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Elevated Atmospheric CO_2 Magnifies Intra-specific Variation in Seedling Growth of Honey Mesquite: An Assessment of Relative Growth Rates

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Abstract

The shrub honey mesquite (*Prosopis glandulosa* var. *glandulosa* Torr.) readily encroaches into rangelands in the southwestern United States that vary in annual rainfall by almost a factor of 5 (200 to 1 000 mm). This occurs partly because mesquite seedlings grow rapidly and become uncoupled from competition with established herbaceous vegetation. Species that occupy such a wide precipitation gradient frequently include plants that differ genetically in seedling growth rate. Whether atmospheric carbon dioxide (CO_2) enrichment affects seedling biomass uniformly across mesquite genotypes or magnifies the expression of genetic variation in seedling growth remains unresolved. We measured the effects of CO_2 enrichment on growth of seedlings derived from 14 adult plants (open-pollinated families), 7 plants each from rangelands located near extremes of the rainfall gradient occupied by the shrub (arid southeastern New Mexico vs. mesic central Texas). Growth was measured over days 10 to 30 following emergence on well-watered seedlings in glasshouses at ambient and elevated CO_2 concentrations (391 and 706 $\mu mol \cdot mol^{-1}$, respectively). Proportional responses of biomass (day 30) to CO_2 enrichment varied from 1.03 to 1.74 among families. CO_2 enrichment did not consistently favor the largest or fastest-growing families at ambient CO_2. Rather, proportional responses of biomass to elevated CO_2 were highly correlated across families with the stimulation of relative growth rate (RGR) at elevated CO_2. Biomass at ambient CO_2 was 19% greater, on average, in families from mesic rangeland than from arid rangeland, but families from extremes of the precipitation gradient did not diverge by seedling size or response to CO_2. Selection for greater RGR could augment the mean growth response of mesquite seedlings to CO_2. Even in the absence of selection, CO_2 enrichment could increase mesquite establishment by enhancing seedling growth and thereby exacerbate the management challenge of minimizing woody encroachment.

Resumen

El “Honey mesquite” (*Prosopis glandulosa* var. *glandulosa* Torr.) invade rápidamente los pastizales del suroeste de E.U.A. que varían en precipitación anual por casi un factor de cinco (200 a 1000 mm). Esto ocurre, en parte, porque las plántulas de “Honey mesquite”crecen rápidamente y llegan a desfasarse de la competencia con la vegetación herbácea establecida. Las especies que ocupan tan amplio gradiente de precipitación frecuentemente incluyen plantas que difieren genéticamente en la tasa de crecimiento de las plántulas. Un aspecto que aun esta sin resolver es si el enriquecimiento de dióxido de carbón (CO_2) atmosférico afecta uniformemente la biomasa de las plántulas entre genotipos de “Honey mesquite” o magnifica la expresión de la variación genética del crecimiento de las plántulas. Medimos los efectos del enriquecimiento de CO_2 en el crecimiento de plántulas derivadas de 14 plantas adultas (familias de polinización abierta), 7 plantas, cada una proveniente de pastizales localizados cerca de los extremos del gradiente de precipitación en el que el arbusto invade los pastizales (el sudeste árido de New México versus la parte central mésica Texas). El crecimiento fue medido del día 10 al 30 después de la emergencia en plántulas con buen suministro de agua creciendo en condiciones de invernadero bajo concentraciones de CO_2 ambientales y elevadas (391 y 706 $\mu mol \cdot mol^{-1}$, respectivamente). Las respuestas proporcionales de la biomasa (día 30) al enriquecimiento de CO_2 variaron entre familias de 1.03 a 1.74. El enriquecimiento de CO_2 no favoreció consistentemente a las familias más grandes o de crecimiento más rápido bajo condiciones ambientales de CO_2. Mas bien, las repuestas proporcionales de la biomasa a la elevación de CO_2 estuvieron altamente correlacionadas entre familias con la estimulación de la tasa relativa de crecimiento (RGR) en la concentración elevada de CO_2. A la concentración ambiental de CO_2, la biomasa de las familias provenientes del pastizal mésico fue en promedio 19% mayor que la de las provenientes del árido, pero las familias de los extremos del gradiente de precipitación no divergieron en el tamaño de plántula o en la repuesta al CO_2. La selección en base a una mayor RGR pudiera aumentar la respuesta de crecimiento medio de las plántulas de “Honey mesquite” al CO_2. Aun, en ausencia de selección, el enriquecimiento de CO_2 pudiera incrementar el establecimiento del “Honey mesquite” al aumentar el crecimiento de las plántulas y por lo tanto exacerbar la meta de manejo de minimizar la invasión de plantas leñosas.

