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The relationship between $^{18}O/^{16}O$ and $^{13}C/^{12}C$ ratios of ambient CO_2 in two Amazonian tropical forests

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ABSTRACT

Oxygen and carbon isotopic compositions of ambient carbon dioxide in two tropical forests in the Amazonian basin were determined at the beginning and the end of the dry season. One site, Fazenda Vitoria, is typical of a seasonal forest with a pronounced dry season from May to November whereas the other site, Reserva Ducke, has a milder dry season. Samples collected at night revealed that in the beginning of the dry season, when the forest is still relatively wet, isotopic exchange between carbon dioxide and wet surfaces or fog is possible. Oxygen isotope ratios of ambient carbon dioxide collected during the day were correlated with the carbon isotope ratios. These correlations are representative of the mixing line between tropospheric and respired CO_2. The mixing lines were used to extrapolate the $\delta^{18}O$ values of soil-respired CO_2. These values were compared with those derived from the isotopic composition of bulk soil water (as reflected by plant stem water) and a diffusion isotopic effect of 8.8‰. It was observed that $\delta^{18}O$ values of respired CO_2 derived by extrapolation were greater than those calculated by using $\delta^{18}O$ values of bulk soil water and the associated diffusion isotope effect. The results show that in tropical forests, assumptions used in previous studies of temperate regions to derive the oxygen isotopic composition of respired CO_2 do not hold.

1. Introduction

The increase in atmospheric CO_2 over the last century has precipitated interest in the carbon budget of the biosphere. It has been particularly important to identify the location of terrestrial sinks, since recent studies have concluded that terrestrial photosynthesis is a substantial CO_2 sink (Dixon et al., 1994; Tans, 1993). There is evidence that the land masses of the southern hemisphere comprise one of the most important CO_2 global sinks (Dixon et al., 1994). This assessment has been supported recently by eddy correlation tower based measurements in an Amazonian forest which indicate a net carbon uptake (Grace et al., 1995).

Isotopic analysis of atmospheric CO_2 is an important tool for the determination of global CO_2 sources and sinks. Pioneering work by Keeling et al. (1979) utilizing carbon isotope ratios of atmospheric CO_2 revealed that the increase in atmospheric CO_2 is due to fossil fuel burning. Other studies by Tans et al. (1990) led to the conclusion that there is a large terrestrial carbon sink. More recently the oxygen isotopic signature

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of atmospheric CO_2 has emerged as a means for assessing the global carbon budget (Francey and Tans, 1987; Farquhar et al., 1993; Friedli et al., 1987). This method takes advantage of the rapid equilibration between ambient CO_2 and leaf water during photosynthesis. The CO_2 absorbed by the leaf equilibrates with leaf water and "leaks" back out. Photosynthesis, therefore, will leave an isotopic signature on ambient CO_2.

The relationship between the $\delta^{18}O$ of leaf water, atmospheric CO_2, and the internal CO_2 concentration in the chloroplast versus the discrimination against ^{18}O of CO_2 during photosynthesis has been established by the following equation (Farquhar and Lloyd, 1993):

$$\Delta_a = \bar{a} + [(C_c/(C_a - C_c))(\delta_c - \delta_a)], \qquad (1)$$

where $\bar{a}$ is the average fractionation during diffusion of CO_2 through the stomatal pores ($\sim 7.4^\circ/_{\infty}$); C_c and C_a are the CO_2 concentrations in the chloroplast and atmosphere respectively, δ_c and δ_a are the oxygen isotopic composition of CO_2 in chloroplast and atmosphere respectively. The discrimination factor (Δ_a) is highly variable and depends on the biome (Farquhar et al., 1993). Discrimination values may range from $-20^\circ/_{\infty}$ in arctic tundra to $+32^\circ/_{\infty}$ in the dry steppes of Kazakhstan and the Ukraine (Farquhar et al., 1993). In conjunction with $\delta^{13}C$ analysis, $\delta^{18}O$ analysis of tropospheric CO_2 may provide an accurate map of terrestrial CO_2 sinks (Farquhar et al., 1993).

For a greater understanding of terrestrial processes affecting the $\delta^{18}O$ values of global CO_2, studies on the interaction of respiration, photosynthesis, and atmospheric mixing on the isotopic signatures of ambient CO_2 in regional ecosystems will be necessary (Farquhar et al., 1993). The authors of this study are aware of three prior studies related to the above mentioned research direction (Flanagan and Varney, 1995; Yakir and Wang, 1996; Buchmann et al., 1997). The first two studies were conducted in temperate ecosystems. Both made 4 assumptions: (1) the $\delta^{18}O$ values of plant stem water reflects the $\delta^{18}O$ values of bulk soil water; (2) respired CO_2 equilibrates with bulk soil water before being released to the atmosphere; (3) there is a diffusion isotope discrimination factor of $8.8^\circ/_{\infty}$ as equilibrated CO_2 diffuses from the soil to the atmosphere, (therefore the $\delta^{18}O$ value of respired CO_2 was calculated by subtracting a diffusion isotope discrimination factor from the isotopic value of the CO_2 equilibrated with plant stem water); (4) the exchange between CO_2 and wet surfaces or clouds and/or fog is negligible. The first three assumptions were based in part on the observations of Hesterberg and Siegenthaler (1991). The validity of their equations, however, have recently been questioned (Tans, 1998).

