Nutrient Uptake and Growth Responses of Virginia Pine to Elevated Atmospheric Carbon Dioxide[1]

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ABSTRACT

One-year-old Virginia pine (*Pinus virginiana* Mill.) seedlings with native or *Pisolithus tinctorius* mycorrhizal associations were grown in pots with soil low in organic matter and in cation exchange capacity and were exposed to one of five atmospheric CO_2 levels in the range of 340 to 940 $\mu L/L$ in open-top field chambers. The mean dry weight of the seedlings increased from 4.4 to 11.0 g/plant during the 122-d exposure period. Significant increases in dry weight and uptake of N, Ca, Al, Fe, Zn, and Sr occurred with CO_2 enrichment. Greater chemical uptake was associated with greater root weight. Specific absorption rates for chemicals (uptake per gram of root per day) were generally not affected by CO_2 enrichment. The uptake of P and K was not increased with elevated CO_2, and these elements showed the greater nutrient-use efficiency (C gain per element uptake). The nutrient-use efficiency for N and Ca was not influenced by atmospheric CO_2 enrichment. Large increases in Zn uptake at high CO_2 suggested an increase in rhizosphere acidification, which may have resulted from the release of protons from the roots, since it was estimated that cation uptake increasingly exceeded anion uptake with CO_2 enrichment. Potassium, P, and NO_3^- concentrations in the pot leachate decreased with higher CO_2 levels, and a similar trend was found for Al and Mg. These results suggest that soil-plant systems may exhibit increased nutrient and chemical retention at elevated atmospheric CO_2.

Additional Index Words: nutrient-use efficiency, root weight duration, chemical leaching, rhizosphere acidification, *Pinus virginiana* Mill.

Increases in atmospheric CO_2 have been occurring over the last seven or eight decades and are projected to continue for several more decades (Council on Environmental Quality, 1981). The responses of temperate forest species under natural conditions to rising levels of atmospheric CO_2 are largely unknown. Temperate forests are a significant component of the global C cycle, since they occupy large land areas and maintain large C storage capacities (Olson et al., 1983). A small increase in forest C storage with elevated CO_2 could reduce the rate of increase in atmospheric CO_2. All temperate forest species exhibit the C_3 form of photosynthesis, and it is well

established that photosynthesis of C₃ plants is enhanced at CO_2 levels up to 1000 $\mu L/L$. This enhancement is usually accompanied by an increase in water-use efficiency (Pearcy and Bjorkman, 1983). Increased photosynthesis may not necessarily translate into increased growth; water and nutrients are also needed in sufficient quantities for a growth response to an increased photosynthate supply (Kramer, 1981).

Temperate forest growth is often limited by the N or P supply (Tamm, 1979; Lavender and Walker, 1979; Cole and Rapp, 1980); thus, the question arises as to whether forests can extract more N and P from soil pools at elevated atmospheric CO_2. Luxmoore (1981) hypothesized that stimulated root, rhizosphere, and mycorrhizal activity at elevated CO_2 could result in increased nutrient uptake by plants as well as increased nutrient retention in soil-plant systems; i.e., the nutrient leaching rate would be reduced. Any such buildup in the nutrient status of a forest community could lead to long-term increases in forest growth, although there may be a lag in response if preferential nutrient accumulation in soil microorganisms occurs. In the longer term, autotrophs may obtain additional nutrients through the mineralization of decaying microbial tissues.

A high proportion of the N and P in temperate forest ecosystems is in the soil (Table 1), and this is a potential source of nutrients needed for a growth response to higher atmospheric CO_2 levels if mechanisms of enhanced nutrient extraction exist. If increased atmospheric CO_2 resulted in a 0.36% increase in net primary production per year for 40 yr, as proposed by G.H. Kohlmaier, C.D. Keeling, and R.R. Revelle of San Diego State Univ., CA (1985, personal communication), only a small shift of N (2%) and P (1%) from the soil to vegetation would be needed during the 40-yr period to maintain current C/N/P ratios in vegetation. Sufficient supplies of N and P exist in many temperate forest ecosystems, and only a small increase in nutrient uptake could sustain a growth response to elevated atmospheric CO_2. It should be noted that such an increase is small in terms of total N or P but much larger in terms of available nutrient supply.

