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Seasonal respiration of foliage, fine roots, and woody tissues in relation to growth, tissue N, and photosynthesis

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Abstract

Autotrophic respiration may regulate how ecosystem productivity responds to changes in temperature, atmospheric [CO_2] and N deposition. Estimates of autotrophic respiration are difficult for forest ecosystems, because of the large amount of biomass, different metabolic rates among tissues, and seasonal variation in respiration rates. We examined spatial and seasonal patterns in autotrophic respiration in a *Pinus strobus* ecosystem, and hypothesized that seasonal patterns in respiration rates at a common temperature would vary with [N] for fully expanded foliage and fine roots, with photosynthesis for foliage, and with growth for woody tissues (stems, branches, and coarse roots). We also hypothesized that differences in [N] would largely explain differences in maintenance or dormant-season respiration among tissues.

For April–November, mean respiration at 15 °C varied from 1.5 to 2.8 μmol kg^{-1} s^{-1} for fully expanded foliage, 1.7–3.0 for growing foliage, 0.8–1.6 for fine roots, 0.6–1.1 (sapwood) for stems, 0.5–1.8 (sapwood) for branches, and 0.2–1.5 (sapwood) for coarse roots. Growing season variation in respiration for foliage produced the prior year was strongly related to [N] ($r^2 = 0.94$), but fine root respiration was not related to [N]. For current-year needles, respiration did not covary with [N]. Night-time foliar respiration did not vary in concert with previous-day photosynthesis for either growing or fully expanded needles. Stem growth explained about one-third of the seasonal variation in stem respiration ($r^2 = 0.38$), and also variation among trees ($r^2 = 0.43$). We did not determine the cause of seasonal variation in branch and coarse root respiration, but it is unlikely to be directly related to growth, as the pattern of respiration in coarse roots and branches was not synchronized with stem growth. Seasonal variations in temperature-corrected respiration rates were not synchronized among tissues, except foliage and branches. Spatial variability in dormant-season respiration rates was significantly related to tissue N content in foliage ($r^2 = 0.67$), stems ($r^2 = 0.45$), coarse roots ($r^2 = 0.36$), and all tissues combined ($r^2 = 0.83$), but not for fine roots and branches. Per unit N, rates for *P. strobus* varied from 0.22 to 3.4 μmol molN^{-1} s^{-1} at 15 °C, comparable to those found for other conifers. Accurate estimates of annual autotrophic respiration should reflect seasonal and spatial variation in respiration rates of individual tissues.

Keywords: nitrogen, phenology, seasonal variation, temperature response, tissue respiration

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Introduction

Autotrophic respiration may regulate how ecosystem productivity responds to altered environmental conditions such as changes in temperature, atmospheric [CO_2], and N deposition. Extrapolating measurements of respiration taken with cuvettes to the canopy or stand is daunting: a typical sample is on the order of $1:10^8$ of the biomass and is taken for just $1:10^5$ of a year. Despite problems, it is necessary to extrapolate leaf or cuvette-level measurements to the canopy in order to assess carbon and water

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balances for a significant period of time (Ryan *et al.* 1997; Saugier *et al.* 1997), to build and evaluate models (Running & Hunt 1993), and to compare with whole-system flux measurements taken by eddy covariance (Goulden *et al.* 1996; Lavigne *et al.* 1997). For a successful extrapolation, we need to know: (i) the variation of flux rates within the canopy at a given point in time for a standard set of environmental conditions; (ii) the response of flux rates to the environment; and (iii) variation of rates through time, independent of environment (phenological change). Instantaneous response to the environment is probably the best understood for photosynthesis and respiration (Landsberg & Gower 1997). For foliage of trees, variation within the canopy is reasonably well described, because leaf N, specific leaf area, photosynthetic capacity and respiration all vary predictably with the light environment (e.g. (Field & Mooney 1986; Leuning *et al.* 1995; Hollinger 1996). Phenological change in physiology and the controls over those patterns remain poorly understood, particularly for autotrophic respiration.

For respiration, changes in phenology are often difficult to separate from a simple response of increased reaction rate with temperature. This is because physiological activity and growth tend to be greater at higher temperatures, and because many studies confound phenology and temperature, by reporting the temperature response of respiration derived from seasonal differences in respiration and temperature (e.g. Brooks *et al.* 1991; Law *et al.* 1999). In order to separate phenology from temperature, the measured respiration rate needs to be adjusted to a common reference temperature, using a temperature response function derived from measurements using either a temperature-controlled cuvette or from contemporaneous diurnal temperature variability. Using this approach, respiration rates at a common reference temperature can vary substantially throughout the year, both for growing tissues, and also for fully expanded foliage and fine roots. For example, woody respiration for boreal conifers normalized to 15 °C can vary fourfold over the course of the growing season (Lavigne & Ryan 1997), as a result of seasonal differences in wood growth. Respiration of fully expanded foliage and fine roots (presumed to be growing only at the root tips) can vary twofold over the growing season in boreal conifers (Ryan *et al.* 1997).

The most widely accepted model of autotrophic respiration is the two-component, functional model (McCree 1970; Amthor 1989), where respiration is partitioned into that used for construction of new tissues, and that used for maintenance of existing tissues. The model implicitly assumes that any difference in respiration rates is derived from changes in plant growth, and is used in most mechanistically based models of ecosystem carbon balance (e.g. McMurtrie *et al.* 1990; Rastetter *et al.* 1991; Running & Gower 1991). The functional model was derived for annual plants, where growth is the dominant source of respiration, but also has been used to understand autotrophic respiration in forests, where large amounts of existing biomass elevate the importance of maintenance respiration. However, little is known about how well the functional model explains the phenology of respiration for trees, especially conifers (Sprugel *et al.* 1995). For expanding tissues, is knowing the instantaneous growth rate and maintenance respiration sufficient to estimate the annual course of respiration? For fully expanded tissue, with no construction respiration, does maintenance respiration stay constant throughout the year?