To date, there are no studies of tropical ecosystems in which the $\delta^{18}O$ values of ambient CO_2 were related to soil water, photosynthetic discrimination, and tropospheric CO_2. Tropical regions may present conditions in which the aforementioned assumptions will not hold. First, the large amount of CO_2 produced by shallow roots and litter on the soil surface may not be subjected to the full expression of the diffusion isotope fractionation factor of $8.8^\circ/_{\infty}$ (Hesterberg and Siegenthaler, 1991) and may contribute a disproportionate amount of respired CO_2 equilibrated with surface water. This surface water would certainly be more isotopically enriched than bulk soil water. Second, high relative humidity and temperature changes may result in a considerable amount of condensation and wet surfaces where ambient CO_2 can exchange.

The purpose of this study was: (1) to compare measured Δ_a values in the Amazon basin with those calculated using the Farquhar et al. model (1993) and (2) to test whether the assumptions put forth in the studies cited (Flanagan and Varney, 1995; Yakir and Wang, 1996) hold for tropical ecosystems.

2. Approach

Values for Δ_a were determined by measuring the oxygen isotope ratios of leaf water and ambient CO_2. Some assumptions about the relationship between chloroplast water and leaf water and about the CO_2 concentration in the chloroplast were necessary in order to calculate the approximate Δ_a for the two locations in the Amazon basin. These assumptions will be discussed in the results.

To test the assumption that CO_2 will not equilibrate with wet surfaces or water droplets, and that the $\delta^{18}O$ of respired CO_2 can be determined by subtracting a diffusion isotopic fractionation factor from the $\delta^{18}O$ values of CO_2 equilibrated with bulk soil water, a graphical approach similar to the one utilized by Bottinga and Craig (1969) in their interpretation of $\delta^{18}O$ values of marine CO_2 was used. There are two major sources of CO_2 in a forest: respired and tropospheric (Fig. 1). These sources will have different carbon and oxygen isotopic composition, with tropospheric CO_2 having higher $\delta^{13}C$ and $\delta^{18}O$ values than those of respired CO_2. Estimation of the $\delta^{18}O$ value of respired CO_2 in other studies were based on the previously mentioned assumptions.

If the effect of photosynthesis or ambient CO_2 equilibration with water droplets or wet surfaces is negligible, then a plot of $\delta^{18}O$ versus $\delta^{13}C$ values of ambient CO_2 should fall along the mixing line shown in Fig. 1. Photosynthesis has the potential of causing a change in isotopic ratios of ambient CO_2. Given a high enough photosynthetic rate, a plot of $\delta^{18}O$ versus $\delta^{13}C$ values of ambient CO_2 can deviate from this mixing line (Fig. 1). The extent of the deviation depends on two factors. First, if the ratio of the oxygen (Δ_a) and carbon (Δ) isotope discrimination factors (Δ_a/Δ) is significantly different from the slope of the mixing line, then photosynthesis can cause a significant deviation from the mixing line. Second, photosynthetic signals are greater if the amount of turbulent mixing between ambient forest CO_2 and the troposphere is low (Sternberg et al., 1997). Thus lower turbulence will cause a greater potential for deviation from the mixing line.

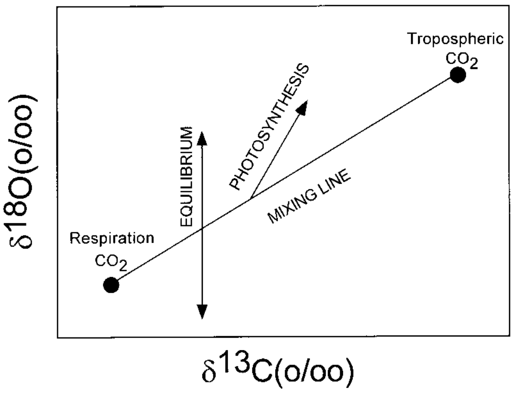

Fig. 1. Hypothetical relationships between $\delta^{18}O$ and $\delta^{13}C$ values of ambient CO_2 in forests. The mixing line is a simple straight line intersecting the values of the two sources of the ambient CO_2 mixture: respired and tropospheric CO_2. Deviations from this mixing line can be caused by photosynthesis and by equilibration between ambient CO_2 and moist surfaces or water droplets.

