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Relating coarse root respiration to root diameter in clonal *Eucalyptus* stands in the Republic of the Congo

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Summary Root respiration is an important component of the carbon balance of a forest ecosystem. We measured CO_2 efflux of excised fine roots and intact coarse roots in 3-, 4- and 13-year-old *Eucalyptus* stands in the region of Pointe-Noire, Republic of the Congo. A transportable and adaptable closed chamber gas exchange system directly measured CO_2 efflux of roots from 0.5 to 32 mm in diameter. Fluxes were corrected for measurement system leaks and normalized to a reference temperature of 30 °C. Mean fine root respiration rates at the reference temperature varied between 8.5 and 10.8 μmol CO_2 kg^{-1} s^{-1} depending on the stand. Coarse root respiration was strongly negatively correlated to root diameter. We propose a model based on a radial gradient of respiratory activity within the root to simulate the exponential decrease in respiration with diameter. Although many sources of uncertainty in the measurements remain, as discussed in this paper, these results provide a basis for scaling up organ-level root respiration measurements to the tree and stand levels.

Keywords: closed chamber gas exchange system, direct measurements, root CO_2 efflux.

Introduction

Ecosystem models of carbon budgets are being applied world-wide to measure the carbon-sequestration strength of a variety of ecosystems, including forests and fast-growing tree plantations (Landsberg and Waring 1997, McMurtrie et al. 2000, Baldocchi and Wilson 2001, Dufrêne et al. 2005). Detailed sub-models have been developed that simulate radiation transfer, photosynthesis and the link between photosynthesis and transpiration over short time scales (von Caemmerer and Farquhar 1981, Verhoef 1984, Baldocchi 1994). In contrast, relatively few studies have focused on carbon allocation by trees to growth and respiration, particularly to the root compartment, despite the importance of root systems in the carbon balance of both soil and trees (Giardina and Ryan 2002). Root respiration (i.e., including rhizomicrobial respiration (Kuzyakov and Larionova 2006)) is estimated to account for between about 30% (Ryan et al. 1996, Xu et al. 2001) and more than 50% (Granier et al. 2000) of total tree respiration, second only to foliage respiration flux (Maier et al. 2004), and it is therefore critical to understand its controls and dynamics in order to simulate tree growth and production correctly. Measurement of root respiration in the field is not easy, however, and a range of techniques has been documented (Hanson et al. 2000, Subke et al. 2006).

Root respiration can be estimated from direct gas-exchange measurements on roots. This approach, involving measurement chambers mounted directly on the plant organ in situ, is the standard method for measuring aboveground woody respiration (Ryan et al. 1996, Clinton and Vose 1999, Ceschia et al. 2002, Meir and Grace 2002, Vose and Ryan 2002, Bolstad et al. 2004, Cavaleri et al. 2006). One can expect good correspondence between above- and below-ground respiration if similar measurement methods are used, which is valuable for scaling up autotrophic respiration to the stand level. Nevertheless, measurement chambers are difficult to use on roots, because of the difficulty of access and of establishing standard conditions in situ. Although the experimental setup has a large influence on what is measured, no standard practice has been established. Factors varying among protocols include location (laboratory, field or greenhouse), the state of the roots (excised or attached, washed or unwashed, whole root system or sample), sampling technique, length of measurement and methods of measuring temperature and [CO_2] (gas-exchange measurement system design). Several factors have been shown to influence root respiration rates, including root diameter (Pregitzer et al. 1998, Desrochers et al. 2002), root nitrogen and total nonstructural carbon concentrations (Ryan et al. 1996, Zogg et al. 1996, Burton et al. 2002, Desrochers et al. 2002), root location and depth (Pregitzer et al. 1998), root age (Bouma et al. 2001, Volder et al. 2005), temperature (Bouma et al. 1997, Atkin et al. 2000, Bryla et al. 2001, Bond-Lamberty et al. 2004), soil water content (Bouma et al. 1997, Bryla et al. 2001) and season (related to above- and belowground growth and phenology) (Gansert 1994, Desrochers et al. 2002, Dannoura et al. 2006, Misson et al. 2006). The need to scale up

organ-level measurements to obtain estimates of tree and stand-level root respiration (Marsden et al. 2008), means that representative measurements must reflect the main variations associated with the key controlling factors at the whole root system level.

Our study, conducted on three stands of plantation *Eucalyptus* in the southern Republic of the Congo, aimed to measure fine and coarse root respiration in the field, and to develop robust relationships for subsequent scaling-up of the organ-based measurements to the stand level. Rates of CO_2 efflux of fine roots (< 2 mm diameter) and coarse roots (2–35 mm diameter) were measured in the field with a closed chamber system and corrected for leakage effects. The variability in respiration rates was examined in relation to root diameter and nitrogen concentration. A model based on a radial gradient in respiratory activity in the root was proposed to describe the strong relationship that was observed between coarse root diameter and respiration rates.

