

# Increased photosynthetic capacity of *Scirpus olneyi* after 4 years of exposure to elevated CO<sub>2</sub>

W. J. ARP & B. G. DRAKE Smithsonian Environmental Research Center, P.O. Box 28, Edgewater, MD 21037, U.S.A.

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**Abstract.** While a short-term exposure to elevated atmospheric CO<sub>2</sub> induces a large increase in photosynthesis in many plants, long-term growth in elevated CO<sub>2</sub> often results in a smaller increase due to reduced photosynthetic capacity. In this study, it was shown that, for a wild C<sub>3</sub> species growing in its natural environment and exposed to elevated CO<sub>2</sub> for four growing seasons, the photosynthetic capacity has actually increased by 31%. An increase in photosynthetic capacity has been observed in other species growing in the field, which suggests that photosynthesis of certain field grown plants will continue to respond to elevated levels of atmospheric CO<sub>2</sub>.

**Key-words:** carbon dioxide; photosynthesis; photosynthetic acclimation.

## Introduction

Short-term exposure to an elevated atmospheric CO<sub>2</sub> concentration increases photosynthesis in most plants by raising the CO<sub>2</sub> concentration at the site of carboxylation, while transpiration is usually reduced by stomatal regulation. Long-term exposure to elevated CO<sub>2</sub> often leads to a reduction in the photosynthetic response to CO<sub>2</sub>. A literature survey by Cure & Acock (1986) showed that, while the average initial response to elevated CO<sub>2</sub> was a 52% increase in carbon exchange, the increase after acclimation to high CO<sub>2</sub> was only 29%. The most commonly given explanation for this acclimation effect is that elevated CO<sub>2</sub> causes an imbalance in the supply and demand of carbohydrates resulting in end product inhibition (Neales & Incoll, 1968; Herold, 1980; Ehret & Joliffe, 1985).

Almost all experiments showing photosynthetic acclimation have been performed on annuals or tree seedlings. While annuals are often sink limited in the vegetative growth stage, plants with large storage organs such as tubers, bulbs or rhizomes may not be sink limited at any time during the growing season. One experiment with potato showed no decrease in photosynthetic capacity (Sage, Sharkey & Seemann, 1989). No other photosynthetic acclimation experiments have been reported for plants with large storage organs. Because most studies were conducted on annuals, no data are

available on acclimation of plants growing in high CO<sub>2</sub> for more than one growing season.

In this study, we determined the photosynthetic capacity of rhizomatous C<sub>3</sub> sedge *Scirpus olneyi* after 4 years of exposure to elevated CO<sub>2</sub> concentration in the field. In many studies, the possible effect of stomatal acclimation to high CO<sub>2</sub> on photosynthesis is not taken into account, although it has been shown that this effect can be significant (Imai & Murata, 1978). To separate a possible stomatal effect on photosynthesis from photosynthetic acclimation, the photosynthesis (A) of elevated and ambient grown plants were compared at identical intercellular CO<sub>2</sub> concentrations (C<sub>i</sub>) by generating A/C<sub>i</sub> curves.

## Materials and methods

*Scirpus olneyi* plants growing in a brackish marsh on the Chesapeake Bay, Maryland, U.S.A., were exposed to elevated CO<sub>2</sub> (ambient CO<sub>2</sub> + 340 μmol CO<sub>2</sub> mol<sup>-1</sup> air) from April 1987 to the present using open top chambers. For a description of the system, see Drake *et al.* (1989). Leaf photosynthesis measurements were made during several afternoons in August 1990 using a mini cuvette control system (Bingham Interspace Co.), a Binos infra-red gas analyser (Leybold Heraeus, Hanau, Germany) and dew point hygrometers (General Eastern, Dew-10). The CO<sub>2</sub> and H<sub>2</sub>O concentrations were regulated by mixing CO<sub>2</sub>-free dry air, CO<sub>2</sub>-free humidified air and dry air with 1% CO<sub>2</sub>, using mass flow controllers. The system was controlled by a custom program on a portable computer. This program automatically performed A/C<sub>i</sub> curves while maintaining a constant vapour pressure deficit (vpd) and temperature of the air around the leaf. The vpd was maintained at 1.5 kPa ± 0.1, the temperature in the leaf chamber was 30°C ± 0.2, and photosynthetic photon flux was supplied by natural sunlight and was always above 1000 μmol m<sup>-2</sup>s<sup>-1</sup>.