This study was undertaken to evaluate the nutrient-uptake and growth responses of Virginia pine (*Pinus virginiana* Mill.) to elevated atmospheric CO_2. Pot culture methods and open-top exposure chambers were used, and the soil was taken from an old field that had not received fertilizer in the last decade. Since mycorrhizae are known to have important effects on the nutrient status of plants growing on nutrient-poor soils, the study included a comparison of mycorrhizal treatments.

METHODS

Six kilograms of air-dried sandy loam topsoil (15 g/kg organic matter, pH = 5.9, cation exchange capacity = 3.5 cmol (+)/kg) from an old field was packed in three stages to a bulk density of 1.3 Mg/m³ into 25-cm diam pots. The soil was a Cecil sandy loam, classified as a Typic Hapludult (clayey, kaolinitic, thermic). A single bare-rooted Virginia pine seedling, obtained from the Indiana State Nursery, was planted in each pot. The seedlings were 1-yr-old; one-half of them had been specifically inoculated with the mycosymbiont *Pisolithus tinctorius,* and the other half formed mycorrhizal associations with undetermined native fungi. The seedlings in the initial planting of 156 pots were grown in the open for 80 d during the spring (8 Apr.–28 June 1982). During this period, active shoot

Table 1. Nutrient distribution (mean and [standard deviation]) in some temperate forests. Data from Kimmins et al. (1979) and Swank and Waide (1980).

Forest component	N, 13 sites	P, 7 sites
A = Shoots and roots (kg ha⁻¹)	499 (219)	54 (28)
B = Litter (kg ha⁻¹)	546 (343)	33 (17)
C = Soil (kg ha⁻¹)	3776 (1364)	943 (1503)
D = Vegetation/total (A/[A + B + C])(%)	10 (3)	21 (19)

elongation growth occurred. Seventy-two pots from the group were selected for exposure to six different atmospheric CO_2 treatments. These pots contained uniform seedlings in three size classes (small, medium, large) from each of the two mycosymbiont types.

Four of the six atmospheric CO_2 treatments (75, 150, 300, and 600 $\mu L/L$ above ambient) were controlled with a CO_2 monitoring and injection system in open-top field chambers (Rogers et al., 1983a). The chambers were cylindrical (4.3-m wide by 3-m high), and air flow was directed through a double-walled lower section of the chamber with holes on the inside, which allowed air passage up through the chamber. The other two CO_2 treatments (fifth and sixth) were at ambient levels, one with and one without an open-top chamber. Six pots (three size classes and two mycorrhizal types) were mounted in a support rack in each of the six CO_2 treatments, which were replicated twice for a total of 72 pots. The positions of the treatments in the support rack were fixed for plant size, but random for the mycorrhizal treatment resulting in a split-split plot design for the experiment. Variability due to position in the support rack was deliberately confounded with the variance due to plant size.

The ambient atmospheric CO_2 concentration averaged 340 $\mu L/L$ during the daylight hours of the experimental period. Thus, the CO_2 treatments ranged from 340 to 940 $\mu L/L$ during the hours of photosynthesis. The ambient concentration increased at night, and all the CO_2-enrichment treatments were superimposed on the natural variation of the ambient air concentration.

At the initiation of CO_2 exposures in late June 1982, the dry weight, height, and stem diameter of the plants in eight pots were measured, along with elemental content (N, P, K, Ca, Mg, B, Al, Fe, Mn, Cu, Zn, Sr, Ba) of leaf, stem, and root tissues. The elemental content of soil leachate (P, K, Ca, Mg, B, Fe, Al, Mn, Cu, Zn, NO₃, NH₄) and solution pH were determined from the same eight pots. Leachate was obtained by adding additional water to each pot after watering the soil to field capacity (0.22 m³/m³). Soil elemental content (P, K, Ca, Mg, NO₃, NH₄, soluble salts), organic matter, and pH of soil samples from six unleached pots were measured. Chemical analyses were performed by the Soil Testing and Plant Analysis Laboratory at the Univ. of Georgia. The element concentrations in plant tissues were obtained from direct-reading emission spectroscopy except for total N, which was measured with a colorimetric autoanalyzer system calibrated against known standards (Isaac and Johnson, 1976). Soil leachate elements were analyzed by inductively coupled plasma emission spectrophotometry. Ammonium and NO₃-N in the soil leachate and extractable in KCl from the soil were determined by a calibrated titration method. Soil organic matter, pH, and soluble salts were measured, using standard methods of soil analysis (Black, 1965). The same plant and chemical measurements were made on leachate, plant tissues, and soil from all pots at the final harvest. (The words *chemical, element,* and *nutrient* are sometimes used interchangeably as appropriate in the context of this paper.)

