Photosynthetic down-regulation in *Larrea tridentata* exposed to elevated atmospheric CO₂: interaction with drought under glasshouse and field (FACE) exposure

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**ABSTRACT**

The photosynthetic response of *Larrea tridentata* Cav., an evergreen Mojave Desert shrub, to elevated atmospheric CO₂ and drought was examined to assist in the understanding of how plants from water-limited ecosystems will respond to rising CO₂. We hypothesized that photosynthetic down-regulation would disappear during periods of water limitation, and would, therefore, likely be a seasonally transient event. To test this we measured photosynthetic, water relations and fluorescence responses during periods of increased and decreased water availability in two different treatment implementations: (1) from seedlings exposed to 360, 550, and 700 μmol mol⁻¹ CO₂ in a glasshouse; and (2) from intact adults exposed to 360 and 550 μmol mol⁻¹ CO₂ at the Nevada Desert FACE (Free Air CO₂ Enrichment) Facility. FACE and glasshouse well-watered *Larrea* significantly down-regulated photosynthesis at elevated CO₂, reducing maximum photosynthetic rate (*A*ₘₐₓ), carboxylation efficiency (CE), and Rubisco catalytic sites, whereas droughted *Larrea* showed a differing response depending on treatment technique. *A*ₘₐₓ and CE were lower in droughted *Larrea* compared with well-watered plants, and CO₂ had no effect on these reduced photosynthetic parameters. However, Rubisco catalytic sites decreased in droughted *Larrea* at elevated CO₂. Operating *C*ₐ increased at elevated CO₂ in droughted plants, resulting in greater photosynthetic rates at elevated CO₂ as compared with ambient CO₂. In well-watered plants, the changes in operating *C*ₐ, CE, and *A*ₘₐₓ resulted in similar photosynthetic rates across CO₂ treatments. Our results suggest that drought can diminish photosynthetic down-regulation to elevated CO₂ in *Larrea*, resulting in seasonally transient patterns of enhanced carbon gain. These results suggest that water status may ultimately control the photosynthetic response of desert systems to rising CO₂.

**Key-words:** *Larrea tridentata* (Creosotebush); A–Cᵢ response; drought; elevated CO₂; FACE; Mojave Desert; NDFF; photosynthetic down-regulation; transient CO₂ response.

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**INTRODUCTION**

It is increasingly important to examine the responses of terrestrial plants to changing levels of atmospheric CO₂ in light of current predictions of a doubling in CO₂ concentration that may occur within the next century (Bazzaz 1990). Increasing atmospheric CO₂ overcomes biochemical limitations by enhancing CO₂ concentration at the site of carboxylation in C₃ plants and by reducing the oxygenase activity of Rubisco, resulting in increased rates of carbon fixation (Sharkey 1985). Thus, for plants exposed to elevated CO₂, leaf photosynthetic rates initially increase (Bowes 1991). However, for many C₃ species, long-term growth at elevated CO₂ eventually results in photosynthetic rates that are only marginally enhanced, or even lower than those observed prior to elevated CO₂ exposure (Sage 1994). This long-term, down-regulation response generally results in a reduced leaf nitrogen content and a decreased amount of protein associated with the photosynthetic apparatus (Sage, Sharkey & Seemann 1989; Kuehny et al. 1991; Sage 1994; Jacob, Greitner & Drake 1995). The mechanisms underlying this long-term downward adjustment of photosynthesis to elevated CO₂ are not well understood, and the developed hypotheses are based on data collected from glasshouse-grown, potted plants. Data from intact plants in field conditions may provide crucial insights into the mechanisms associated with photosynthetic down-regulation and growth in elevated CO₂ (Jacob et al. 1995; Lewis, Tissue & Strain 1996).

Large increases in leaf carbohydrate amounts (up to two times for starch in tobacco) occur in plants exposed to elevated CO₂ (Long & Drake 1992), and carbohydrate accumulation in source tissues is often associated with inhibition of photosynthesis (Azcon-Bieto 1983; Tissue, Thomas & Strain 1993). Increased carbohydrate metabolism in source leaves may produce a regulatory signal that results in decreased photosynthetic gene expression readjusting source–sink imbalance (Van Oosten & Besford 1996). When sink activity is enhanced, such as when root growth restrictions are removed (typical characteristics associated with the down-regulation response), photosynthetic acclimation typically decreases (Thomas & Strain 1991). Therefore, the inability
of sink tissues to maintain a sufficient demand for use of photosynthetic end products potentially explains the down-regulation response of photosynthesis to elevated CO$_2$ (Stitt 1991). Additionally, environmental conditions such as drought may reduce both sink activity and leaf water status to such low levels that accumulation of photosynthetic end products does not occur, resulting in a decrease in photosynthetic down-regulation (Oechel et al. 1995). Data from intact plants exposed to elevated CO$_2$ in the field may provide additional insight on the mechanisms of long-term photosynthetic response to elevated CO$_2$, as plants grown in the field provide a normal relationship between sink and source tissues.

