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Discrimination against $C^{18}O^{16}O$ during photosynthesis and the oxygen isotope ratio of respired CO_2 in boreal forest ecosystems

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Abstract. Our objective was to analyze factors that influence changes in the oxygen isotope ratio ($\delta^{18}O$) of atmospheric CO_2 within boreal forest ecosystems. We made measurements in the three major forest types (black spruce, jack pine, and aspen) at the southern and northern ends of the boreal forest in central Canada. This research was part of a larger study, the Boreal Ecosystem-Atmosphere Study (BOREAS). In terrestrial ecosystems the $\delta^{18}O$ value of atmospheric CO_2 is strongly influenced by isotope effects that occur during photosynthesis and respiration. Of primary importance is an equilibrium isotope effect that occurs between oxygen in CO_2 and oxygen in soil water and plant chloroplast water. During the equilibrium reaction the oxygen isotope ratio of CO_2 becomes enriched in ^{18}O relative to that of water. We measured seasonal changes in the oxygen isotope ratio of (1) water input to the ecosystems (precipitation), (2) water taken up by the major plant species from the soil (plant stem water), and (3) water in plant leaves. We used this information in calculations of isotope discrimination during photosynthesis and soil respiration. Discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) (influenced by equilibration with chloroplast water) averaged approximately 21‰ at midday and was similar for all forest types. In contrast, CO_2 released during plant and soil respiration had an average $\delta^{18}O$ value of -14.4‰ but was less depleted in ^{18}O than would be expected for respired CO_2 in isotopic equilibrium with soil water. This effect was most pronounced in black spruce sites because of the extensive coverage of moss on the ground surface and the observation that water in the upper moss layers can have an oxygen isotope ratio substantially different from water in deeper soil layers.

1. Introduction

Measurements of the stable isotope ratio of atmospheric CO_2 can provide information useful for studies of the global carbon budget, although much of the previous work with the stable isotope ratios of atmospheric CO_2 has focused only on carbon isotopes [*Keeling et al.*, 1989; *Ciais et al.*, 1995; *Francey et al.*, 1995]. Recently, substantial progress has been made in understanding the mechanistic basis for oxygen isotope effects that occur during vegetation and soil CO_2 exchange [*Hesterberg and Siegenthaler*, 1991; *Farquhar et al.*, 1993; *Flanagan et al.*, 1994; *Flanagan and Varney*, 1995; *Williams et al.*, 1996]. The $\delta^{18}O$ value of atmospheric CO_2 is strongly influenced by an equilibrium isotope effect that occurs between oxygen in CO_2 and oxygen in plant water, soil water and ocean water [*Francey and Tans*, 1987; *Friedli et al.*, 1987; *Farquhar et al.*, 1993]. During the equilibrium reaction the oxygen isotope ratio of CO_2 becomes enriched in ^{18}O relative to that of water [*Bottinga and Craig*, 1969]. The ^{18}O content of ocean water is relatively constant throughout the globe, varying only slightly in association with ocean salinity [*Craig and Gordon*, 1965]. In contrast, the ^{18}O content of fresh water (precipitation, soil water, plant water) is very different from that of ocean water, and it also varies substantially across the globe [*Dansgaard*, 1964; *International Atomic Energy Agency (IAEA)*, 1986]. Carbon dioxide exchange between the terrestrial biosphere and atmosphere therefore has a different effect on the oxygen isotopic composition of atmospheric CO_2 than does CO_2 exchange with the oceans [*Farquhar et al.*, 1993; *Ciais et al.*, 1996a, b]. Within terrestrial ecosystems, photosynthesis and respiration have contrasting effects on the $\delta^{18}O$ of atmospheric CO_2 because of the difference in the oxygen isotope ratio of water in soils and plant chloroplasts [*Farquhar et al.*, 1993; *Ciais et al.*, 1996a, b]. The different isotope effects associated with photosynthesis and respiration allow separation of these components of net CO_2 exchange on local and regional scales using simultaneous measurements of changes in CO_2 concentration and isotopic ratio [*Yakir and Wang*, 1996].

While much progress has been made in understanding the oxygen isotope effects that occur during leaf gas exhange, and in the modeling of the effects of CO_2 exchange on the oxygen isotope ratio of atmospheric CO_2 at the global scale, only a few experimental studies have been conducted to examine the factors influencing the $\delta^{18}O$ value of CO_2 within plant canopies. Such information is necessary in order to constrain modeled estimates so that ecosystem influence on the oxygen isotopic composition of atmospheric CO_2 on large spatial scales can be modeled with confidence and compared to atmospheric observations. Our objective was to analyze

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factors that influence changes in the oxygen isotope ratio of atmospheric CO_2 within boreal forest ecosystems. We made measurements in the three major forest types at the southern and northern limits in the Canadian boreal forest. This research was part of a larger study, the Boreal Ecosystem-Atmosphere Study (BOREAS), that aims to improve our understanding of the interactions between the atmosphere and the boreal forest, a globally important biome [*Sellers et al.*, 1995].

2. Materials and Methods

A description of the study sites, air sample collection procedures, and isotopic analysis was provided in a previous paper [*Flanagan et al.*, 1996]. We describe below the methods that were specifically used in the analysis of the ^{18}O content of atmospheric CO_2.

2.1 Meteorological Measurements

Fine-wire copper-constantan thermocouples, shielded from direct solar radiation, were used for air temperature measurement at heights of 9 and 0.5 m within the canopy. Photon flux density was measured with either quantum sensors (LI-190SA, Li-Cor, Lincoln, Nebraska) or GaAsP photodiodes (G1118, Hamamatsu Corp., Bridgewater, New Jersey) calibrated relative to a Li-Cor quantum sensor. The light sensors were positioned at the same heights as the air temperature thermocouples. Relative humidity (RH) was measured at the midpoint within the forest canopy (middle of the live crown, approximately 6.5 m in jack pine and black spruce sites, and at 9-m in the Aspen site in the south) using a 207 temperature and RH probe (Campbell Scientific, Logan, Utah). Temperature, light, and relative humidity sensors were located at different heights in the canopy by attachment to a 9 m mast (Rohn E20 telescoping mast). All sensors were connected to a solid-state data logger (CR10 or CR21X, Campbell Scientific, Logan, Utah). The data logger scanned the sensors at 30-s intervals and averaged readings for 30-min intervals throughout the day.

2.2 Water Sample Collection

Water samples from tree stem xylem tissue, leaf tissue, atmospheric water vapor, and precipitation were collected for stable isotope analysis. Small nongreen stem samples (approximately 7 X 60 mm) were cut from trees near the top of the canopy and immediately placed in a glass tube which was sealed with a rubber stopper and wrapped with Parafilm. One sample from each of 4 or 5 trees was collected at midday on each sampling date. Foliage samples were collected from a variety of positions within the tree crown and combined for one tree. The foliage was immediately placed in a glass tube which was sealed with a rubber stopper and wrapped with Parafilm. The glass tubes containing plant samples were placed on dry ice in a small cooler in the field and returned to the laboratory where they were stored in a freezer until water was extracted from the tissue using a cryogenic vacuum distillation apparatus [*Ehleringer and Osmond*, 1989]. Atmospheric water vapor was collected by pumping (TD-4N pump, Brailsford & Co. Inc., Rye, New York) air through two glass traps (Vacuum traps 8746, Ace Glass, Vineland, New Jersey) cooled with an ethanol-dry ice slurry. Precipitation was collected in Atmospheric Environment Service (AES) standard collectors at the AES meterological station at the zoo in Thompson, Manitoba, and in the private yard of a residence in Prince Albert, Saskatchewan. Precipitation samples from a series of separate collections made close together in time were combined in one tight sealing Nalgene bottle (500 mL) and analyzed as a bulk sample for oxygen isotopic composition. Small stem water samples were prepared for measurements of oxygen isotopic composition by converting the oxygen in water to CO_2 using the guanidine hydrochloride method [*Wong et al.*, 1987]. Precipitation samples were analyzed using the standard CO_2 equilibrium method [*Ehleringer and Osmond*, 1989].

2.3 Modeling Isotopic Compositions

A mass balance approach was used to determine the relative importance of discrimination during photosynthetic gas exchange, the isotope ratio of respired CO_2, and turbulent exchange on the oxygen isotope ratio of atmospheric CO_2 within plant canopies. The influence of the three major fluxes is weighted by their associated isotope effects, as shown in the equation below [*Lloyd et al.*, 1996]:

$$M_i C_i \frac{d\delta_i}{dt} = A\Delta_A + R(\delta_R - \delta_i) + F_{oi}(\delta_o - \delta_i) \quad (1)$$

where M_i is the molar density of air inside the canopy (mol m^{-3}); C_i is the average CO_2 concentration inside the canopy (μmol mol^{-1}); A is the net CO_2 assimilation rate (μmol m^{-2} s^{-1}); Δ_A is discrimination during photosynthetic gas exchange (‰); R is the respiration rate of plants and soil (μmol m^{-2} s^{-1}); δ_R is the isotope ratio of plant and soil respired CO_2 (‰); δ_i is the average isotope ratio of atmospheric CO_2 inside the canopy (‰); δ_o is the average isotope ratio of CO_2 in the atmosphere above (outside) the canopy (‰); and F_{oi} is the one-way turbulent flux of CO_2 (μmol m^{-2} s^{-1}) into the canopy from the atmosphere above the canopy.