Another factor which causes a deviation from this mixing line is the equilibration of ambient CO_2 with wet surfaces or water droplets from condensation. This deviation is controlled by two factors: (1) the difference between the $\delta^{18}O$ of the CO_2 equilibrated with the water surfaces or droplets relative to expected values from the mixing line and (2) the rate of turbulent mixing relative to the equilibration rate.

In this paper, we will show that in the tropics the observed relationships between $\delta^{18}O$ and ^{13}C values of ambient CO_2 can deviate from theoretical mixing lines constructed on the basis of the aforementioned assumptions. Further, we will show that this deviation is caused by CO_2 equilibrating with wet surfaces. We will also demonstrate that the assumptions used to estimate the $\delta^{18}O$ values of respired CO_2 in temperate studies do not hold for tropical forests.

3. Methods

3.1. Sites of study

Air samples were collected in two sites: Fazenda Vitoria in the state of Pará (2°59′S, 47°31′W) and Reserva Ducke in the state of Amazonas (3°8′S, 60°1′W). These two sites have contrasting climates. The Fazenda Vitoria site is typical of the dryer and more seasonal climate found in the eastern Amazon basin and the Reserva Ducke site is typical of the wetter and more aseasonal climate of the central and western Amazon.

The forest structure at these two sites differ in part because of climatic differences and anthropogenic effects. The Fazenda Vitoria site is located in an area of extensive deforestation; thus, it is a somewhat smaller forest parcel than that of the Reserva Ducke. Higher wind speeds at the Fazenda Vitoria site coupled with a smaller forest parcel cause erasure of any isotopic signals associated with photosynthesis (Sternberg et al., 1997). Other characteristics of these sites are described in Moreira et al. (1997) and Sternberg et al. (1997).

3.2. Collection of samples

3.2.1. Air samples. Air samples were collected on two dates at each site: at the beginning of the dry season on 1 and 7 July at the Reserva Ducke and Fazenda Vitoria site, respectively, and at the end of the dry season on 7 and 15 December at the Reserva Ducke and Fazenda Vitoria respectively. Samples were collected at approximately 0 and 6 hours (night samples) and at 10, 14, and 17 hours (day samples). Ambient air samples were collected in 500 cm^3 pre-heated and pre-evacuated flasks similar to previous studies (Moreira et al., 1997; Sternberg et al., 1997).

In order to desiccate the sample before collection, a magnesium perchlorate trap (2 cm diameter by 20 cm length) was placed between the collection flask and the manifold (Flanagan and Varney, 1995; Buchmann et al., 1997). This trap was evacuated before sample collection. Although the trap did not completely eliminate moisture, it was sufficient to prevent any condensation on the vessel walls during transport from the field to the laboratory. Samples were collected at 0.5, 18, 45 and ~900 m elevation at the Reserva Ducke site and collected at 0.5, 18 and ~300 m elevations at the Fazenda Vitoria site in the manner described in Sternberg et al. (1997).

3.2.2. Stem, leaf and soil samples. Within one to two days of air sample collection, stem, leaf and soil samples were also collected. Soil samples were collected only at the Fazenda Vitoria site. Stem and leaf samples were collected at the approximate height of 18m after 10:00 AM when plants are actively transpiring. Soil samples were collected at the surface and at 20 cm intervals below the surface down to 2 m. Sections of stems (approximately 1 cm diameter and 15 cm length), leaves and soil samples were placed in glass tubes and sealed with parafilm. Water was distilled from these samples as described in Moreira et al. (1997).

3.2.3. Isotopic analysis of samples. Oxygen isotopic composition of water extracted from leaves and stems was analyzed by the CO$_2$ equilibration method as described by Epstein and Mayeda (1953).

Air samples were taken to the University of Miami. CO$_2$ from air samples was then cryogenically distilled and sealed in 6 mm by 15 cm Vycor ampoules containing 0.3 g of copper and baked at 450°C for 1 h to remove nitrous oxides. Vycor ampoules were used because oxygen from CO$_2$ can exchange with Pyrex at high temperatures, but does not exchange with Vycor (Burk, 1979).

All extracted gas samples were analyzed in a PRISM mass spectrometer with a precision of $\pm 0.1°/_{\infty}$. δ^{18}O values are expressed either on the SMOW scale for isotopic composition of plant water and soil water or on the PDB–CO$_2$ scale for ambient CO$_2$.

4. Results

Average daytime oxygen isotope ratios of ambient CO$_2$ at 18 m elevation, of CO$_2$ at equilibrium with chloroplast water, the ratio C_c/C_a, and the oxygen isotope discrimination factor (Δ_a) for each site are shown in Table 1. δ^{18}O values of the chloroplast water were derived by measuring δ^{18}O values of leaf water and using a mass balance equation. The equation assumes that 20% of the water in the leaf has the isotopic identity of stem water and 80% (which includes the chloroplast water) has the identity of water undergoing evaporation (Leaney et al., 1985).