Seasonal patterns of photosynthetic down-regulation may have been affected by environmental conditions in *Pinus taeda* and *Scirpus olneyi* grown at elevated CO$_2$ in open-top chambers (Jacob et al. 1995; Lewis et al. 1996). Environmental stress may impose modifications on the responses of terrestrial plants to increased CO$_2$ (Field et al. 1992; Curtis 1996; Sage 1996). Because sink strength can interact with the stimulation of photosynthesis to regulate whole plant responses to elevated CO$_2$ (Thomas & Strain 1992; Barrett & Gifford 1995) and water availability can be quite important in regulating sink strength (Krizek et al. 1985), the relationship between water availability and photosynthetic response to elevated CO$_2$ is especially important to evaluate. Drought may reduce the carbohydrate build-up in leaves, and in one study eliminated increased photosystem II efficiency associated with growth at elevated CO$_2$ (Roden & Ball 1996b). In water-limited systems, sink demands are driven by highly variable seasonal precipitation patterns, and photosynthetic responses to elevated CO$_2$ may be directly or indirectly controlled by variation in water availability (Oechel et al. 1995).

Because the down-regulation of photosynthesis may be a key physiological process that limits increases in productivity of a high CO$_2$ world, we evaluated the pattern of photosynthetic down-regulation to elevated CO$_2$ in a glasshouse and in the field during periods of seasonal water stress and increased water availability in the Mojave Desert shrub *Larrea tridentata* Cav. We expected that drought would result in reductions in the degree of photosynthetic down-regulation to elevated CO$_2$ as compared with non-droughted conditions in both glasshouse and field conditions. This follows from the influence of drought, which can reduce photosynthetic rates and leaf carbohydrate accumulation to levels such that feedback inhibition of photosynthesis may not occur (Oechel et al. 1995). In addition, we expected the glasshouse-grown plants to exhibit a greater down-regulation response to elevated CO$_2$ than their field-grown counterparts as a result of the restriction on root growth imposed by pots. To test these hypotheses, we evaluated gas exchange and fluorescence characteristics under droughted and well-watered conditions on fully established adult plants in a Free Air Carbon Dioxide Enrichment (FACE) system and seedlings in a glasshouse. This approach allows for a comparison of results from two CO$_2$ treatment implementation techniques that may lead to different source–sink aspects under contrasting moisture conditions.

### MATERIALS AND METHODS

#### Study species

*Larrea tridentata* Cav. (Zygophyllaceae) is a dominant evergreen, distributed throughout the warm deserts of the arid New World (Smith, Monson & Anderson 1997). The distribution of *Larrea* is a primary determinant of the boundaries of the North American warm deserts. *Larrea* has been classified as a true drought-resisting species that remains metabolically active during dry seasons and has the potential to exhibit opportunistic flowering and growth in response to rainfall at any time of the year (Oechel, Strain & Odening 1974; Bowers & Dimmitt 1994). *Larrea* can reach a relatively high rate of positive net photosynthesis as compared with most desert evergreens and shows remarkable photosynthetic acclimation to temperature variation (Mooney, Bjorkman & Collatz 1978). The seedlings used in this study were taken as seed from a population on the Nevada Test Site, near the location of the Nevada Desert FACE Facility (NDFF). Adult plants at NDFF were undisturbed during construction of the FACE facility and remain intact.

#### CO$_2$ treatment facilities

*Larrea* seedlings were exposed to three CO$_2$ treatment levels (ambient – 360 μmol mol$^{-1}$; 1.5 × ambient – 550 μmol mol$^{-1}$; and 2 × ambient – 700 μmol mol$^{-1}$) in a controlled-environment glasshouse at the University of Nevada, Las Vegas for a year prior to plant sampling (ambient CO$_2$ concentration fluctuated between 360 and 390 μmol mol$^{-1}$ in this urban area). The seedlings were planted in 15 cm diameter by 1 m tall PVC pipes in a 20:80 silt:sand mix. All *Larrea* received 400 cm$^3$ 1:40 Hoaglands solution every 2 weeks until 2.5 months prior to sampling, when water and Hoagland’s solution were withheld from randomly selected individuals in each CO$_2$ treatment. The glasshouse facility received natural light (maximum photosynthetic photon flux density (PPFD) = 1600 μmol m$^{-2}$ s$^{-1}$) and roughly tracked external temperature with limits of 0 °C and 45 °C.