The model of *Farquhar and Lloyd* [1993] was used to estimate the influence of discrimination during photosynthetic gas exchange (Δ_A) on the isotopic ratio of forest air, as shown below (equation (2)). During photosynthetic gas exchange a portion of the CO_2 that enters the leaf and equilibrates with chloroplast water is not fixed and diffuses back out of the leaf with an altered oxygen isotopic ratio. The amount of CO_2 that escapes from the leaf depends on the partial pressure of CO_2 in the chloroplast and conductance (mesophyll, stomatal, and boundary layer) to CO_2 within and outside of the leaf:

$$\Delta_A = a + \frac{C_c}{C_a - C_c}(\delta_c - \delta_o) \quad (2)$$

where a is the average fractionation during diffusion of CO_2 into and out of a leaf (7.4‰); C_c and C_a are the partial pressures of CO_2 in the chloroplast and ambient air, respectively; δ_c is the oxygen isotope ratio of CO_2 in the chloroplast (Pee Dee belemnite (PDB) scale); and δ_o is the oxygen isotope ratio of CO_2 in the atmosphere above the forest (see *Flanagan et al.* [1996, Table 1] for the values that were used for δ_o). Measurements of the carbon isotope ratio of leaf tissue were used to estimate the average partial pressure of CO_2 in the chloroplast, based on the *Farquhar et al.* [1989]

model of isotopic fractionation during photosynthetic gas exchange. We make the simplifying assumption that C_c is constant during the day because including diurnal variation in C_c would require us to use a detailed photosynthesis model in our calculations. Our primary purpose is to use equation (2) to estimate diurnal variation in $A\Delta_A$, which is mainly controlled by diurnal variation in A and δ_c. Changes in δ_c are controlled by changes in the isotope ratio of leaf water and temperature effects on the CO_2 and H_2O equilibration reaction, as described below.

We assume that CO_2 in the chloroplast (δ_c) is at complete isotopic equilibrium with leaf (chloroplast) water. At equilibrium the oxygen isotope ratio of CO_2 becomes enriched relative to that of chloroplast water, with the extent of the enrichment dependent on a temperature- sensitive fractionation factor (α_{BC}). Taking into account the conversion between PDB and SMOW scales, the isotopic composition of CO_2 (PDB-CO_2) in equilibrium with leaf water (δ_l) (SMOW) can be calculated as

$$\delta_c\ (\text{PDB–CO}_2) = \frac{\alpha_{BC}\,[\,1 + \delta_l\,(\text{SMOW})\,]}{1.04142} - 1 \qquad (3)$$

The equation presented by *O'Neil and Adami* [1969] [see also *Bottinga and Craig*, 1969] was used for calculating the temperature dependence of the CO_2-H_2O equilibration fractionation factor (α_{BC}). The temperature dependence of α_{BC} is approximately -0.2‰ per °C in the range 0-45°C [*O'Neil and Adami*, 1969; *Bottinga and Craig*, 1969]. Leaf water (δ_l) isotopic ratio can be estimated based on a model of isotopic fractionation during transpiration [*Craig and Gordon*, 1965; *Dongmann et al.*, 1974; *Flanagan et al.*, 1991]:

$$R_l = \alpha^*\left[\,\alpha_k\,R_s\left(\frac{e_i - e_a}{e_i}\right) + R_a\left(\frac{e_a}{e_i}\right)\right] \qquad (4)$$

$$\delta_l = \left[\frac{R_l}{R_{SMOW}} - 1\right] \qquad (5)$$

where R is the molar ratio of the heavy to light isotope ($^{18}O/^{16}O$) and the subscripts l, s, and a refer to leaf water, stem water and atmospheric water vapor, respectively; the R value used for SMOW was 0.0020052 [*Ehleringer and Osmond*, 1989]; e is the partial pressure of water vapor and the subscripts i and a refer to the leaf intercellular air spaces and the ambient air, respectively; α^* is the equilibrium fractionation factor between liquid water and water vapor. The regression equation listed by *Majoube* [1971] was used to calculate values for α^* at different temperatures. The parameter α_k is the kinetic fractionation factor which has a value of 1.0285 for $^{16}O/^{18}O$ [*Merlivat*, 1978]. In order to calculate the partial pressure of water vapor in the intercellular air spaces we assumed that leaf temperature was equal to air temperature and that the leaf air spaces were saturated.

We used a simple mixing model developed by *Keeling* [1958, 1961] to calculate the oxygen isotope ratio of CO_2 respired by a forest ecosystem:

$$\delta_f = \frac{[CO_2]_o}{[CO_2]_f}\,(\delta_o - \delta_R) + \delta_R \qquad (6)$$

where $[CO_2]$ is concentration of CO_2 and δ is the stable isotope ratio of CO_2, and the subscripts f and o represent the atmosphere within a forest and the atmosphere above (outside) the forest, respectively. It can be seen from equation (6) that a plot of $1/[CO_2]_f$ versus δ_f gives a straight line relationship with a slope, $[CO_2]_o\,(\delta_o - \delta_R)$, and an intercept, δ_R. We used this model to estimate δ_R, the isotope ratio of CO_2 respired by plants and soils. Estimates were obtained from the y intercept of a geometric mean linear regression [*Sokal and Rohlf*, 1981] between $\delta^{18}O$ and $1/[CO_2]$ values measured on air samples collected at 2 heights in the forest canopy during the night when photosynthesis was not active.

Carbon dioxide in the soil (soil CO_2) is assumed to be in isotopic equilibrium with soil water [*Hesterberg and Siegenthaler*, 1991]. The $\delta^{18}O$ value of CO_2 released from the soil (soil respired CO_2), however, should be depleted by approximately 8.8‰ from that in equilibrium with soil water because of fractionation that occurs during diffusion [*Hesterberg and Siegenthaler*, 1991]. When we make reference to the isotopic composition of "respired CO_2," we imply that we have included fractionation associated with diffusion, in addition to fractionation occurring during equilibration with a water pool. We use the tree stem water isotopic composition (δ_{stem}) to estimate the $\delta^{18}O$ value of soil water, since no fractionation occurs during water uptake by plant roots [*White et al.*, 1985; *Ehleringer and Dawson*, 1992]. We estimate the $\delta^{18}O$ value of CO_2 released from the soil ($\delta_{R\text{-}soil}$) with the following equation:

$$\delta_{R\text{–}Soil}\ (\text{PDB–CO}_2) = \frac{\alpha_{BC}\,[\,1 + \delta_{stem}\,(\text{SMOW})]}{1.04142} - 1.0088 \qquad (7)$$

It is important to recognize that the isotope ratio of soil respired CO_2 ($\delta_{R\text{-}soil}$, calculated using equation (7)) is only one component of the isotope ratio of CO_2 respired by an entire ecosystem (δ_R), which is calculated using equation (6).

During our collection of air samples for isotope analysis in the southern black spruce and jack pine sites, simultaneous measurements were made by other research groups of net ecosystem CO_2 exchange using the eddy covariance technique [*Baldocchi et al.*, 1996]. The net CO_2 exchange measurements made at night were used to estimate R, the respiration rate of plants and soil. The net CO_2 assimilation rate during the day was then calculated as the sum of the net ecosystem exchange during the day and the ecosystem respiration rate. By combining the CO_2 flux measurements with our isotope measurements and calculated values (Δ_A and δ_R), we obtained values for all terms in equation (1) except $F_{oi}\,(\delta_o - \delta_i)$, which was calculated by difference.

3. Results

The $\delta^{18}O$ (SMOW) value of precipitation showed the expected seasonal cycle, with low values recorded during the winter and values more enriched in ^{18}O observed during the summer months (Figure 1). Similar values and patterns of

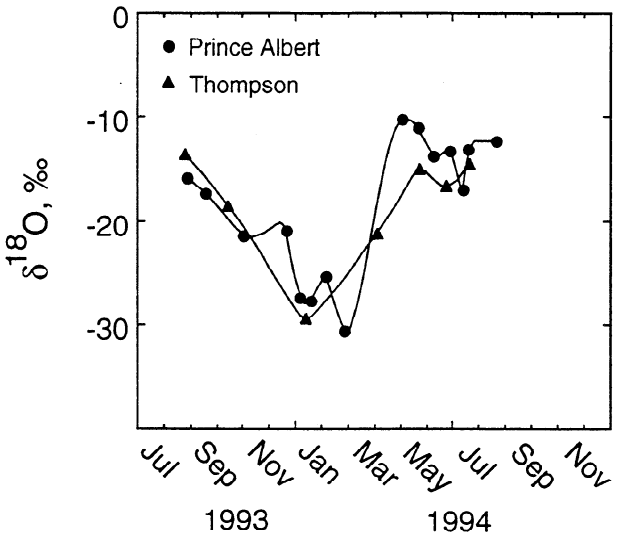

Figure 1. Seasonal variation in the oxygen isotope ratio of precipitation in the northern (Thompson, Manitoba) and southern (Prince Albert, Saskatchewan) BOREAS study areas. The $\delta^{18}O$ values are expressed relative to SMOW.

seasonal change were observed at both Prince Albert and Thompson. However, there was substantial variation (approximately 7‰) in the $\delta^{18}O$ values of precipitation samples collected during the main growing season (May-September) in Prince Albert (Figure 1). The volume-weighted average $\delta^{18}O$ value for precipitation collected from August 1993 to September 1994 was -18.1‰ and -18.6‰ for Prince Albert and Thompson, respectively. These values were more negative than the -16.3‰ measured for groundwater collected at the jack pine site in the southern study area (SSA) near Prince Albert.