The ambient CO<sub>2</sub> concentration was altered in eight to 10 steps between 0 and 1200 μmol mol<sup>-1</sup>. The direction of change of the CO<sub>2</sub> concentration had no effect on the results. Before and after each A/C<sub>i</sub> curve, measurements were made at the growth CO<sub>2</sub> concentration. Measurements of assimilation and stomatal conductance were made when all variables were steady (photosynthesis ± 0.2 μmol CO<sub>2</sub> m<sup>-2</sup>s<sup>-1</sup>, transpiration

$\pm 0.2 \text{ mmol H}_2\text{O m}^{-2}\text{s}^{-1}$ , and light  $\pm 25 \mu\text{mol m}^{-2}\text{s}^{-1}$  for at least 30 s). Measurements with empty leaf chambers were made before and after each A/C<sub>i</sub> curve to check the system for leaks. The leaf chamber was attached to *Scirpus* shoots growing in the elevated and ambient chambers, at 25 cm from the top of the shoot. Performing an A/C<sub>i</sub> curve took 40 min on average.

Leaf A/C<sub>i</sub> curves were fitted to the nonlinear model  $A = A_{\infty}[1 - e^{-k(C_i - x_0)}]$ , where  $A_{\infty}$ , the asymptotic level, is the predicted saturated rate of CO<sub>2</sub> uptake, k is the rate of increase in photosynthesis with increase in CO<sub>2</sub>, and  $x_0$  is the CO<sub>2</sub> compensation point (Potvin, Lechowicz & Tardif, 1990). The curves were fitted using the SAS NLIN (nonlinear regression) procedure and the statistical analysis of the model parameters was performed using the SAS analysis of variance procedure. Relative stomatal limitation of photosynthesis was calculated as  $(1 - (A_{ci}/A_{ca})) * 100$ , where  $A_{ci}$  is the estimated assimilation value for C<sub>i</sub> corresponding to a certain atmospheric CO<sub>2</sub> concentration (C<sub>a</sub>), and  $A_{ca}$  is the estimated assimilation value when C<sub>i</sub> is assumed to be equal to this C<sub>a</sub> (no stomatal limitation).

## Results

The A/C<sub>i</sub> curves for six ambient and six elevated plants are shown in Fig. 1. Symbols represent the measured data points and lines show the curves fitted to the data for each plant. The R<sup>2</sup> value for all curves was above 0.993. On average, elevated-CO<sub>2</sub>-grown plants had a higher photosynthesis rate than ambient-CO<sub>2</sub>-grown plants at any C<sub>i</sub> above the compensation point. In CO<sub>2</sub>-free air the CO<sub>2</sub> release was higher in plants grown in elevated CO<sub>2</sub>, but the compensation point was not affected by CO<sub>2</sub> treatment. Table 1 shows a comparison of the properties of the A/C<sub>i</sub> curves for ambient- and elevated-CO<sub>2</sub>-grown plants. Elevated-CO<sub>2</sub>-grown

plants show a significant 31% increase in assimilation at both low and high C<sub>i</sub> compared with plants grown in normal ambient CO<sub>2</sub>. The internal CO<sub>2</sub> concentration was not affected by the CO<sub>2</sub> concentration during growth when compared at ambient CO<sub>2</sub>, and was somewhat higher in the elevated-CO<sub>2</sub>-grown plants when measured at elevated CO<sub>2</sub> (Table 1). The percentage limitation of photosynthesis by stomatal resistance is reduced in plants grown in elevated CO<sub>2</sub> at both measurement concentrations, but the decrease is largest when measured at elevated CO<sub>2</sub>.

The parameter values giving the best fit in the model are also included in Table 1. The rate of increase (the initial slope of the A/C<sub>i</sub> curves) was increased by the elevated CO<sub>2</sub> treatment. Although the mean  $A_{\infty}$  is higher in elevated-CO<sub>2</sub>-grown plants, this is not statistically significant. The assimilation of two elevated-CO<sub>2</sub>-grown plants did not saturate at high CO<sub>2</sub> which caused a large variation in the  $A_{\infty}$  of plants grown in elevated CO<sub>2</sub>. This theoretical asymptotic level may not be as biologically relevant as the assimilation rates at the actual growth conditions.

Plants grown in ambient CO<sub>2</sub> all show a similar response with a maximum stomatal conductance occurring between 0 and 200  $\mu\text{mol mol}^{-1}$  CO<sub>2</sub> and a rapid decline in conductance with increasing CO<sub>2</sub> concentration. The same response is found in three of the plants grown in elevated CO<sub>2</sub>, but the conductance of three other plants is less affected by the measurement CO<sub>2</sub> concentration. The stomatal limitation of photosynthesis, and therefore, the C<sub>i</sub>, is not affected by the CO<sub>2</sub> treatment when measured at ambient CO<sub>2</sub> (Table 1). Plants grown in elevated CO<sub>2</sub> display a reduction in stomatal limitation when measured at high CO<sub>2</sub>, but this effect is not statistically significant.