Mature *Larrea* shrubs were exposed to either ambient (360 μmol mol$^{-1}$) or 1.5 × ambient (550 μmol mol$^{-1}$) CO$_2$ at NDFF beginning in April 1997. NDFF is located on the Nevada Test Site (36°49’ N, 115°55’ W, 970 m), an environmental research park operated by the US Department of Energy. This area is a pristine portion of the Mojave Desert, which has been free from grazing for more than 40 years. The average precipitation is 138 ± 62 mm, falling mostly as rain during winter months (Hunter 1994), but the Mojave Desert can receive unpredictable rain during any portion of the year (Bowers 1987). NDFF consists of nine ringed plots (491 m$^2$), each encompassing = 12 *Larrea*. Three plots are maintained at 550 μmol mol$^{-1}$ CO$_2$, while the remaining six rings are controls for CO$_2$ enhancement and application assembly. The NDFF maintains continuous CO$_2$ enrichment except when the 5 min wind speed average exceeds 6.0 m s$^{-1}$, resulting in elevated CO$_2$ treatment conditions occurring greater than
95% of the time. The plants are accessed from an overhead moveable walkway system that allows sampling without disturbing soil in the plots. A more extensive facility description is available in Jordan et al. (1998).

**Plant sampling**

*Larrea* were organized into three blocks within each CO₂ treatment room in the glasshouse, and individuals within each block were randomly assigned a droughted or well-watered condition. Two to three individuals in each block for each treatment combination were randomly selected for sampling. The block (a mean of these two to three individuals) was used as the replicate for statistical analysis. Similarly, one to three individuals were randomly selected from each treatment ring at the FACE site, and the ring was used as the replicate for statistical analysis. This organization led to a sample size of three for each CO₂ treatment/water status combination at both the glasshouse and FACE site. *Larrea* were sampled at the FACE site over 1 week in June 1997 during a pronounced drought, and well-watered individuals were measured in October 1997 following a series of late summer rainfall events. All plants were sampled between 0900 and 1200 h, a period that has the least diurnal fluctuation in gas exchange and fluorescence patterns. All measurements were performed on the terminal newest expanded leaves. For gas exchange, leaf area was determined by regressions between leaf number, stem diameter and length, and leaf area from harvested individuals.

The $A–C_i$ (assimilation rate–internal CO₂ concentration) response of *Larrea* was determined with a programmable, open-flow gas exchange system (Li 6400, LiCOR Inc., Lincoln, NE, USA). Block temperature was held at 30 °C and relative humidity at 20%, typical of environmental conditions in the field. All measurements were made with a red light source at PPFD values of 1500 μmol m⁻² s⁻¹. The CO₂ concentration of the cuvette ($C_a$) was initially maintained at 200 μmol mol⁻¹ for 5 min to stimulate stomatal opening and then reduced to 75 μmol mol⁻¹. $C_a$ was incrementally increased to 150, 250, 350, 550, 700, 800, 900, 1200, and 2000 μmol mol⁻¹, and gas exchange properties were logged at each $C_a$ once the system had reached a predetermined stability point (coefficient of variation = 1%). The data from the $A–C_i$ response was fitted to a non-linear least-squares regression using an exponential model from which maximum photosynthetic rate ($A_{max}$) and carboxylation efficiency (CE) were determined (Jacob et al. 1995).

Chlorophyll fluorescence was used to assess the photochemical efficiency of photosystem II ($F_v/F_m$), non-photochemical (QNP) and photochemical (QP) quenching. Terminal shoots were dark adapted in aluminium foil for 1 h, and then fluorescence was measured with a pulse-amplitude-modulated (PAM) fluorimeter (Model MFMS/2S, Hansatech Instruments Ltd, King’s Lynn, UK) under the control of a personal computer. Initial fluorescence ($F_o$) was taken to be the average of 20 s fluorescence upon exposure to a low level light source (583 nm; 2 μmol m⁻² s⁻¹). Maximal fluorescence ($F_m$) was determined by exposing the shoots to 0.5 s pulses of saturating pulse-modulated light (8500 μmol m⁻² s⁻¹ PPFD; 1 Hz). $F_v/F_m$ was determined where $F_v = F_m – F_o$, QNP was determined by $1 – F_v/F_m$, where $F_v = F_m – F_0$ ($F_m$ is maximum fluorescence following an initial pulse of 8500 μmol m⁻² s⁻¹ and $F_0$ is initial fluorescence under actinic light). QP was calculated as $1 –$ QNP.