The stem water $\delta^{18}O$ values were similar for aspen and jack pine trees at both the southern and northern study sites in all three study periods (Figure 2). In contrast, the stem water $\delta^{18}O$ values observed for black spruce were more variable; at times they were very similar to the other tree species and at times were significantly different. For example, stem water in the black spruce trees in the SSA showed a progressive 3.3‰ change (a change in $\delta^{18}O$ values from -18.0 to -14.7‰) during an 11-day period (Julian day 202-213) following 3 days of rain (Julian days 200-202) (Figure 2). The rain accumulated over days 200-202 had a $\delta^{18}O$ value of -13.1‰. Jack pine trees in the SSA showed no significant change in stem water $\delta^{18}O$

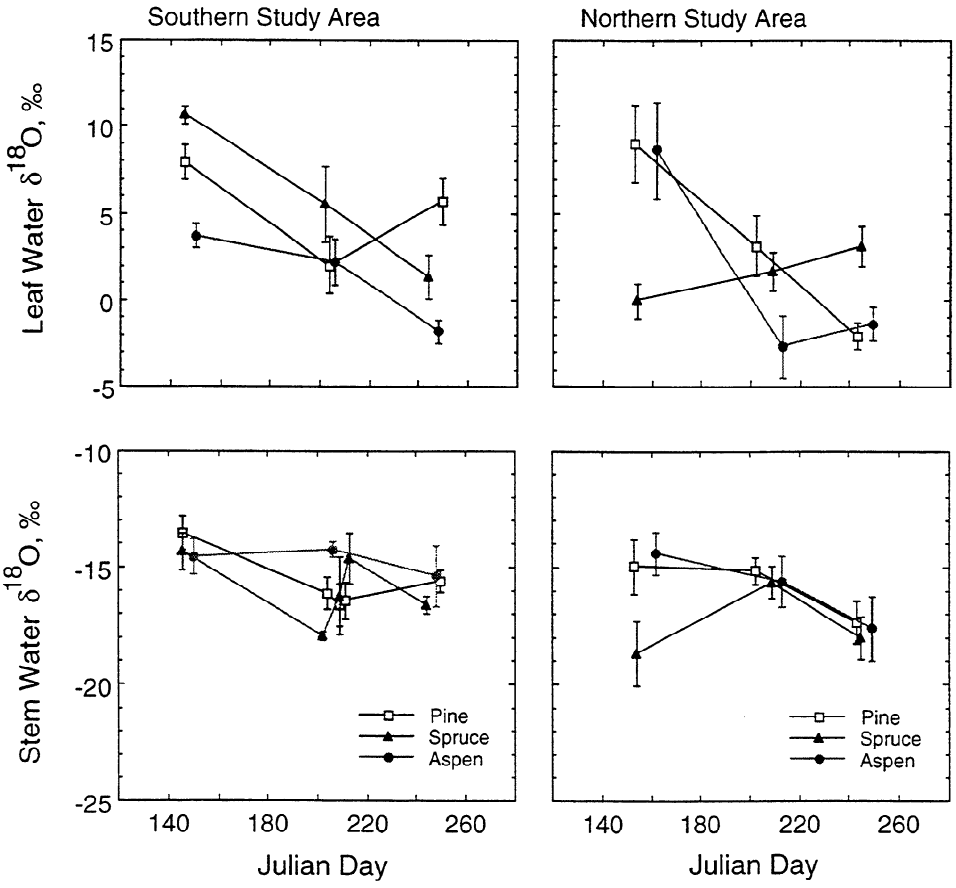

Figure 2. Variation in the oxygen isotope ratio of water extracted from plant leaves and stems from the dominant tree species in the northern and southern BOREAS study areas during 1994. Values are the mean ± 1 standard deviation, n=5. The $\delta^{18}O$ values are expressed relative to SMOW.

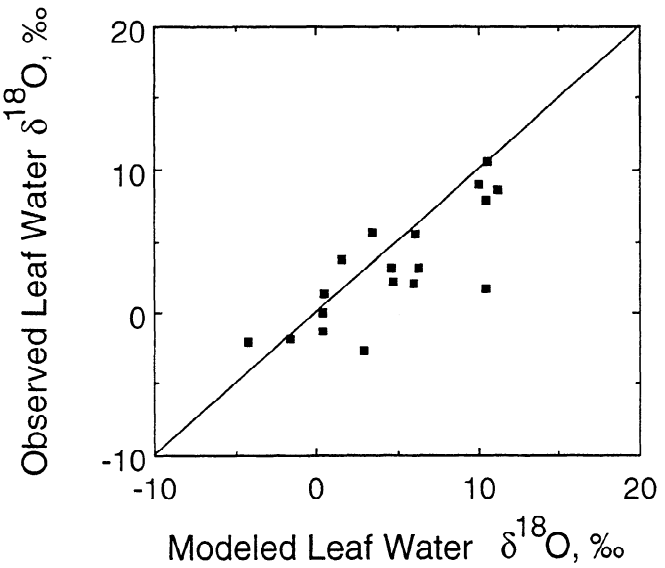

Figure 3. Comparison between observed and modeled leaf water oxygen isotope ratio. Observed values are the average of 5 separate leaf collections. The modeled values were averages calculated for a 2-hour interval just preceeding the time the foliage was collected. The $\delta^{18}O$ values are expressed relative to SMOW.

values during a comparable period of time (Julian Days 204-211), however, and had average stem water $\delta^{18}O$ values of -16.4 ± 0.3‰, which were very similar to the isotopic composition of groundwater (-16.3‰) at the jack pine site. This suggests that the isotope ratio of stem water in black spruce trees was more likely to be influenced by short-term changes in the $\delta^{18}O$ values of surface inputs (summer precipitation and melting snow) than the other two tree species (forest types).

The oxygen isotope ratio of leaf water was enriched in ^{18}O relative to stem water and showed substantial variation (approximately 14‰) among species sampled at different times during the growing season (Figure 2). There was a good correlation (correlation coefficent, r=0.799) between average observed leaf water $\delta^{18}O$ values and values predicted by the evaporative enrichment model (equations (4) and (5)) (Figure 3). The modeled values were averages calculated for a 2-hour interval just preceeding the time the foliage was collected. In general, there was a tendency for leaf water to be less enriched in ^{18}O later in the season (Figure 2), because of lower temperatures and higher relative humidity, although environmental conditions and leaf water $\delta^{18}O$ values varied substantially among sites from day to day.

The modeled leaf water data were used to calculate the isotope ratio of CO_2 in equilibrium with chloroplast water.

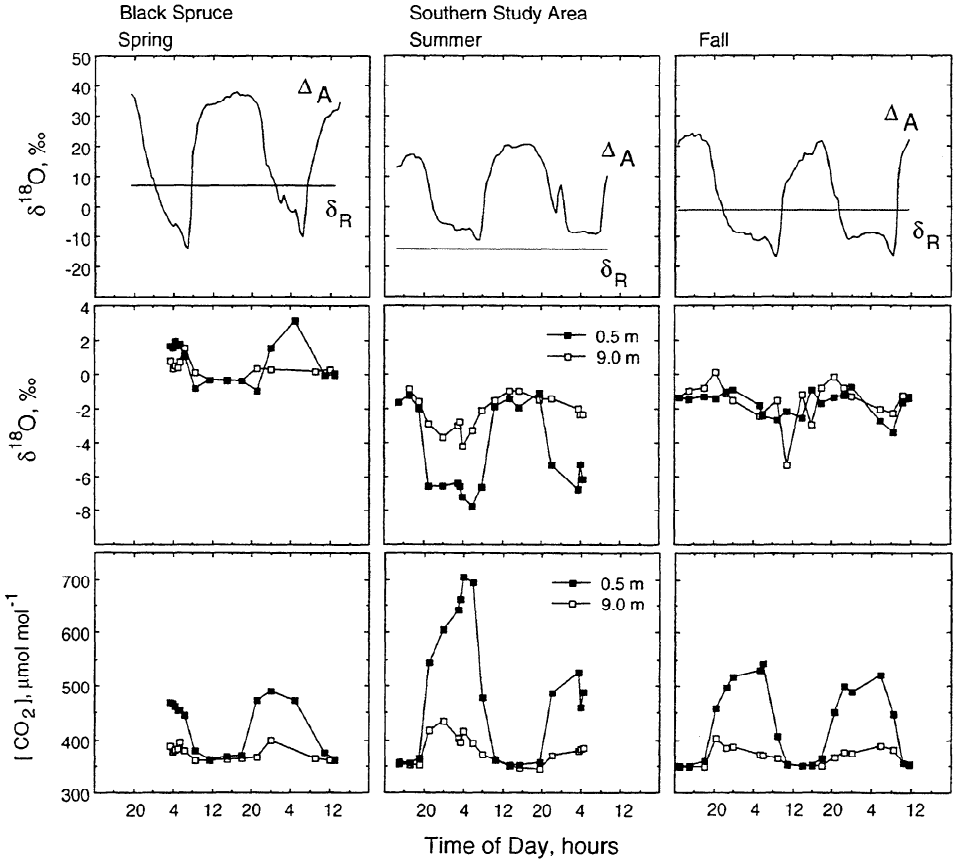

Figure 4. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Picea mariana* forest in the BOREAS southern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of plant and soil respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

