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Root growth and physiology of potted and field-grown trembling aspen exposed to tropospheric ozone

M. D. COLEMAN,[1] R. E. DICKSON,[1] J. G. ISEBRANDS[1] and D. F. KARNOSKY[2]

[1] *USDA Forest Service, North Central Forest Experiment Station, Forestry Sciences Laboratory, 5985 Highway K, Rhinelander, WI 54501, USA*

[2] *School of Forestry and Wood Products, Michigan Technological University, Houghton, MI 49931, USA*

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Summary We studied root growth and respiration of potted plants and field-grown aspen trees (*Populus tremuloides* Michx.) exposed to ambient or twice-ambient ozone. Root dry weight of potted plants decreased up to 45% after 12 weeks of ozone treatment, and root system respiration decreased by 27%. The ozone-induced decrease in root system respiration of potted plants was more closely correlated with decreased root dry weight than with specific root respiration, suggesting that aspen root metabolism was less affected by ozone than root growth. We used minirhizotrons to study the appearance and disappearance of roots in the field. Length of live roots of field-grown trees increased rapidly early in the season and peaked by midseason in association with a decrease in root production and an increase in root disappearance. In the twice-ambient ozone treatment, live root lengths were 17% less than those of controls, but the effect was not statistically significant. Seasonal soil CO_2 efflux of field-grown trees decreased significantly in the ozone treatments, but because differences in live root length were not significant and root dry weights were not available, the effect on CO_2 efflux could not be attributed directly to decreased root growth.

Keywords: fine root dynamics, growth respiration, maintenance respiration, Populus tremuloides, root respiration, soil CO_2 efflux.

Introduction

Both trees and agronomic plants exposed to ambient concentrations of ozone show negative growth and metabolic responses (Heck et al. 1983, Pye 1988). The negative response of root growth is typically equal to or even greater than the response of shoots (Tingey 1974, Cooley and Manning 1987, Mooney and Winner 1988). Decreased root growth is attributed to decreased carbon translocation to roots (Tingey 1974, Blum et al. 1983, McCool and Menge 1983, Ito et al. 1985, Cooley and Manning 1987, Adams et al. 1990, Spence et al. 1990, Andersen et al. 1991, Gorissen et al. 1991*a*, 1991*b*). We have found up to a 40% decrease in root growth and up to a 70% decrease in the total amount of carbon allocated to roots of potted aspen (*Populus tremuloides* Michx.) exposed to ozone (Karnosky et al. 1995, Coleman et al. 1995). In addition to decreased growth and carbon allocation, root respiration per unit root length decreases in plants exposed to ozone (Hofstra et al. 1981, Ito et al. 1985, Edwards 1991).

Most studies on the effects of ozone on root growth of trees have been conducted on seedlings grown in pots. Because tree age may affect the response to gaseous pollutants as a result of the many morphological and physiological differences between potted and field-grown trees (Cregg et al. 1989), it may not be valid to extrapolate the experimental data collected on seedlings to predict adult tree responses to ozone (cf. Samuelson and Edwards 1993, Edwards, et al. 1994, Grulke and Miller 1994, Hanson et al. 1994). To test the relevance of results with seedlings in predicting adult tree responses to ozone, we investigated ozone effects on belowground responses in both potted plants and field-grown trees.

Methods

Pot experiment

Rooted cuttings of three aspen clones (216, Bayfield Co., WI, ozone tolerant; 271, Porter Co., IN, ozone tolerant; 259, Porter Co., IN, ozone sensitive) were grown in 1.7-m^3 exposure chambers. Air entering the chambers was either supplemented with ozone applied in a square wave (150 ppb O_3, 8 h daily) or left untreated (controls). The ozone application resulted in 10 ppm h accumulated each week compared to 3 ppm h for the controls. Two chambers were used per treatment. Plants were grown under these conditions for 12 weeks in 6-liter pots (15-cm diameter, 35 cm tall) containing peat/sand/vermiculite (2/1/1, v/v). Each pot received 1 liter of balanced nutrient solution daily. The nutrient solution concentration increased by 4% day^{-1} from a starting concentration of 1.22 mg N l^{-1} (McDonald et al. 1991). From each clone, one to two plants per chamber (n = 2 to 4) were measured and then harvested every 3 weeks.