The issue of δ^{18}O values of chloroplast water is controversial and still unresolved. On the one hand Farquhar et al. (1993) have evidence that chloroplast water has the isotopic identity of evaporative water; on the other hand Yakir (1992) has evidence that chloroplast water has the isotopic identity of a mixture of non-evaporated and evaporated water. For the purpose of our study, it was assumed that chloroplast water has the isotopic signature of water undergoing

Table 1. *$\delta^{18}O$ values $\pm$ SEM (CO_2-PDB scale) of ambient CO_2 (δ_a) and CO_2 equilibrated with chloroplast water (δ_c), ratio of CO_2* concentration at the chloroplast relative to atmospheric concentration (C_c/C_a), and the oxygen isotopic discrimination against oxygen-18 in ambient carbon dioxide during photosynthesis (Δ_a)at the two sites

Site and date	δ_a	δ_c	C_c/C_a	Δ_a
Ducke 7/96	-0.9 ± 0.2	-0.8 ± 0.3	0.53	7.5
Vitoria 7/96	-0.6 ± 0.4	$+9.4 \pm 1.0$	0.53	18.7
Ducke 12/96	-2.7 ± 0.4	$+1.8 \pm 0.3$	0.53	12.5
Vitoria 12/96	-2.0 ± 0.3	$+11.9 \pm 0.6$	0.53	23.1

evaporation. If our assumption were incorrect, Δ_a values would be less than those shown in Table 1.

Values of C_c/C_a were calculated by first calculating the ratio of CO_2 concentration in the substomatal cavity (C_{st}) to that of ambient (C_a) using the following equation (Farquhar et al., 1982):

$$\delta_p = \delta_{amb} - a + (a - b)C_{st}/C_a\,, \qquad (2)$$

where δ_p and δ_{amb} are the $\delta^{13}C$ values of leaf biomass and ambient CO_2, respectively. The constants a and b are the values of discrimination associated with stomatal diffusion (4.4‰) and the carboxylation reactions (29‰), respectively. The average $\delta^{13}C$ value of −28‰ and −8‰ for leaf biomass and ambient CO_2 at 18 m elevation, respectively was used in the above equation (Merwe and Medina, 1989, Sternberg et al., 1997, Lloyd et al., 1996). The concentration of ambient CO_2 at 18 m elevation during the day was measured to be approximately 340 ppm (Buchmann et al., 1997, Sternberg et al.,1997). C_c/C_a was then calculated by using the relationship $(C_{st} - C_c)/C_a = 0.1$ (Farquhar et al., 1993).

Photosynthetic discrimination against ^{18}O of ambient CO_2 is lower at the beginning of the dry season (7.5 and 18.7‰ for the Ducke and Vitoria site, respectively) compared to the end of the dry season (12.5 and 23.1‰ for the Ducke and Vitoria site, respectively).

The $\delta^{13}C$ and $\delta^{18}O$ values of the endpoints for the mixing line between respired and tropospheric CO_2 calculated according to the assumptions to be tested here are shown in Table 2. According to these assumptions $\delta^{18}O$ values of respired CO_2 were derived by calculating the $\delta^{18}O$ values of CO_2 equilibrated with bulk soil water (as reflected in plant water) and subtracting a diffusion isotopic effect of 8.8‰ (Flanagan and Varney, 1995; Yakir and Wang, 1996). Values of plant water were fairly constant at the beginning and end of dry season ranging from −3.6 to −4.4‰. Consequently the calculated values of respired CO_2 did not vary much between sampling dates and sites. The $\delta^{18}O$ values of respired CO_2 ranged from −13.0 to −12.2‰. $\delta^{18}O$ and $\delta^{13}C$ values of high altitude CO_2 samples are shown in the fourth and fifth column on Table 2. $\delta^{18}O$ and $\delta^{13}C$ values of tropospheric CO_2 tended to be less negative in the samples taken during the beginning of the dry season compared to those taken at the end of the dry season.

The relationship between $\delta^{18}O$ versus $\delta^{13}C$ values of air samples collected at the Reserva Ducke and Fazenda Vitoria site are shown in Fig. 2, with samples collected at night represented by empty circles and those collected during the day represented by full circles. The theoretical mixing line is also shown. This line was drawn by connecting points representing the hypothetical $\delta^{18}O$ and $\delta^{13}C$ values of respired CO_2 with those representing the $\delta^{18}O$ and $\delta^{13}C$ values of tropospheric CO_2 (as shown in Table 2). The $\delta^{13}C$ value of respired CO_2 of −27‰ as measured by several investigators was used (Buchmann et al., 1997; Sternberg et al.,1997; Quay et al., 1989; Lloyd et al., 1996).