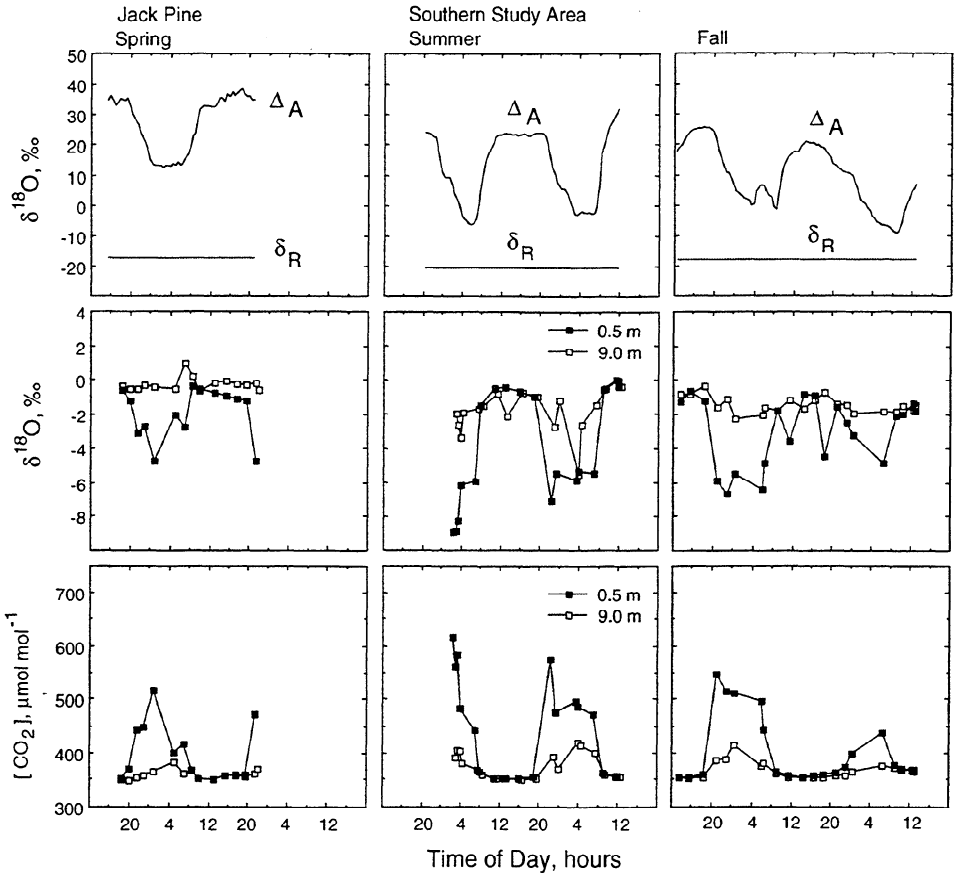

Figure 5. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Pinus banksiana* forest in the BOREAS southern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

The modeled values of discrimination during photosynthetic gas exchange (influenced by equilibration with chloroplast water) are shown in Figures 4-9 for all sites and sampling periods. During the day, discrimination during photosynthesis was predicted to enrich atmospheric CO_2 in ^{18}O (Figures 4-9). The calculated Δ_A values were similar for all forest types with an overall average of 21‰. However, the magnitude of the influence of discrimination during photosynthetic gas exchange on the atmosphere is also dependent on CO_2 assimilation rate (see below).

During the majority of sampling periods there were large changes in CO_2 concentration and $\delta^{18}O$ value observed during diurnal time courses (Figures 4-9). When CO_2 concentration increased at night in response to respiration inputs, the $\delta^{18}O$ value of atmospheric CO_2 normally declined. Usually, there were increases in $\delta^{18}O$ values associated with declines in CO_2 concentration during the day. In addition, there was a strong vertical stratification of CO_2 concentration and isotope ratio, with higher concentrations and lower $\delta^{18}O$ values observed closer to the soil surface (Figures 4-9). An exception to this pattern was observed in the SSA black spruce site during the spring sampling period, when an increase in CO_2 concentration at night resulted in an increase in the $\delta^{18}O$ value of atmospheric CO_2 at low levels within the canopy (Figure 4).

We estimated the oxygen isotope ratio of CO_2 released during plant and soil respiration (δ_R) by calculating the y intercept from a linear regression of atmospheric $\delta^{18}O$ values and $1/[CO_2]$ for samples collected at night (see equation (6)) (Figures 10 and 11). In some cases there was no significant linear relationship between $\delta^{18}O$ values and $1/[CO_2]$ for air samples collected within the canopy (e.g., Figures 10c, 11a, and 11g). In these cases the isotope ratio of respired CO_2 was taken as the mean of the measured atmospheric $\delta^{18}O$ values. The estimates we obtained of δ_R were, on average, enriched in ^{18}O relative to $\delta_{R\text{-}Soil}$ values calculated with equation (7) (Figure 12). Two occasions occurred when measured δ_R values were more negative than the predicted $\delta_{R\text{-}Soil}$ values; these corresponded to times when samples were collected during

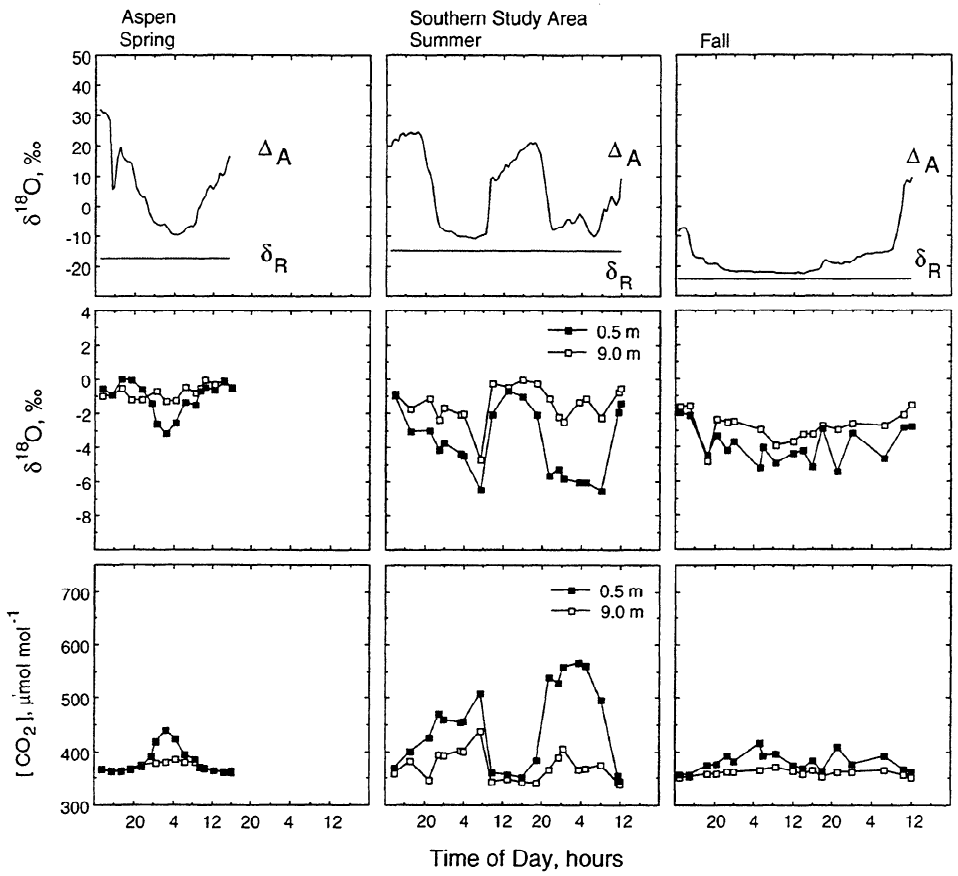

Figure 6. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Populus tremuloides* forest in the BOREAS southern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

mild rain. The observation of measured δ_R values being generally higher than predicted $\delta_{R\text{-}Soil}$ values was particularly noticeable in the black spruce site in the SSA and was primarily due to the fact that the $\delta^{18}O$ value of water taken up by tree roots was not necessarily representative of the composition of all water in the soil profile, particularly water in the shallow soil layers. In the black spruce sites this effect was most pronounced because of the extensive coverage of moss on the ground surface. Evaporation of water from the moss can result in significant changes to the oxygen isotopic composition of moss water. Any CO_2 diffusing out of the moss/soil surface has the potential to equilibrate isotopically with this moss water pool, which can be substantially enriched in ^{18}O and very different in composition from the rest of the water in the soil profile.

Examples to illustrate the influence of the moss layer in the black spruce site in the SSA are discussed below. During the midsummer (July) sampling period at the black spruce site, the moss had been recently wetted by precipitation and had a relatively negative $\delta^{18}O$ (SMOW) value of -7.3 ± 1.3‰ (mean±standard deviation, n=5), which was enriched in ^{18}O relative to black spruce stem water (-18.0 ± 0.2‰), but still significantly lower than the black spruce leaf water $\delta^{18}O$ (SMOW) value of 5.5 ± 2.2‰, measured on leaf samples collected at midday. The δ_R value calculated using the linear regression technique (-14.4‰, Figure 10b) was very similar to the $\delta_{R\text{-}Soil}$ value predicted for CO_2 in isotopic equilibrium with moss water (-13.4‰) but enriched in ^{18}O relative to that of $\delta_{R\text{-}Soil}$ predicted for CO_2 in equilibrium with bulk soil water (as estimated by tree stem water, Figure 12). In contrast, there was no significant linear relationship between changes in CO_2 concentration and the $\delta^{18}O$ value for atmospheric CO_2 during the fall (September) sampling period (Figure 10c). Prior to the fall sampling, the moss had not received any precipitation for some time, and moss water must have been relatively enriched in ^{18}O with a similar $\delta^{18}O$ (SMOW) value to that of tree leaf water (1.3 ± 1.3‰, measured at midday). The increase in CO_2 concentration within the canopy at night, resulting from plant and soil respiration, did not cause a significant change in isotope ratio because CO_2 released from the soil/moss surface had equilibrated with moss water and so was similar in isotopic composition to that of atmospheric CO_2 above the forest (Figure 4). If moss water had a $\delta^{18}O$