The CO_2 exposures were maintained for 16 weeks (28 June–29 Oct. 1982), and the pots were watered periodically (approximately twice per week) to bring soil water close to a volumetric water content of 0.22 m³/m³ (field capacity). Prior to harvest, the 72 pots were watered to field capacity and leached with additional water. The leachate was subsampled for chemical analysis.

Data Analysis

All data anaysis was conducted with the Statistical Analysis System (SAS, 1979) using the general linear model (GLM) procedure. The procedure was suitable for conducting analysis of
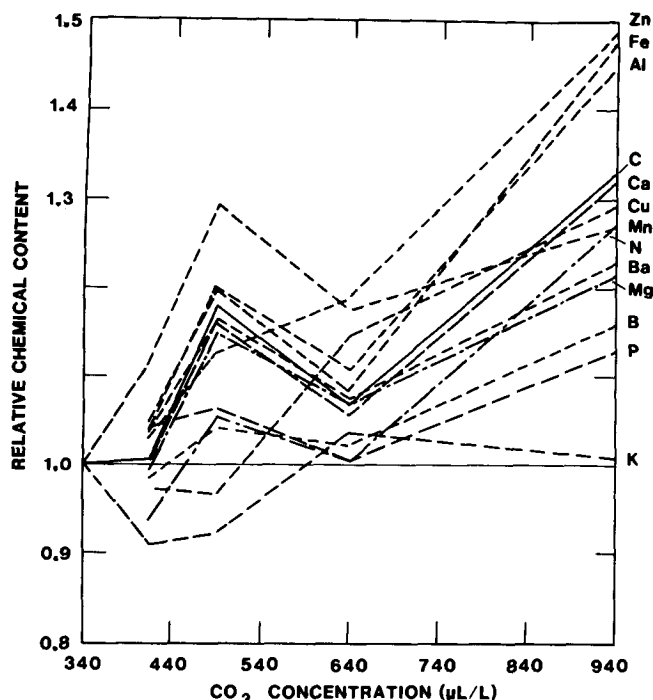
Fig. 1. Relative content of 13 plant elements as a function of atmospheric CO_2 concentration. The significance level at which the least squares linear slope differs from the zero slope is given in Table 4. (Data for Al and Fe are approximate; see text. Data for Sr are not presented since they are very close to that for C.)

however, this was not a rigorous test, since there were only two replicates in the experiment. There were significant effects due to plant size: the large plant-size class showed greater growth and nutrient uptake than the small size class. This result was expected. The main purpose of separating the plants into size classes was to increase the significance of mycorrhizal and CO_2 treatment comparisons by separation of variance due to size class.

RESULTS

The primary data obtained from the initial and final harvests have been documented in a published report (CDIC, 1985) and are available as a computer file from the Carbon Dioxide Information Center at Oak Ridge National Laboratory, Oak Ridge, TN. The major findings from these data are summarized in this section.

Plant Growth

The mean (average for all treatments) total dry weight of the pine seedlings increased from 4.4–11.0 g/plant during the 16-week experimental period. This was due to a greater than fivefold increase in root weight and smaller relative increases in needle (19%) and stem (64%) dry weight. The major increase in elongation growth occurred during the spring prior to the CO_2-enrichment exposures. This is characteristic of determinant growth of *Pinus* species where current year elongation growth is determined by the previous year's storage reserves. During the experimental period, the main stem showed small increases in height, diameter, and the number of growing points (candles). Diameter growth continues through the growing season and would be expected to be higher under more favorable growing conditions. A large increase in the root/shoot ratio was obtained for all treatments as would be expected for plants transferred from fertile to nutrient-poor soil conditions (Agren et al., 1980). The larger root systems were visually longer and had a greater number of roots than the smaller systems, indicating that increases in root size were associated with increases in root-soil interface area and to a lesser extent in root radial growth. In the mycorrhizal treatments, some statistically significant effects (ANOVA data analysis) on plant growth were seen. For example, the *P. tinctorius* mycorrhizal plants were taller and had lower needle and stem weights than the plants with native mycorrhizal associations (Table 2). The *P. tinctorius* symbiont generally had inhibiting effects on plant dry weight under the pot culture methods used. No significant