Construction respiration may be simpler to extrapolate than maintenance respiration, because measurements of growth are routine, several methods exist that relate easily measured characteristics of tissue to construction cost (McDermitt & Loomis 1981; Vertregt & Penning de Vries 1987; Williams *et al.* 1987), and variation in construction respiration rates are small (max = 1.3 × min, Chung & Barnes 1977) compared with the reported variation in maintenance rates (max = 100 × min, Ryan 1991). One hypothesis proposed to explain differences in maintenance respiration is that tissue N varies within and between tissues (Ryan 1991). In this model, N is used as an easily measured surrogate for protein concentration because most of the nitrogen in plant cells is associated with protein (Lexander *et al.* 1970). Maintenance respiration and protein are linked because maintenance respiration may support protein repair and replacement (Penning de Vries 1975; but see Bouma *et al.* 1994), and because other maintenance processes such as ion transport may be correlated with protein content. To our knowledge, the use of tissue N to scale maintenance respiration for all the tissues of a forest ecosystem has never been tested.

In this paper, we measured respiration of foliage, branches, stems, and coarse and fine roots from the beginning of the growing season through dormancy. The objectives were to determine: (i) if tissue growth, tissue N content, temperature, or photosynthetic efficiency were related to the phenology of respiration; (ii) if seasonal variation in respiration rates is synchronous among tissues; and (iii) whether the relationship between tissue N concentration and dormant season respiration rates (an estimate of maintenance respiration) is conservative across all tissues for a white pine (*Pinus strobus* L.) forest.

Materials and methods

Study site and stand description

The study was conducted in Watershed 1 (WS1) at the Coweeta Hydrologic Laboratory, located in the Southern Appalachian region of Western North Carolina, USA. The

watershed is 16.1 ha, has a southerly aspect, and spans an elevation range of 705–988 m. Soils are mesic Typic Hapludults of the Fannin soil series. Mean annual precipitation is 1786 mm and is distributed evenly throughout the year. The mean annual temperature is 12.6 °C and ranges from an average of 6.7 °C in the dormant season to 18.5 °C in the growing season. WS1 was planted to white pine (*Pinus strobus* L.) in 1957 at a 1.8 × 1.8 m spacing. Competing species were removed with herbicides and by cutting. Basal area of WS1 during the respiration measurements was 54 m^{-2} ha^{-1}, density was 1000 stems ha^{-1}, and leaf area index was 5.5 (Vose & Swank 1990). The high leaf area of the pines precluded growth of understorey vegetation on the study plots.

Respiration measurements were conducted at two locations within WS1. The first location (Plot 1) was located near the bottom of the watershed (elevation 725 m). Plot 1 was also used for a study of nutrient cycling (EPRI Integrated Forest Study; Johnson & Lindberg 1992) and had a 31-m scaffold tower extending through the canopy that was used to sample branches and foliage. Trees in the vicinity of the tower were used for measurements of stems and coarse and fine roots. Plot 2 was at 825 m elevation, and only stem and root respiration were measured.

Climate was measured from a weather station located at the top of the 31-m walk-up tower and from the main Coweeta weather station located approximately 400 m from WS1. Measurements included hourly air temperature, relative humidity, photosynthetically active radiation and precipitation.

Procedures common to gas exchange measurements

All gas exchange measurements were made using an open system (Field *et al.* 1991) with an ADC LCA3 infrared gas analyser (ADC, Hoddeston, Herts, UK). Foliar measurements were made 2–3 h before sunrise, and all other measurements were made during the day. Measurements on foliage and fine roots used the Parkinson Leaf Chamber (ADC); measurements on other tissues used custom cuvettes, constructed of Lexan® plastic and sealed with closed-cell foam, held tightly with spring clamps or elastic cords. Before measurements all cuvettes were tested periodically for leaks by comparing airflow entering and exiting the cuvette. Any leaks were repaired before continuing with measurements. Temperature was measured for all samples using thermistors or thermocouples. Foliage and fine roots were harvested after each sample period to determine nitrogen concentration ([N]); we extracted samples of woody tissues with a corer after the final sample period to measure [N] and sapwood thickness. The nitrogen concentrations of all tissues were determined using a Perkin-Elmer CHN analyser (Perkin-Elmer, Norwalk, CT), on samples dried at 60 °C for 72 h.

Foliar measurements

Foliar respiration (R_F) was measured on attached needles in the dark (2–3 h before sunrise) on 1–2 shoots of 3–4 randomly selected branches in the upper (L1), middle (L2), and lower (L3) thirds of the canopy; the same branches were used for all measurements. R_F was measured monthly from early May to late October. When only one age-class of foliage was present in the canopy, 15 measurements (5 per level × 3 levels) were made per sample period. When two age classes were present, 24 measurements (4 per age class × 2 age classes × 3 levels) were made per sample period. For developing foliage, needles were large enough (about 75% of full length) to be placed in the cuvette in early July. Needles > 1-y-old had abscised by late October. Each sample included 10–15 needles (approximately 0.10–0.20 g dwt). The flow rate through the chamber was 103 $\mu mol\ s^{-1}$.

We measured photosynthesis and predawn leaf water potential (Ψ_P) to determine if they affected R_F. Photosynthesis and Ψ_P were measured on attached needles within a day of respiration measurements on the same branches used to measure leaf respiration. Photosynthesis was measured under ambient conditions of light, temperature and humidity between 11.00 and 13.00 hours. We measured 10 samples on each canopy third (2–4 per branch). When two age-classes of foliage were present, the 10 samples were split between the two age-classes. Photosynthesis was calculated on an area basis, and converted to mass using estimates of specific leaf area for each age-class per canopy level. Area in the cuvette was estimated using age class, canopy position, and sample period specific measurements from 10 random samples (area determined using CID Image Analyser; CID Inc., Moscow, ID). Ψ_P was measured with a Scholander Pressure Bomb (PMS Inc., Corvallis, OR) on four to five needles per branch (by age-class).