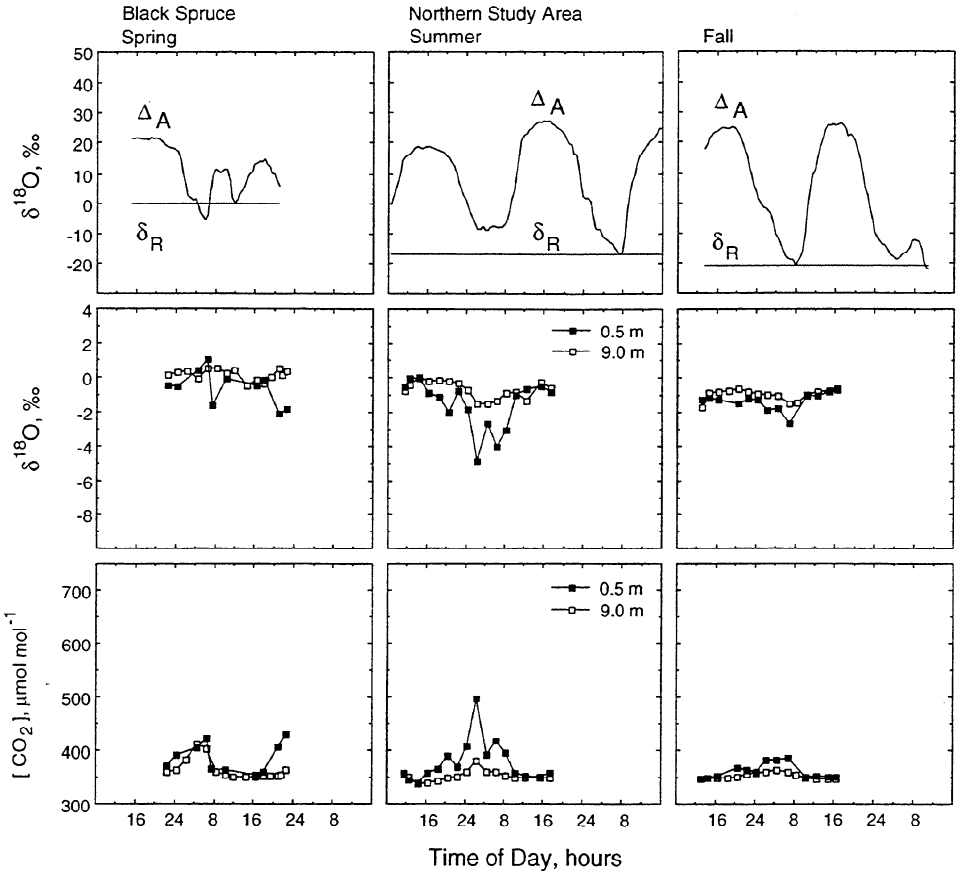

Figure 7. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Picea mariana* forest in the BOREAS northern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

(SMOW) value of approximately 4‰, the expected $\delta_{R\text{-}Soil}$ value for CO_2 in equilibrium with moss water (-1.4‰) would be in close agreement with the average of $\delta^{18}O$ measurements made on CO_2 samples collected at night (Figures 4 and 10c). During the spring sampling period in the black spruce site (May) we observed a significant increase in the $\delta^{18}O$ value of atmospheric CO_2 at night in association with the increase in CO_2 concentration (Figure 4). This suggested that soil respired CO_2 had equilibrated with water that was very enriched in ^{18}O, with the linear regression analysis indicating a δ_R value of 6.6‰ (Figures 10a and 12). Black spruce leaf water had $\delta^{18}O$ (SMOW) values at midday of 10.6‰ during the spring sampling. Assuming that moss water had a similar oxygen isotope ratio as tree leaf water, the expected $\delta_{R\text{-}Soil}$ value for CO_2 in equilibrium with moss water was 6.6‰, in complete agreement with the δ_R value calculated using the linear regression technique.

Our isotope analyses were used in combination with simultaneous CO_2 flux measurements to determine the relative influence of photosynthesis, respiration, and turbulent exchange on the isotope ratio of CO_2 in canopy air. During the day, photosynthetic CO_2 exchange can have the dominant effect on the isotope ratio of atmospheric CO_2, acting to enrich canopy CO_2 in ^{18}O, followed closely by turbulent exchange with the atmosphere above the canopy, which acts in an opposite direction (Figure 13). At night, input of respiratory CO_2 can cause substantial changes (usually depletions) in the ^{18}O content of atmospheric CO_2 within plant canopies as discussed above. The influence of the isotope effects are weighted by the magnitude of the associated CO_2 flux. For example, despite similar Δ_A values during the July and September sampling periods in the southern jack pine site, the influence of discrimination during photosynthetic gas exchange was reduced during September because of a low CO_2 assimilation rate (Figure 14), likely associated with water stress effects on the vegetation. In contrast, discrimination during photosynthetic gas exchange in the black spruce site had similar effects during July and September because no reduction in CO_2 assimilation was apparent during the fall sampling period (Figure 14).

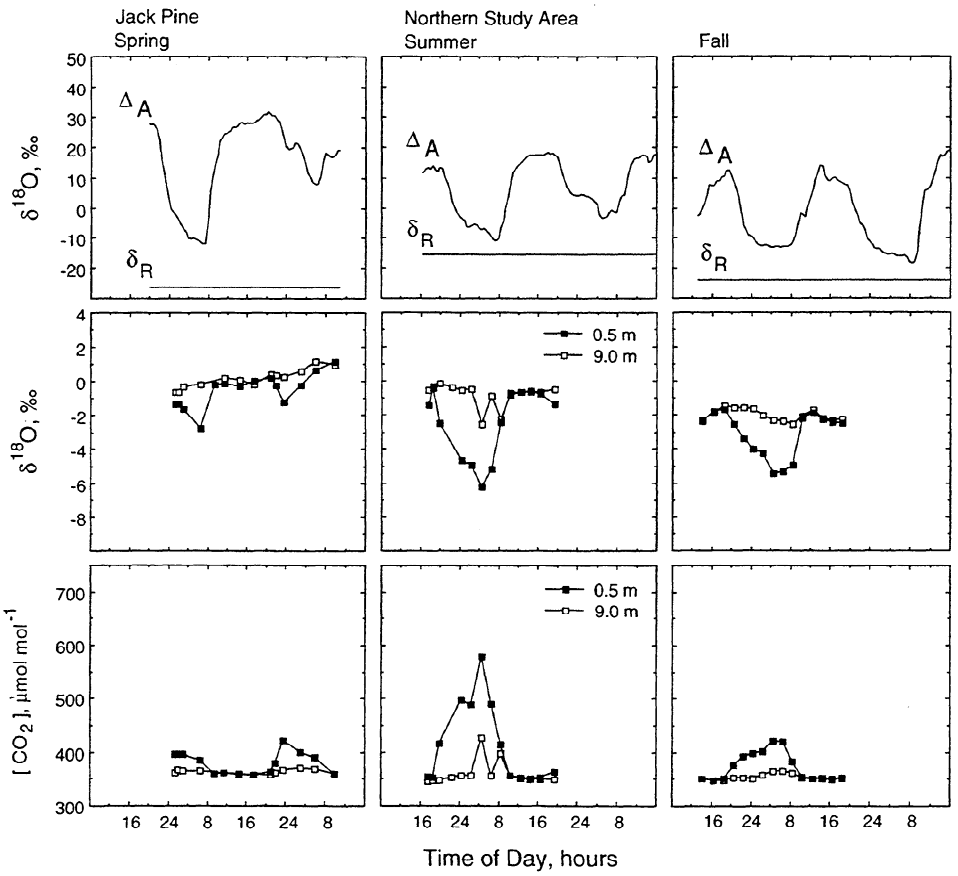

Figure 8. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Pinus banksiana* forest in the BOREAS northern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

4. Discussion

Model calculations indicate that the terrestrial biosphere has the dominant influence on the oxygen isotope ratio of atmospheric carbon dioxide [*Farquhar et al.*, 1993, *Ciais et al.*, 1996a, b]. This strong influence is associated with the very different isotope effects predicted for photosynthesis and respiration in terrestrial ecosystems. Our measurements and associated calculations confirm the contrasting effects of photosynthetic and respiratory CO_2 exchange in boreal ecosystems. Our modeled values of discrimination during photosynthetic gas exchange (Δ_A) at midday were on average approximately 21‰, while CO_2 released during plant and soil respiration was significantly depleted in ^{18}O, with an average δ_R value of -14.4‰. However, our results illustrate a number of factors that could affect estimates of the $\delta^{18}O$ of atmospheric CO_2 in northern regions that are predicted by global models [*Farquhar et al.*, 1993; *Ciais et al.*, 1996a, b].

First, our modeled Δ_A values were slightly higher than values predicted by the *Farquhar et al.* [1993] global model for boreal regions. The difference between our Δ_A values and those predicted by *Farquhar et al.* [1993] resulted because of the low ratio of chloroplast CO_2/ambient CO_2 modeled by *Farquhar et al.* [1993] that was not consistent with the carbon isotope discrimination values we observed in boreal forests [see *Flanagan et al.*, 1996]. Our modeled values for Δ_A were also based on calculations of diurnal variation in the isotopic composition of leaf water. In general, we observed good agreement between measured and predicted leaf water isotopic compositions (Figure 3), providing support for our calculation of Δ_A. There was a tendency, however, for the evaporative enrichment model to slightly overestimate leaf water $\delta^{18}O$ values, likely because of differences between water at the evaporative sites within leaves and the total water extracted from leaf tissue, as has been observed in a number of other studies [*Dongmann et al.*, 1974; *Farris and Strain*, 1978; *Zundel et al.*, 1978; *Flanagan et al.*, 1991; *Yakir et al.*, 1994; *Wang and Yakir*, 1995] and is discussed in detail by *Flanagan* [1993]. However, a number of recent laboratory measurements of Δ_A under a variety of controlled environmental conditions showed strong correlation between observed and modeled Δ_A values, and the results were consistent with the assumption that chloroplast water has an isotopic signature close to that of water at the evaporative

