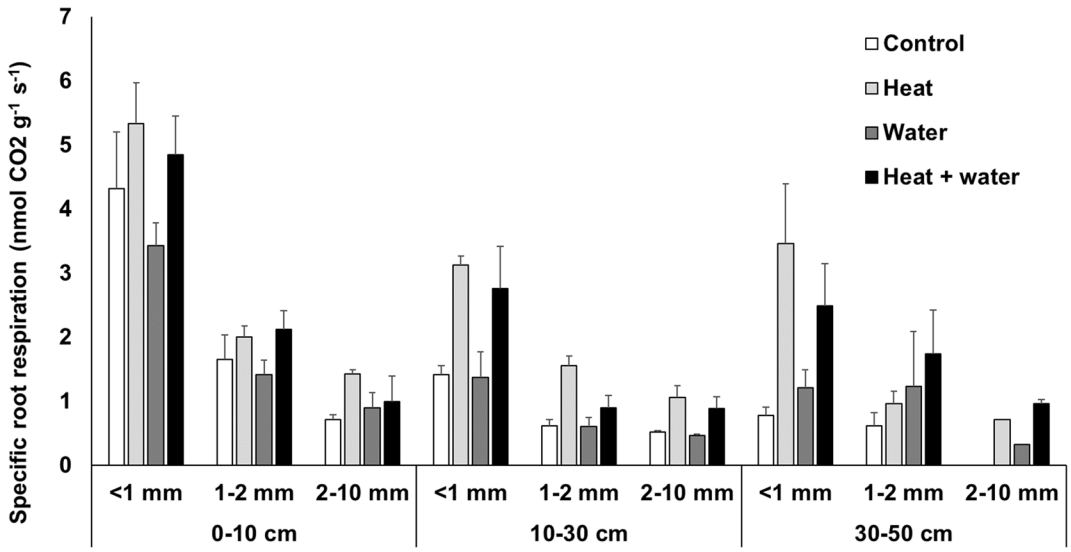

Fig. 2 Average specific respiration (nmol CO_2 g^{-1} s^{-1}) at ambient soil temperature (16.4 °C and 21.8 °C for the non-heated and heated treatments respectively). Respiration is categorized by soil depth increment (0–10, 10–30, and 30–50 cm) and root diameter class (<1, 1–2, 2–10 mm). Error bars are one standard error of the mean

temperature acclimation is present. This effect is most apparent for fine-roots at the 0–10 cm depth, as well as in 1–2 mm roots at the same depth (Fig. 3). The 0–10 cm soil depth is where the majority of fine-roots associated with acquisition of nutrients are located (Joslin et al. 2006; Makita et al. 2011; Pregitzer et al. 1998). In contrast to our hypothesis, acclimation does not appear to have occurred for any root diameter class at deeper depths (Fig. 3).

Melillo et al. (2011) found no evidence of temperature acclimation in specific root respiration in a soil warming experiment at the Harvard Forest after seven years of experimental soil warming. However, they did observe a decrease in fine-root biomass in the 0–10 cm depth that was sufficient to constrain the amount of total C released back to the atmosphere by root respiration at the ecosystem level. In our study, partial acclimation in surface roots, with no change in root biomass, somewhat