There was a significant correlation between $\delta^{18}O$ and $\delta^{13}C$ values for samples collected during the day for both sites and dates of collection. The slope, intercept, r^2, and P values for the fitted relationships are shown in Table 3. We used a model I regression analysis since the parameters

Table 2. *Isotope ratios ± SEM of the two end points of the mixing line between respired CO_2 and tropospheric CO_2; table shows $\delta^{18}O$ values of plant stem water, of CO_2 at equilibrium with plant stem water at 25°C discriminated by a diffusion isotope fractionation factor of 8.8‰ (theoretical value of respired CO_2), of CO_2 from high altitude air samples, and $\delta^{13}C$ values of CO_2 from high altitude air samples*

Site and date	$\delta^{18}O$* of plant water	$\delta^{18}O^{\#}$ of resp. CO_2 theoretical	$\delta^{18}O^{\#}$ of CO_2 high altitude	$\delta^{13}C^{\#}$ of CO_2 high altitude
Ducke 7/96	−3.6 ± 0.3	−12.2 ± 0.3	−0.3 ± 0.4	−8.1 ± 0.2
Vitoria 7/96	−3.7 ± 0.7	−12.3 ± 0.3	−1.2 ± 0.3	−8.0 ± 0.2
Ducke 12/96	−4.4 ± 0.2	−13.0 ± 0.2	−3.4 ± 0.2	−9.0 ± 0.1
Vitoria 12/96	−3.9 ± 0.3	−12.5 ± 0.3	−2.2 ± 0.9	−8.8 ± 0.4

* $\delta^{18}O$ values given relative to SMOW.

$\delta^{18}O$ and $\delta^{13}C$ values given relative to PDB-CO_2.

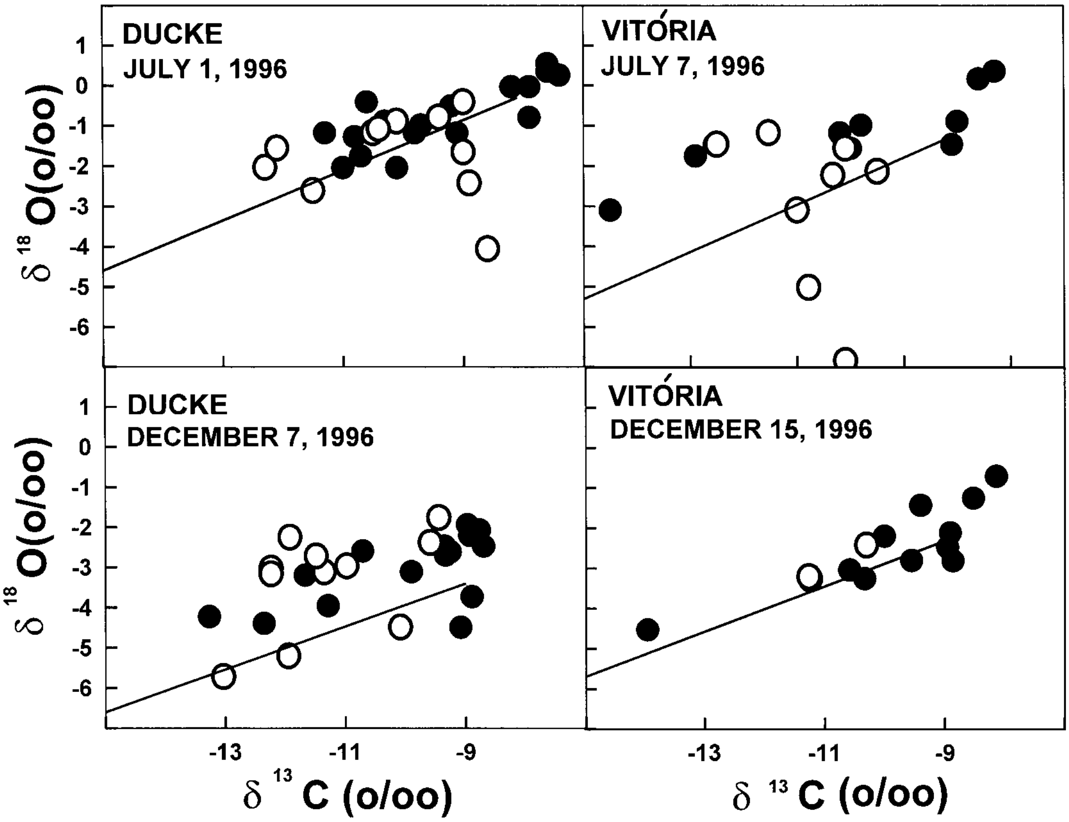

Fig. 2. Observed relationship between $\delta^{18}O$ and $\delta^{13}C$ of ambient CO_2 for samples collected at Reserva Ducke and Fazenda Vitoria at the beginning (July, 1996) and end (December, 1996) of the dry season. Full circles represent samples collected during the day and empty circles represent samples collected during the night. The figure also shows the mixing lines given the theoretical isotopic values of respired and observed isotopic values of tropospheric CO_2.