Leaf samples for chlorophyll content and Rubisco analysis were taken concurrent with $A–C_i$ measurements from shoots adjacent to gas exchange measurements, and immediately preserved in liquid nitrogen. Stems and dead leaves were removed prior to grinding in a mortar. Approximately 0.15 g of leaves were ground in 1.5 cm³ of buffer containing 100 mM Bicine pH 7.8, 5 mM MgCl₂, 1 mM ethylene diamine tetraacetic acid (EDTA), 5 mM dithiothreitol (DTT), 0.1% (v/v) Triton X-100, 25 mM KCl, and 20% (w/w) polyvinylpyrrolidone. Extracts were filtered through Miracloth and centrifuged for 1 min at 10 000 g. The chlorophyll content was determined following extraction with ethanol (Wintemans & DeMots 1965). Rubisco catalytic sites were determined by immunoprecipitation as described by Evans & Seemann (1984).

Xylem water potential ($Ψ_{xylem}$) was determined with a Scholander-type pressure chamber (Soil Moisture Stress Inc, Santa Barbara, CA, USA) on terminal shoots taken at midday from the *Larrea* on which the $A–C_i$ response was determined.

**Statistical analysis**

A multivariate analysis of variance (MANOVA, Statistica, StatSoft Inc., Tulsa, OK, USA) was used to compare the overall responses of *Larrea* to different CO₂ treatment techniques, CO₂ growth concentrations, and drought. Initially the 700 μmol mol⁻¹ treatment was excluded so that the design was factorial; inclusion of the 700 μmol mol⁻¹ growth condition was applied to the glasshouse data independently using a MANOVA so that pairwise comparisons could be constructed. Six response variables were used in the MANOVA, including $A_{max}$, CE, $Ψ_{xylem}$, $F_v/F_m$, QNP, and QP. Post-hocs were Newman–Kuels tests at $α = 0.05$. MANOVA data distribution assumptions were met by arcsin square root transformation of the data.

The effect of CO₂ treatment, drought, and their interaction on leaf chlorophyll content and Rubisco catalytic sites were analysed separately for the FACE and glasshouse plants with a two factor ANOVA and Newman–Kuels tests as post-hocs, with significance at $α = 0.05$.

**RESULTS**

When the six response variables ($A_{max}$, CE, $Ψ_{xylem}$, $F_v/F_m$, QNP, and QP) in the MANOVA model were considered, the effects of exposure to elevated CO₂ and drought were significant for both the glasshouse and FACE treatment techniques (Table 1). In addition, the interactions between drought and fumigation technique, as well as drought and CO₂ growth condition, were significant. There was no
interaction between the fumigation technique and the effect of CO₂ growth environment upon the response variables, suggesting that the pattern of response to elevated CO₂ was consistent. There were no significant differences in the patterns of Larrea response for the plants grown in the glasshouse as compared with the plants exposed at the FACE site.

Overall, elevated CO₂ had little effect on the response of midday $\Psi_{xylem}$. $\Psi_{xylem}$ in Larrea varied from –6 MPa during drought to –2 MPa when soil moisture was high at the FACE site. $\Psi_{xylem}$ in droughted FACE plants were much more negative than droughted glasshouse plants, and their well-watered counterparts were less negative than well-watered glasshouse plants (Fig. 1). There was a CO₂-by-drought interaction for plants grown in the glasshouse, leading to an amelioration of drought in the 2·ambient CO₂ concentration. There were no pairwise significant differences in $\Psi_{xylem}$ in FACE plants with respect to growth at elevated CO₂.

Well-watered Larrea grown in glasshouse conditions showed a progressive decline in the initial slope of the $A$–$C_i$ response (CE), along with a decrease in the asymptote ($A_{max}$) when grown exposed to 1·5 × and 2 × ambient CO₂ as compared with those grown in ambient conditions, and the degree of response appeared to be a gradient with 

<table>
<thead>
<tr>
<th>Factor</th>
<th>Wilks’ lambda</th>
<th>d.f.</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fumigation technique</td>
<td>0·581</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>CO₂ concentration</td>
<td>0·235</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>Drought</td>
<td>0·036</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>Fumigation × CO₂</td>
<td>0·618</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>Fumigation × drought</td>
<td>0·091</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>Drought × CO₂</td>
<td>0·360</td>
<td>6, 11</td>
<td>–</td>
</tr>
<tr>
<td>Fumigation × drought × CO₂</td>
<td>0·448</td>
<td>6, 11</td>
<td>–</td>
</tr>
</tbody>
</table>

$A_{max}$, maximum photosynthetic rate; CE, carboxylation efficiency; $\Psi_{xylem}$, xylem water potential; QNP, non-photochemical quenching; QP, photochemical quenching; FACE, Free Air CO₂ Enrichment.