Materials and methods

Study site and duration

The study site is located in the Atlantic coastal zone of the Republic of the Congo (Congo-Brazzaville), in central Africa. About 40,000 ha of *Eucalyptus* plantations have been managed for pulpwood production around the city of Pointe Noire (4° S, 12° E, 100 m a.s.l.) for over 30 years. The original vegetation on these poor sandy soils was a savannah dominated by the C_4 Poaceae, *Loudetia arundinacea* (Hochst.) Steud. The climate is characterized by high mean annual air humidity and temperature (85% and 25 °C, respectively), with low seasonal variation (2% and 5 °C, respectively). Mean annual precipitation was 1274 mm between 1998 and 2005, with a dry season between May and September. Measurements were carried out from April to the end of June 2005. April is the last month of the Congolese wet season, which is characterized by high temperatures, precipitation and global radiation, and May is a transition month; whereas the beginning of the dry season in June is marked by cloudiness, lower temperatures and no rainfall. In 2005, however, April was uncharacteristically dry.

Soils in the area are deep and sandy, classified as ferralic arenosols according to the FAO classification (Trouvé et al. 1994), i.e., with high sand (80–90%) and low clay (8–10%) and silt (2–2.5%) contents. They are characterized by low water retention, a low organic matter content (the top 5 cm of mineral soil contains from 0.45 to 1.1% of carbon, with a mean carbon content of 0.77% in a 3-year-old stand (Trouvé et al. 1994)) and poor cationic exchange capacity (Nzila et al. 2002).

Three stands (A, B and C) were selected for study. Stand A in the Kissoko area was aged 3 years in 2005 and consisted of clones of the hybrid *E. urophylla* × *E. grandis* (UG) at a density of 750 stems ha^{-1}. This is currently the most commonly planted clone in Congo-Brazzaville because of its rapid growth potential. The other stands, located on the Kondi plateau, were 4 years (Stand B) and 13 years (Stand C) old at the time of measurement (a normal rotation lasts 7 years). These stands were planted at densities of 560 and 506 trees ha^{-1}, respectively, with the most productive clone (1-41) of the hybrid *E. PF1* (result of natural crosses of a *Eucalyptus alba* mother tree and poorly identified father trees thought to be crosses of *E. grandis, E. robusta, E. urophylla* and *E. botryoides*) that is widely used in this area.

Fine root sampling

Fine root samples comprised roots less than 2 mm in diameter, taken from the top 15 cm of the soil (where 75% of all root impacts to a depth of 2 m was observed in a neighboring 6.5-year-old eucalypt stand by Laclau et al. (2001)), within a radius of 3 m from the tree trunk. On each measurement date, 20 fine root samples from one plot were analyzed. A portion of surface soil of 30 × 30 cm and 15 cm deep was removed and root segments carefully separated by hand from the sand and organic matter. Brittle roots with a brown center were discarded. A few of the longest and least damaged roots of varying sizes were selected, rinsed in water, blotted and placed in the gas-exchange measurement chamber. Time between excision and the beginning of the measurement was about 5 min. After measurement, the sample was oven-dried at 60 °C to constant mass. Each sample had a dry mass of about 0.5 g. Subsamples were analyzed for root carbon and nitrogen concentrations (NCS2500 analyzer, CE Instrument Thermo Quest).

Coarse root sampling

Diameters of coarse roots ranged from 2 to 32 mm. Because of the shape of the measurement cuvette, we chose regular cylindrical roots with few lateral branches. The soil was excavated from around the tree trunk until a main root was located and carefully exposed. The measured root portions were located between 0.3 and 2 m from the trunk. The diameter of each end of the segment enclosed in the cuvette was estimated as the mean of two measurements made with a vernier caliper. The central part of the measured segment (9 cm long) was cut off after measurement and processed as for the fine root samples. The volume of root segments was computed from the diameter of each extremity assuming a cone-shaped root.

The effect of excision on coarse root respiration was determined on five samples. Respiration was first measured on the intact root, then the root segment was cut and placed in a sealed chamber, and CO_2 efflux was measured at regular intervals for 3 h. For all samples, measured CO_2 efflux rate decreased rapidly in the minutes immediately after excision, reaching a minimum after about 30 min, and then increased to a fairly stable rate. All CO_2 efflux rates measured after excision were higher than the initial rate of the intact root, and the final rate attained was 1.9–2.7 times higher than the rate measured in the intact root. We therefore chose to measure only intact coarse roots.