Root respiration

Fine root (< 2 mm) respiration (R_{FR}) was measured monthly at 10 random locations on both plots. The litter layer was removed carefully to expose *P. strobus* fine roots growing along the litter/soil interface, without detaching them from the root system. Soil was rinsed from the roots with deionized water, and excess water removed by patting the roots dry with a paper towel. We placed 0.2–0.4 g dwt of roots into the cuvette, and measured R_{FR} at the CO_2 concentration ([CO_2]) of air above the forest floor. Because [CO_2] can alter CO_2 evolution from respiration (Qi *et al.* 1994; Burton *et al.* 1997) and soil [CO_2] is higher

than in the air above the forest floor, *in situ* R_{FR} was estimated as measured $R_{FR}/2.5$, the mean correction determined for *P. strobus* (Clinton & Vose 1999) at the same site.

Coarse root respiration (R_{CR}) was measured monthly (May to October) on eight roots at both plots ($n = 8$ for Plot 1, $n = 8$ for Plot 2). Litter was removed from the base of randomly selected trees and the upper surface of coarse roots exposed. The root surface was rinsed with deionized water, dried, and a 20-cm^2 chamber was permanently installed with silicon caulk. After the caulk dried, the soil and litter were replaced. Inlet and outlet tubing extended through the soil and litter for sampling with the gas analyser. Between sample periods, glass wool placed in the tubing prevented insects from entering the cuvettes, but allowed gas exchange. After the final measurement period, soil and litter were removed, chambers were removed, and the condition of the root beneath the chamber assessed. Data from two chambers were discarded from analyses because of obvious changes in the condition of the tissue beneath the chambers. Root respiration did not differ by plot, so we combined data for analysis.

Branch and stem respiration

Ten branches were selected at the three canopy levels used to measure leaves ($n = 3$ branches in L1 and in L3, $n = 4$ branches in L2) for monthly (May to October) measurements of branch respiration (R_B). Branch sizes varied from 0.9 to 3.2 cm diameter, with dry weights from 0.0082 to 0.087 kg. We installed two closed-cell foam collars 20 cm apart on each branch; during measurements, a plexiglass chamber encircled the branch. The chamber was shaded to prevent heating and bark photosynthesis.

Stem respiration (R_S) was measured using techniques developed by Ryan (1990). Ten trees at each plot (total $n = 20$) were selected to maximize the variation in the ratio of sapwood volume to stem surface area. For each sample tree, we used putty to attach a chamber plate 1.2 m above the ground on the north side of the tree after removing loose bark. R_S was measured monthly (May to October) by attaching a small plexiglass chamber (8.5 × 25 cm) to the chamber plate with an elastic cord. Flow through the chamber was 205 μmol s^{-1} and we allowed 300 s equilibration time before recording. R_S was measured between 06.00 and 09.00 hours, when stem temperature had generally been stable for 1–3 h. Stem respiration did not differ by plot, so we combined data for analysis.

Temperature response

The temperature response of foliar and fine-root respiration was estimated using a temperature-controlled cuvette (Hubbard *et al.* 1995) and the ADC LCA3 analyser during December. December measurements were used to ensure that growth activity was minimal, thus limiting the temperature response to tissue maintenance. The temperature response of R_F was measured for shoots of two branches at each of the three canopy positions, using 5 °C temperature increments from 0 to 35 °C. Temperature response was estimated as β_1 in:

$$R_F = \beta_0 e^{(\beta_1 T)} \quad (1)$$

where β_o and β_1 are regression coefficients and T is leaf temperature. Temperature response of R_{FR} was determined on roots (0.7–1.2 g dry weight) sampled from six random locations in Plot 1. Roots were isolated and prepared as above, root respiration was measured at 5 °C increments from 5 to 25 °C, and (1) used to estimate the temperature response. Stem and coarse-root temperatures varied < 5 °C during the day, because of the energy storage of the biomass and soil. Therefore, we were unable to estimate 'instantaneous' temperature responses for R_{CR} and R_S. We used $\beta_1 = 0.07$ (Q_{10} of 2) for the temperature response of all woody tissue (Amthor 1989), similar to that found for a variety of conifers (Ryan *et al.* 1995).

Stem growth and growth and maintenance respiration

In order to develop a relationship between R_S *and* stem growth and to estimate growth respiration (R_g), we measured periodic growth at ~30 day intervals with dendrometer bands placed 5 cm below the stem respiration chambers ($n = 10$ on plot 1, $n = 10$ on plot 2). Change in circumference over a period was converted to specific growth rate (μmol m^{-3} sapwood s^{-1}), assuming a density of 340 kg m^{-3} (USDA Wood Handbook), a C content of 0.48 kg C kg^{-1}, and uniform growth over the period. Annual diameter growth on the respiration measurement trees was measured with a diameter tape and specific growth rate (μmol m^{-3} sapwood volume s^{-1}) determined using the same assumptions.

Maintenance respiration (R_m) and R_g were estimated monthly and annually by linear regression of specific respiration vs. specific growth rate (Amthor 1989) – the intercept estimates R_m and the slope, R_g. In order to estimate specific respiration rates for each month, we extrapolated values measured each month to the period using the equation:

$$R_S = R_{15} e^{(T_s - 15)} \quad (2)$$

where R_{15} is the respiration rate per unit sapwood at 15 °C, and T_S = sapwood temperature. Sapwood temperature for each hour was estimated as:

$$T_S = \frac{T_{S(h-1)} + (T_A - T_{S(h-1)})}{8.5} \quad (3)$$

where T_A is air temperature at the at the top of the canopy access tower, and $T_{S(h-1)}$ is sapwood temperature of the previous hour. The equation was developed using T_A and average sapwood temperature for 10 trees for two periods (days 195–200 and 336–343; $P<0.001$, $r^2=0.92$). Annual respiration was estimated by summing respiration totals for each period.