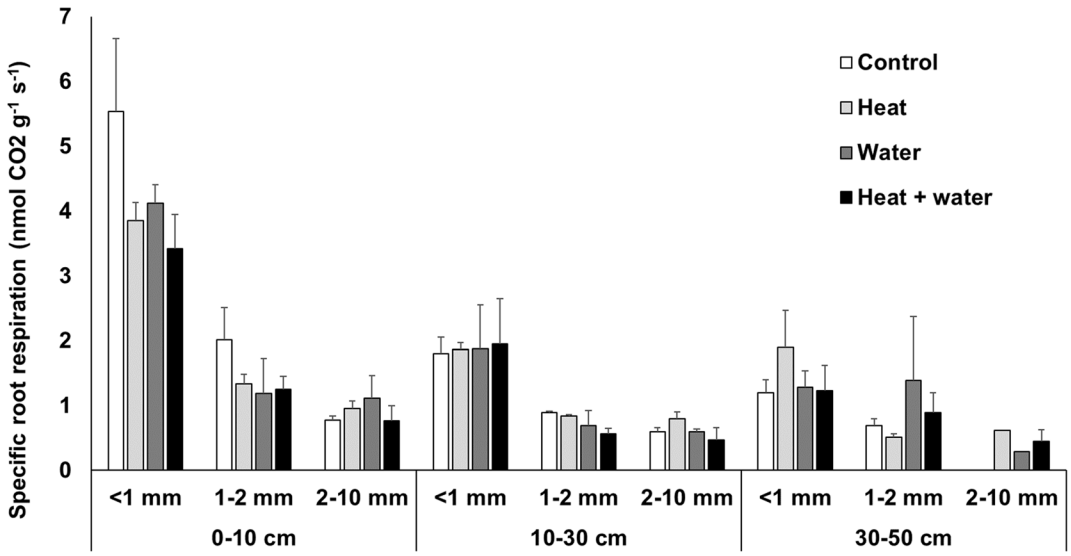

Fig. 3 Average specific respiration (nmol CO_2 g^{-1} s^{-1}) at a common reference temperature of 18 °C. Respiration is categorized by soil depth increment (0–10, 10–30, and 30–50 cm) and root diameter class (<1, 1–2, 2–10 mm). Error bars are one standard error of the mean

Table 2 Analysis of variance (ANOVA) table for effects on root biomass of soil warming and water addition, by root diameter class and soil depth increment

	d.f.	Sum Sq	Mean Sq	F value	*P* value
Heat	1	82,839	82,839	1.148	0.285
Water	1	135,975	135,975	1.884	0.171
Root diameter	2	1,589,823	794,911	11.013	**<0.001**
Soil Depth	2	1,073,396	536,698	7.435	**0.06**
Heat x Water	1	4003	4003	0.055	0.814
Heat x Root diameter	2	93,859	46,929	0.650	0.523
Water x Root Diameter	2	47,595	23,797	0.330	0.719
Heat x Soil depth	2	248,646	124,323	1.722	0.180
Water x Soil depth	2	105,491	52,745	0.731	0.482
Root diameter x Soil depth	4	844,748	211,187	2.926	**0.012**
Heat x Water x Root diameter	2	52,673	26,336	0.365	0.695
Heat x Water x Soil depth	2	226,163	113,082	1.567	0.211
Heat x Root diameter x Soil Depth	4	294,956	73,739	1.022	0.396
Water x Root diameter x Soil depth	4	123,808	30,952	0.429	0.788
Heat x Water x Root diameter x Soil depth	4	504,051	126,013	1.746	0.134
Residuals	*288*	*20,788,156*	*72,181*		

constrained ecosystem root system respiration. However, lack of acclimation for deeper roots and no change in biomass led to overall increases in ecosystem root respiration (Table 4).

Earth system models often use a Q_{10} of 2 for plant tissue respiration in response to elevated temperatures. We have found that that the respiration of roots from unwarmed soil at our study location responds with a Q_{10} of 2.7 to both changes in soil temperature across the growing season and for measurements made on a single date at temperatures ranging from 6 to 24 °C (Burton et al. 2002). This would create an expected increase in root respiration of 71% on the heated versus the non-heated treatments for the temperature increase of 5.4 °C existing during the sampling period. Respiration of roots in the top 10 cm of the soil increased only 32, 34 and 51% with the 5.4 °C of soil warming for the <1, 1–2, and 2–10 mm classes, respectively. In contrast, the same temperature increase caused root respiration across all size classes to more than double at the 10–30 and 30–50 cm depths (Fig. 2).

Table 3 Average fine-root biomass (<1 mm, g m^{-2}) for the soil depth increments of 0–10, 10–30, 30–50 cm across experimental soil treatments with standard error of the mean in parentheses. Root biomass was not altered by either the heat or water addition treatments at any depth (see Table 2)

	Root biomass (g m^{-2})		
Treatment	0–10 cm	10–30 cm	30–50 cm
Control	316 (19)	227 (43)	55 (9)
Heat	304 (104)	267 (154)	117 (38)
Water	311 (101)	225 (38)	34 (1)
Heat + water	295 (88)	190 (52)	51 (1)

Previous exploration of how far the heat transfers to these deeper soil depths on the heated treatments has shown that we achieve our target of 4 to 5 °C above ambient to at least 30 cm, and most likely beyond that (Fig. 1). To our knowledge, this was the first time respiration of larger diameter roots and roots at different soil depths was measured for a forest soil warming experiment. Wei et al. (2017) found strong evidence of thermal acclimation of leaf respiration in three broadleaf tree species commonly found in sugar maple forests (*Acer rubrum* L., *Betula papyrifera* Marsh., and *Populus tremuloides* Michx.), and this acclimation was found throughout different habitats, sites, and time of year. This finding is important for total tree productivity because in the presence of acclimation less C is respired and more can be allocated elsewhere for purposes that include biomass production. We have