Estimation of the *Scirpus* biomass shortly after the photosynthesis measurements were made show a 13.2% higher shoot density in the elevated chambers, but only a

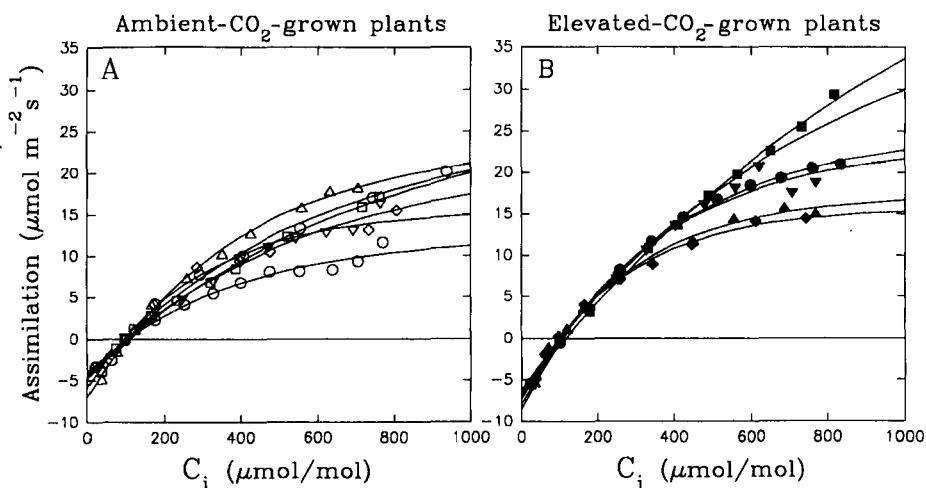


Figure 1. Assimilation versus internal CO<sub>2</sub> concentration (C<sub>i</sub>) for (A) six plants grown in ambient and (B) six plants grown in double ambient CO<sub>2</sub>. The symbols show the individual measurements while the lines show the best fitting curve for each plant using the model  $A = A_{\infty}[1 - e^{-k(C_i - x_0)}]$  ( $A_{\infty}$  = predicted assimilation rate at saturating CO<sub>2</sub>; k = rate of increase;  $x_0$  = compensation point).

**Table 1.** A comparison of properties of A/C<sub>i</sub> curves of plants grown in ambient and elevated CO<sub>2</sub>. Presented are the mean and standard error (SE) of six curves from each treatment, the percentage increase as an effect of high CO<sub>2</sub>, and the level of significance. Properties shown are: the estimated assimilation rates at C<sub>i</sub> of 283 and 526 μmol mol<sup>-1</sup> (which are respectively the mean C<sub>i</sub>s of all plants measured at ambient and elevated CO<sub>2</sub>, the rate of CO<sub>2</sub> release in CO<sub>2</sub>-free air, the C<sub>i</sub> and percentage stomatal limitation to photosynthesis at ambient CO<sub>2</sub> concentrations of 350 and 700 μmol mol<sup>-1</sup>); and the parameter values giving the best fit in the model  $A = A_{\infty}[1 - e^{-k(C_i - x_0)}]$  ( $A_{\infty}$  = predicted rate of assimilation at saturating CO<sub>2</sub>; k = rate of increase; x<sub>0</sub> = compensation point).

	Concentration during growth					
	350 μmol mol <sup>-1</sup>		690 μmol mol <sup>-1</sup>		Percent increase	Probability (<)
	Mean	SE	Mean	SE		
Assimilation at C <sub>i</sub> = 283 μmol mol <sup>-1</sup> (μmol m <sup>-2</sup> s <sup>-1</sup> )	6.60	0.50	8.67	0.23	31.1	0.0061
Assimilation at C <sub>i</sub> = 526 μmol mol <sup>-1</sup> (μmol m <sup>-2</sup> s <sup>-1</sup> )	12.15	0.86	16.03	0.91	31.9	0.0177
CO <sub>2</sub> release at C <sub>a</sub> = 0 (μmol m <sup>-2</sup> s <sup>-1</sup> )	3.66	0.28	5.36	0.09	46.4	0.0003
C <sub>i</sub> at C <sub>a</sub> = 350 (μmol mol <sup>-1</sup> )	283.1	4.6	283.5	5.2	0.1	0.9625
C <sub>i</sub> at C <sub>a</sub> = 700 (μmol mol <sup>-1</sup> )	517.1	14.1	534.5	19.4	3.4	0.5223
Percent stomatal limitation at C <sub>a</sub> = 350	22.00	2.22	21.40	1.36	-2.7	0.8377
Percent stomatal limitation at C <sub>a</sub> = 700	18.28	2.18	15.32	1.95	-16.2	0.3757
<b>Model parameters</b>						
A <sub>∞</sub> (μmol m <sup>-2</sup> s <sup>-1</sup> )	21.24	2.27	32.00	7.29	50.7	0.227
x <sub>0</sub> (compensation point) (μmol mol <sup>-1</sup> )	102.0	2.0	105.3	2.4	3.2	0.356
k (rate of increase) (μmol mol <sup>-1</sup> )	0.00223	0.00028	0.00247	0.00044	10.6	0.690

4.4% increase in above ground biomass. Percentage senescence was reduced in high CO<sub>2</sub> (4.6%) when compared with ambient CO<sub>2</sub> (8.6%).