variance on unbalanced data sets (using type III sum of squares) as well as for linear regression analysis. Statistical analysis by the analysis of variance method (ANOVA) provided insensitive comparisons of main treatment effects due to the low replication (two times) in the experiment. In particular, there was no significant plant growth response to CO_2 enrichment according to this method of analysis. Significant effects due to mycorrhizal treatments were found with ANOVA data analysis and these results are presented. Visual inspection of growth and nutrient data suggested that there were approximately linear responses to elevated CO_2. The five CO_2 treatments given in the open-top chambers provided a suitable range for evaluating linear responses for each of the six plant size-mycorrhizal combinations by regression analysis. The data were further analyzed, using the following regression relationship:

$$Y = A + BX$$

where Y = dependent variable (plant growth or nutrient data), A = intercept (ambient response), B = slope (enrichment response per $\mu L/L$ CO_2), and X = CO_2 concentration above ambient ($\mu L/L$).

In this procedure, the slope values (B) were tested to identify cases significantly different from zero slope indicating a response to CO_2 enrichment. Treatment effects due to mycorrhizae and plant size were identified by statistically significant differences in intercept values (ambient responses) of linear regressions with CO_2. These were determined for the six plant size-mycorrhizal combinations. Any such treatment effects were maintained over the range of CO_2 treatments since the slope values (B) for the response of the six treatment combinations are identical in this method of regression analysis. It is also noted that the testing for a linear trend is a more powerful test than the ANOVA, thus significant linear relationships found by regression are not contradictory to nonsignificant effects in ANOVA procedures.

Three of the *P. tinctorius* mycorrhizal plants died during the experimental period resulting in unequal class sizes in the ANOVA data analysis. There was no statistically significant effect (ANOVA) of the open-top chamber on growth or chemical content of the plants at ambient CO_2 relative to plants grown without a chamber;

Table 3. Intercept and slope values for linear regressions between the content of each of 14 plant elements and atmospheric CO_2 concentration as the independent variables.

Element	Intercept element/ plant	Slope Element/plant/ (μL/L CO_2)	R^2	Probability (%) of slope = 0
Carbon, g/plant†	4.32	0.00284	0.45	4.3
Macro, mg/plant				
N	61	0.04191	0.44	2.6
P	13	0.00383	0.48	>5
K	30	0.00678	0.43	>5
Ca	35	0.02368	0.48	3.5
Mg	6	0.00307	0.50	>5
Minor, μg/plant				
B	130	0.05336	0.55	>5
Al	5809	5.0447	0.39	1.6
Fe	5082	4.6754	0.38	1.5
Mn	1521	0.61134	0.39	>5
Cu	54	0.04277	0.37	>5
Zn	384	0.40075	0.45	1.4
Sr	97	0.06589	0.50	1.6
Ba	306	0.13837	0.43	>5

† 0.45 of total dry weight. Data for Al and Fe are approximate.

Table 4. Chemical-use efficiencies for five macro and eight minor elements, calculated from intercept data (ambient response) and slope data (enrichment response), along with the ratio of the enrichment to ambient responses

Element	Chemical-use efficiency		Enrichment response ratio
	Ambient response	Enrichment response	
	340 μL/L	340–940 μL/L	
Macro			
N	71	68	0.96
P	332	741	2.23
K	144	418	2.91
Ca	123	120	0.98
Mg	720	924	1.28
Minor			
B	33 200	53 200	1.60
Al†	744	562	0.76
Fe†	850	607	0.71
Mn	2 840	4 640	1.63
Cu	80 000	66 300	0.83
Zn	11 300	7 080	0.63
Sr	44 500	43 100	0.97
Ba	14 100	20 500	1.45

† Data for Al and Fe are approximate.

mycorrhizal × CO_2 interactions were shown in the ANOVA procedure.