Table 3. *Regression equations expressing the relationship between $\delta^{18}O$ and $\delta^{13}C$ values of ambient forest CO_2 sampled during the day; table shows the slope, intercept, correlation coefficient, and the probability for the regression equations for the two sites sampled in this study; also shown are the estimated $\delta^{18}O$ values of respired and tropospheric CO_2 from the regression equation assuming the $\delta^{13}C$ value of respired and tropospheric CO_2 is $-27°/_{oo}$ and $-7.8°/_{oo}$, respectively; all $\delta^{18}O$ values are given relative to PDB-CO_2.*

Site and date	Sl.	Int.	r^2	P	$\delta^{18}O$ of resp. CO_2	$\delta^{18}O$ of trop. CO_2
Ducke 7/96	0.48	3.7	0.64	<0.01	-9.3	0.0
Vitoria 7/96	0.33	2.2	0.71	<0.01	-6.7	-0.4
Ducke 12/96	0.37	0.7	0.38	<0.05	-9.3	-2.2
Vitoria 12/96	0.56	3.0	0.72	<0.01	-12.1	-1.4

for the regression equation were used for the purposes of prediction (Sokal and Rohlf, 1981). Using the respective regression equations and the assumption that respired and tropospheric CO_2 have $\delta^{13}C$ values of $-27°/_{\infty}$ and $-7.8°/_{\infty}$ respectively, we extrapolated the regression equations to $\delta^{18}O$ values of respired and tropospheric CO_2. These values are shown in the sixth and seventh column on Table 3.

$\delta^{18}O$ values of the water in the soil profile at the Fazenda Vitoria site for both sampling dates are shown in Fig. 3. $\delta^{18}O$ values of water from surface soil samples differed by only $0.5°/_{\infty}$ between the two sampling dates ($-0.9°/_{\infty}$ for July and $-1.4°/_{\infty}$ for December). $\delta^{18}O$ values of water from the soil profile between surface and 1.2m depth ranged from $-10.0°/_{\infty}$ to $-5.1°/_{\infty}$ for the July samples and between $-9.6°/_{\infty}$ and $-5.2°/_{\infty}$ for the December samples. Therefore, differences in isotopic composition of soil water between the two sampling dates were small for soil samples collected on the above depth interval. There were greater differences between the $\delta^{18}O$ values of soil water collected at the two dates at depths below 1 m, with the July samples having an average $\delta^{18}O$ value of -9.9 ± 1.7 (SDEV) and the December samples having an average $\delta^{18}O$ value of -6.9 ± 0.95.

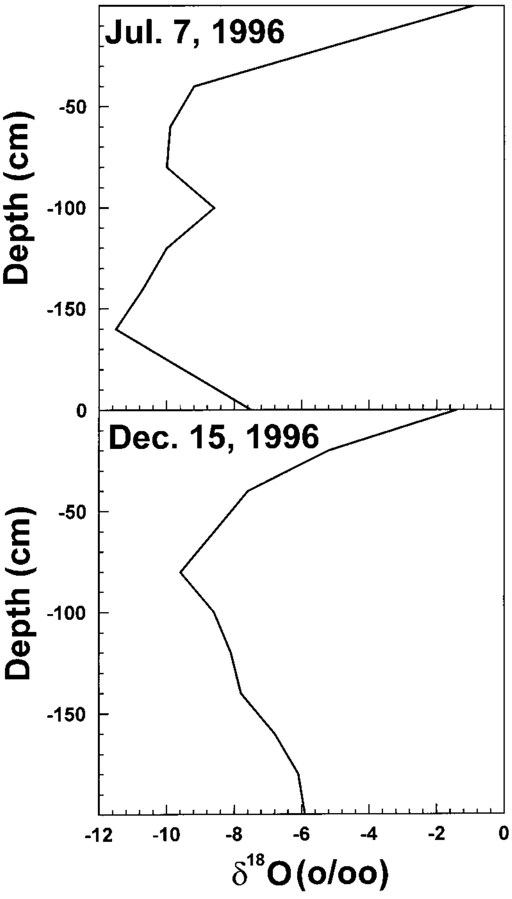

Fig. 3. $\delta^{18}O$ values of water extracted from soil samples throughout the soil profile at Fazenda Vitoria in 7 July and 15 December 1996.

5. Discussion

5.1. Calculation of the photosynthetic discrimination factor

The discrimination factors (Δ_a) for the Reserva Ducke site (7.5 and $12.5°/_{\infty}$, Table 1) are low compared to the one estimated by Farquhar et al. (1993) for the Amazon region ($18–20°/_{\infty}$). Note that Figs. 2 and 3 are reversed in the paper published by Farquhar et al. (1993). The discrimination factor for Fazenda Vitoria during the beginning of the dry season ($18.7°/_{\infty}$) is similar to that estimated by Farquhar et al. (1993) but is higher at the end of the dry season ($23.1°/_{\infty}$).