Measurement system and design of the cuvettes

All respiration measurements were made by circulating gas from the respiration chamber through the sample cell of an in-

frared gas analyzer (EGM-4, PP Systems, Hitchin, U.K.) operating in closed cycle mode. Excised fine roots were placed in the chamber (volume 0.25 l) equipped with a ventilator and a thermocouple probe. Terostat putty (Terostat-7, Teroson, Germany) was used to minimize leaks around the chamber seal and tubing connections. A homemade PVC chamber, similar to that used on stems by Damesin et al. (2002), was placed around the coarse roots, which were kept intact and attached to the root system during measurement. The PVC chamber comprised two half-boxes with semicircular openings to accommodate the root, and removable rubber adapters to accommodate roots of different diameters. The two half-boxes, equipped with a ventilator, thermocouple probe and inlet and outlet ports, were screwed into place and sealed with putty.

Flux calculations

During measurement, the CO_2 concentration was recorded every 5 s for 225 s, starting at a concentration of between 370 and 400 ppm. The concentration increased almost linearly. To account for the slight loss of CO_2 resulting from leakage (see Appendix), we fitted Equation A3 (which describes the increase in chamber [CO_2] over time when there are both respiration and leakage fluxes) to these curves by nonlinear regression. The result of this fit was compared with the fit of a linear equation that assumes that there are no leaks (see Appendix, Equation A1). Compared with Equation A1, Equation A3 showed a better fit to the data, generally almost halving the root mean squared error. We computed the difference between the leakage-corrected fluxes and the non-corrected fluxes. The mean relative difference was 4.52% for coarse roots and 5.11% for fine roots. We used the corrected fluxes for the rest of the study. Coarse root respiration was expressed on a dry mass basis (R_m, μmol kg^{-1} s^{-1}) or on a volume basis (R_v, mmol m^{-3} s^{-1}) by dividing CO_2 efflux (F_c) by the dry mass or the volume of the sample, respectively. Density of fine root tissues was estimated on a subsample, and used to convert rates per unit mass to rates per unit volume.

Temperature normalization

Chamber temperature (T), which varied by up to 10 °C during the day, was recorded during each measurement with a thermocouple probe. We normalized the respiration data (R) to 30 °C (R_{30}) with a simple Q_{10} relationship:

$$R(T) = R_{30} Q_{10}^{\frac{1}{10}(T-30)} \quad (1)$$

We corrected for short-term temperature sensitivity based on a Q_{10} value of 2.2, as observed for *Eucalyptus* grown in root boxes in controlled conditions (Thongo M'Bou, personal communication).

Model of coarse root respiration as a function of diameter

We applied the Levy-Jarvis approach (Levy and Jarvis 1998, Cavaleri et al. 2006) to determine whether R was more closely related to root volume or to root surface area. According to this approach, a positive and linear correlation between respiration per unit area (R_a) and diameter (D) means that respiration is related to volume, whereas a similar correlation between R_v and $1/D$ implies that respiration is proportional to organ surface area. We compared the correlations and found a strong relationship between R_v and $1/D$ (see Figure 1). From an upscaling perspective, such a relationship is unsatisfactory because it implies infinite respiration rates for small root diameters.

To simulate the decrease in coarse root R_v or R_m with increasing D, we propose another equation (Equation 2), which assumes that CO_2 production in a certain tissue volume decreases exponentially from the root surface toward the center, instead of assuming that only the surface elements of the root produce CO_2. In our model, $R_v(x)$ represents the F_c of a given elementary volume at a distance x (mm) from the root surface, R_{vm} (mmol m^{-3} s^{-1}) is the maximum respiration rate, i.e., the respiration rate per volume of the tissues containing the highest proportions of living cells, supposedly localized close to the surface, and k is the exponential coefficient (mm^{-1}):

$$R_v(x) = R_{vm} e^{-kx} \quad (2)$$

Because living cells are absent at depth 0 due to the presence of bark, the thickness of which increases with root D, we tested modifying the equation by including a threshold distance. However, the adjusted parameter was too small to have biological relevance, and was not significantly different from the zero reference. The integration of Equation 2 over all radial directions yields the mean respiration rate of a root segment of radius r and of elementary thickness:

$$\begin{aligned} \bar{R}_v &= \frac{1}{\pi r^2} \int_0^r R_{vm} e^{-kx} 2\pi (r-x)\, dx \\ &= R_{vm} \frac{2}{k^2 r^2} \left(e^{-kr} + kr - 1 \right) \end{aligned} \quad (3)$$

An empirical power coefficient α was introduced to account for other D-related factors that may affect F_c (see Discussion), and we subsequently determined whether its adjusted value was significantly different from 1. The volumetric F_c rate of a root segment of radius r is thus finally expressed as:

$$\bar{R}_v = R_{vm} \left(\frac{2}{k^2 r^2} \left(e^{-kr} + kr - 1 \right) \right)^{\alpha} \quad (4)$$

The values of this function range from R_{vm} to 0 when r ranges from 0 to $+\infty$.

Statistical analysis

The respiration rates of fine roots of each stand were tested for normality by a D'Agostini Pearson test (Trujillo-Ortiz and Hernandez-Walls 2003) which assesses the normality of a sample based on skewness and kurtosis. At a significance level of 0.05, the data were not normally distributed. We therefore tested the existence of a stand effect on fine root respiration rates by the non-parametric Kruskal-Wallis test.