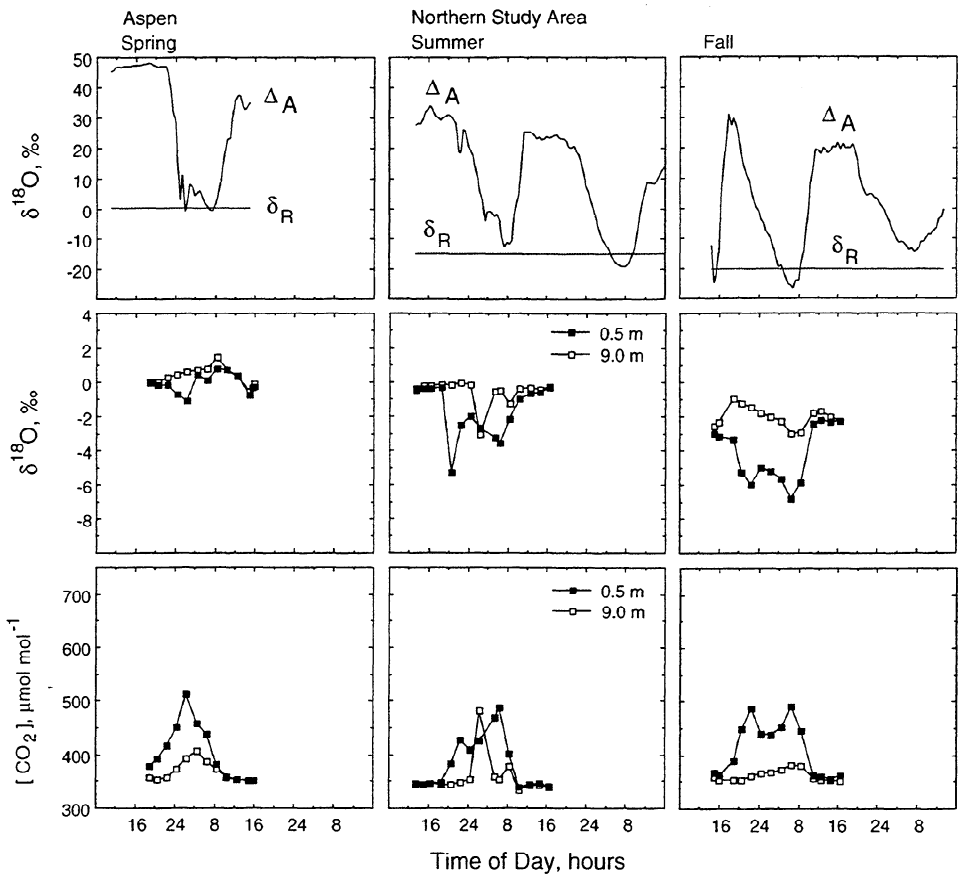

Figure 9. Diurnal variation in the concentration and oxygen isotope ratio of CO_2 in air samples collected within the canopy of a *Populus tremuloides* forest in the BOREAS northern study area during 1994. Air samples where collected at two heights within the canopy, 9 m (open symbols) and 0.5 m (closed symbols) aboveground. Also shown is the diurnal variation in modeled discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (Δ_A) and measured values for the isotope ratio of respired CO_2 (δ_R). The $\delta^{18}O$ values are expressed relative to PDB-CO_2.

sites within leaves [*Flanagan et al.*, 1994; *Williams and Flanagan*, 1996; *Williams et al.*, 1996]. We feel justified therefore in using the Craig-Gordon evaporative enrichment model to estimate the ^{18}O content of chloroplast water in studies of factors influencing the isotope ratio of atmospheric CO_2 within plant communities.

Our estimates for the oxygen isotope ratio of respired CO_2 were generally consistent with average values predicted for boreal regions by the global models of *Farquhar et al.* [1993] and *Ciais et al.* [1996a]. However, our calculated values of δ_R were usually enriched in ^{18}O relative to predictions for soil respired CO_2 ($\delta_{R\text{-}Soil}$). A number of factors could potentially influence this discrepancy between $\delta_{R\text{-}soil}$ and δ_R values as discussed below.

The oxygen isotope ratio of respired CO_2 (δ_R) is dependent on the isotope content of water in soil and plant components. The isotope ratio of water in soils is influenced by precipitation inputs, canopy interception and evaporation, groundwater transport, and the amount of evaporation that occurs from the ground surface [*Ehleringer and Dawson*, 1992]. Water extracted from plant xylem tissue is indicative of the water taken up by plant roots because no fractionation occurs during water uptake by most plants [*White et al.*, 1985; *Ehleringer and Dawson*, 1992]. However, water taken up by tree roots may differ substantially from water in surface layers of the soil (or moss), as was demonstrated particularly by our studies in the black spruce ecosystems, but is also evident in the other forest types as well. Our measurements of δ_R made at night also include the effects of CO_2 respired from plant tissue, in addition to the CO_2 released from the soil surface. Depending on the relative magnitude of plant and soil respiration components, δ_R could be enriched in ^{18}O at night relative to predictions for $\delta_{R\text{-}soil}$. This is possible because leaf water does not always return to the same isotope ratio as source water inputs during a diurnal cycle but can remain enriched in ^{18}O relative to source water at night [*Flanagan and Ehleringer*, 1991; *Flanagan et al.*, 1993]. In addition, green, unsuberized stem tissue can also have higher ^{18}O contents than source water because of evaporative enrichment [*Dawson and Ehleringer*, 1993]. M. Ryan (unpublished data, 1996) has determined that foliage, stem, and soil respiration contribute to total nighttime respiration in the following proportions at BOREAS study sites: SSA jack pine site:

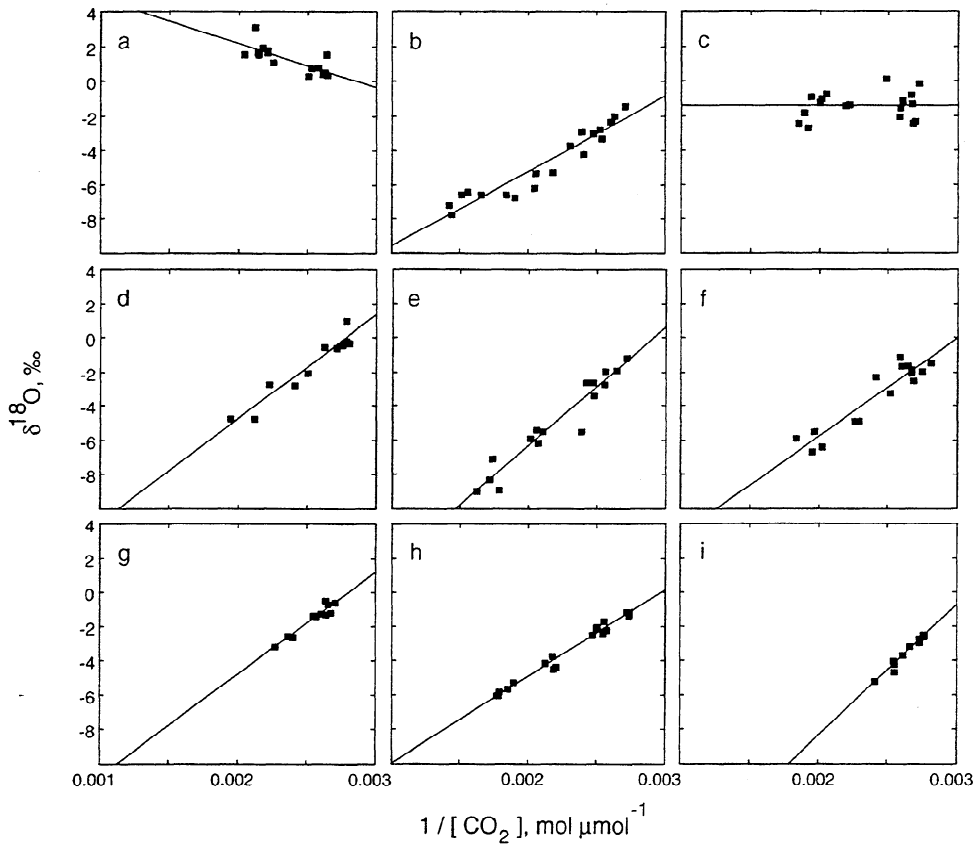

Figure 10. Comparison of the relationships between $1/CO_2$ concentration and the oxygen isotope ratio of CO_2 in air samples collected at night in the BOREAS southern study area during 1994. The black spruce site during (a) spring, (b) summer, and (c) fall sampling periods. The jack pine site during (d) spring, (e) summer, and (f) fall sampling periods. The aspen site during (g) spring, (h) summer, and (i) fall sampling periods.

foliage 0.315, stem 0.079, soil 0.606; NSA black spruce site: foliage 0.39, stem 0.15, soil 0.46. Given the large contribution of leaf to total system respiration, respired CO_2 will have a heterogeneous isotopic signature if leaf water and soil water pools have very different oxygen isotope ratios. This is consistent with the greater variability observed in the oxygen isotope ratio of respired CO_2 compared to the carbon isotope ratio of respired CO_2 calculated using the same technique [*Flanagan et al.*, 1996].

In our studies we have assumed that discrimination during diffusion of CO_2 out of the soil results in an isotope effect of -8.8‰ because of fractionation during molecular diffusion. While the magnitude of the discrimination factor during diffusion through the soil is approximately 8.8‰, its precise value depends on the relative rates of (1) CO_2 production, (2) equilibration between CO_2 and H_2O, and (3) diffusion [*Hesterberg and Siegenthaler*, 1991]. It is also possible that our values of δ_R are enriched in ^{18}O relative to $\delta_{R\text{-}soil}$ because of incomplete isotopic equilibrium between CO_2 and H_2O in soils. Alternatively, CO_2 production could occur near to the surface which would also reduce the influence of fractionation during diffusion out of the soil [*Hesterberg and Siegenthaler*, 1991].