Respiration was measured by sealing the pots in a cuvette, with internal air circulation, attached to an open-flow gas exchange system (MPH-1000, Campbell Scientific Inc., Logan, UT) with an infrared gas analyzer to measure CO_2

concentration (LI-6251, Li-Cor Inc., Lincoln, NE). Before harvest, CO_2 efflux from the pot gave a measure of total root and soil respiration. After all roots were removed at plant harvest, the soil was returned to the pots for measurement of soil respiration. To achieve stable readings, the soil was equilibrated for 5 days and was watered 18 h before CO_2 efflux from the pot was measured. Root system respiration (nmol plant^{-1} s^{-1}) was calculated as the difference between pre- and post-harvest CO_2 efflux rates. Harvested roots were dried and weighed. Specific root respiration was calculated by dividing root system respiration by root dry weight. Respiration was measured at 350 ppm CO_2 at temperatures between 14 and 22 °C. Respiration rates were adjusted to 20 °C assuming $Q_{10} = 2$.

Plantation experiment

In autumn 1991, dormant first-year aspen plants grown in 7-liter pots from rooted greenwood cuttings were planted in 2-m diameter circular trenches. Twelve plants were spaced 0.5 m apart in each of nine trenches. Each trench was backfilled with extrinsic sandy-loam topsoil. A 3-m diameter open-top chamber was placed over the experimental material in each circular trench. Six individuals of two aspen clones (216, ozone tolerant; 259, ozone sensitive) were arranged randomly in each circular plot. The genotypes were selected to assess variation in sensitivity within the species, but we have considered only the overall species response because it was not possible to distinguish between roots of the individual clones.

The experiment consisted of three treatments applied in standard 3-m diameter open-top chambers (Heagle et al. 1973). There were three replicate chambers per treatment arranged in a completely random design. The gaseous treatments included: (1) charcoal-filtered air (CF), (2) charcoal-filtered air with simulated ambient ozone added (1×), and (3) charcoal-filtered air with simulated twice-ambient ozone added (2×). The 1× treatment was derived from hourly ambient profiles constructed from 1987 ozone data collected in Michigan's Lower Peninsula. A sigmoidal weighting of the ambient profile (Karnosky et al. 1995) was used to produce the 2× ozone treatment. The 1992 growing season was the first year of treatment. The seasonal ozone exposures spanned the period from June 10 to September 14, 1992, from June 1 to September 15, 1993, and from June 6 to September 1, 1994. The average seasonal exposure (± standard deviation) was CF = 10 ± 3, 1× = 58 ± 4 and 2× = 71± 6 ppm h in 1992, CF = 13 ± 5, 1× = 53 ± 1 and 2× = 91 ± 3 ppm h in 1993, and CF = 14 ± 6, 1× = 68 ± 5 and 2× = 104 ± 9 ppm h in 1994. At the end of the 1992 season, six plants were harvested from each chamber. After the 1993 season, four more plants were harvested. Before the 1994 season, chambers were doubled in height to accommodate the growing trees.

In May 1993, two acrylic minirhizotron observation tubes (5 cm inside diameter) were installed in each of the open-top chambers for a total of 18 tubes. Two tubes were installed within each circular trench next to one individual of each clone. Tubes were installed at a 45° angle to an average vertical depth of 29 cm. To ensure that only aspen roots were observed from minirhizotrons, the plots were kept free of ground vegetation by hoeing. Root observations were made with a microvideo camera designed to travel down the clear acrylic tubing while recording images of the exterior surface (Bartz Technology, Santa Barbara, CA). The indexing handle allowed the camera to travel down the tubes while observing successive 13.5-mm deep × 18-mm wide frames on the exterior surface. Root observations were recorded on June 21, July 14, July 29, August 3 and August 16.

Video images of the frames were assessed with the ROOTS video-image analysis program (Hendrick and Pregitzer 1992*a*, 1992*b*). Only roots smaller than 2 mm in diameter were considered in the analysis. The lengths of fine roots that were either new, previously measured, dead or missing (disappeared) were totaled for each observation time in each tube. Given that roots appearing in minirhizotrons are a sample of the fine root population, fine root production can be measured as the appearance of roots, and survivorship can be determined through their disappearance. New roots seen at each observation time were considered to be a cohort distinguished by their similar age.