Although we were not able to calculate all parameters for the model reported in Farquhar et al. (1993), we note that the equation used to calculate soil water (equation 3 in their paper) gave $\delta^{18}O$ values of soil water in the Amazon region of $+12.3°/_{\infty}$ (we used the following parameters for this region: temperature $T(°C) = 25°C$; precipitation $Pa(m) = 2.0m$; and elevation $Ev(m) = 30m$). This value is highly enriched compared with our measurements of -4 to $-3°/_{\infty}$ for plant water and $-5.0°/_{\infty}$ for water from soil samples below the surface. We suspect, however, that another error in the model lies in the estimates of leaf water. Leaf water had low enrichment in the Reserva Ducke site whereas at the Fazenda Vitoria site, leaf water may have been more enriched than predicted by the model. Furthermore, the circulation model used to predict $\delta^{18}O$ values of ambient vapor (Jouzel et al., 1987) underestimates $\delta^{18}O$ values of ambient vapor. Our measurements (Moreira, M. Z., personal communication) at the end of October and beginning of November gave δ^{18}O values of ambient vapor averaging −10 to −9‰. The Jouzel et al. model predicts values ranging from about −19 to −14‰.

5.2. *Deviation between δ^{18}O versus δ^{13}C values of ambient CO$_2$ and the theoretical mixing lines*

A plot of δ^{18}O versus δ^{13}C values of daytime ambient CO$_2$ deviates considerably from the theoretical mixing lines for the Reserva Ducke samples (Fig. 2). Values for the Fazenda Vitoria site samples fall above the theoretical mixing line at the beginning of the dry season and along the theoretical mixing line at the end of the dry season.

At the beginning of the dry season, when humidity is usually high, large deviations on the δ^{18}O values ambient night time CO$_2$ are noticeable without a concomitant variation on the δ^{13}C values (Fig. 2). This pattern is typical of CO$_2$ exchange with moist surfaces or water droplets (Fig. 1). Since this occurred at both sites only at night, it may represent exchange which occurs with water droplets during the condensation and formation of fog, often seen over tropical forests. It is interesting to note that the 1 July sampling occurred as a cold front ("friagem") moved in, bringing unusually low temperatures for the tropics (Minimum temperature on 1 July at the Reserva Ducke was 17°C). This could have led to condensation resulting in the equilibration of the ambient CO$_2$ with water droplets or wet surfaces.

Although evidence of equilibration between CO$_2$ and water droplets cannot account for global and tropospheric patterns of δ^{18}O values (Francey and Tans, 1987; Friedli et al., 1987), it may explain local and regional patterns. The assumption that there is no equilibration between ambient CO$_2$ and water droplets or wet surfaces cannot be applied to the tropics. Under certain conditions (particularly during the wet season) equilibration between CO$_2$ and water droplets or wet surfaces may be an important factor modulating δ^{18}O values of ambient CO$_2$.

There is a significant relationship between δ^{18}O and δ^{13}C of ambient CO$_2$ for gas collected during the day at both sites on the two sampling dates (Table 2). By extrapolating with the regression equations fitted to data collected at each site and time of sampling to δ^{13}C values of respired CO$_2$, the δ^{18}O values of respired CO$_2$ can be estimated (Table 2). δ^{18}O values of respired CO$_2$ were −9.3‰ for the Reserva Ducke site both at the beginning and end of the dry season (Table 3). These values are greater then the values of −12.2 to −13.0‰, which were derived with the assumptions: (1) that isotope ratios of plant water reflect that of the bulk soil; (2) respired CO$_2$ equilibrates with bulk soil water; (3) that the diffusion isotope fractionation factor of 8.8‰ is fully expressed. It would appear that in wet tropical forests at least one of these assumptions do not hold.

For the seasonal site, the δ^{18}O values of respired CO$_2$ varied. At the beginning of the dry season, the δ^{18}O value of respired CO$_2$ was extrapolated to be −6.7‰. At the end of the dry season, the δ^{18}O value of respired CO$_2$ was calculated to be −12.1‰, nearly the same as that expected in which the following conditions exist: plant water reflects bulk soil water, respired CO$_2$ equilibrates with bulk soil water, and the diffusion isotope fractionation factor is fully expressed (Table 2).

Given the discrepancy between our observed δ^{18}O values of respired CO$_2$ and those calculated using the previously mentioned assumptions, we can think of four possible explanations which could cause these discrepancies.

First, photosynthesis alters the oxygen and carbon isotopic composition of ambient CO$_2$ in such a way that its δ^{18}O and δ^{13}C values deviate from the theoretical mixing line. This possibility is highly unlikely. At the Reserva Ducke site the average ratio of oxygen to carbon isotopic discrimination is approximately 0.5. Thus any effect of photosynthesis on the relationship between δ^{18}O and δ^{13}C values of ambient CO$_2$ would hardly deviate from the theoretical mixing line which as an average slope of 0.57. For the Vitoria site, previous measurements indicate that turbulence at this site erases any photosynthetic signal (Sternberg et al., 1997).