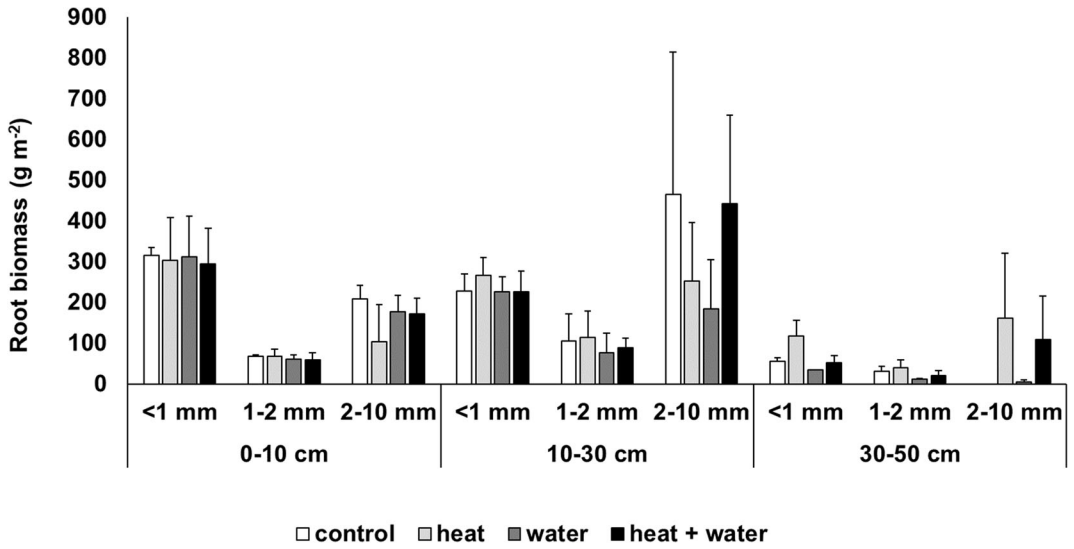

Fig. 4 Average root biomass across treatments and root diameters for depth increments of 0–10 cm, 10–30 cm, and 30–50 cm. Error bars are standard error of the mean

found acclimation of fine-root respiration in roots from the top 0–10 cm of soil in this study, but found no temperature acclimation for fine roots at deeper depths or for coarse roots at any soil depth (Fig. 2). This apparent lack of acclimation of deeper roots could have negative effects on total tree productivity as a result.

The approximate growing season at the study site is 124 days. Extrapolating the rates of ecosystem root respiration in the top 30 cm of soil (Table 4) for 124 days suggests the heated treatments (heat, heat + water) would release 8.8 more mol CO_2 m^{-2} over the growing season than the non-heated treatments (control, water), with 30.1 and 21.3 mol CO_2 m^{-2} for the heated and non-heated treatments, respectively. Averaged across treatments, root respiration during the sample period contributed a CO_2 flux to the atmosphere equivalent to 66% of the soil respiration measured two weeks later. Soil temperatures on the soil respiration date were 3 °C cooler than those during root respiration measurements. Adjustment for this temperature difference could increase soil respiration by about 33% ($Q_{10} \geq 2.6$, Burton et al. 2004), which would cause our estimated ecosystem root respiration to be, on average, about 49% of soil respiration. Regardless of this adjustment, root respiration clearly contributed a larger proportion of soil respiration for the heated treatments (Table 4).