Soil CO_2 efflux was determined with a 6000-09 soil respiration chamber attached to the LI-6200 portable photosynthesis system (Li-Cor Inc.). Permanently installed plastic sampling collars (10 cm diameter) were inserted to a depth of 2.5 cm. The chamber was placed on the collar, and circulating air in this closed system was scrubbed of CO_2 for 3 to 6 s. Six observations were collected at 5-ppm intervals as the CO_2 concentration increased beyond the ambient treatment CO_2 concentration (Norman et al. 1992). Two early afternoon measurements were taken in each chamber directly below treatment trees on four occasions during the 1994 growing season. Soil water was not controlled, but temperature corrections were made to 20 °C assuming $Q_{10} = 2$.

Data analysis of pot experiment

Least-squares regression analysis was used to examine the relationship between respiration and root dry weight. The suitability of a linear versus a nonlinear model was examined with an *F*-test for a significant improvement following the addition of a quadratic term (Kleinbaum and Kupper 1978). Relative growth rate in stem volume (diameter squared × height) was calculated from data collected 1 and 8 days before harvest. We used stem volume relative growth rate to estimate root relative growth rate after confirming stem volume was a good predictor of root dry weight (R^2 from 0.73 to 0.97). The hypotheses that ozone or clonal effects resulted in different relationships between specific root respiration versus root relative growth rate were tested by comparing linear regressions according to the *F*-test method of Ratkowsky (1983).

A three-way factorial analysis of variance was used to test for treatment differences. The three factors in the design were ozone, clone and harvest time. Because ozone treatment was applied within chambers, a split-plot analysis was employed. The main-plot error (chamber within treatment) was used to test ozone treatment effects.

Data analysis of plantation experiment

Several fine root growth parameters were measured and calculated (Cheng et al. 1991). Briefly, total root production was the total cumulative new root length that appeared. Root death or disappearance was the length of previously observed roots that disappeared. The difference between total root production and root disappearance is the standing live root length. Specific growth rate and specific death rate are the rates of roots appearing or disappearing between any two observations normalized by the average standing live root length for that period. The root turnover index is the average of specific growth rate and specific disappearance rate. The rates of growth and disappearance are averaged to estimate the rate of replacement on a standing crop basis for each observation period (Odum 1975).

A split-plot repeated-measures analysis of variance was used to test for ozone treatment differences in response parameters. As in the pot studies, the design had treatments nested within chambers so the main-plot error was used to test ozone treatment effects. Orthogonal polynomials were used to examine the form of the response over time.

Results

Pot experiment

Root respiration increased during the experimental growth period (Figure 1). The increase in root system respiration over time is explained by increased root dry weight (Figure 2A) rather than by a major change in soil respiration (Figure 1). However, the increase in root system respiration was not directly proportional to the increase in root dry weight, hence there was a nonlinear decline in specific root respiration with increasing root dry weight (Figure 2B). There was a direct proportionality between specific root respiration and root relative growth rate according to the functional model of respiration (Hesketh et al. 1971, Amthor 1989, Sprugel et al. 1995) (Table 1).

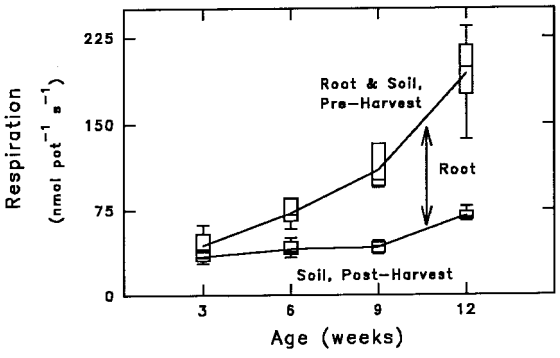

Figure 1. Changes in root and soil respiration rates with plant age. Box plots show the range for all treatments and all pot-grown aspen clones combined at each of four plant ages. The respiration rate for the intact plant–soil system is indicated by the pre-harvest line. The soil respiration rate after removal of the plant root system is indicated by the post-harvest line. The difference between the two lines (arrow) is the root respiration rate fraction of total soil respiration.