## Discussion

*Scirpus olneyi* plants growing under natural conditions showed a positive photosynthetic acclimation after exposure to elevated CO<sub>2</sub> for 4 years. The assimilation rate was significantly higher in the elevated-CO<sub>2</sub>-grown plants when compared with the ambient grown plants at both low and high C<sub>i</sub>. The absence of down regulation of photosynthetic capacity of *Scirpus olneyi* is consistent with other findings for these plants: There was no evidence of down regulation of maximum leaf photosynthesis (Ziska, Drake & Chamberlain, 1990), quantum yield (Long & Drake, 1991), or whole canopy photosynthesis (Drake & Leadley, 1991) after several years of growth in high CO<sub>2</sub>.

Positive acclimation of photosynthesis has also been observed in experiments in which soybean and cotton were grown in the field (Campbell, Allen & Bowes, 1988; Kimball *et al.*, 1986), while Radin *et al.* (1987) found no effect of CO<sub>2</sub> treatment on photosynthetic capacity of field grown cotton. A survey of the literature revealed that the size of the pots in which plants were grown is a dominating factor in acclimation to elevated CO<sub>2</sub>; the larger the pot, the smaller the acclimation effect (Arp, 1991). The unrestricted root growth of field grown plants may help prevent sink limitation and end

product inhibition in plants exposed to an elevated CO<sub>2</sub> concentration. The availability of a large carbohydrate sink in the rhizomes of *Scirpus* may provide another important factor in preventing negative acclimation (Ziska *et al.*, 1990). In a study of another plant with large storage organs, Sage *et al.* (1989) found no negative acclimation in potato plants, while in the same study negative acclimation of photosynthesis was observed in bean, cabbage and eggplants grown in high CO<sub>2</sub>.

Acclimation may consist of a redistribution of resources towards the most limiting process (Mott, 1990) and morphological changes which optimize the performance of the plants under the prevailing environmental conditions. According to this hypothesis, the elevated-CO<sub>2</sub>-grown plants measured at high CO<sub>2</sub> are expected to show an improved performance when compared with ambient-CO<sub>2</sub>-grown plants, but when measured at low CO<sub>2</sub>, the plants grown at ambient CO<sub>2</sub> should perform better. The increased photosynthetic capacity of plants grown in elevated CO<sub>2</sub> when measured at high CO<sub>2</sub> is consistent with this hypothesis. However, in this and in other field experiments (Campbell *et al.*, 1988; Kimball *et al.*, 1986), the increase in photosynthetic capacity in elevated-CO<sub>2</sub>-grown plants is as high when measured at low CO<sub>2</sub> as it is when measured at high CO<sub>2</sub>. It may be that acclimations of the plants to elevated CO<sub>2</sub> which enhance photosynthetic capacity (e.g. an increase in enzyme activity, or an increase in the number of palisade cells) also increase the rate of photosynthesis at lower CO<sub>2</sub> concentrations.

Although the above ground biomass of *Scirpus olneyi* was not significantly different between elevated and ambient chambers at the time of measurement, it is possible that other changes may have occurred which affect the photosynthetic capacity. The rate of senescence is reduced in elevated CO<sub>2</sub> (Curtis *et al.*, 1989), and it cannot be excluded that the green shoots used for the photosynthesis measurements were affected. The improved water status of plants grown in elevated CO<sub>2</sub> (Rozema *et al.*, 1991) may also have had a positive effect on photosynthesis. Earlier measurements show a reduction of the nitrogen concentration of *Scirpus* plants grown in high CO<sub>2</sub> (Curtis, Drake & Whigham, 1989) which is not consistent with the increase in photosynthetic capacity. However, it is conceivable that, with the improvement of the water status, nitrogen may have been reallocated from solutes for osmotic adjustment towards the photosynthetic apparatus.

Even in field-grown plants, downward regulation of photosynthetic capacity is reported to occur late in the season (Kimball *et al.*, 1986; Radin *et al.*, 1987). This may be due to a reduction of growth, limiting the sink for carbon in growing tissues, to a gradual filling of the available sinks, to lower temperatures, or to reallocation of nutrients.

After 4 years of exposure to elevated CO<sub>2</sub>, the photosynthetic capacity of *Scirpus olneyi* growing in the field was preserved. These results show that a down regulation of the photosynthetic capacity in response to elevated CO<sub>2</sub> may not occur in field-grown plants, and suggest that natural ecosystems and field grown crops may continue to respond to increasing atmospheric levels of CO<sub>2</sub>.

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