Table 1. Results of a general three-way MANOVA with six response variables ($A_{max}$, CE, $\Psi_{xylem}$, $F_v/F_m$, QNP, and QP) and main factors of fumigation technique (glasshouse versus FACE), drought (well-watered versus droughted), and CO₂ concentration (360 µmol mol⁻¹ versus 550 µmol mol⁻¹). Significance (*) is at $\alpha = 0·05$

Figure 2. Net assimilation rate ($A$) versus internal CO₂ concentration ($C_i$) for Larrea tridentata exposed to various levels of CO₂ in a glasshouse (a & b) and at a FACE (Free Air CO₂ Enrichment) facility (c & d) under wet (well-watered; a & c) and dry (droughted; b & d) conditions. Curves plotted are the outputs of a non-linear least-squares regression model fitted to the actual data (Jacob et al. 1995). Open circles upon the regression line indicate operational $C_i$ for ambient-grown Larrea, while open squares are for 1·5 × ambient and open triangles are for 2 × ambient CO₂ Larrea. Coefficients of determination are indicated within each plot, identified by CO₂ environment concentration.

Figure 1. Midday xylem water potentials ($\Psi_{xylem}$) of Larrea tridentata exposed to elevated levels of CO₂ under wet (well-watered) and dry (droughted) conditions in a glasshouse (a) and at a FACE (Free Air CO₂ Enrichment) facility (b). Letters indicate significant differences among treatments within panels at $\alpha = 0·05$. © 1998 Blackwell Science Ltd, Plant, Cell and Environment, 21, 1153–1161
Larrea maintained \( C_i \) in such a manner that \( A_{\text{net}} \) was consistent between CO\(_2\) growth environments (Fig. 2a & c). In droughted Larrea, the increase in \( C_i \) resulted in greater increases in \( A_{\text{net}} \) in elevated plants as compared with ambient plants (Fig. 2b & d).

\( A_{\text{max}} \) was reduced by 25% in non-water-stressed plants exposed to elevated CO\(_2\) compared with ambient CO\(_2\) non-water-stressed plants for both treatment facilities. Drought was a significant factor in determining \( A_{\text{max}} \), regardless of CO\(_2\) growth concentration, typically reducing \( A_{\text{max}} \) by \( \approx 50\% \) (Fig. 3). Therefore, \( A_{\text{max}} \) was significantly higher in well-watered plants exposed to ambient CO\(_2\) as compared with the \( A_{\text{max}} \) of well-watered plants exposed to elevated CO\(_2\), and both were higher than the \( A_{\text{max}} \) of all droughted plants. CE exhibited a similar pattern as \( A_{\text{max}} \) in glasshouse-grown plants, in that drought led to a decline in CE (Fig. 3). An interaction between drought and CO\(_2\) concentration resulted in the largest CE values in the ambient CO\(_2\), well-watered condition (Fig. 3). In FACE-exposed plants, the pattern was similar, but there were no pairwise differences between the wet and dry plants but rather a strong effect of CO\(_2\), leading to a 40% decrease in CE at elevated CO\(_2\) in Larrea (Fig. 3).

The chlorophyll content of Larrea leaves responded to changes in water availability (Table 2). There was \( a = 10\% \) increase from drought to well-watered conditions. The number of Rubisco catalytic sites decreased by 25% with increasing CO\(_2\) concentration, similar to the decrease in CE for well-watered plants in both treatment facilities (Table 2). However, for droughted plants, the number of Rubisco catalytic sites did not differ between ambient and elevated CO\(_2\) treatments at NDFF, but the greenhouse plants did show a decrease in Rubisco catalytic sites even in droughted plants (Table 2). It is important to note that chlorophyll content and Rubisco active sites were determined on a fresh mass basis, and interpretation should be made with caution. However, the fresh mass to dry mass ratio of Larrea leaves from glasshouse-grown conditions increased by only 14% from droughted to well-watered conditions and relative water content increased by less than 10% (data not shown). The direction of change in water content with increasing drought and CO\(_2\) environment suggests that the differences reported here in chlorophyll content and Rubisco active sites represent a conservative estimate of actual differences associated with elevated CO\(_2\).