Key Words: genotypic variation, grassland ecology, rangeland, shrub invasion, woody encroachment

INTRODUCTION

The shrub honey mesquite (*Prosopis glandulosa* var. *glandulosa* Torr.) is an aggressive invader of rangelands in the southwestern United States that vary in annual rainfall from 200 to 1000 mm. Mesquite is successful as an invader partly because

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seedlings of the species grow rapidly. By quickly extending a taproot into moist soil at depth, seedlings of honey mesquite may rapidly become uncoupled from competition with established herbaceous vegetation for water and other resources in surface soils (Brown and Archer 1989). Rapid seedling growth also may enhance tolerance of disturbances such as fire and herbivory (e.g., Bond and Midgley 2000).

Atmospheric carbon dioxide (CO_2) enrichment usually increases the biomass or growth rate of honey mesquite seedlings (Polley et al. 1999; Derner et al. 2005) and other plant species (Poorter 1993; Bunce 1997), and has been found to exaggerate interspecific differences in plant growth and other traits (Groninger et al. 1995; Arp et al. 1998; Catovsky and Bazzaz 1999). Indeed, CO_2 enrichment often preferentially stimulates the relative growth rate (RGR; Poorter 1993) or biomass (Bunce 1997) of species that already grow rapidly.

Genotypes within a species also may respond differently to CO_2 (Curtis et al. 1994; Zhang and Lechowicz 1995; Ward and Strain 1997), but this pattern is not consistent (e.g., Lüscher et al. 1998). Roumet et al. (2002) noted that genotypes differed in response to CO_2 enrichment in only 7 of the 21 species that had been studied.

Plant traits that promote rapid growth in mesic sites are thought to be a liability in drier environments (Orians and Solbrig 1977; Smith and Huston 1989). Rapidly growing plants invest heavily in leaves (Tilman 1988; Poorter and Remkes 1990) and may exhibit high rates of transpiration per unit of leaf dry mass (Orians and Solbrig 1977), traits that may not be conducive to growth or survival when soils dry. If a tradeoff exists between rapid growth and tolerance of water limitation, mesquite from extremes of the precipitation gradient would be anticipated to differ genetically in seedling growth rate.

We grew progeny from adult mesquite plants (open-pollinated families) from mesic (central Texas) and arid (southeastern New Mexico) rangelands in glasshouses at near ambient (391 $\mu mol \cdot mol^{-1}$) and at elevated CO_2 concentrations (706 $\mu mol \cdot mol^{-1}$). Well-watered seedlings of 7 families from each extreme of the precipitation gradient were harvested over days 10 to 30 after emergence to measure growth. With these data, we addressed 2 primary questions: 1) Does honey mesquite exhibit intraspecific variation in biomass or growth rate under favorable growth conditions? 2) How will intraspecific variation be impacted by CO_2 enrichment? Consistent with recent studies indicating that CO_2 enrichment often exaggerates interspecific growth differences (Poorter 1993; Bunce 1997), we hypothesized that increasing CO_2 would reinforce any differences in biomass among mesquite families by stimulating biomass proportionally more in families with the greatest RGR at ambient CO_2. If greater biomass enhances seedling establishment or survival, CO_2 enrichment thus could accelerate the rate at which larger or more responsive genotypes dominate mesquite populations and exacerbate the challenge faced by rangeland managers to minimize mesquite encroachment.

METHODS

Plant Culture

Seeds were collected during the summers of 1995 and 1996 from 7 honey mesquite plants located near Temple in central Texas (mean precipitation = 880 mm) and from 7 plants located on the Jornada Experimental Range, north of Las Cruces in southeastern New Mexico (mean precipitation = 200 mm). To ensure that different genotypes (families) were sampled in each rangeland, trees from which seeds were collected were located at least 2 km from major roads and were separated by at least 5 km. Honey mesquite is an outcrosser, so seeds from a given plant are half-sibs (Simpson 1977). To ensure that seeds from each family were of uniform size, seeds were individually weighed. Seeds were discarded if mass differed by more than 1 standard deviation from the family mean. We had not previously worked with the families investigated in this experiment.