Statistical analyses

Relationships among variables were determined using simple linear regression analyses, and multiple regression analyses when more than one independent variable was considered. The statistical significance ($\alpha=0.05$) of the regression line slope parameter (b) was assessed using analysis of variance testing the hypothesis $\beta=0$. The coefficient of determination, r^2, was used to assess the proportion of total variation in the dependent variable explained by the independent variable(s).

Results

Foliar respiration

R_F at 15 °C varied nearly twofold throughout the year for foliage (Fig. 1a). For foliage produced in the prior-year (*'mature'*), R_F was lowest in August (1.40 μmol kg^{-1} s^{-1}) and highest in May (2.83 μmol kg^{-1} s^{-1}). For new foliage, lowest rates (1.66 μmol kg^{-1} s^{-1}) were in August and highest rates (2.86 μmol kg^{-1} s^{-1}) in September. New foliage had a 27% greater respiration rate than 1-y foliage. The foliage temperature response had a Q_{10} of 3.0.

Leaf N, Ψ_P, and A also varied throughout the year, and leaf N and Ψ_P were correlated with R_F of mature foliage. Leaf N varied from 0.86 to 1.00 mol kg^{-1} (Fig. 1b) and peak values occurred in May for mature foliage and in September and October for new foliage. Seasonal R_F of mature foliage was related to seasonal variation in leaf N through late summer ($r^2=0.94$; $P<0.05$; $n=4$). When mature foliage began to senesce in September (needles were beginning to yellow during this measurement period), R_F increased while foliar N remained the same as in August. Foliar N and R_F also covaried by canopy thirds, for measurements from early spring through late summer ($r^2=0.64$; $P<0.05$, $n=12$; Fig. 2). For mature foliage, Ψ_P was lowest in the spring and early summer (Fig. 1c), and was greater for new foliage for all sample periods. Canopy water stress, estimated with Ψ_P, did not explain any additional variation in the relationship between R_F and foliar N ($P=0.54$). For new foliage, R_F was not related to either leaf N or Ψ_P ($P=0.94$ and $P=0.91$, respectively). R_F and A were not related for either new or mature foliage ($P=0.30$ and $P=0.61$, respectively); the ratio of A to R_F varied from 10 to 27 (Fig. 1e) and peaked in August.

Root respiration

R_{FR} at 15 °C was nearly constant throughout the year, except for very high rates in early autumn (Fig. 3a). Fine root N varied seasonally (Fig. 3b), but seasonal differences in mean R_{FR} were not related to fine root N ($P=0.83$). R_{FR} and fine root N were also not related for individual samples ($P=0.09$), which reflect both temporal and spatial variation. However, if we exclude the period of very high respiration in the fall (Day 267) where we suspect fine root growth promoted additional respiration unrelated to N, R_{FR} and fine root N were related ($P<0.01$; $n=78$) (data not shown). The temperature response of R_{FR} had a Q_{10} of 2.0.

R_{CR} at 15 °C was highest in the early spring (1.50 μmol kg^{-1} s^{-1}), and lowest in the late autumn (0.30 μmol kg^{-1} s^{-1}, Fig. 4a). R_{CR} was not synchronized with seasonal patterns of stem diameter growth or R_{FR} (Fig. 4a). To prevent tissue damage, seasonal variation in

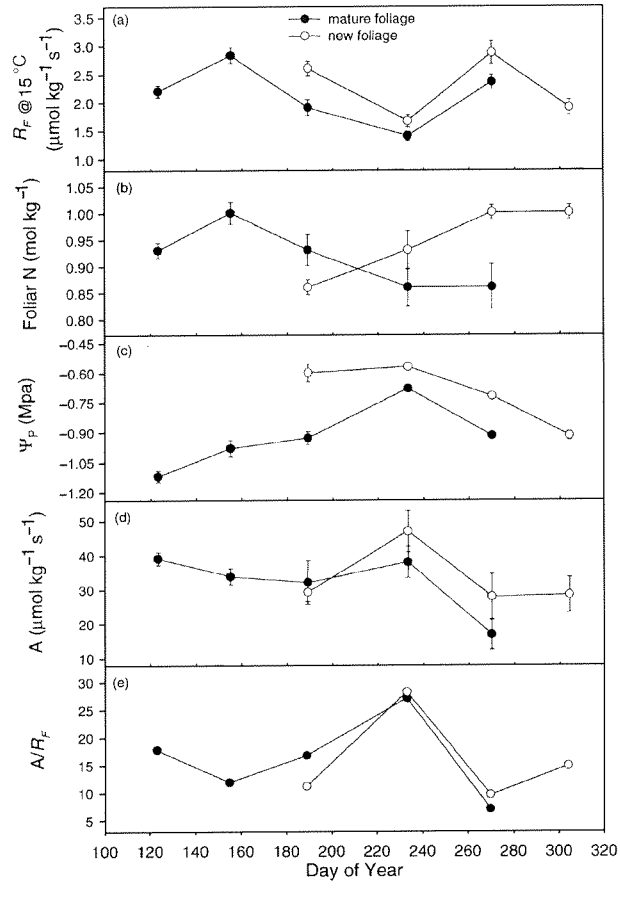

Fig. 1 (a)–(e) Seasonal variation in *Pinus strobus* canopy physiology for mature and new foliage. Data are averaged across three canopy positions (upper 1/3, mid 1/3, and lower 1/3). Bars on data represent SEs. R_F, foliage respiration; N, nitrogen; Ψ_P, predawn leaf water potential; and A, net photosynthesis.

[N] was not measured. R_{CR} measured during the final sample period (late autumn) was related to ($r^2 = 0.36$; $P < 0.05$; $n = 14$) coarse root sapwood N concentration (Fig. 5a).

Branch and stem respiration

R_B and R_S at 15 °C were lower than foliage, fine root, or coarse root respiration. R_B at 15 °C ranged from 0.05 to 0.18 μmol kg^{-1} (sapwood) s^{-1}, and R_S stemwood at 15 °C ranged from 0.06 to 0.11 μmol kg^{-1} (sapwood) s^{-1} (Fig. 4a). Like coarse roots, branch respiration rates were greatest in the early part of the growing season and declined in the late summer and autumn. As with coarse roots, N was measured only at the end of the study. R_B in the autumn and wood N were not related for branches ($P = 0.88$); but R_S and sapwood N were significantly related in autumn ($r^2 = 0.45$; $P < 0.05$; $n = 20$; Fig. 5*b*).