Table 4 Average ecosystem root respiration (μmol CO_2 m^{-2} s^{-1}) across treatments and soil depths and soil respiration on 8 September by treatment, with one standard error of the mean within parentheses. Ecosystem root respiration was lower for heated treatments for the 10–30 cm depth $P = 0.05$) and for the total to 30 cm ($P = 0.10$). Ecosystem root respiration in the top 10 cm and soil respiration did not differ in response to either the heat or water addition treatments

Treatment	Root respiration for soil depth (μmol CO_2 m^{-2} s^{-1})			Soil respiration (μmol CO_2 m^{-2} s^{-1})
	0–10 cm	**10–30 cm**	**Total to 30 cm**	
Control	1.55 (0.18)	0.63 (0.23)	2.18 (0.16)	3.74 (0.31)
Heat	1.82 (0.39)	1.30 (0.54)	3.12 (0.92)	3.70 (0.42)
Water	1.33 (0.41)	0.46 (0.18)	1.79 (0.47)	3.40 (0.51)
Heat + Water	1.61 (0.22)	0.88 (0.22)	2.49 (0.25)	3.71 (0.33)

Under climatic warming a longer growing season will occur, allowing more time for canopy C assimilation. However, the extra growing season length may be insufficient to enable enough C gain to offset a potential warming-induced increase in ecosystem root respiration. This region is predicted to become warmer, but without much change in precipitation (IPPC 2013). This suggests a greater likelihood of moisture deficits during the growing season than currently occur, and C assimilation in sugar maple forests is reduced during portions of the growing season when moisture availability is insufficient (Collins et al. 2018). Increased root system respiration leading to reduced C availability for net

primary production, including aboveground woody biomass increment, is thus a distinct possibility for this forest type.

Acclimation is apparent for the smaller diameter roots at the 0–10 cm depth but is not for large diameter surface roots or any root diameter at deeper soil depths (Fig. 3). We postulate that differences among root classes in the relative proportions of respiration associated with ion uptake versus maintenance may play a role in their differential ability to thermally acclimate. The surface very fine-roots in our study, which are akin to the absorptive fine-roots of McCormack et al. (2015), function primarily for nutrient acquisition (Trocha et al. 2017), and thus most of their respiration during the growing season should be for ion uptake. Respiration for ion uptake would not necessarily increase greatly with temperature. Both plant demand for nutrients and availability of nutrients in soil can increase with temperature, but in a mature forest, neither is likely to increase to the same degree that respiratory capacity could increase if it were to follow an exponential response (Q_{10} of 2 or more). Jarvi and Burton (2018) provide evidence that partial temperature acclimation of surface fine-root respiration in this forest is due to adenylate control, likely associated with root requirements for ATP for ion uptake being less than the root's respiratory capacity to produce ATP in experimentally warmed soil.

The larger diameter roots are not involved in nutrient absorption (Trocha et al. 2017), and the deeper fine-roots, although absorptive, are typically in soil horizons with lower nutrient availability, especially for N and P, and perform much of their nutrient absorption through mass flow during water uptake. As a result, respiration rates in the deeper and larger diameter roots are lower (Fig. 2) and a greater proportion of their respiration would be maintenance respiration than for surface fine roots. A large proportion of maintenance respiration is associated with protein turnover (Bouma et al. 1994; Bouma et al. 1996), which is temperature sensitive (Atkin et al. 2000). Membrane leakage also contributes to increases in maintenance respiration with temperature (Rachmilevitch et al. 2006). The increased soil temperatures on the heated treatments thus could cause an increase in root maintenance respiration, and if this is the dominant type of respiration in large diameter roots and deep fine-roots, a lack of measurable acclimation to warmer temperature would be a possible consequence.

Fine-root respiration (<1 mm) showed an indication that it had partially acclimated to soil warming at the 0–10 cm depth, somewhat limiting the increase in ecosystem root respiration with soil warming. However, there were large increases in root respiration at field soil temperatures in response to soil warming in the deeper (10–30 and 30–50 cm) soil depths. The evidence for acclimation at ambient temperature is apparent in the fine roots (<1 mm) from the 0–10 cm depth with only 32% more respiration on the heated treatments versus the non-heated treatments, which is far less than the expected respiration increase of about 71% for a Q_{10} of 2.7 and a temperature difference of 5.4 °C. However, similar acclimation did not occur for deeper fine-roots and roots of greater diameter. As result, at the ecosystem level, there was 41% more C being released to the atmosphere from root system respiration for the heated treatments for the 5.4 °C differential in soil temperature. Such a response to a warmer climate could reduce C availability for other plant uses, including aboveground woody increment.

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Compliance with ethical standards

Conflict of interest The authors have no conflict of interest to declare.