We fitted Equation 4 to the temperature-corrected coarse-root respiration rates, constraining the fit by assuming the respiration rate of roots of $D = 1$ mm (mean D of the fine roots) to be $R_{fr,v}$, the mean rate (on a volume basis) measured in fine roots (4.35 mmol m^{-3} s^{-1}). The normality of the residuals was tested by the d'Agostini Pearson test. The correlations of the residuals with root D and nitrogen concentration were also examined. Model fits were performed by estimating the coefficients of a nonlinear function based on the Gauss-Newton algorithm for least squares estimation.

Results

Fine root respiration

Inter-stand variability in fine root respiration rates was highly significant ($P < 0.0001$). Mean respiration rates varied from 8.5 to 10.8 μmol kg^{-1} s^{-1}, with the lowest rates in the oldest C stand and the highest rates in Stand B (Table 1). There was a weak but significant correlation between nitrogen concentration and fine root respiration, with r^2 values of 0.33, 0.51 and 0.50 in Stands A, B and C, respectively.

Coarse root respiration

The standard deviations of measured coarse root fluxes were high, often more than half of the mean value (Table 1). Measured coarse root respiration rates correlated poorly with tissue nitrogen concentration, whether data were considered separately or pooled across stands (data not shown). Coarse root R_a showed no relationship with D, but standard deviations were relatively high (means of 1.41, 1.21 and 0.94 μmol m^{-2} s^{-1} and standard deviations of 0.75, 0.98 and 0.43 μmol m^{-2} s^{-1} in Stands A, B and C, respectively). Coarse root R_m and R_v were strongly correlated with $1/D$ (Figure 1).

We used Equation 4 to describe the relationship between R_v and D and obtained a good fit to the data, which explained 85% of the variability in respiration rates in the oldest stand (Stand C) and 77% in the 3-year-old UG stand (Stand A) (Table 2, Figure 2) when data for both stands were fit by the equation simultaneously. The α parameter was adjusted to 1.24, significantly different from 0. The use of stand-specific k and α parameters did not improve the goodness of fit. In Stand B, the nonlinear regressions did not converge, possibly because of insufficient data for roots with small diameters (only three roots in the 2–5 mm diameter class in Stand B were sampled, compared with 12 and 10 sampled roots in this diameter class for Stands A and C, respectively; Table 1). However, when Equation 4 was applied to Stand B data based on the parameter values estimated in Stands A and C ($k = 3.67$ mm^{-1} and $\alpha = 1.24$), the simulated versus measured respiration rates were distributed on the 1:1 line as were those of Stands A and C (Figure 3), suggesting that respiration rates followed the same pattern as a function of D in all stands.

The relative residuals of the fit were normally distributed, and the absolute residuals showed a definite heteroscedasticity (higher for smaller diameter roots than for coarser roots, Figure 3b), and a negatively skewed distribution (skewness of –1.30 for Stands A and C). The heteroscedasticity of the residuals may reflect a stronger natural variability in respiration rates of roots of smaller D, or an increased measurement error associated with roots of smaller D. The residuals showed no correlation with nitrogen concentration, indicating that root nitrogen concentration explained no additional variation in respiration rates that was not already explained by D.

Table 1. Number of samples (n), means and standard deviations (σ) of measured respiration rates (μmol kg^{-1} s^{-1}) of roots of five diameter classes (mm), normalized to 30 °C (R_{30}) with a Q_{10} of 2.2.

	Fine roots	Coarse roots			
	< 2	2–5	5–10	10–20	> 20
Stand A: UG, 3 years					
Mean	10.32	4.24	2.37	0.89	0.58
σ	2.54	1.97	1.34	0.47	0.36
n	95	12	23	42	18
Stand B: PF1, 4 years					
Mean	10.84	2.22	1.84	0.80	0.52
σ	3.57	0.92	1.51	0.60	0.35
n	80	3	17	30	13
Stand C: PF1, 13 years					
Mean	8.50	3.54	0.98	0.59	0.35
σ	1.98	2.52	0.55	0.40	0.13
n	78	10	11	20	17

Discussion

Our root respiration measurements were characterized by high variability, some of which was likely associated with measurement artifacts. The causes of uncertainty in our measurements are numerous, including disturbance and wounding effects, and [CO_2] and instrumentation effects (analyzer accuracy and leaks).