Stem growth and growth and maintenance respiration

Dendrometer band measurements of stemwood growth indicated two peak growth periods, one in early summer (JD 170) and another in late summer (JD 230) (Fig. 4b). R_S for a given 30-day period was significantly correlated with growth within each period ($P < 0.05$). The relationship was stronger ($r^2 > 0.32$) during periods of rapid growth (days 170–263) than during periods of slower growth ($r^2 < 0.18$; days 120–156 and 297–346). Average annual stem respiration was significantly related to average annual growth ($r^2 = 0.43$; $P < 0.05$; $n = 20$, Fig. 6). Using the parameters of the regression model, we estimated average maintenance respiration as 30.0 μmol m^{-3} (sapwood) s^{-1} and growth respiration as 0.17 mol mol^{-1}. We also estimated maintenance respiration as dormant season respiration, and estimated growth respiration by subtracting maintenance respiration from total respiration. Using this approach, average annual maintenance respiration was 20.4 μmol m^{-3} (sapwood) s^{-1} and growth respiration was 0.38 mol mol^{-1}.

Effect of temperature response on seasonal patterns in respiration

Because we were unable to measure the response of respiration to temperature for every sampling period, we checked whether the use of a single temperature response function would have influenced the seasonal patterns in rates at 15 °C. Regressions of R_{15} vs. measurement temperature were not significant ($P = 0.11$ for fine roots, 0.62 for foliage, and 0.06 for stems), indicating that temperature likely had a small effect on seasonal variation. For stems and fine roots, high values at warmer temperatures likely indicate increased growth rather than an effect of temperature acclimation (Table 1).

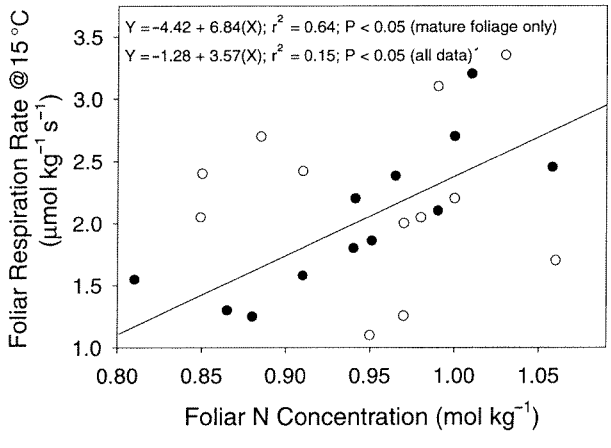

Fig. 2 Linear relationship between seasonal and spatial variation in *Pinus strobus* foliar respiration (corrected to 15 °C) and foliar N concentration. Closed circles represent mature foliage and open circles represent new foliage. The regression line is drawn for the relationship between mature foliage and foliar N.

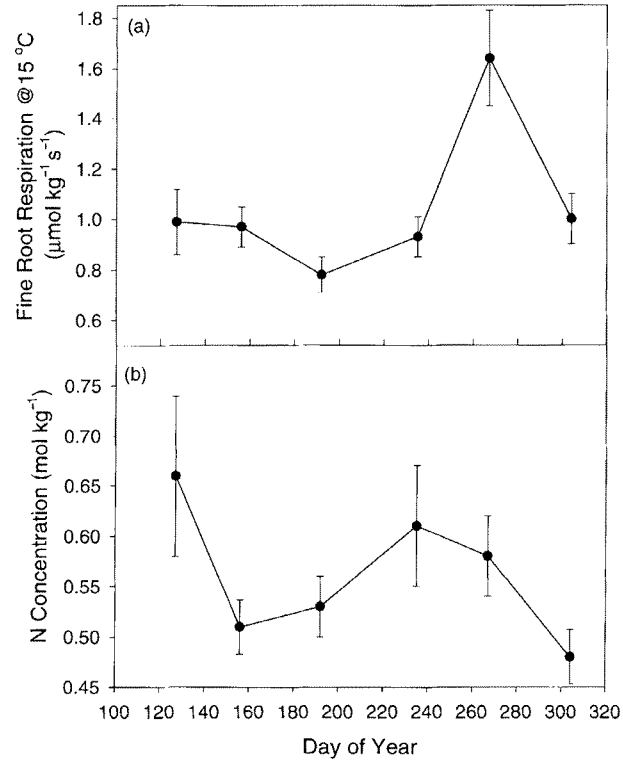

Fig. 3 Seasonal variation in *Pinus strobus* (a) *in situ* fine root respiration (corrected to 15 °C and for ambient measurement [CO_2]) and (b) fine root N concentration. Bars on data represent SEs.

Table 1 Mean respiration rates for fine roots, mature foliage, and stem wood for each measurement date. Rates are given at measurement temperature and adjusted to 15°C. Values in parentheses are SEs

Tissue	Day	Measurement temperature (°C)	Flux at 15 °C ($\mu mol\ kg^{-1}\ s^{-1}$)	Flux at measurement temperature ($\mu mol\ kg^{-1}\ s^{-1}$)
Fine roots	127	21.5 (0.2)	0.99 (0.13)	1.56 (0.22)
	156	18.0 (0.2)	0.97 (0.08)	1.19 (0.09)
	192	23.4 (0.3)	0.78 (0.07)	1.40 (0.12)
	235	22.1 (0.4)	0.93 (0.08)	1.52 (0.13)
	267	17.7 (0.5)	1.64 (0.19)	2.00 (0.27)
	304	19.7 (0.7)	1.00 (0.10)	1.37 (0.14)
Mature foliage	123	15.4 (0.2)	2.20 (0.16)	2.31 (0.18)
	155	23.3 (1.3)	2.83 (0.19)	6.18 (0.55)
	189	20.1 (0.5)	1.90 (0.21)	3.04 (0.36)
	233	14.9 (0.5)	1.40 (0.12)	1.43 (0.16)
	270	9.4 (0.4)	2.35 (0.19)	1.44 (0.13)
Stem	123	12.7 (0.3)	0.081 (0.006)	0.094 (0.006)
	154	18.7 (0.2)	0.124 (0.007)	0.096 (0.006)
	189	20.0 (0.1)	0.115 (0.008)	0.081 (0.005)
	232	14.1 (0.4)	0.096 (0.008)	0.103 (0.010)
	267	14.8 (0.3)	0.084 (0.009)	0.086 (9.4)
	302	13.8 (0.6)	0.054 (0.005)	0.058 (4.9)