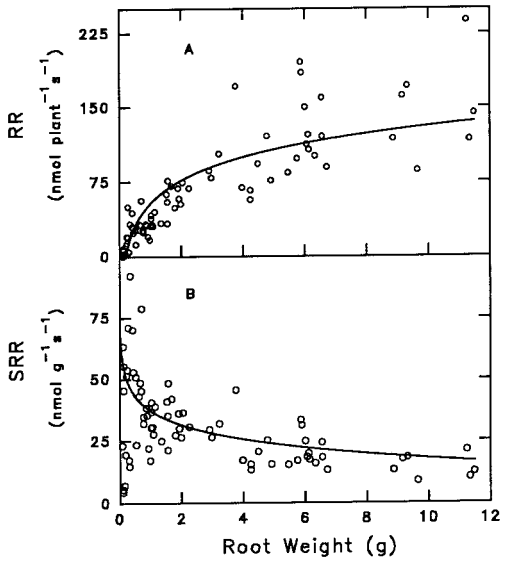

Figure 2. Changes in root respiration rates with root dry weight. (A) Root respiration rate (RR) for the entire plant (R^2 = 0.79) and (B) specific root respiration rate (SRR) expressed on a gram dry weight basis (R^2 = 0.22) are presented for each individual tree in the pot study. Data include values for all ozone treatments, aspen clones and sampling dates during the 12-week experiment.

Ozone significantly decreased root system respiration and root dry weight ($P < 0.033$) but the treatment effect did not differ among clones ($P > 0.66$) (Figures 3A and 3B) and there were no significant second- or third-order interactions in the three-way analysis of variance for root system respiration or root dry weight. There were no significant treatment effects attributable to either ozone ($P = 0.48$) or clone ($P = 0.15$) on specific root respiration (Figure 3C). There were two significant interactions (harvest × ozone, $P = 0.038$, and harvest × clone, $P = 0.019$) that were mainly the result of atypical trends in ozone and clonal responses at the Week 3 harvest, where measurements were the least precise.

Table 1. Regression statistics for the functional model of respiration. Specific root respiration (SRR, nmol g^{-1} s^{-1}) was analyzed as a linear function of relative growth rate (RGR, s^{-1})[1] of pot-grown aspen. The slope coefficient (g), multiplied by RGR, estimates the growth respiration component, and the intercept coefficient estimates the maintenance (m) respiration component; thus, SRR = gRGR + m (Hesketh et al. 1971, Amthor 1989, Sprugel et al. 1995). Data are from root systems of three aspen clones (216, 259 and 271). Ozone-treated plants were exposed to an ozone concentration of 150 ppb for 8 h each day.

Treatment	g (nmol g^{-1})	m (nmol g^{-1} s^{-1})	R^2	P
Control	1.42×10^7	15.3	0.37	< 0.001
Ozone	2.12×10^7	13.0	0.28	0.001
Mean	1.67×10^7	14.8	0.28	< 0.001

[1] Seconds are used for RGR units to correspond with respiration time units.

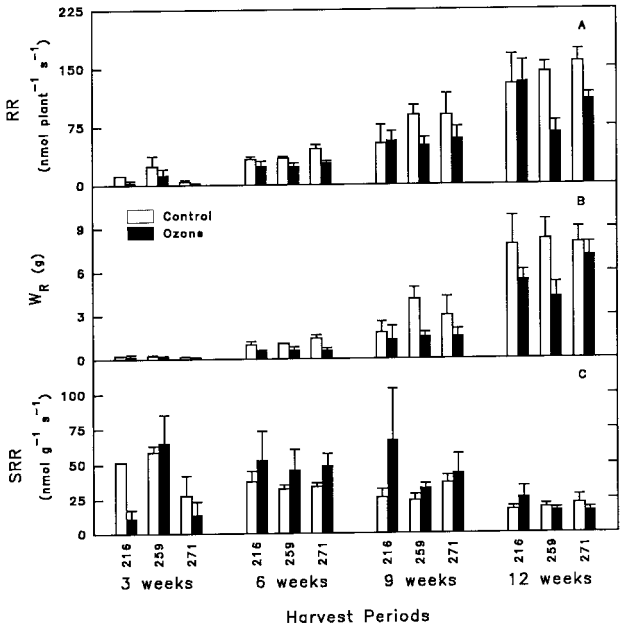

Figure 3. Clonal root respiration and dry weight responses to ozone. (A) Root respiration rate (RR) for the entire plant, (B) root system dry weight (W_R), and (C) specific root respiration rate (SRR) are presented for the three aspen clones (216 and 271, ozone tolerant; 259, ozone sensitive) in the pot study. The ozone-treated plants were exposed to an ozone concentration of 150 nl l^{-1} for 8 h each day. The mean and standard error are presented for one to two plants in each of two replicate chambers.