For technical reasons, we followed the common practice of measuring fine root respiration in excised roots (Fitter et al. 1998, Pregitzer et al. 1998, Clinton and Vose 1999, Burton et al. 2002, Bahn et al. 2006); however, the validity of measurements on excised organs as a means of estimating respiration of intact organs remains open to question. We made several measurements of respiration rates of fine root samples in rapid succession following excision and found that CO_2 efflux declined rapidly after excision, contrary to observations by Lipp and Andersen (2003) on *Pinus ponderosa* Dougl. ex P. Laws, but confirming results obtained by Rakonczay et al. (1997*a*, 1997*b*) for fine roots of *Pinus strobus* L. For a subsample of coarse roots, respiration rates were systematically higher after excision, even after several hours of stabilization, compared with rates measured before excision. Increased F_c following excision of coarse roots may reflect wound respiration resulting from the sudden liberation of CO_2 transported in the xylem sap flow. Teskey and McGuire (2005) demonstrated that stem F_c is strongly related to xylem CO_2 concentration, particularly in wounded stems. The same effect likely exists in roots. Based on our results and the studies cited, we conclude that (1) if res-

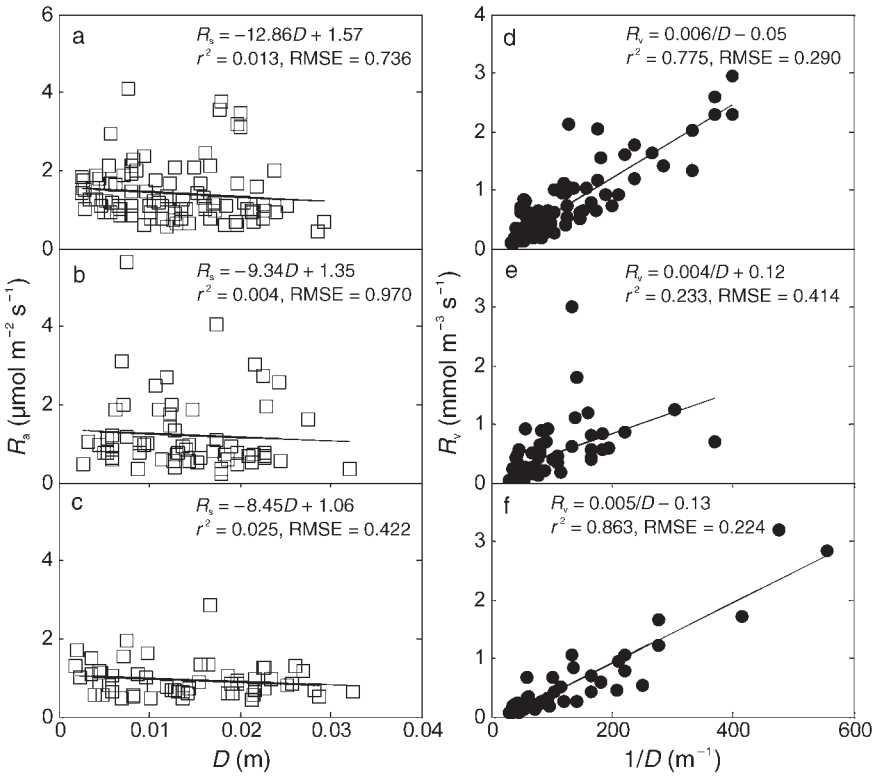

Figure 1. (a, b and c) Root respiration per unit surface area (R_s) compared with diameter (D), and (d, e and f) respiration per unit volume (R_v) compared with the inverse of D in *Eucalyptus* Stands A, B and C, respectively, at a reference temperature of 30 °C.

piration is estimated by measurements on excised organs, the timing of measurement after excision is critical, and (2) excision or any form of wounding of roots should be avoided, even if it means working with fewer samples.

The flux calculation is another possible source of error. There are three potential reasons for the slightly nonlinear rate of increase in CO_2 concentration in the chamber: (1) leaks, for which we corrected our data; (2) the effect of chamber CO_2 concentration on diffusion through the root tissues; and (3) a direct inhibition of cellular respiration at high ambient CO_2 concentration, which has been reported by several authors in the 1990s (Ryan et al. 1996, Burton et al. 1997, Clinton and Vose 1999, McDowell et al. 1999). No recent work has confirmed these results, however, and it has since been shown that leaks in the measurement systems could be responsible for this apparent inhibitory effect of high CO_2 concentration (Amthor et al. 2001, Burton and Pregitzer 2002). A putative effect of chamber CO_2 concentration on diffusion through the root tissues is more difficult to dismiss. When roots are uncovered to measure their respiration, the equilibrium between the CO_2 concentrations inside and outside the root tissues is disturbed and F_c by diffusion through the root surface can be momentarily enhanced because of this larger gradient, so that it no longer reflects the rate of CO_2 production by the root. Later,

Table 2. Parameters and results of the fit of Equation 4 (respiration as a function of root diameter) to coarse root respiration data of *Eucalyptus* Stands A and C. Abbreviations: R_{vm}, calculated maximum value of root tissue respiration on a volume basis (mmol m^{-3} s^{-1}); k (mm^{-1}) and α, adjusted parameters ± 95% confidence interval; and r^2 and root mean squared error (RMSE, mmol m^{-3} s^{-1}) of the regression of simulated against measured values, analyzing each stand separately and pooling both stands.