For each family, we planted 1 scarified seed into each of 80 pots (6.2 cm diameter and 25 cm deep) containing commercial potting soil on 17 May 1997. Following emergence of most seedlings 3 days later, we selected 60 pots per family with seedlings of uniform height and cotyledon size. These 60 pots then were randomly divided between 2 glasshouse bays that were maintained at nominal CO_2 concentrations of either 390 or 700 $\mu mol \cdot mol^{-1}$, the latter representing the CO_2 level predicted by the end of the century. Position of pots within each bay was randomized every 2 days. To minimize any effect of glasshouse bay on plant performance, plants and the appropriate CO_2 treatment were switched on day 15 between bays. We added 100 ml of Hoagland's nutrient solution (Hoagland and Arnon 1950) to each pot on days 3, 10, 17, and 24 post-emergence. Distilled water was applied as necessary to maintain soil near drip capacity.

Plants were harvested on days 10, 15, 20, 25, and 30 following emergence ($n = 4$ plants per family and CO_2 treatment, with a double harvest [$n = 8$] on days 10 and 30). At each harvest, we measured leaf area per plant (Model LI-300A, Li-Cor, Inc., Lincoln, NE) and washed soil from roots. All plant material was weighed after oven-drying at 60°C for 72 hours. The stable carbon (C) isotope composition of the sixth true leaf of each plant harvested on day 30 was measured by mass spectrometry and expressed as $\delta^{13}C$, ‰ (parts per thousand) ^{13}C relative to a PeeDee belemnite reference standard (Isotope Services, Inc, Los Alamos, NM). Leaf $\delta^{13}C$ values have been shown to correlate negatively with transpiration rates per unit of leaf area (Poorter and Farquhar 1994) and positively with leaf water-use efficiency, the ratio of leaf net photosynthetic rate to transpiration rate (Farquhar et al. 1989).

Growth and Statistical Analyses

For each family and CO_2 treatment, we calculated means of standard growth analysis parameters with data collected over the 10–30 day interval. RGR, which represents the change in biomass per unit of biomass and time, was calculated as the slope of the linear relationship between the natural logarithm (ln) of total biomass and time. RGR is divisible into components of net assimilation rate (NAR) and leaf area ratio (LAR), the latter of which is divisible into components of specific leaf area (SLA) and leaf weight ratio (LWR). NAR (the change in biomass per unit of leaf area and time) was calculated with a formula from classical growth analysis appropriate for the linear relationship that existed between leaf area and biomass (Radford 1967; Hunt 1982). LAR (the ratio of leaf area to total biomass) and LWR (the

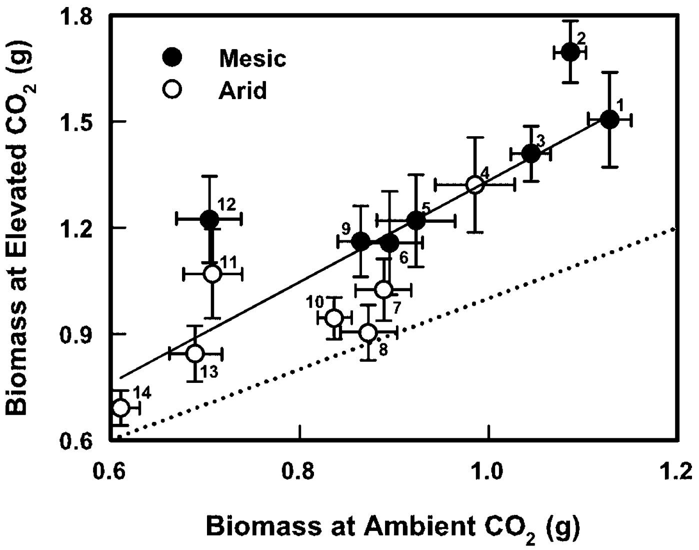

Figure 1. Mean ± SE ($n = 8$) values of total biomass of seedlings from 14 families of honey mesquite (7 families each from arid and mesic rangelands) at elevated and ambient CO_2 concentrations on day 30 following emergence. Numbers indicate the rank order of families by biomass at ambient CO_2. The dashed line denotes the 1:1 relationship. The solid line is a linear regression of biomass at elevated CO_2 on biomass at ambient CO_2 ($P = 0.0003$, $r^2 = 0.67$).

ratio of leaf weight to total biomass) were calculated as the slopes of linear regressions (with intercepts through the origin) between leaf area and leaf weight and total biomass. SLA (the ratio of leaf area to leaf biomass) was calculated only for plants harvested on day 30. Regression slopes between ln (biomass) and time and between both leaf area and leaf weight and total biomass were compared with the F statistic (Weisberg 1980).