Plant Elemental Content

The total nutrient content of the plants at the final harvest was calculated from the dry weight and chemical concentration data for needles, stems, and roots. There were some significant differences in P, K, Mn, Zn, and Ba content in the plants grown under the two different mycorrhizal treatments (Table 2). In each case, the native mycorrhizal plants had greater nutrient content than the *P. tinctorius* plants.

The C content of each plant at the final harvest was assumed to be 0.45 of dry weight for the purposes of data analysis. A clear trend was apparent in many of the data, as can be seen in Fig. 1, which shows the relative chemical content as a function of CO_2 concentration in the open-top chambers.

Many data for the chemicals were shown to have linear relationships with CO_2 enrichment, with the slope values (*B*) being significantly greater than zero (shown by the probability values) even though the R^2 values for the regression relationships (from the GLM procedure) were not high (Table 3). The intercept vlaues (*A*) (Table 3) represent the plant-chemical content at ambient CO_2. Six elements (P, K, Mg, B, Mn, Cu) were not present in significantly greater amounts with CO_2 enrichment, and several others (N, Ca, Al, Fe, Zn, Sr) were present in larger quantities with elevated CO_2. The results for Al and Fe are approximate. The concentration of these elements in roots exceeded the upper limit of the instrument's measurement range, so the upper-limit concentration was used as an index. The pattern of these results is compared with data for the other elements and evaluated for consistency of results, as discussed in a later section.

The ratio of C content per unit of chemical content (chemical-use efficiency) was calculated from the intercept and slope values given in Table 3. The chemical-

use efficiencies for ambient conditions (based on *A* values) were sometimes less than and sometimes greater than the efficiencies calculated for the range of CO_2-enrichment treatments (*B* values) (Table 4). The ratio of the enrichment response to the ambient response (called "enrichment response ratio" in Table 4) was <1 for Al, Fe, Cu, and Zn, >1 for P, K, Mg, B, Mn, and Ba, and essentially unchanged for N, Ca, and Sr. Potassium showed the greatest increase in enrichment response ratio (2.91) followed by P (2.23), indicating that these two nutrients may have been the most limiting for growth in the nutrient-deficient soil. The chemical-use efficiencies for N and Ca were essentially the same over the CO_2 treatment range as that for ambient conditions.

The reduced chemical-use efficiencies for Al, Fe, Cu, and Zn indicate a greater content of these elements with elevated CO_2 relative to the increase in C with elevated CO_2. The Cu response to higher CO_2 levels was not significant (Table 3); however, C (plant dry weight) had a significant response to CO_2 enrichment, which suggests that the chemical-use efficiency for Cu should be >1. There was high variability in the Cu data (lowest R^2 value in Table 3). The responses for Al and Fe are consistent with the response of Zn and may be related to increased solubility at lower pH (Lindsay, 1979). This supposition leads to the idea that rhizosphere acidification may have resulted from the elevated CO_2 treatments. Rhizosphere acidification may have been associated with a greater uptake of cations than anions (Jarvis and Robson, 1983), and this was elevated from the cation-anion charge balance on a per plant basis. The calculation of the cation-anion balance was made by determining the quantity of positive charges (Ca^{2+}, Mg^{2+}, K^+, and NH_4^+) and of negative charges ($H_2PO_4^-$ and NO_3^-) taken up per plant for each CO_2 treatment. It was assumed in this analysis that the uptake of NH_4^+ and NO_3^- by the plants was in the same proportion as that measured in the leachate waters. The mean NH_4^+-N concentration was (0.7 ± 0.1 mg/L), whereas the NO_3-N decreased significantly with CO_2

Table 5. Mean values of plant cation and anion uptake, and cation excess calculated for five atmospheric CO₂ treatments.

CO₂	Cation uptake	Anion uptake	Cation excess
μL/L	mmol(+)/plant	mmol(−)/plant	mmol(+)/plant
340	4.84	3.84	1.00
415	4.76	3.55	1.21
490	5.60	3.71	1.89
640	5.39	3.49	1.90
940	6.73	4.09	2.64

enrichment (discussed in the section on leachate response) and mean NO_3^- values for each CO_2 treatment were used. The uptake of SO_4^{2-} and Cl^- were not included, since such data were not obtained; however, other studies (Jarvis and Robson, 1983) suggest that these anions are in low concentration and cannot account for the cation excess. It can be seen that cation uptake is larger than anion uptake and that cation excess increased with elevated CO_2 (Table 5). These calculations lend support to the hypothesis that rhizosphere acidification was a contributor to the enhanced uptake of Zn with CO_2 enrichment. The calculations lead to the same inference if it is assumed that NH_4^+ and NO_3^- are taken up in equal proportions.