Exposure to elevated CO\(_2\) increased the overall quantum yield of photosystem II (\( F_v/F_m \)) by 10%, regardless of CO\(_2\) growth concentration.

### Table 2. Chlorophyll content and Rubisco active sites extracted from leaves of Larrea tridentata exposed to elevated or ambient CO\(_2\) at a FACE facility and in a glasshouse under three levels of CO\(_2\) fumigation.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Treatment</th>
<th>Chlorophyll (( \mu g g^{-1} \text{FW} ))</th>
<th>Rubisco protein (nmol catalytic sites g(^{-1}) FW)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FACE</td>
<td>Dry</td>
<td>360: 620 ± 125(^a)</td>
<td>61-4 ± 4-8(^a)</td>
</tr>
<tr>
<td></td>
<td>550</td>
<td>839 ± 100(^a)</td>
<td>57-1 ± 2-9(^a)</td>
</tr>
<tr>
<td></td>
<td>Wet</td>
<td>360: 1402 ± 125(^a)</td>
<td>112-4 ± 9-4(^a)</td>
</tr>
<tr>
<td></td>
<td>550</td>
<td>1397 ± 198(^a)</td>
<td>91-1 ± 7-0(^a)</td>
</tr>
<tr>
<td>Glasshouse</td>
<td>Dry</td>
<td>360: 822 ± 38(^a)</td>
<td>31-2 ± 2-9(^a)</td>
</tr>
<tr>
<td></td>
<td>550</td>
<td>590 ± 88(^b)</td>
<td>6-4 ± 1-2(^a)</td>
</tr>
<tr>
<td></td>
<td>700</td>
<td>557 ± 21(^b)</td>
<td>4-8 ± 0-8(^b)</td>
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<tr>
<td></td>
<td>Wet</td>
<td>360: 875 ± 60(^b)</td>
<td>35-2 ± 3-5(^b)</td>
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<td></td>
<td>550</td>
<td>1189 ± 156(^b)</td>
<td>26-5 ± 3-3(^b)</td>
</tr>
<tr>
<td></td>
<td>700</td>
<td>780 ± 198(^a)</td>
<td>13-7 ± 5-1(^b)</td>
</tr>
</tbody>
</table>

FACE, Free Air CO\(_2\) Enrichment; SD, standard deviation; FW, fresh weight.
of atmospheric CO₂ under wet (well-watered) and dry (droughted) growth in elevated CO₂ does not offset a stress response (QP) of individuals of photochemical quenching (QPN) and photochemical quenching from FACE showed a Larrea Fv/m that were not significantly different, suggesting that elevated CO₂ may partially down-regulation of photosynthesis upon exposure to elevated CO₂ in controlled-environment conditions, but drought diminishes the down-regulation response (Oechel et al. 1995). The photosynthetic responses of L. tridentata to elevated CO₂ indicate that this long-lived, relatively slow-growing Mojave Desert shrub adjusts resource investment to photosynthesis when exposed to elevated CO₂ under conditions of potentially optimum resource availability, either in the field or under controlled glasshouse conditions. However, this down-regulation of photosynthesis to elevated CO₂ diminishes following exposure to drought. In fact, drought converged all values of A_max in each treatment combination to very low levels. Similarly, the chaparral shrub Adenostoma fasciculatum exhibits down-regulation of photosynthesis to elevated CO₂ under conditions of potentially optimum resource availability, either in the field or under controlled glasshouse conditions. However, this down-regulation of photosynthesis to elevated CO₂ diminishes following exposure to drought. In fact, drought converged all values of A_max in each treatment combination to very low levels. Similarly, the chaparral shrub Adenostoma fasciculatum exhibits down-regulation of photosynthesis to elevated CO₂ upon exposure to elevated CO₂ and drought experienced some degree of increased instantaneous photosynthetic rates and biomass production as compared with those grown in ambient conditions, even though there was still some reduction in A_max in elevated CO₂-grown plants (Oechel et al. 1995). Currently, there is no known mechanism that explains the drought-induced loss of down-regulation of photosynthesis to elevated CO₂. Drought may act to reduce A_max to such a level that the amount of photosynthate supplied to the plant is insufficient to invoke a form of end product inhibition or feedback response of photosynthesis (Oechel et al. 1995). Therefore, the idea that down-regulation may be a general response to elevated CO₂ in suboptimal environments (Curtis 1996) may need to be re-evaluated. In this respect, inter- and intra-annual patterns of water availability may be very important in integrating photosynthetic and productivity patterns of desert systems in future climate scenarios, including elevated CO₂.