For each family and CO_2 treatment, we also calculated allometric coefficients to describe variation in the biomass of leaves or roots with changes in the total biomass of seedlings (e.g., Stirling et al. 1998). The allometric coefficient was calculated as the slope of the linear regression between the natural logarithm of the biomass of leaves or roots and the natural logarithm of total biomass. Regression slopes were compared with the F statistic (Weisberg 1980).

Data collected on days 10 and 30 were analyzed with a 2-way analysis of variance (ANOVA) that included CO_2 treatment, family identity, and the CO_2 × family interaction (Sokal and Rohlf 1981). Significant differences among means were assessed with Student–Newman–Keuls multiple range test.

CO_2 Control and Environmental Conditions

The CO_2 concentration of air in each glasshouse bay was measured at 4-minute intervals with a Li-Cor Model LI-6262 infrared gas analyzer (Li-Cor) that was calibrated daily against CO_2 gas standards. Air temperature was measured in the center of each bay and outside the glasshouse with fine-wire (25-μm diameter) thermocouples. Photosynthetic photon flux density (PPFD) was measured on the glasshouse roof with a silicon photodiode (LI-190SB, Li-Cor) and above plants in each bay with silicon detectors along a 1-m-long sensing surface (LI-191SA, Li-Cor).

Pure CO_2 gas was injected into the appropriate bay as required to maintain the elevated concentration. Air temperature within air-conditioned bays was maintained near that of outdoors by manually adjusting thermostatic controls.

The CO_2 concentration of air in 'ambient' and 'elevated' CO_2 treatments averaged 391 and 706 $\mu mol \cdot mol^{-1}$, respectively. Standard deviations of CO_2 concentration were calculated daily, and averaged 8.3 $\mu mol \cdot mol^{-1}$ at elevated CO_2 and 23.6 $\mu mol \cdot mol^{-1}$ at the ambient CO_2 where concentration was not directly controlled. Means of daytime temperature were 27.9°C at elevated CO_2 (range among days = 27.0° to 29.3°C) and 28.7°C at ambient CO_2 (range among days = 27.1° to 29.8°C). The PPFD inside bays during daylight averaged 700 and 686 $\mu mol \cdot m^{-2} \cdot s^{-1}$ at elevated and ambient CO_2, respectively, approximately 82% of the daytime mean measured above the glasshouse during the experiment (839 $\mu mol \cdot m^{-2} \cdot s^{-1}$).

RESULTS

Seedling biomass varied considerably among families (Fig. 1). Biomass on day 30 differed by almost a factor of 2 among the 14 families at ambient CO_2 (range = 0.61 to 1.13 g). Seedling RGR differed little among the 14 families at ambient CO_2 (range = 0.120 to 0.141 $g \cdot g^{-1} \cdot d^{-1}$; Table 1). Consequently, much of the biomass variation among families at ambient CO_2 was determined by differences in seed mass (Table 2). Families from extremes of the precipitation gradient clearly did not segregate by size (Fig. 1), but the inclusion of families from both arid and mesic rangelands increased variation for seedling biomass. Families with the largest seedlings originated from mesic grassland in central Texas, whereas families with the smallest seedlings originated from arid rangeland in southeastern New Mexico.