References

Atkin OK, Holly C, Ball MC (2000) Acclimation of snow gum (Eucalyptus pauciflora) leaf respiration to seasonal and diurnal variations in temperature: the importance of changes in the capacity and temperature sensitivity of respiration. Plant, Cell & Environment 23:15–26

Ballantyne AP, Andres R, Houghton R, Sotcker BD, Wanninkhof R, Aneregg W, Cooper LA, DeGrandpre M, Tans PP, Miller JB, Alden C, White JWC (2015) Audit of the global carbon

budget: estimate errors and their impact on uptake uncertainty. Biogeosciences 12:2565–2584

Bouma TJ, De Visser R, Janssen JHJA, De Kock MJ, Van Leeuwen PH, Lambers H (1994) Respiratory energy requirements and rate of protein turnover in Vivi determined by the use of an inhibitor of protein synthesis and a probe to assess its effect. Physiologia Plantarum 92:585–594

Bouma TJ, Broekhuysen AGM, Veen BW (1996) Analysis of root respiration of Solanum tuberosum as related to growth, ion uptake and maintenance of biomass. Plant Physiology and Biochemistry 34(6):795–806

Burton AJ, Pregitzer KS, Zogg GP, Zak DR (1998) Drought reduces root respiration in sugar maple forests. Ecological Applications 8:771–778

Burton AJ, Pregitzer KS, Ruess RW, Hendrick RL, Allen MF (2002) Root respiration in north American forests: effects of nitrogen concentration and temperature across biomes. Oecologia 131:559–568

Burton AJ, Pregitzer KS, Crawford JN, Zogg GP, Zak DR (2004) Simulated chronic NO_3^- addition reduces soil respiration in northern hardwood forests. Global Change Biology 10: 1080–1091

Burton AJ, Jarvey JC, Jarvi MP, Zak DR, Pregitzer KS (2012) Chronic N deposition alters root respiration-tissue N relationship in northern hardwood forests. Global Change Biology 18:258–266

Canadell JG, Quere CL, Raupach MR, Field CB, Buitenhuis ET, Ciais P, Conway TJ, Gillett NP, Houghton RA, Marland G (2007) Contributions to accelerating atmospheric CO_2 growth from economic activity, carbon intensity, and efficiency of natural sinks. PNAS USA 104:18866–18870

M Collins, et al. (2013): Long-term climate change: projections, commitments and irreversibility. In: climate change 2013: the physical science basis. Contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change [stocker, T.F., D. Qin, G.-K. Plattner, M. Tignor, S.K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex and P.M. Midgley (eds.)]. Cambridge University press, Cambridge, United Kingdom and New York, NY, USA

Collins AR, Burton AJ, Cavaleri MA (2018) Effects of experimental soil warming and water addition to the transpiration of mature sugar maple. Ecosystems 21(1):98–111

Finér L, Helmisaari HS, Lõhmus K, Majdi H, Brunner I, Børja I, Eldhuset E, Godbold D et al (2007) Variation in fine root biomass of three European tree species: beech (Fagus sylvatica L.), Norway spruce (Picea abies L. karst.) and scots pine (Pinus sylvestris L.). Plant Biosyst 141:394–405

Hanson PJ, Edwards NT, Garten CT, Andrews JA (2000) Separating root and soil microbial contributions to soil respiration: a review of methods and observations. Biogeochemistry 48:115–146

Helmisaari HS, Derome J, Nöjd P, Kukkola M (2007) Fine root biomass in relation to site and stand characteristics in Norway spruce and scots pine stands. Tree Physiology 27:1493–1504

Hendrick RL, Pregitzer KS (1993) The dynamics of fine root length, biomass, and nitrogen content in two northern hardwood ecosystems. Canadian Journal of Forest Research 23: 2507–2520

Hursh A, Ballantyne A, Cooper L, Meneta M, Kimball J, Watts J (2017) The sensitivity of soil respiration to soil temperature, moisture, and carbon supply at the global scale. Global Change Biology 23:2090–2103

IPCC, 2013: Annex I: Atlas of Global and Regional Climate Projections [van Oldenborgh, G.J., M. Collins, J. Arblaster, J.H. Christensen, J. Marotzke, S.B. Power, M. Rummukainen and T. Zhou (eds.)]. In: Climate Change 2013: The Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change [Stocker, T.F., D. Qin, G.-K. Plattner, M. Tignor, S.K. Allen, J. Boschung, A. Nauels, Y. Xia, V. Bex and P.M. Midgley (eds.)]. Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA

Jarvi MP, Burton AJ (2013) Acclimation and soil moisture constrain sugar maple root respiration in experimentally warmed soil. Tree Physiology 33(9):949–959