	R_{vm}	k	α	r^2	RMSE
Stand A				0.77	0.303
Stand C	8.36	3.67 ± 1.71	1.24 ± 0.20	0.85	0.263
Both stands				0.78	0.289

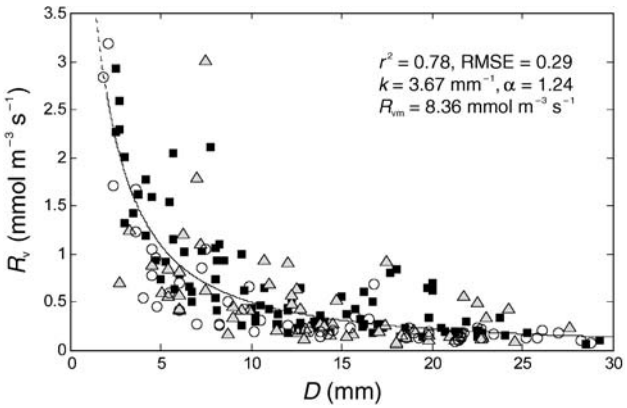

Figure 2. Measured root respiration per unit volume (R_v) of coarse roots as a function of diameter (D) in *Eucalyptus* Stands A (■), B (▲) and C (○), at a reference temperature of 30 °C. The solid line represents the fit of Equation 4 to the data for Stands A and C. The dashed line represents the extrapolation to roots 1–2 mm in diameter.

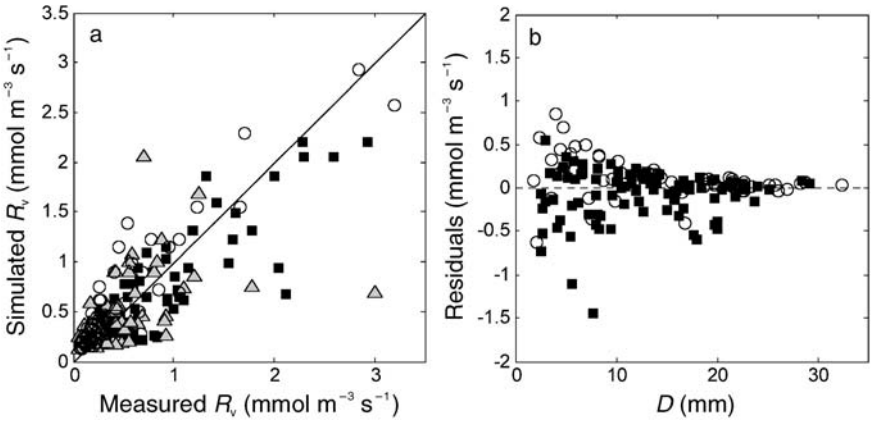

Figure 3. (a) Measured values of coarse root respiration on a volume basis (R_v) versus values simulated with Equation 4 adjusted on Stands A and C simultaneously, for *Eucalyptus* Stands A (■), B (▲) and C (○), at a reference temperature of 30 °C; the 1:1 line is also represented. (b) Residuals of the model (Equation 4) applied to Stands A (■) and C (○) compared with root diameter (D).

however, as the CO_2 concentration in the chamber increases, the gradient decreases and the efflux rate tends to decrease, resulting in a curvature of the increase in chamber CO_2 concentration with time, which could be mistaken for the effect of a leak.

If measurements are made during this transitory period, and are corrected for an apparent leak, the respiration flux would be doubly overestimated. Unfortunately, it is difficult to know when equilibrium is regained and fluxes reflect the true rate of CO_2 production rather than a transitory, artificial rate. We are therefore cautious about the leak correction we applied to our data, because it may be responsible for a 5% overestimation of respiration.

Growth is variable during the year and is responsible for cellular activity that necessarily induces increased respiration (Amthor 2000, Cannell and Thornley 2000), hence different growth rates of our samples could be responsible for some of the variability in our data. The commonly made distinction between maintenance and growth respiration (Thornley 1970)—which is not based on knowledge of plant functioning—has often been criticized because it is correlative and may be physiologically misleading (Hansen et al. 1998). Respiration should instead be related to individual processes, several of which are involved in both growth and tissue maintenance (Amthor 2000, Cannell and Thornley 2000, Thornley and Cannell 2000). Nevertheless, in field studies it is reasonable to make measurements on growing and non-growing organs with the aim of assessing the variability in respiration associated with growth (Desrochers et al. 2002).

We attempted to compare respiration during a period of active tree growth (end of the wet season) with that during a period of little or no growth (beginning of the dry season). However, because of the unusually dry conditions between March and May 2005, the entire experimental period can be considered to represent a time of little or no root growth in the superficial root layers that we sampled, although we have no experimental data to support this statement. Reliable monitoring of root growth is difficult to achieve, and cannot be done simply by installing permanent dendrometers as is the case for stems. In a rhizotron study on *Pinus taeda*, Sword et al. (1996) reported that growth of fine roots was concomitant with aboveground growth, and that during a period of soil water deficits, new root initiation was displaced to deeper soil layers.