Seedling biomass on day 30 was greater at elevated than at ambient CO_2 (Fig. 1). The positive effect of CO_2 on biomass was evident even by day 10, however. Elevated CO_2 increased seedling biomass on day 10 by a mean of 23% across families (mean [± SE] biomass = 0.065 [0.001] and 0.081 [0.001] g at ambient and elevated CO_2, respectively; $P < 0.0001$, $n = 112$). This implies that CO_2 enrichment also increased RGR during the initial 10 days following emergence. Indeed, when calculated using mean seed mass per family as initial biomass, RGR during the initial 10 days of seedling growth increased across families by 39% at elevated compared to ambient CO_2 (0.075 and 0.054 $g \cdot g^{-1} \cdot d^{-1}$, respectively). The family × CO_2 interaction for day 10 biomass was not significant ($P = 0.86$), but the effects of elevated CO_2 on biomass differed among families on day 30 (the family × CO_2 interaction was significant, $P = 0.02$). The ratio of biomass at elevated CO_2 to biomass at ambient CO_2 on day 30 varied from 1.03 to 1.74 among families and was highly correlated with the stimulation of RGR over days 10 to 30 ($P < 0.0001$, $r^2 = 0.78$; Fig. 2). The increase in RGR was, in turn, better correlated with the increase in NAR ($P = 0.02$, $r^2 = 0.37$; Fig. 3) than with change in the LAR ($P = 0.24$, $r^2 = 0.12$; Table 2), although the proportional change from smallest to largest values among families was similar for NAR and LAR (Table 1). CO_2 enrichment increased both the absolute and proportional differences in biomass between the families with the largest and smallest seedlings (Fig. 1), but stimulation of biomass at

Table 1. Mean values of growth analysis parameters for seedlings from 14 mesquite families grown at ambient CO_2 (391 $\mu mol \cdot mol^{-1}$) for 30 days following emergence. Parentheses contain ratios of values measured at elevated CO_2 (706 $\mu mol \cdot mol^{-1}$) to values measured at ambient CO_2. Growth analysis parameters include relative growth rate (RGR; $g \cdot g^{-1} \cdot d^{-1}$), net assimilation rate (NAR; $g \cdot m^2 \cdot d^{-1}$), leaf area ratio (LAR; $m^2 \cdot g^{-1}$), specific leaf area (SLA; $m^2 \cdot g^{-1}$), and leaf weight ratio (LWR; $g \cdot g^{-1}$). Numbers assigned to families indicate the rank order of families by biomass at ambient CO_2. Families originated from arid (A) or mesic (M) rangeland. Note that the implied equality between a growth analysis parameter and the product of the parameter's multiplicative components (i.e., RGR = NAR × LAR) does not hold for mean values reported here (Radford 1967).

Family	Origin	RGR	NAR	LAR	SLA	LWR
1	M	0.141 (0.96)	12.87 (1.18)	0.0108 (0.84)	0.0180 (0.85)	0.622 (1.02)
2	M	0.133 (1.09)	14.36 (1.27)	0.0089 (0.85)	0.0166 (0.81)	0.567 (1.04)
3	M	0.135 (1.02)	14.56 (1.35)	0.0097 (0.75)	0.0180 (0.78)	0.560 (1.00)
4	A	0.133 (0.98)	15.38 (1.15)	0.0084 (0.87)	0.0173 (0.87)	0.536 (0.98)
5	M	0.136 (0.99)	13.03 (1.11)	0.0107 (0.94)	0.0191 (0.93)	0.601 (0.99)
6	M	0.120 (1.02)	12.08 (1.29)	0.0111 (0.74)	0.0201 (0.85)	0.593 (0.94)
7	A	0.128 (0.93)	15.44 (1.01)	0.0086 (0.98)	0.0179 (1.00)	0.499 (1.00)
8	A	0.126 (0.98)	13.04 (1.32)	0.0094 (0.83)	0.0188 (0.83)	0.527 (1.03)
9	M	0.132 (1.01)	13.73 (1.17)	0.0097 (0.87)	0.0179 (0.87)	0.565 (0.99)
10	A	0.118 (0.99)	11.14 (1.24)	0.0104 (0.83)	0.0195 (0.89)	0.563 (0.97)
11	A	0.124 (1.03)	13.36 (1.46)	0.0097 (0.75)	0.0193 (0.85)	0.532 (0.92)
12	M	0.126 (1.14)	13.07 (1.34)	0.0103 (0.86)	0.0181 (0.84)	0.587 (1.01)
13	A	0.124 (1.00)	12.30 (1.15)	0.0105 (0.83)	0.0207 (0.85)	0.552 (1.01)
14	A	0.123 (0.96)	12.26 (1.13)	0.0094 (0.95)	0.0198 (0.88)	0.540 (0.99)

elevated CO_2 was not related to either RGR over days 10 to 30 or day 30 biomass at ambient CO_2 ($P > 0.38$; Table 2).