Jarvi MP, Burton AJ (2018) Adenylate control contributes to thermal acclimation of sugar maple fine-root respiration in experimentally warmed soil. Plant, Cell & Environment 41(3):504–516

Joslin JD, Gaudinkski JB, Torn MS, Riley WJ, Hanson PJ (2006) Fine-root turnover patterns and their relationship to root diameter and soil depth in a C_{14}-labeled hardwood forest. The New Phytologist 172:523–535

Jurgensen MF, Larsen MJ, Harvey AE (1977) A soil sampler for steep, rocky sites. Publ. INT-RN-217. Intermountain Forest and range Experimental Station. Ogden, UT. 5p

Litton CM, Giardina CP (2008) Below-ground carbon flux and partitioning: global patterns and response to temperature. Functional Ecology 22:941–954

Litton CM, Raich JW, Ryan MG (2007) Review: carbon allocation in forest ecosystems. Global Change Biology 13:2089–2109

Makita N, Hirano Y, Mizoguchi T, Kominami Y, Dannoura M, Ishii H, Finér L, Kanazawa Y (2011) Very fine roots respond to soil depth: biomass allocation, morphology, and physiology in a broad-leaved temperate forest. Ecological Research 26:95–104

McCormack ML, Dickie IA, Eissenstat DM, Fahey TJ, Fernandez CW, Guo D et al (2015) Redefining fine roots improves understanding of below-ground contributions to terrestrial biosphere processes. The New Phytologist 207(3):505–518

Meier IC, Leuschner C (2008) Belowground drought response of European beech: fine root biomass and carbon partitioning in 14 mature stands across a precipitation gradient. Global Change Biology 14:1–15

Melillo JM, Callaghan TV, Woodward FI, Salati E, Sinha ESK (1990) Effect on ecosystems in climate change: the IPCC scientific assessment, eds Houghton JT. Ephraums JJ (Cambridge University Press, New York), Jenkins GJ, pp 283–310

Melillo JM, Kicklighter DW, McGuire AD, Peterjohn WT, Newkirk K (1995) In: Zepp R, Sonntag C (eds) Global change and its effects on soil organic carbon stocks. Report of the Dahlem workshop on the role of nonliving organic matter in the Earth's carbon cycle. John Wiley & Sons, Oxford, pp 175–189

Melillo JM, Butler S, Johnson J, Mohan J, Steudler P, Lux H, Burrows E, Bowles F, Smith R, Scott L, Vario C, Hill T, Burton A, Zhou Y, Tang J (2011) Soil warming, carbon-nirtogen interactions, and forest carbon budgets. PNAS 108(23):9508–9512

Pregitzer KS, Laskowski MJ, Burton AJ, Lessard VC, Zak DR (1998) Variation in sugar maple root respiration with root diameter and soil depth. Tree Physiology 18(10):665–670

R Development Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3–900051–07-0, URL http://www.R-project.org

Rachmilevitch S, Lambers H, Huang B (2006) Root respiratory characteristics associated with plant adaptation to high soil temperature for geothermal and turf-type Agrostis species. Journal of Experimental Botany 57(3):623–631

Schimel DS, Braswell BH, Holland EA, McKeown R, Ojima DS, Painter TH, Parton WJ, Townsend AR (1994) Cimatic, edaphic, and biotic controls over storage and turnover of carbon in soils. Global Biogeochemical Cycles 8:279–293

Shaver GR, Canadell J, Chapin FS, Gurevitch J, Harte J, Henry G, Ineson P, Jonasson S, Melillo J, Pitelka L, Rustad L (2000) Global warming and terrestrial ecosystems: a conceptual framework for analysis. Bioscience 50:871–882

Trocha LK, Bulaj B, Kutczy'nska P, Muchal J, Rutkowski P, Zadworny M (2017) The interactive impact of root branch order and soil genetic horizon on root respiration and nitrogen concentration. Tree Physiology 37:1055–1068

Wei X, Sendall KS, Stefanski A, Zhao C, Hou J, Rich RL, Montgomery RA, Reich PB (2017) Consistent leaf respiratory response to experimental warming of three north American deciduous trees: a comparison across seasons, years, habitats and sites. Tree Physiology 37(3):285–300

Xia M, Guo D, Pregitzer KS (2010) Ephemeral root modules in Fraxinus mandshurica. The New Phytologist 188(4):1065–1074