Our fine root respiration rates of about 10 μmol kg^{-1} s^{-1} at 30 °C, are in the same range as the mean of published values for several tree species (Zogg et al. 1996, Rakonczay et al. 1997*b*, Bryla et al. 2001, Desrochers et al. 2002, Burton and Pregitzer 2003, Ryan et al. 2006). Published respiration rates for coarse roots are scarce. The mean summer respiration rate of coarse and medium roots of a few 10-year-old beech saplings was about 1.5 μmol kg^{-1} s^{-1} (Gansert 1994). Rates corresponding to 1.7 μmol kg^{-1} s^{-1} at 30 °C have been reported for *Pinus taeda* L. coarse root samples extracted from soil cores (Maier and Kress 2000), but these samples included roots of varying diameters. Desrochers et al. (2002) studied respiration of long segments excised from coarse roots from a 55-year-old aspen (*Populus tremuloides* Michx.) stand, with diameters ranging from 5 to 50 mm, and reported a mean respiration rate equivalent to 1.84 to 2.61 μmol kg^{-1} s^{-1} at 30 °C (using their Q_{10} value of 2.57), depending on season, which is higher than our estimates (means of 1.6, 1.1 and 1.1 μmol kg^{-1} s^{-1} over all coarse root samples in Stands A, B and C, respectively). Desrochers et al. (2002) did not report on the distribution of diameters in their samples, so we do not know whether our samples are comparable. Pregitzer et al. (1998) measured O_2 consumption of excised sugar maple roots and found a value corresponding to a CO_2 efflux of about 3.5 μmol kg^{-1} s^{-1} at 30 °C for roots in the 2–10 mm diameter range, which agrees well with our measurements in Stand A (3 μmol kg^{-1} s^{-1} for 2–10 mm roots). Studies of pines (Ryan et al. 1996, Vose and Ryan 2002, Wieser and Bahn 2004) also gave comparable values, ranging from 0.2 to 5 μmol kg^{-1} s^{-1}.

In most of these studies, a negative correlation between F_c and root D was observed. Based on five coarse root samples, Ryan et al. (1996) proposed a decreasing exponential relationship between coarse root R_v and D, and we have now confirmed this relationship. Furthermore, we found that the relationship was robust and the shape of the curve of the exponential decrease was conservative among the three stands.

Measurements on stems have also shown a negative relationship between D and stem R_v, of similar shape to the one we present (Ceschia 2001, Ceschia et al. 2002, Bosc et al. 2003,

Cernusak et al. 2006, Kim et al. 2007). Empirical exponential, inverse or power functions were proposed to describe the relationship, and Bosc et al. (2003) included stem age in their model. These relationships are valuable because they provide a good simulation of the measured trend, and can be used for scaling-up provided that they are not extrapolated to diameters outside the measurement range: the main disadvantage of these relationships is that they imply infinite respiration rates for stems of small diameter.

Levy and Jarvis (1998) deduced from their stem respiration measurements that the F_c was mainly due to tissues located close to the stem surface, rather than to tissues present throughout the whole stem volume, i.e., R_v was related to $1/D$, as observed in our study. A sequence of studies on the potential respiration rate of differently localized tree stem tissues (Pruyn et al. 2002*a*, 2002*b*, Spicer and Holbrook 2007*a*) showed that the respiratory potential decreased from inner bark (near the surface) to outer then inner sapwood. Inspired by these results, and since roots that have undergone secondary radial growth are similar in structure to stems, we propose that radial differences in root tissue respiration rate can be simplistically described by an exponential decrease in rate with increasing distance from the root surface. Integrated over the volume of a root segment, this yields an equation relating segment R_v to D, with two parameters: R_{vm}, the maximal respiration rate of tissues close to the surface; and k, the exponential coefficient.

The distribution of living cells may partly explain the radial variations in tissue respiration rates. Stockfors and Linder (1998) showed that the proportion of living cells in Norway spruce (*Picea abies* (L.) Karst.) stem sapwood decreases exponentially with distance from the cambium. This distribution could be species specific: Ceschia (2001) and Spicer and Holbrook (2007*b*) noted the same exponential decrease in *Picea abies*, *Pinus strobus* and *Tsuga canadensis* (L.) Carr., but a constant distribution in four angiosperms. Another likely explanation of radial variations in tissue respiration rates resides in the radial differentiation of specific tissues inside woody organs, which, because of their different functions, must vary in energy requirements.