Root Weight Duration

The previous section described results for chemical content of the plants at the final harvest and these data included the initial chemical content of the plants prior to the CO_2 exposure period. Chemical uptake by the plants during the CO_2 exposure period was obtained by taking the difference between chemical contents of the plants at the initial and final harvests.

The effect of CO_2 enrichment on chemical uptake by plants may be due to greater root exploration associated with the increase in roots and/or to greater chemical uptake per unit of root. In order to evaluate these two possibilities, the total chemical uptake per plant during CO_2 enrichment was partitioned into two components, using growth analysis, as follows:

Chemical   root weight   specific
uptake   =   duration   ×   absorption rate
(mg)   (g root × d)   [mg/(g root × d)]

In this approach, root weight duration (RWD) is a measure of root exploration given by the integration of root weight with time. Root weight is viewed as an approximate index of root surface area, since greater root weight is usually associated with greater surface area. The RWD was calculated as follows:

$$RWD = \frac{(R_2 - R_1)(T_2 - T_1)}{\ln R_2 - \ln R_1},$$

where R_2 is the root weight at the final harvest T_2, and R_1 is the initial root weight at the initial harvest T_1 (Hunt, 1978).

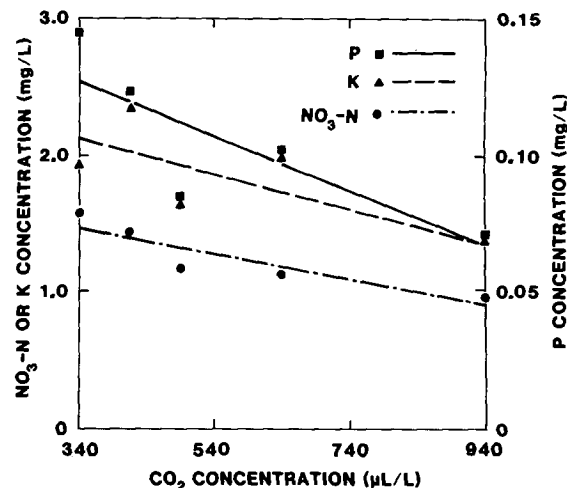The mean RWD was 320 g × d at ambient CO_2 and was correlated with the CO_2 treatments, showing an in-

Fig. 2. Mean nutrient concentrations in leachate from soil for the five atmospheric CO₂ treatments, and the least squares regression lines with CO₂ concentration as the independent variable.

crease of 0.1824 g × d per μL/L CO_2 enrichment. There was a 99.2% probability that this slope value was greater than zero slope. There were no significant effects of mycorrhizal treatment on RWD; however, the largest plant size had a significantly larger RWD than the other smaller plant sizes.

Specific Absorption Rate

The mean rate of chemical uptake by roots was estimated, using the ratio of the chemical uptake during the CO_2 exposure period (122 d) to the RWD. Chemical uptake was determined from the difference in plant chemical content at the final and initial harvests. The initial harvest data were taken as the mean of values obtained for the eight plants sampled at the commencement of the CO_2 treatments. Insufficient samples were taken to obtain suitable mean data for the small, medium, and large plant sizes at the initial harvest. Thus, a small bias was introduced into plant size effects on specific absorption rate and these effects were not considered. There were no significant effects of CO_2 enrichment on the specific absorption rate of any of the chemicals, although the trends for Zn, Fe, and Al were much stronger than for the other elements. A significant increase in specific absorption rate for Zn, Fe, Al, and Cu with CO_2 enrichment would confirm the inferences of rhizosphere acidification discussed earlier.

There were significant mycorrhizal treatment effects on the specific absorption rates for P, Mn, and Zn. In each case, the native mycorrhizal treatment vlaues were significantly greater. This examination of the chemical uptake data suggests that greater root weight has a much more significant effect than specific absorption rate on chemical uptake from nutrient-poor soil under CO_2-enrichment conditions.