Tissue N and maintenance respiration

We used dormant season respiration to estimate maintenance respiration (R_m) for mature and new foliage, branches, stems, and coarse and fine roots. Values for foliage were averaged by canopy position and branch. Tissue [N] varied from $<0.05\ mol\ kg^{-1}$ for stems to $>1.0\ mol\ kg^{-1}$ for foliage, and R_m from $<0.2\ \mu mol\ kg^{-1}\ s^{-1}$ for woody tissue to $>3.0\ \mu mol\ kg^{-1}\ s^{-1}$ for foliage. For all tissues combined, R_m was related linearly to tissue [N] ($r^2 = 0.83$; $P < 0.001$; $n = 54$; Fig. 7). Per unit N, R_m for branches was very low, while R_m for coarse roots was high.

Discussion

Foliar respiration

The twofold variation in R_F corrected to a reference temperature suggests that physiological or biochemical changes throughout the year influence leaf respiration. *Pinus strobus* foliage produced in the previous growing season contributes about 75% of canopy photosynthesis. For this foliage, growing season variation in R_F varied in concert with foliar N content. Retranslocation of N from older to developing foliage (Nambiar & Fife 1987) and an increase in weight per leaf area caused by an increase in nonphotosynthetic tissue (Schoettle & Smith 1999) likely caused the decrease in N concentration as leaves aged. Relationships between foliar N and R_F in conifers have also been seen within crowns (Ryan 1995), between fertility treatments (Ryan *et al.* 1996), for *P. sylvestris* from different latitudes (Reich *et al.* 1996), and for conifers from different biomes (Reich *et al.* 1998). N and respiration may be related because most of N is in protein

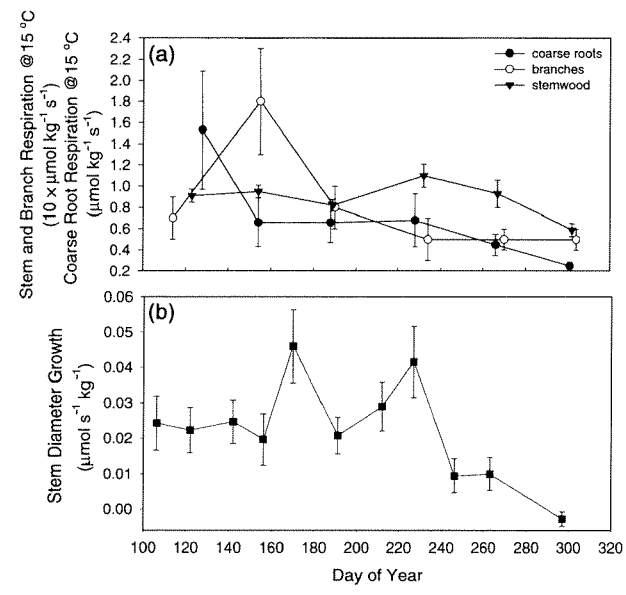

Fig. 4 (a) & (b). Seasonal variation in *Pinus strobus* (a) stem, branch, and coarse root respiration (corrected to 15 °C) and (b) stem diameter growth measured with dendrometer bands. Bars on data represent SEs.

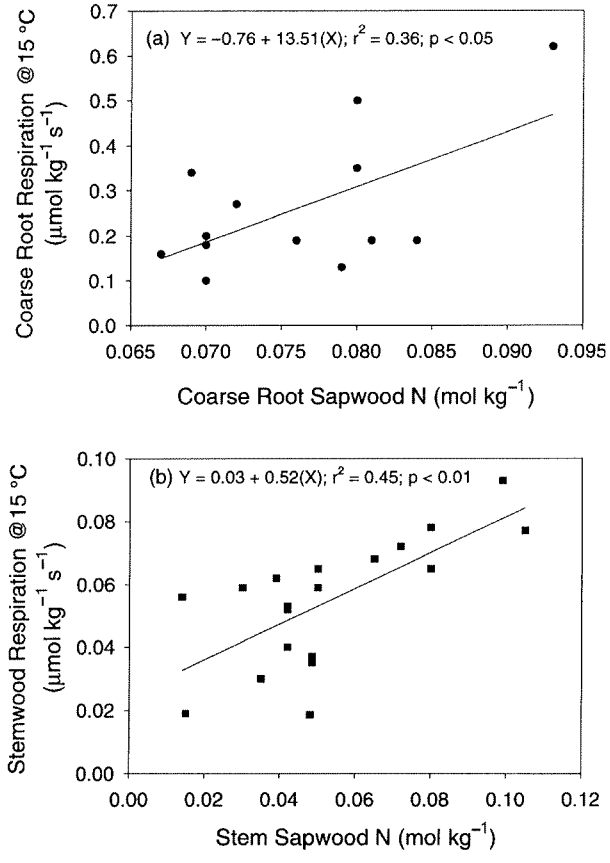

Fig. 5 Relationships between *Pinus strobus* (a) dormant season coarse root respiration (corrected to 15 °C) and sapwood N, and (b) dormant season stemwood respiration (corrected to 15 °C) and sapwood N.