While down-regulation of photosynthesis occurs, growth at elevated CO₂ improves the overall quantum yield of photosystem II (Fv/Fm) in all conditions. This was primarily due to the increase in Fv/Fm in elevated CO₂ during drought, suggesting that elevated CO₂ may partially offset stress to photosystem II associated with growth in droughted conditions. Similarly, Fv/Fm increased at elevated CO₂ for Arbutus unedo, presumably associated with amelioration of drought (Jones et al. 1995). In Eucalyptus, when plants were exposed to elevated CO₂, high temperature and light, there was a decrease in the quantum yield of photosystem II, presumably related to a down-regulation of electron transport associated with increased photosynthetic end products (Roden & Ball 1996a). These differences observed in the current experiment and others invoking temperature- and light-induced photoinhibition coupled with increases in CO₂ do not lead to mutually exclusive conclusions concerning the effects of elevated CO₂ on plants, but highlight the importance of considering the state of photosynthetic down-regulation when stress

leaf chlorophyll contents, which increased from droughted to well-watered treatments.

**DISCUSSION**

The photosynthetic responses of L. tridentata to elevated CO₂ indicate that this long-lived, relatively slow-growing Mojave Desert shrub adjusts resource investment to photosynthesis when exposed to elevated CO₂ under conditions of potentially optimum resource availability, either in the field or under controlled glasshouse conditions. However, this down-regulation of photosynthesis to elevated CO₂ diminishes following exposure to drought. In fact, drought converged all values of A_max in each treatment combination to very low levels. Similarly, the chaparral shrub Adenostoma fasciculatum exhibits down-regulation of photosynthesis upon exposure to elevated CO₂ in controlled-environment conditions, but drought diminishes the down-regulation response (Oechel et al. 1995). Adenostoma exposed to elevated CO₂ and drought experienced some degree of increased instantaneous photosynthetic rates and biomass production as compared with those grown in ambient conditions, even though there was still some reduction in A_max in elevated CO₂-grown plants (Oechel et al. 1995). Currently, there is no known mechanism that explains the drought-induced loss of down-regulation of photosynthesis to elevated CO₂. Drought may act to reduce A_max to such a level that the amount of photosynthate supplied to the plant is insufficient to invoke a form of end product inhibition or feedback response of photosynthesis (Oechel et al. 1995). Therefore, the idea that down-regulation may be a general response to elevated CO₂ in suboptimal environments (Curtis 1996) may need to be re-evaluated. In this respect, inter- and intra-annual patterns of water availability may be very important in integrating photosynthetic and productivity patterns of desert systems in future climate scenarios, including elevated CO₂.

While down-regulation of photosynthesis occurs, growth at elevated CO₂ improves the overall quantum yield of photosystem II (Fv/Fm) in all conditions. This was primarily due to the increase in Fv/Fm in elevated CO₂ during drought, suggesting that elevated CO₂ may partially offset stress to photosystem II associated with growth in droughted conditions. Similarly, Fv/Fm increased at elevated CO₂ for Arbutus unedo, presumably associated with amelioration of drought (Jones et al. 1995). In Eucalyptus, when plants were exposed to elevated CO₂, high temperature and light, there was a decrease in the quantum yield of photosystem II, presumably related to a down-regulation of electron transport associated with increased photosynthetic end products (Roden & Ball 1996a). These differences observed in the current experiment and others invoking temperature- and light-induced photoinhibition coupled with increases in CO₂ do not lead to mutually exclusive conclusions concerning the effects of elevated CO₂ on plants, but highlight the importance of considering the state of photosynthetic down-regulation when stress

leaf chlorophyll contents, which increased from droughted to well-watered treatments.

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treatments are implemented in conjunction with changes in CO₂ environment.

The down-regulation response of Larrea in the field was rather rapid considering the plants were exposed to elevated CO₂ for only 5 months. To date, the down-regulation response to elevated CO₂ has taken several to many growing seasons to occur in other field studies (Korner & Diemer 1994; Jacob et al. 1995). Predictions of plant responses based on functional types suggest that Larrea should not have shown a down-regulation response over such a short time period. This is based on the slow growth rate and high stress tolerance demonstrated by this long-lived desert plant. However, Larrea photosynthesis and growth respond rapidly to the return of optimal conditions following drought (Oechel et al. 1974; Brisson & Reynolds 1994), suggesting that a rapid response to changing CO₂ concentration should not be unexpected. Indeed, analysis by gas exchange 1 month prior to the rainfall event, which saturated soils and improved water availability, indicated no down-regulation response (T. E. Huxman, unpublished results). In the greenhouse, watering of droughted Larrea increases Aₘₐₓ rapidly (2–4 d) in all CO₂ conditions, and the typical decreased A–Cₚ asymptotes associated with photosynthetic down-regulation occurs within several days in elevated as compared with ambient CO₂-grown Larrea (T. E. Huxman, unpublished results).