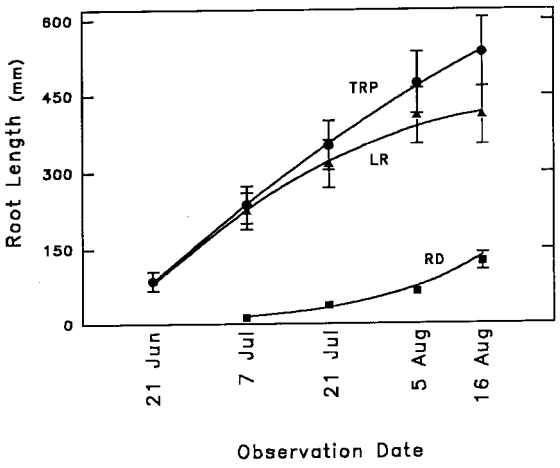

Figure 4. Time course of total root production (TRP), root disappearance (RD) and standing live root length (LR) per minirhizotron. Trees were grown in open-top chambers ventilated with charcoal-filtered air, charcoal-filtered air with simulated ambient ozone, or charcoal-filtered air with simulated twice-ambient ozone. Means and standard errors are presented for 18 minirhizotrons and include all ozone treatments combined.

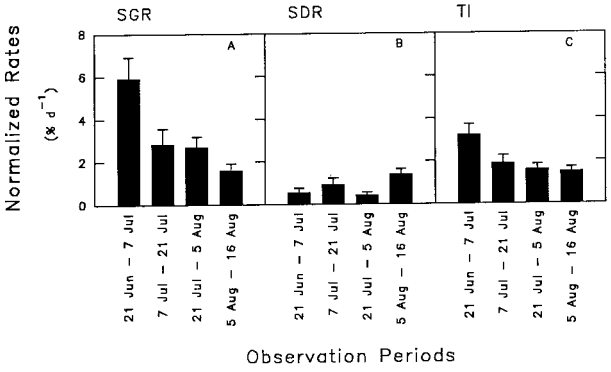

Figure 5. Normalized rates of aspen fine root dynamics measured at different times during the growing season. (A) Specific root growth rate (SGR), (B) specific disappearance rate (SDR) and (C) the turnover index (TI) calculated from minirhizotron observations (Cheng et al. 1991). Trees were grown in open-top chambers ventilated with charcoal-filtered air, charcoal-filtered air with simulated ambient ozone, or charcoal-filtered air with simulated twice-ambient ozone. Means and standard errors are presented for 18 minirhizotrons and include all ozone treatments combined.

Plantation experiment

There were distinct seasonal patterns in fine root dynamics of the field-grown trees (Figure 4). Total root production (TRP), root disappearance or death (RD), and the standing live root length (LR) per minirhizotron tube all increased during the growing season. Total root production was greatest for the period from June 21 to July 7 (9.5 mm day^{-1}) and steadily declined to 5.6 mm day^{-1} by the August 5–16 period. In contrast, RD was minimal during June (0.8 mm day^{-1}), but increased to 5.5 mm day^{-1} during August. Consequently, live root length equaled total root production in June, when root disappearance rate was low, but it decreased with respect to total root production during August as total root production slowed and root disappearance accelerated. The nonlinear shape of these parameters with time was confirmed by significant ($P < 0.008$) linear and quadratic contrasts in the repeated measures analysis.

Root production and disappearance rates normalized to a live root length basis also showed highly significant responses over time (Figure 5). During the observation period from June 21 to July 7, specific growth rate was 5.9% day^{-1}, whereas the specific disappearance rate was only 0.6% day^{-1}. By August 5–16, the specific growth rate of 1.5% day^{-1} was similar to the disappearance rate of 1.3% day^{-1}. Specific disappearance rate between July 21 and August 5 was less than during any other period, and this was associated with increased survival (see Table 2). Because these normalized parameters are rates of change for nonlinear functions, they were expected to be linear with respect to time. Linearity was confirmed by polynomial contrasts analysis. Thus, the linear contrast was significant ($P < 0.029$), whereas the quadratic contrast was not significant ($P > 0.192$).