Our data suggest that the isotope ratio of CO_2 respired from the moss/soil surface in the black spruce sites was significantly influenced by short-term changes in precipitation and the oxygen isotope ratio of moss water. Since black spruce ecosystems and other vegetation types with an abundance of moss dominate northern regions, moss water enriched in ^{18}O could have a substantial influence on the oxygen isotope ratio of atmospheric CO_2 in these areas. The ^{18}O-enriched water signal in the moss can get passed on to respired CO_2 diffusing through the moss layer to the forest floor surface and could also influence the atmosphere during moss photosynthetic gas exchange. In addition, both trees and moss in the black spruce ecosystems are strongly influenced by short-term changes in the isotope ratio of summer precipitation, which, in general, is more enriched in ^{18}O than groundwater. This would tend to cause terrestrial biosphere-atmosphere CO_2 exchange to have less of a depletion effect on the ^{18}O content of the atmosphere than global-scale model calculations using average annual precipitation inputs as an estimate for soil water isotopic composition.

Ciais et al. [1996a, b] used a three-dimensional atmospheric transport model in conjunction with estimates of surface fluxes to calculate the influence of terrestrial photosynthesis and respiration, ocean CO_2 exchange, and anthropogenic emissions on the $\delta^{18}O$ of atmospheric CO_2. Their results illustrated the important contrasting effects of terrestrial photosynthesis and respiration. In particular, the

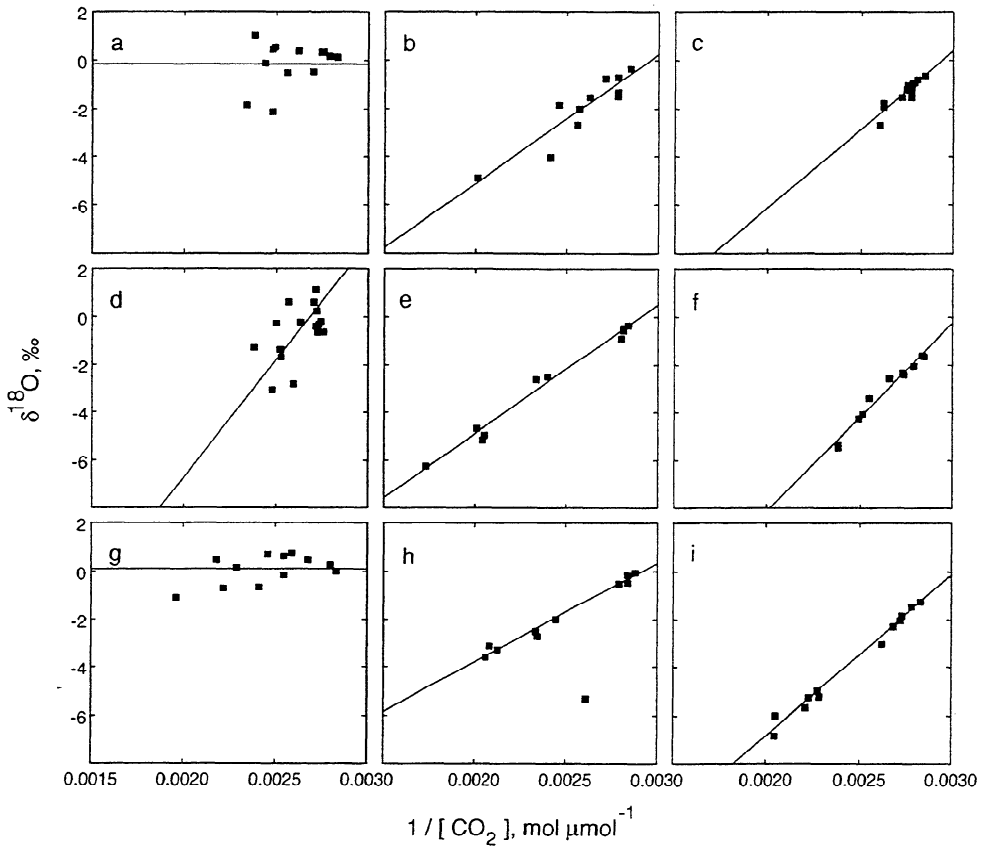

Figure 11. Comparison of the relationships between $1/CO_2$ concentration and the oxygen isotope ratio of CO_2 in air samples collected at night in the BOREAS northern study area during 1994. The black spruce site during (a) spring, (b) summer, and (c) fall sampling periods. The jack pine site during (d) spring, (e) summer, and (f) fall sampling periods. The aspen site during (g) spring, (h) summer, and (i) fall sampling periods.

strong influence of terrestrial respiration was shown because of its effect on the shape of the latitudinal change in the $\delta^{18}O$ of atmospheric CO_2 and on the lag between seasonal changes in CO_2 concentration and $\delta^{18}O$ observed at northern locations. *Ciais et al.* [1996a, b] concluded that measurements of the $\delta^{18}O$ of atmospheric CO_2 could be used to provide information on the large-scale distribution of gross CO_2 fluxes, although they noted that their model could not yet be used for quantitative flux estimates because they did not examine the sensitivity of their calculations to model parameters. In order to adequately interpret global scale differences in ecosystem net CO_2 exchange it is necessary to have knowledge of the source of variation in the gross CO_2 fluxes. This is because photosynthesis and respiration respond differently to important environmental changes such as temperature and CO_2 concentration. *Farquhar et al.* [1993] previously suggested that measurements of the $\delta^{18}O$ of atmospheric CO_2 could be used in large scale studies of the partitioning of anthropogenic emissions between the ocean and the terrestrial biosphere. Some simple mass balance calculations are shown below to help illustrate this claim.

The change in the oxygen isotope ratio of atmospheric CO_2 over time can be modeled as [*Farquhar et al.*, 1993; *Ciais et al.*, 1996a]:

$$\frac{d\delta_a}{dt} = \frac{1}{C_a}\left[F_{oa}(\delta_o - \delta_a) + \varepsilon_w (F_{ao} - F_{oa}) + R(\delta_s - \delta_a - \varepsilon_s) + A\Delta_A + F_{an}(\delta_{an} - \delta_o)\right] \quad (8)$$

where C_a is the total pool of atmospheric carbon (739 Gt), F_{oa} is the gross flux of CO_2 from the ocean to the atmosphere (90 Gt yr^{-1}), δ_o is the isotope ratio of CO_2 released from the ocean (1.75‰), δ_a is the isotope ratio of atmospheric CO_2, ε_w is the fractionation during diffusion and hydration of $C^{18}O^{16}O$ in water (0.8‰), F_{ao} is the gross flux of CO_2 from the atmosphere to the ocean (91.69 Gt yr^{-1}), R is the total terrestrial ecosystem respiration (105 Gt yr^{-1}), δ_s is the isotope ratio of CO_2 in isotopic equilbrium with soil water, ε_s is the fractionation during diffusion of $C^{18}O^{16}O$ through the soil to the atmosphere (-3.29‰), A is CO_2 assimilation by vegetation (equal to gross primary productivity (GPP) at the regional or global level; 106.69 Gt yr^{-1}), Δ_A is discrimination against $C^{18}O^{16}O$ during photosynthetic gas exchange (7.22‰), F_{an} is the flux of CO_2 released by anthropogenic emissions (7.7 Gt yr^{-1}), and δ_{an} is the isotope ratio of anthropogenic emissions (-17‰). The above parameters for ocean and terrestrial fluxes

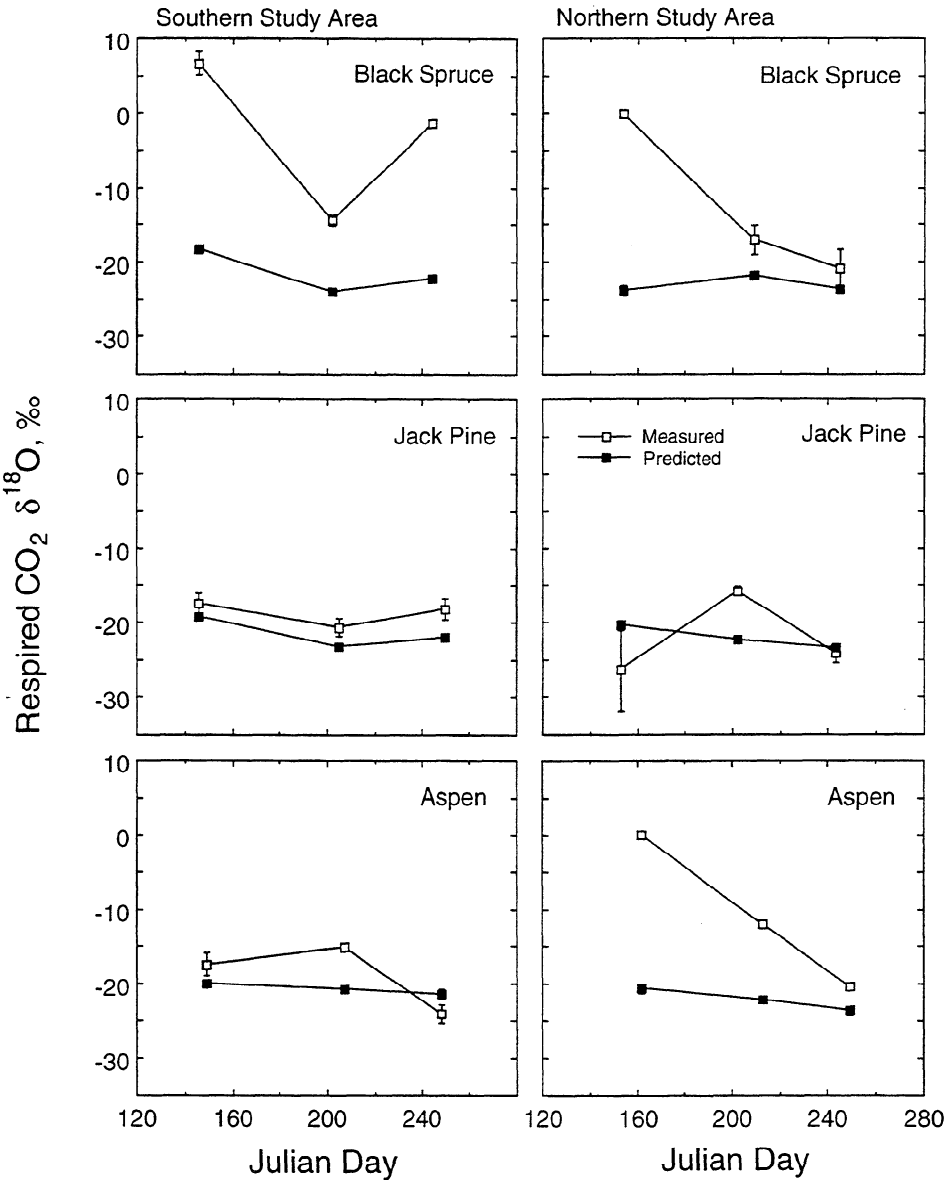

Figure 12. Relationship between measured (δ_R) and predicted ($\delta_{R\text{-}Soil}$) values for the oxygen isotope ratio of respired CO_2 at BOREAS study sites during 1994. The δ_R values were determined from the y intercept from the relationships shown in Figures 10 and 11. The $\delta_{R\text{-}Soil}$ values were calculated using equation (7).