In greenhouse conditions, Larrea exhibits changes in root growth upon exposure to elevated CO₂. Total root system biomass increased when exposed to elevated CO₂, and the proportion and absolute amount of fine roots to coarse roots decreased, resulting in a smaller number of fine roots (BassiriRad, Reynolds & Virginia 1997). However, there appear to be no differences in the ability of root systems of Larrea to take up nutrients (BassiriRad et al. 1997) or to conduct water to stems (Huxman, Smith & Neuman 1998) in elevated versus ambient CO₂.

Combining results showing down-regulation of photosynthesis with no significant change in the potential rates of nutrient and water delivery to the shoot (albeit from different studies) suggest that the pattern of photosynthetic readjustment may be adaptive in terms of linking above- and below-ground processes, ensuring long-term productivity. However, this idea is based upon the assumption that root characteristics in the field do not change with changing CO₂ concentration, which remains to be validated. Whole plant nitrogen and phosphorus budgets have yet to be determined under field conditions at elevated CO₂, which may also help our understanding of the significance of the down-regulation response to whole plant function.

While it appears from these data that the seasonal course of drought is quite important in determining plant gas exchange response to elevated CO₂ for arid and semi-arid systems, typified by Larrea and Adenostoma, respectively, temperature may also be important and potentially help to explain the results from the FACE Larrea in the current experiment. The relative increases in photosynthetic responses of Pinus taeda to elevated CO₂ were much higher during summer periods as compared with winter (Lewis et al. 1996). This may be due to the counteractive effects of increasing temperature and CO₂ on photorespiration (Long 1991). Increasing temperature reduces the specificity of Rubisco for CO₂ by enhancing the oxygenase activity of Rubisco more than the carboxylase activity, thus increasing the proportion of potential photosynthesis lost to photorespiration (Brooks & Farquhar 1985). Increasing CO₂ overrides oxygenase activity, thus providing relief for plants growing in elevated atmospheric CO₂ from the inefficient use of Rubisco active sites. Because deserts are typified by extreme seasonality in temperature and episodic precipitation patterns, the photosynthetic response of desert plants to elevated CO₂ may be more difficult to predict, and the changes in temperature between field measurements in this study may help to explain the patterns observed. In spite of this, the data from greenhouse-grown Larrea, which remove the temperature differences between field sampling dates, suggest that drought is still important.

Taken together, these results suggest that the photosynthetic response of L. tridentata to increasing CO₂ concentration may be controlled by seasonal patterns of resource availability. Specifically, the down-regulation response in Larrea, and perhaps in other desert perennials, may be seasonally transient, occurring primarily in wet seasons but largely absent during the extensive dry seasons that typify the Mojave Desert. Therefore, the potential for interaction between drought-controlled and nutrient-controlled down-regulation response may exist, as periods of high water availability are often concurrent with periods of nitrogen limitation on plant growth in North American deserts (Smith et al. 1997). Because the down-regulation response may be seasonally transient, future research will be designed to evaluate the potential for seasonal patterns of photosynthetic adjustment in Larrea and other desert perennials of the Mojave Desert FACE site. Many ecosystem-level processes are affected by the characteristics of carbon fixation and soil resource utilization in plants, suggesting the pattern of down-regulation response throughout a year may be very important in terms of understanding how plants adjust to the new resource levels exhibited in a future high CO₂ world. Because the down-regulation response of Larrea is related to water availability and deserts are systems that are driven by episodic patterns of precipitation, the pattern of down-regulation over a season may be extremely important. Specifically, if the down-regulation response occurs in tandem with precipitation patterns, the accumulation of carbon-based end products of photosynthesis and dilution of leaf nitrogen by carbon may also occur seasonally, influencing litter quality, decomposition rates, and nutrient cycling in the desert. In addition, because we have observed no evidence of stomatal acclimation to elevated CO₂ (Hamerlynck, unpublished results), the down-regulated photosynthetic rates may reduce the usually observed benefit of increased water-use efficiency associated with growth in elevated CO₂.

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