The turnover index decreased over time (Figure 5C) as a result of the predominate effect of declining specific growth rate. We calculated fine root population replacement time by taking the inverse of turnover index (Odum 1975). During June, the fine root population replacement time was approximately 31 days, whereas in August it was 71 days.

Few dead roots were observed; most roots simply disappeared from the minirhizotron tube surface. Therefore, we refer to this category as disappearing roots. Disappearance of roots was expressed as a survival percentage of the initial root length for each cohort (Table 2). In most cohorts, the greatest change in survival occurred during the first 2-week observation period (a change of 16% of the initial root length). During subsequent observation periods, the change in survival did not exceed 7% for most cohorts. For example, survival in the July 7 cohort was 80% for the observation period from July 7 to 21, a decrease of 20%. After 2 more weeks, survival only changed by 7%. The exception was in the July 21 cohort where survival dropped by 9% after 2 weeks and then by 21% in the following 2 weeks. Because of the initial high survival of the July 21 cohort, survival was highest and specific disappearance rate was lowest for the July 21 to August 5 observation period. The average change in survival percentages for all cohorts between July 21 and August 5 was only 6%, compared to more than 12% for the other periods.

Ozone had no statistically significant effects on any of the fine root parameters examined because of the high variability in live root length (coefficient of variation ranging from 50 to 100%) observed in the minirhizotrons. However, standing live root length in the 1× ozone treatment was consistently greater, and live root length in the 2× ozone treatment was lower than in other treatments except on July 21 (Figure 6).

Soil CO_2 efflux was affected by both season and ozone treatment (Figure 7). Efflux measured on July 12 was more than twice that measured at any other time during the rest of the season. On each measurement date, ozone treatment decreased soil CO_2 efflux by more than 41%. Although there was a highly significant effect associated with measurement date ($P < 0.001$ for both univariate and multivariate test statistics), there was also a significant time by treatment interaction ($P < 0.02$ for both univariate and multivariate test statistics) because the effects of the ozone treatment varied during the growing season. Univariate F-tests showed highly significant ozone treatment effects ($P < 0.006$) on July 20 and August 16, but the treatment effect was less significant ($P = 0.051$) on July 12, and ozone treatment effects were nonsignificant ($P = 0.15$) on August 31.

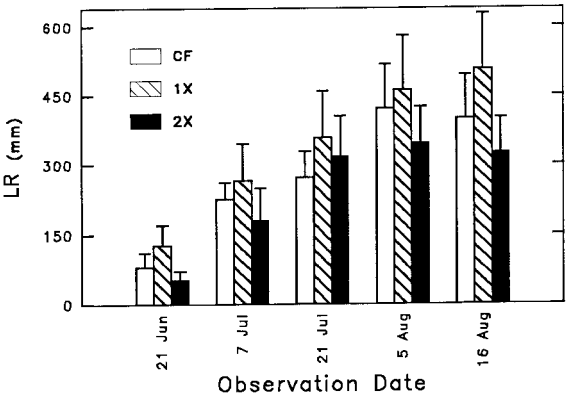

Figure 6. Standing live root length (LR) observed in minirhizotron tubes at different times during the growing season. Trees were grown in open-top chambers ventilated with charcoal-filtered air (CF), charcoal-filtered air with simulated ambient ozone (1×), or charcoal-filtered air with simulated twice-ambient ozone (2×). Means and standard errors are presented for two minirhizotrons per chamber and three chambers per treatment.

Discussion

Pot experiment

The specific root respiration rates reported here are greater than those found for other woody plants. Respiration rates range from 0.2 to 34 nmol g^{-1} s^{-1} for coniferous species (Sprugel et al. 1995) and from 1.2 to 24 nmol g^{-1} s^{-1} for several

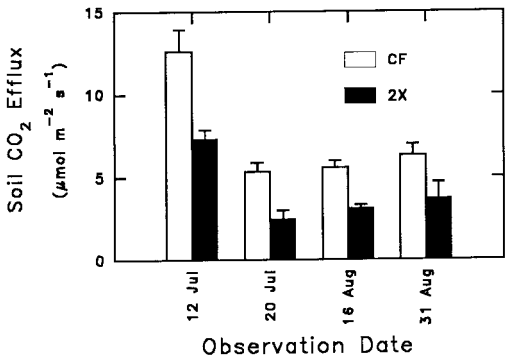

Figure 7. Soil CO_2 efflux measured at different times during the growing season. Data from aspen trees in the plantation study are presented for two ozone treatments: charcoal-filtered air (controls, CF) and charcoal-filtered air with simulated twice-ambient ozone (2×). Means and standard errors are presented for two measurement locations per chamber and three chambers per treatment.