When Equation 4 was fitted to the data from Stands A and C, and constrained by respiration rates measured on fine roots of mean diameter 1 mm, it yielded a value of 8.4 mmol CO_2 m^{-3} s^{-1} for R_{vm}, i.e., about double the mean fine root respiration rate. This value is higher, but roughly of the same order as, the maximal respiration rate of *Prunus* seedling roots in non-limiting conditions of O_2 supply at 20 °C (2.5 mmol CO_2 m^{-3} s^{-1}) presented by Bidel et al. (2000), which would equate to about 5 mmol CO_2 m^{-3} s^{-1} at 30 °C. The k parameter was adjusted to a value of about 3.5 mm^{-1}. According to Equation 4, this would mean that the respiration rates of tissues situated 1 mm from the root surface were only about 5% of R_{vm}. The adjusted α parameter of Equation 4 was significantly greater than one, and the model including the α parameter displayed smaller root mean squared errors and better distribution of residuals than a model without the power function. This implies that the simulated respiration of fine roots was enhanced compared with that of thicker roots to better fit to the data. The slight growth may have occurred more quickly in finer roots than in coarser roots, resulting in higher respiration rates.

We found that respiration of all roots of $D > 2$ mm could be estimated as root surface area multiplied by a constant. Although Equation 4 did not perform better than this simple method of scaling up our dataset, it may be valid in a variety of situations; e.g., where respiration is more closely correlated to volume than to surface area, or to both volume and surface area (as was found for CO_2 efflux of wood less than 2 m above the ground by Cavaleri et al. (2006)). In Equation 4, when k is close to zero, R_v approaches a constant R_{vm}, meaning that R_a is a linear function of root radius, whereas higher values of k apply when respiration rates are highest in tissues close to the root surface.

Improved methods for monitoring root growth in the field are needed to develop a more mechanistic description of root F_c and its seasonal variations. More work is also needed on the characterization of root tissues at different developmental stages. The influence of sap flow on measured root respiration should also be considered because there is evidence that respiration rates of certain tissues are limited by hypoxia when xylem sap flow is low (Bidel et al. 2000, Pruyn et al. 2002*a*). Others have found dissolved CO_2 in xylem sap (Teskey and McGuire 2005, 2007, McGuire et al. 2007), and suggested that the root system could be a large source of the CO_2 measured in stem F_c. In this case, a higher xylem sap flow may, on the one hand, boost respiration by ensuring a non-limiting oxygen supply, and on the other hand, diminish apparent respiration by favoring the escape of respired CO_2 through sap flow rather than through radial diffusion.

To gain more confidence in the estimations of autotrophic respiration, CO_2 efflux measurement techniques must be improved to eliminate wound respiration and the effects of CO_2 gradients. Nevertheless, our demonstration of a strong relationship between coarse root D and F_c is a first step toward the scaling up of direct measurements. Our model presents a good fit to measured coarse root F_c values, and predicts realistic respiration rates for roots of smaller D, and seems, therefore, to be a good candidate for scaling-up root F_c measurements. Whether Equation 4 proves broadly applicable to other species remains to be seen.

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Appendix: Pattern of change in [CO_2] in the chamber during measurement

In a perfect closed chamber system, the rate of change in chamber CO_2 concentration as a function of time (t, s) can be described as:

$$C_c(t) = t\frac{F_c}{\rho V} + C_c(t_0) \quad \text{(A1)}$$

where C_c is CO_2 molar concentration inside the chamber (μmol mol^{-1}), F_c is efflux of CO_2 from the enclosed organ (μmol s^{-1}), ρ is molar density of air (mol m^{-3}), and V is volume of air inside the chamber (m^3).

It is critical when using closed gas-exchange systems to ensure that leaks are negligible, as they can introduce a parasitic flux that depends on the CO_2 concentration gradient between the inside and outside air (Kimball 1990, Tingey et al. 2000, Baker et al. 2004). The pattern of change in chamber CO_2 concentration during the measurement is governed by the difference between respiration and leakage fluxes:

$$\frac{dC_c}{dt} = \frac{F_c}{\rho V} - \frac{L_c(t)}{\rho V} = \frac{F_c}{\rho V} + \frac{g}{\rho V}\left(C_a - C_c(t)\right) \quad \text{(A2)}$$

where C_a is CO_2 molar concentration of outside the chamber (μmol mol^{-1}), L_c is leakage flux (μmol s^{-1}), and g (mol s^{-1}) is the inverse of the chamber "resistance to loss of CO_2" (following the term used by Baker et al. (2004)).

A solution of the differential equation is given by:

$$C_c(t) = \frac{1}{g}\left(g\left(C_{c0} - C_a\right) - F_c\right)e^{-\frac{g}{\rho V}t} + \frac{1}{g}\left(F_c + gC_a\right) \quad \text{(A3)}$$

where C_{c0} is CO_2 molar concentration in the chamber at the beginning of the measurement (after an initial mixing phase). This equation can be fitted to experimental data to estimate both CO_2 efflux from the organ and leakage flux.