Second, the δ^{18}O value of plant water does not reflect the δ^{18}O values of water with which CO$_2$ equilibrates during its residence time in the soil profile. According to our results, CO$_2$ equilibrates with water which is isotopically more enriched than that reflected by plant water. In a study of the global CO$_2$ budget (Farquhar et al., 1993) observed that on a global basis the average δ^{18}O value of the respired CO$_2$ pool was 1.2‰ higher than the value expected if the discrimination factor of 8.8‰ was fully expressed on the CO$_2$ equilibrated with bulk soil water. One of the factors to which this discrepancy was attributed was similar to this explanation, i.e., the soil water at the evaporation front is slightly more enriched than that of bulk soil water.

Third, the diffusion isotope fractionation factor is not fully expressed as CO_2 from respiration is liberated from the surface layer of the soil. This explanation follows from the observation that respiration in tropical forest soils can come from very shallow litter and root layers.

Fourth, there are other above ground sources of respired CO_2 in the forest liberating CO_2 which equilibrates with water having a higher $\delta^{18}O$ value then soil water.

Observations on the $\delta^{18}O$ values of water from the soil profile collected at the Fazenda Vitoria site during the sampling dates (Fig. 3) may help us determine which of the latter three explanations is more plausible. Recall that $\delta^{18}O$ values of respired CO_2 (−6.7‰) during the July sampling at the Fazenda Vitoria sites were higher than those predicted by previously used assumptions (−12.3‰, Table 3). To test the second explanation, we consider the extreme case in which all the respired CO_2 is equilibrated with soil surface water. This water is the most enriched water available in the soil profile for equilibration. CO_2 equilibrated with surface water and subjected to the diffusion isotopic fractionation factor of 8.8‰ would have a $\delta^{13}C$ value of −9.5‰. The value is still considerably lower than the observed $\delta^{18}O$ value of respired CO_2 (−6.7‰). If the discrimination factor is not expressed, then the $\delta^{18}O$ value of the equilibrated CO_2 would be −0.7‰ and substantially greater than that observed. This explanation cannot fully explain the discrepancy between observed and calculated $\delta^{18}O$ values of respired CO_2, since only small differences in the isotopic composition of water in the Fazenda Vitoria soil profile to 1m depth were observed between July and December, yet there are large differences between $\delta^{18}O$ values of respired CO_2 sample at these two dates (Table 3).

The fourth explanation can be eliminated by the observation that there are no differences in $\delta^{18}O$ values of rainfall between July and December (Victoria et al., 1991). Since December is the end of the dry season one would expect that any above ground water source available for CO_2 equilibration would have a higher $\delta^{18}O$ value in December compared to July. Using the above reasoning the $\delta^{18}O$ values of the respired CO_2 from a hypothetical above ground respired source would be higher in December compared to July. Yet the opposite is observed; $\delta^{18}O$ value of respired CO_2 in July is higher than that collected during December.

By the process of elimination we must conclude that a lower than 8.8‰ diffusion isotopic fractionation factor contributes to the discrepancy between calculated and observed $\delta^{18}O$ values of respired CO_2. This explanation is consistent with the phenomenon of litter decay and surface root respiration during periods when the soil surface is relatively wet (July samples). Because this source of CO_2 is so close to the surface the diffusion isotope fractionation factor may not be fully expressed. Therefore the $\delta^{18}O$ values of respired CO_2 in July are higher than expected. Whereas at the end of the dry season (December samples), litter decay and surface root respiration decreases, the source of respired CO_2 is from lower depths of the soil profile, and the diffusion isotopic fractionation factor is fully expressed. Therefore $\delta^{18}O$ values of respired CO_2 in December are as expected. This variation in the $\delta^{18}O$ values of respired CO_2 is not seen in the Ducke reserve since this site does not have a pronounced seasonality as the Fazenda Vitoria site.

6. Conclusion

Our observations indicate an agreement between measured discrimination against ^{18}O of CO_2 during photosynthesis in an eastern Amazonian site to that previously modeled in the Amazon basin. However, large discrepancies between measured and modeled ^{18}O discrimination was observed in a central Amazonian site. The reasons for these discrepancies could be miss-calculation of $\delta^{18}O$ values of leaf water and/or vapor by the previous model. The determination of the various factors responsible for tropical ecosystem's isotopic signature on the oxygen of CO_2 will be more complicated than temperate systems. Ambient CO_2 can equilibrate with wet surfaces and fog during nighttime condensation. Further, the isotopic identity of respired CO_2 cannot be calculated based on bulk soil water and a constant isotopic fractionation factor.

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