Table 2. Survival (%) of four aspen root cohorts during the growing season. Each cohort is distinguished by the observation date when it first appeared. The root length visible at subsequent observations is expressed as a percent of the initial length. Means ± standard errors are presented for 18 minirhizotrons and include all ozone treatments combined in the plantation study.

Cohort	June 21	July 7	July 21	August 5	August 16
June 21	100	84 ± 4	79 ± 6	77 ± 5	71 ± 6
July 7		100	80 ± 5	73 ± 5	64 ± 5
July 21			100	91 ± 2	70 ± 6
August 5				100	80 ± 4

woody plant species (Ledig et al. 1976). Reported rates for *P. tremuloides* range between 4.4 nmol g^{-1} s^{-1} (Lawrence and Oechel 1983) and 8 nmol g^{-1} s^{-1} (Ledig et al. 1976), whereas our rates ranged from 5 to 92 nmol g^{-1} s^{-1} with an average of 31 nmol g^{-1} s^{-1} (Figure 2B), and average maintenance respiration rates were greater than 13 nmol g^{-1} s^{-1} (Table 1). These rates are similar to those of crop species where respiration rates range from less than 30 to more than 130 nmol g^{-1} s^{-1} (Hansen 1977, Bloom et al. 1992, Cramer and Lewis 1993). We found that the highest specific root respiration values were associated with high root relative growth rates (> 15% day^{-1}), confirming the findings of Poorter et al. (1991) that high specific root respiration is related to high inherent growth rates. The maintenance respiration rates and the total specific respiration rates for plants growing at lower relative growth rates (< 5% day^{-1}) are typical of the rates reported for woody plants. It is probable, therefore, that the high specific respiration rates of the aspen plants are the result of rapid growth rates.

Root system respiration increased during the growth period as root dry weight increased, but the increase was not linear (Figure 2A). Respiration typically declines in roots as diameter increases (Sprugel et al. 1995). In our plants, the fraction of roots less than 1 mm in diameter decreased from 90% after 3 weeks of growth to 50% after 12 weeks, indicating a functional shift from metabolically active absorbing roots to less active woody conductive tissue. A metabolic shift was also indicated by the drop in specific root respiration rate with increasing root dry weight (Figure 2B). Specific root respiration rates were correlated with relative growth rates by the functional model of respiration (Table 1) (Hesketh et al. 1971, Amthor 1989, Sprugel et al. 1995), and root weight and root relative growth rate were inversely proportional (data not shown). Therefore, we conclude that the nonlinear decline in root system respiration can be attributed to the combined effects of a proportional increase in large diameter roots and a decrease in growth respiration as a result of a declining relative growth rate.

Ozone decreases specific root respiration in both agronomic and woody plant species (Hofstra et al. 1981, Ito et al. 1985, Edwards 1991), but the effect is not always associated with decreased root dry weight (Edwards 1991). Because root respiration and root dry weight (Figures 3A and 3B) showed proportionally similar declines in response to ozone, there were no statistically significant ozone effects on specific root respiration. We conclude, therefore, that the decline in root system respiration in response to ozone was largely due to an ozone-induced decrease in root growth rather than to a change in metabolic functioning per unit root weight.

Plantation experiment

Seasonal patterns of fine root growth indicated that total root production peaked early in the season and root disappearance peaked late in the season; as a result, standing live root length peaked by early August (Figure 4). This pattern agrees with other studies on fine root dynamics in temperate deciduous forest species (Hendrick and Pregitzer 1992*a*, 1993, Burke and Raynal 1994, Fahey and Hughes 1994), and the perennial vine *Actinidia deliciosa* (A. Chev.) C.F. Liang et A.R. Ferguson (Reid et al. 1993).