(Lexander *et al.* 1970), which requires maintenance respiration for repair and replacement (Ryan 1991). R_F in developing foliage was unrelated to N, Ψ_P, or A, even though N increased as the leaves developed. When measurements began on current year foliage, needles were approximately 75% expanded, so some of the unexplained variation in seasonal patterns of R_F for current year foliage may be caused by the contribution of growth respiration in developing foliage.

While the seasonal patterns in A and R_F were consistent with what would be expected with developing foliage (Radoglou & Teskey 1997), there was no relationship between A and R_F for either current or 1-y-old foliage. Others (McCree 1982; Azcon-Bieto & Osmond 1983) have suggested that A and R_F are related because carbohydrate concentrations increase with photosynthetic activity, carbohydrates promote respiration (Thomas & Griffin 1994), and photosynthetic capacity increases with foliar N (Field & Mooney 1986; Evans 1989). However, the lack of a relationship between A and R_F in *Pinus strobus* on N-rich sites (Reich & Schoettle 1988) may preclude such a relationship. Alternatively, instantaneous A may be a poor indicator of total A.

Fine and coarse root respiration

Seasonal variation in R_{FR} has also been observed in boreal tree species (Ryan *et al.* 1997), but the pattern differed: temperature-corrected R_{FR} in boreal species was highest in the spring, and lowest in the autumn. Neither seasonal nor spatial variation in root N explained variation in R_{FR}. Other studies that have shown a relationship between fine root N and R_{FR} had differences in N enhanced by

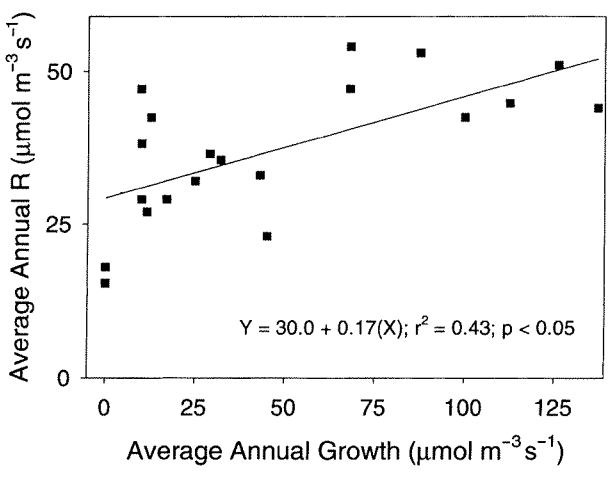

Fig. 6 Relationship between average *Pinus strobus* annual stemwood respiration (corrected to 15 °C) and average annual growth for each measurement tree.

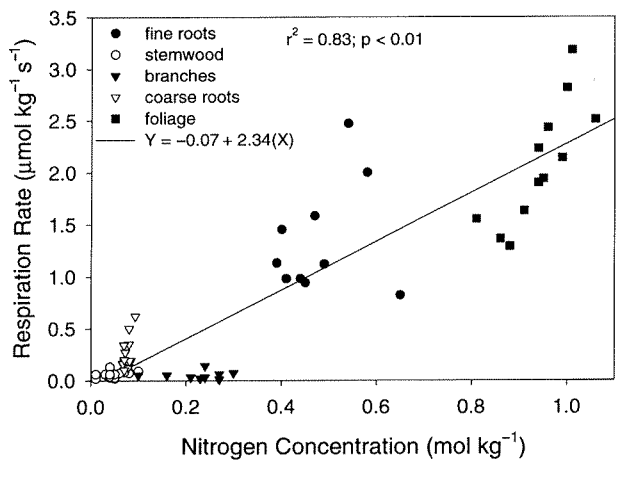

Fig. 7 Relationship between *Pinus strobus* dormant season respiration rate (corrected to 15 °C) for all stemwood, branches, foliage, fine roots, and coarse roots vs. dormant season tissue-specific N concentration.

fertilization (Ryan *et al.* 1996), different soil N availability (Zogg *et al.* 1996), or different root size (Pregitzer *et al.* 1998). Root growth in *Pinus* occurs in response to environment (soil moisture, soil temperature, soil nutrient supply) and assimilate availability (Ericsson & Persson 1980; Eissenstat & Van Rees 1994), both of which can vary considerably over short time scales.

Pinus strobus R_{FR} values were comparable to those of *P. radiata* (0.5–3.0 $\mu mol\ kg^{-1}\ s^{-1}$ at 15 °C), for a similar range of fine root N (0.3–0.9 $mol\ kg^{-1}$), when the *P. radiata* rates were corrected for soil [CO_2] (Ryan *et al.* 1996). *Pinus strobus* R_{FR} values were lower than reported for conifers in general (Ryan *et al.* 1994; Sprugel *et al.* 1995) and two to four times lower than *P. banksiana* (Ryan *et al.* 1997). Rates in the literature surveys were not corrected for soil [CO_2], but R_{FR} for *P. banksiana* were measured at soil [CO_2]. The temperature response of R_{FR} ($Q_{10} = 2.0$) was comparable to those found in other studies (Sowell & Spomer 1986; Cropper & Gholz 1991; Ryan *et al.* 1996).

Peak R_{CR} at Day 128 was perhaps coincident with a flush of coarse root activity (e.g. mobilization of starch reserves) at the onset of soil warming (Eissenstat & Van Rees 1994). Coarse root respiration rates were about 10-fold greater than for branches and stems (Fig. 4a), which is consistent with, but somewhat higher than has been observed in other studies (two- to sevenfold; Ryan *et al.* 1996; Ryan *et al.* 1997). We did not correct R_{CR} for soil [CO_2], but sapwood [CO_2] can be quite high (1.5–8%; Hari *et al.* 1991), and greater than the 0.05–1% in soils, so it is unlikely that soil [CO_2] would depress R_{CR}.

Branch and stem respiration

Seasonal variation in R_S was small compared to branches and stems, and most of the variation was related to periodic growth, where the lowest values occurred in early winter and highest values in mid-summer. R_S in boreal trees had a similar pattern (Lavigne & Ryan 1997). Variation in growth was also important for explaining differences in R_S among trees (Fig. 6). Seasonal variation in R_B (at 15 °C) matched that of foliage, suggesting that R_B may be linked to R_F, perhaps through changes in carbohydrate concentrations or nitrogen metabolism.