assume that 22% of the anthropogenic emissions released to the atmosphere are taken up by terrestrial photosynthesis (1.69 Gt yr^{-1}), 22% is taken up by the ocean (1.69 Gt yr^{-1}), and 56% remains in the atmosphere. By ignoring the anthropogenic emissions, F_{ao} is reduced to 90 Gt yr^{-1}, A is reduced to 105 Gt yr^{-1}, and the calculated $d\delta_a/dt$ is -0.006‰ yr^{-1}, using the parameter values listed above. The values listed above for Δ_A, δ_s, are global average estimates calculated by *Ciais et al.* [1996a]. The only fractionation factor not well characterized is ε_s [*Farquhar et al.*, 1993; *Ciais et al.*, 1996a]. The value for ε_s has been adjusted to give essentially no net change in $d\delta_a/dt$ in the absence of anthropogenic emissions [*Ciais et al.*, 1996a]. This value (-3.29‰) for ε_s is lower than that expected for pure molecular diffusion (-8.8‰) but is not unrealistic given the uncertainty about diffusion distances from the site of CO_2 production in the soil and the possibility of incomplete isotopic equilibrium between CO_2 and soil water, as was discussed above.

Table 1 lists the calculated change in $d\delta_a/dt$ for a variety of scenarios for changes in photosynthesis, respiration, and ocean CO_2 fluxes. The magnitude of the change in $d\delta_a/dt$ is similar to the changes that occur in the $\delta^{13}C$ values of atmospheric CO_2 for equivalent changes in surface fluxes [*Francey et al.*, 1995]. In addition, alterations in the terrestrial CO_2 fluxes have a stronger effect on $d\delta_a/dt$ than do the equivalent changes in oceanic CO_2 fluxes, thus supporting the

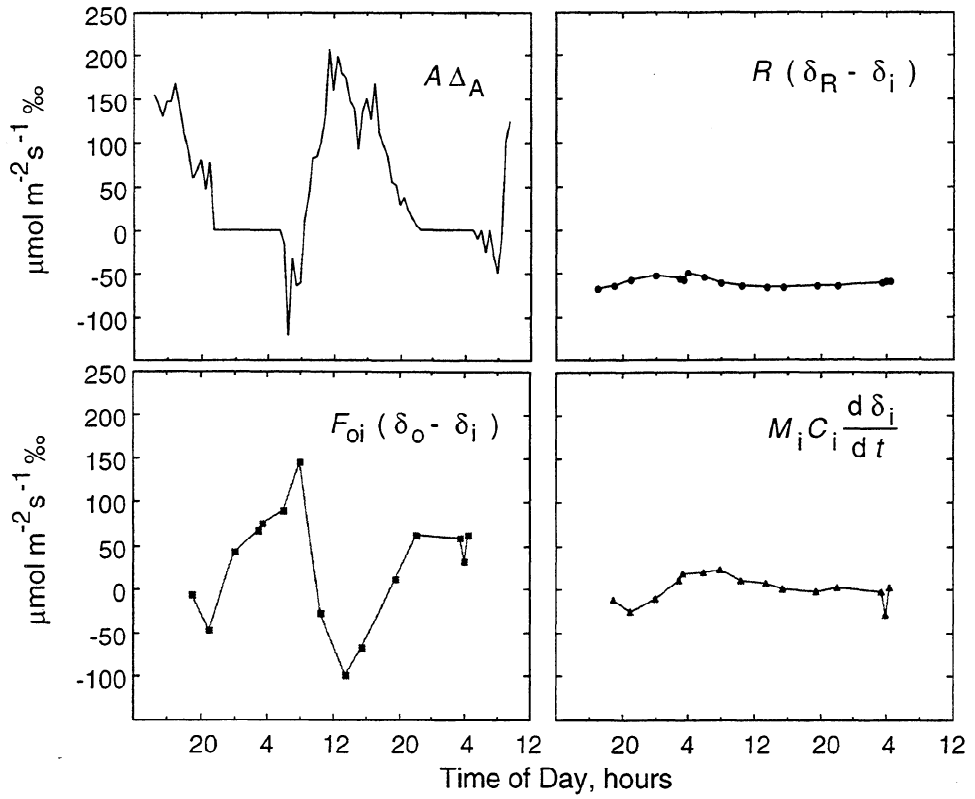

Figure 13. Comparison of the relative influence of photosynthesis, respiration, and turbulent exchange on changes in the oxygen isotope ratio of atmospheric CO_2 within a black spruce canopy during July 1994 in northern Canada (see equation (1)).

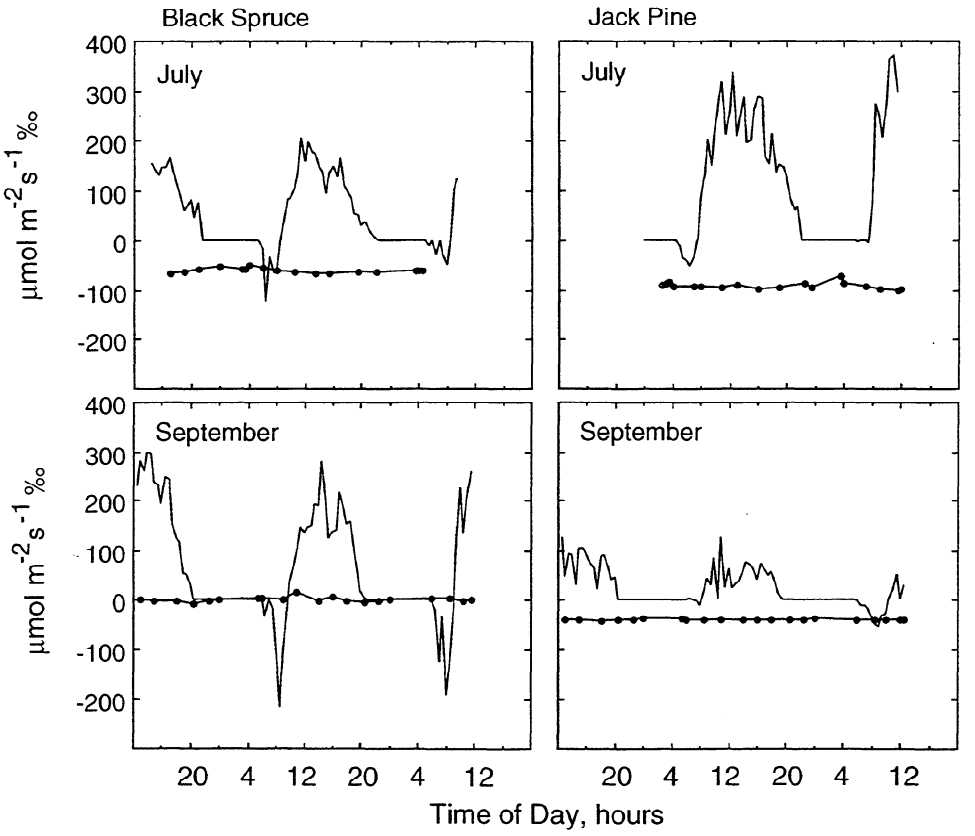

Figure 14. Seasonal changes in the influence of photosynthesis and respiration on the oxygen isotope ratio of atmospheric CO_2 within boreal forest canopies during July 1994 in northern Canada (see equation (1)). The solid line shows the $A\Delta_A$ values. The solid line with dots shows the R (δ_R - δ_i) values.

Table 1. Effect of Changes in Surface CO_2 Fluxes on the Calculated Rate of Change in the Oxygen Isotope Ratio of Atmospheric CO_2

CO_2 Flux, Gt yr^{-1}			
F_{ao}	A	R	$d\delta_a/dt$, ‰ yr^{-1}
91.69	106.69	105	-0.185
91.69	110.69	105	-0.146
91.69	106.69	101	-0.138
87.69	106.69	105	-0.189
95.69	106.69	105	-0.180
90.69	108.69	105	-0.166

Calculations were done using equation (8). Values for all other parameters in equation (8) are listed in the text.

idea that it should be possible to separate oceanic and terrestrial fluxes by monitoring changes in the oxygen isotope ratio of atmospheric CO_2. Carbon stable isotope techniques have contributed greatly to global studies of CO_2 partitioning between the ocean and the terrestrial biosphere [*Ciais et al.*, 1995; *Francey et al.*, 1995]. It will likely be possible in the near future to conduct similar global CO_2 partitioning studies using the ^{18}O composition of atmospheric CO_2 (P. Ciais, personal communication, 1996). Since processes controlling the ^{18}O composition of atmospheric CO_2 are completely independent from those affecting ^{13}C, the ^{18}O data could provide important checks on the results of ^{13}C studies.

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