Leachate and Soil Chemical Responses

A statistically significant decrease in P, K, and NO_3^--N concentration with increase in atmospheric CO_2 was shown for soil leachate (Fig. 2). The R^2 values for the regression lines were low, being 0.16 for P, 0.22 for

NO_3-N, and 0.29 for K. High variability in the chemical concentration of leachates from soil was found and in spite of this, a statistically significant reduction inP, K, and NO_3-N leaching with CO_2 enrichment was demonstrated. The slope of each regression line (Fig. 2) was statistically different from zero slope. It was shown earlier that nutrient-use efficiency was large for P and K, and that these may have been the most limiting nutrients in this study. This observation of reduced nutrient leaching with CO_2 enrichment supports one of the Luxmoore (1981) hypotheses of soil-plant system responses to elevated atmospheric CO_2. Similar trends were observed for Mg and Al (not statistically significant). There were no significant relationships between soil pH or soil-chemical content and the CO_2 treatments. Soluble salts were significantly higher for the native (58 g/L) than for the *Pisolithus* (52 g/L) mycorrhizal treatment. There were no significant plant size effects on the measured soil chemical characteristics.

DISCUSSION

A significant amount of growth occurred during the experiment. The average plant dry weight more than doubled during the 122-d exposure period for all CO_2 treatments. A mean growth response to CO_2 enrichment of 2.9 mg C/(μL L) was estimated from the linear regression analysis. This is a significant finding, since the plants were growing in soil with very low organic matter and very low cation exchange capacity. Growth responses to CO_2 enrichment can be expected for plants supplied with adequate water and nutrients. Rogers et al. (1983b) showed increased growth of sweetgum (*Liquidamber styraciflua*) and loblolly pine (*Pinus taeda*) with elevated CO_2 in an open-top chamber experiment. These plants were grown in pots with soil amended with nutrients and watered frequently. Canham and McCavish (1981) investigated seedling growth responses of three other coniferous species in CO_2-enriched glasshouses and observed increases in shoot dry weight of Sitka spruce (*Picea sitchensis*) after 20 weeks of exposure to 1320 μL/L CO_2. A growth response in lodgepole pine (*P. contorta*) was obtained after 16 weeks at the same exposure. They also showed increased shoot growth of lodgepole pine with an increase in nutrient supply at the high CO_2 treatments (1320, 2620 μL/L) but not at ambient concentration. In their experiment, there was no significant growth response in Corsican pine (*P. nigra* var. maritima).

The inoculation of Virginia pine with *P. tinctorius* resulted in a shoot weight that was lower than that of the pines with the natural mycorrhizal associations. The native mycorrhizal fungi were apparently more favorably associated with the pine seedlings under the experimental conditions. This was also reflected in some of the nutrient results. The uptake of P and Mn was 25% greater and that of Zn was 40% greater for the native mycorrhizal treatment than for the other plants (Table 2). The plants with the *P. tinctorius* treatment also tended to have a lower diameter. An inhibitory effect of inoculation of loblolly pine with *P. tinctorius* was shown in a field study (Walker et al., 1985) in which the growth of inoculated plants was less vigorous than that of noninoculated plants.

The close correspondence between the chemical uptake by plants and the RWD indicates that the rate of root growth is more important than the specific absorption rate in the extraction of nutrients from soil. This is one of the major findings of this study. Barber and MacKay (1985) recently demonstrated that the greater uptake of P and K by seedling maize (*Zea mays* L.) roots receiving a greater water supply was largely due to an increase in root growth and to a small extent due to an increase in the transport of solutes to roots. Nitrogen uptake by perennial ryegrass (*Lolium perenne* L.) increased with increase in root weight duration (Luxmoore and Millington, 1971) and an increase in water supply significantly increased RWD but reduced specific absorption rate, an effect attributed to dilution of the dissolved N. An increase in irradiance greatly increased the RWD of the perennial ryegrass and was associated with reduced specific absorption rate. The maize and ryegrass studies illustrate the much larger increase in RWD than specific absorption rate to more favorable environmental conditions. A similar pattern was established with CO_2 enrichment effects on Virginia pine.