Substantial fine root turnover occurred throughout the growing season (Figure 5, Table 2). Replacement times of between 30 and 70 days suggest that the fine root population was completely replaced more than once during the growing season. Such high turnover rates have been commonly observed (cf. Lyr and Hoffmann 1967, Kolesnikov 1971, Head 1973, Vogt and Bloomfield 1991, Reid et al. 1993). The life span of nearly 20% of the roots observed was less than 2 weeks (Table 2), which represents a high cost to the plant. Possible explanations for this high turnover include herbivory, and rapid death and decomposition (Atkinson 1992, Hendrick and Pregitzer 1992*a*). Our observations support both explanations. Many arthropods, which may be involved in root herbivory, were observed on the surface of the minirhizotrons. Because most roots just disappeared and few were observed to be in a state of death or decay, herbivory is a reasonable explanation. On the other hand, daily observations of aspen roots growing in pots indicated that the finest roots (< 0.1 mm) tend to fade and disappear within just a few days even though no insects were observed. The lack of conclusive evidence about the cause of early fine root mortality and the importance of such rapid turnover to the overall carbon budget warrant further investigation.

There were no statistically significant effects of ozone on fine root length in the plantation study (Figure 6) although root length was usually less in the 2× ozone treatment than in the CF and 1× ozone treatments. The lack of a root response to ozone is contrary to the results observed in our pot study (Figure 3) and other pot experiments (Tingey 1974, Cooley and Manning 1987, Mooney and Winner 1988). In contrast, aboveground dry weight decreased more than 30% in the 2× treatment compared with the CF treatment during the first 2 years of treatment (Karnosky, unpublished data).

Soil CO_2 efflux decreased during the growing season and with ozone treatment (Figure 7). Although CO_2 efflux rates measured after July 12 were more typical of those reported for temperate forest ecosystems (Garrett and Cox 1973, Edwards 1975, Singh and Gupta 1977, Ewel et al. 1987, Hanson et al. 1993), all rates, including the July 12 value, were within previously reported ranges (Singh and Gupta 1977). The similarity in seasonal patterns of soil CO_2 efflux (Figure 7) and fine root specific growth rate (Figure 5A) suggests that these processes are positively correlated as described for the pot studies (Table 1). This approach indicates that high rates of soil CO_2 efflux early in the season may result from high rates of growth respiration in association with high specific growth rates. Ozone typically decreases soil CO_2 efflux of potted plant material (Figure 3; Ito et al. 1985, Edwards 1991). Unfortunately, there are no published soil respiration data on older trees exposed to ozone with which to compare our results. Because measurement of CO_2 efflux provides an estimate of soil respiration that includes respiration from roots and other soil organisms, and the main source of carbon to the belowground system is through the autotrophic plant, any aboveground factors affecting carbon transport to the roots will

proportionally alter the soil CO_2 efflux. Therefore, we conclude that ozone had a significant negative effect on the amount of carbon allocated to the belowground system of aspen.

Comparison of cultural environments

Based on the ozone response data, we compared the pot study and the plantation study with respect to three belowground processes: (1) root growth, (2) root system respiration, and (3) specific root respiration. In the pot study, ozone caused root growth (dry weight) of Clones 216 and 259 to decline an average of 39% at 12 weeks (Figure 3B). In August, for the same clones, root growth in the plantation study (standing live root length) was 17% less in the 2× ozone treatment than in the control treatment (Figure 6), although this effect was not statistically significant. Thus, ozone decreased root dry weight in young potted plants more than it decreased root length in older field-grown trees.

The effects of ozone on root system respiration in the pot study (Figure 3A) and soil CO_2 efflux in the plantation study (Figure 7) were similar. The decline in respiration rate in response to ozone averaged 26% (Clones 216 and 259 at 12 weeks) in the pot study and more than 41% in the plantation. Although the ozone-induced decreases in respiration rates were not equal between cultural environments, the response of young pot-grown plants was similar to that of older field-grown plants.

Comparing specific root respiration of the plants in the pot and plantation studies is difficult. In the plantation, it was not possible to normalize the soil CO_2 efflux by root length or root weight because of the lack of data on heterotrophic soil respiration and the relationship of surface flux with the root density depth profile. In the pot studies, the decrease in respiration in response to ozone was attributed to differences in root dry weight because there were no detectable differences in specific root respiration between treatments (Figure 3). It was not possible to draw similar conclusions about the ozone-induced decrease in soil CO_2 efflux in the plantation study, because root dry weights were not available.

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