R_S for *P. strobus* (0.05–0.11 $\mu mol\ kg^{-1}\ s^{-1}$ or 13–55 $\mu mol\ m^{-3}\ s^{-1}$ at 15 °C) is similar to values reported for other species and ecosystem types. For example, Ryan *et al.* (1997) reported average annual stemwood respiration rates (15 °C) ranging from 18 to 110 $\mu mol\ m^{-3}\ s^{-1}$ across eight boreal forest stands in Canada. A linear relationship between sapwood N content and stemwood R_S in autumn was also found in *P. taeda* (Maier *et al.* 1998), but much of the variation in sapwood N in that study derived from fertilization treatments. In this study, the larger, dominant trees had more sapwood N, suggesting that dominant trees out compete smaller trees for nutrients as well as for light.

Stem growth and maintenance respiration

Average annual stemwood maintenance respiration derived from dormant season R_S was 20 $\mu mol\ m^{-3}\ s^{-1}$ at 15 °C. This value is similar to *P. radiata* (15–39 $\mu mol\ m^{-3}\ s^{-1}$; Ryan *et al.* 1996) and boreal conifers (14–50 $\mu mol\ m^{-3}\ s^{-1}$; Ryan *et al.* 1997), but lower than observed for some temperate hardwood species (15–70 $\mu mol\ m^{-3}\ s^{-1}$; Edwards & Hanson 1996), *Abies amabilis* (86 $\mu mol\ m^{-3}\ s^{-1}$; Sprugel 1990) and *P. taeda* (44–86 $\mu mol\ m^{-3}\ s^{-1}$ at 20 °C; Maier *et al.* 1998). This considerable variation in stemwood maintenance respiration among temperate species indicates that developing stand-level C budgets will require species-, and perhaps stand-level specific estimates of stemwood R_m. Some of the variation in sapwood R_m among species might be related to differences in sapwood [N], and age and growth rate also influence dormant-season R_m (Lavigne & Ryan 1997).

Our average annual growth respiration estimate (the difference between maintenance respiration predicted from dormant season temperature response and total measured stemwood respiration) was 0.38 $mol\ mol^{-1}$. This value is similar to those observed for boreal trees (Lavigne & Ryan 1997) and for a variety of woody species (Griffin 1994). In contrast, the regression of specific respiration and specific growth on an annual basis gave an estimate of 0.17 $mol\ mol^{-1}$ for stemwood growth respiration. This difference suggests caution in applying any single methodology for estimating growth respiration.

All tissues

Differences in R_m (estimated from dormant-season respiration) among *P. strobus* tissues are strongly related to differences in N concentration (Fig. 7). Average respiration per unit of biomass (at 15 °C) varied nearly 40-fold, while respiration per unit N ($\mu mol\ mol\ N^{-1}\ s^{-1}$ at 15 °C) was 1.9 for fine roots, 2.0 for foliage, 3.4 for coarse roots, 1.2 for stem sapwood, and 0.22 for branches. We suspect that branch photosynthesis may have promoted the low branch respiration rates, even though we shaded branches to prevent photosynthesis. Any photosynthetic activity prior to our measurement might have lowered sapwood [CO_2], apparent R_B, and $R_m : N$ because the diffusion of CO_2 in wood and through bark is very slow. Although maintenance respiration may not directly support protein repair and replacement (Bouma *et al.* 1994), tissue N concentration does appear to be a good, general predictor of cellular activity and respiration for the variety of tissues in a *P. strobus* forest.

These results suggest that, in absence of tissue-specific rates, an N-based model may be useful for estimating maintenance respiration budgets for forest ecosystems. Even if tissue-specific rates are available, differences in R_m within a tissue are generally related to N concentration, at least for *P. strobus*. The use of an N-based approach to estimate respiration may be less precise for woody tissue, given the large range of R_m for woody tissue and the large amount of woody biomass in an ecosystem. However, total N in biomass is likely low for wood.

Respiration per unit N for *P. strobus* was similar to those found in other coniferous ecosystems, but lower than those in other functional groups. In order to compare values for *P. strobus* with literature values at a common temperature, we assumed a Q_{10} of 2, and adjusted all values to 15 °C. Compared to 0.22–3.4 μmol mol N^{-1} s^{-1} at 15 °C for *P. strobus*, respiration per N was 1.7 for foliage and 2.6 for fine roots (corrected for [CO_2]) for *P. radiata* (Ryan *et al.* 1996), 3.2 for foliage of boreal conifers (Ryan 1995), 2.1 for a literature survey of various temperate species and tissues (Ryan 1991), 1.9–2.4 for stems and 2.8–3.2 for branches of *P. taeda* (Maier *et al.* 1998), and 3.0 for needle-leaf trees in various biomes (Reich *et al.* 1998). R_m:N for broadleaf trees and shrubs and forbs varied from 4.0 to 7.0 (Reich *et al.* 1998).

Conclusions

Estimates of annual autotrophic respiration will need to reflect seasonal variation in respiration rates of individual tissues to be accurate, as respiration at 15 °C varied two- to threefold from spring to late autumn in all tissues. Seasonal variation in foliage respiration of tissue produced the prior year was related to [N], but only during the active growing season. For current year needles, [N] did not covary with respiration. Foliar respiration did not vary in concert with photosynthesis. Stem growth explained much of the seasonal variation in stem respiration. We did not determine the cause of seasonal variation in branch and coarse root respiration, but it is unlikely to be directly related to growth, as growth patterns in coarse roots and branches were not synchronized with stem growth. With the exception of foliage and branch respiration, seasonal variations in respiration were not synchronized among tissues. Spatial variability in dormant-season respiration rates was correlated with tissue N content in foliage, stems, fine roots, coarse roots, and all tissues combined.

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