The selection of 0.45 of total dry weight for plant C content may not have been appropriate as a constant value independent of CO_2-enrichment treatment. It is possible that the proportion of C may have increased at elevated CO_2. If this was the case, the analysis of chemical-use efficiency was based on conservative data for the nutrient chemicals. In other words, increases in chemical-use efficiency with elevated CO_2 for P, K, Mg, B, Mn, and Ba would have been larger, with an increasing C content in plant tissues, with CO_2 enrichment. On the other hand, the enrichment response ratios of Al, Fe, Cu, and Zn (Table 4) would have been higher and closer to unity. It is unlikely that there would be sufficient changes in the type of C compounds formed with CO_2 enrichment to change the pattern of results presented.

The increases in nutrient-use efficiency for several nutrient elements (P, K, Mg, B) suggests that plants may be able to increase the internal recycling of deficient nutrients to new meristems with CO_2 enrichment. While such mechanisms could support growth increases, there may be some energy requirements that limit the efficiency of this process as nutrient concentration in tissues is reduced. This aspect of plant response to CO_2 enrichment requires further evaluation. The mean concentrations of N, P, K, Ca, and Mg in the needles at the final harvest were 11.4, 1.4, 4.4, 5.4, and 0.8 g/kg, respectively. These values correspond to data for other pine species, as documented by Kimmins et al. (1979), and do not indicate any nutrient imbalance in the foliage.

The cation-anion charge balance calculations (Table 5) show cation excess values up to 2.6 mmol(+)/plant. This value is small in comparison with the soil cation exchange capacity (CEC) of 227 mmol(+)/pot, and a small release of H^+ from the roots to balance excess cation uptake would have little or no influence on bulk soil pH. Since there was one plant per pot, the cation excess was <2% of the soil CEC. Nye (1981) has shown that the release of protons from roots can decrease rhizosphere pH one or two units below the bulk soil pH. A restricted zone of influence for protons released from roots could account for the increased solubility and uptake of Al, Fe,

Cu, and Zn. Sarkar and Wyn Jones (1982a, b) have shown increased uptake of Fe, Mn, and Zn by dwarf French bean (*Phaseolus vulgaris*) in growth situations with lower rhizosphere pH. Lutz et al. (1972) have shown increased Al, Mn, andZn uptake by maize with decrease in soil pH. Data presented by Harmsen (1977) indicate that up to a fourfold increase in Zn solubility can occur with a decrease in pH from 6.1 to 5.9. Zinc may be a useful indicator element for pH changes at the root-soil interface.

Organic acids have been shown to increase the weathering rate of soil minerals. Boyle and Voigt (1973) presented results showing the contributions of tree seedling rhizospheres to the weathering of silt-sized particles of K minerals. In further work, Boyle et al. (1974) demonstrated the weathering of biotite with a range of organic acids. They suggested that K^+ ions can be released from mineral edges by replacement with H^+ ions. If CO_2 enrichment is associated with rhizosphere acidification, an increase in mineral weathering can be expected to provide a source of some additional plant nutrients.

Nutrient leaching was suggested by O'Neill et al. (1977) to be a sensitive indicator of distress in terrestrial ecosystems. They compared results from a range of studies in which trace metals (Pb, Cd, Cu, Zn, and As) were added to various experimental units. Soil cores were the smallest size and field plots were the largest. They concluded that with increse in stress due to toxic metals, there was an increase in nutrient leaching. The opposite effect may occur with elevated CO_2. The reduction in P, K, and NO_3^- concentration in leachate with CO_2 enrichment and a similar trend for Mg provide some supportive data for increased nutrient retention by soil-plant systems at elevated CO_2. This would represent an example of eustress (Selye, 1973) in which plant processes are enhanced as a result of a change in environmental conditions.

SUMMARY

This experiment establishes the following responses of Virginia pine to CO_2 enrichment under nutrient-deficient soil conditions: (i) greater growth, particularly of roots; (ii) greater nutrient uptake associated with greater root weight; (iii) greater nutrient-use efficiency, particularly for P and K; and (iv) reduced P, K, and NO_3^- concentrations in leachate from the soil. An increase in rhizosphere acidification associated with an increase in cation excess may explain the observed increase in the Zn content of the pines at elevated atmospheric CO_2.

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