The benefits of greater NAR to RGR at elevated CO_2 were partly offset by declines in LAR that resulted mostly from reductions in SLA (Fig. 4). Changes in LWR were not significantly correlated with the response of NAR to CO_2 enrichment (Table 2). Because of offsetting changes in NAR and LAR, CO_2 enrichment did not significantly affect the RGR of seedlings over days 10 to 30 when assessed across families ($P = 0.50$). Seedling RGR was slightly, but significantly, greater across CO_2 treatments for families from mesic than from arid rangelands, as determined by comparing the slopes of linear regressions of ln (biomass) on time ($P < 0.01$; 0.134 and 0.124 $g \cdot g^{-1} \cdot d^{-1}$, respectively).

NAR was linearly correlated with leaf $\delta^{13}C$ values at both ambient ($P = 0.03$, $r^2 = 0.34$) and elevated CO_2 concentrations ($P = 0.02$, $r^2 = 0.36$), such that greater values of NAR were associated with less negative values of $\delta^{13}C$ (Fig. 5). Elevated CO_2 increased the allometric coefficient of biomass partitioning to roots from 0.93 to 0.97 ($P < 0.005$), but did not alter the allometry of biomass partitioning to leaves ($P > 0.75$). For a given value of total biomass, therefore, seedlings grown at elevated CO_2 had greater root biomass.

Table 2. P values and r^2 values from linear regression analyses of relationships among seedling biomass (day 30), seed mass, and growth analysis parameters (relative growth rate, RGR; leaf area ratio, LAR; net assimilation rate, NAR; leaf weight ratio, LWR) of seedlings from 14 mesquite families grown at elevated (706 $\mu mol \cdot mol^{-1}$) or ambient (391 $\mu mol \cdot mol^{-1}$) CO_2 concentrations for 30 days following emergence.

Dependent variable	Independent variable	r^2	P value
Biomass (ambient)	Seed mass	0.66	0.0005
RGR (elevated/ambient)	LAR (elevated/ambient)	0.12	0.24
Biomass (elevated/ambient)	RGR (ambient)	0.06	0.39
Biomass (elevated/ambient)	Biomass (ambient)	0.005	0.82
NAR (elevated/ambient)	LWR (elevated/ambient)	0.004	0.33

DISCUSSION

Intra-specific Variation in Biomass and CO_2 Response

The 14 families of honey mesquite we studied varied considerably in both seedling biomass (0.61 to 1.13 g) and in the proportional

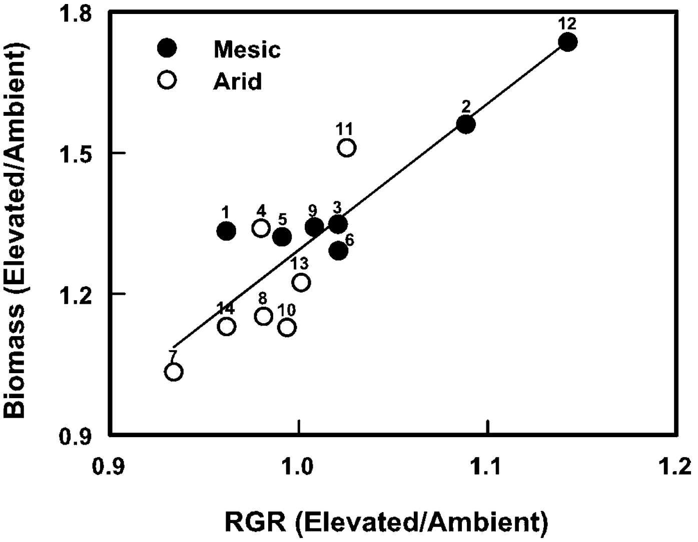

Figure 2. The relationship between the mean relative increase in total biomass of seedlings from 14 families of honey mesquite (7 families each from arid and mesic rangelands) at elevated CO_2 and the proportional increase in relative growth rate (RGR) of seedlings at elevated CO_2. Biomass was measured on day 30 following emergence. RGR was calculated over days 10 to 30. The line is a linear regression between the stimulation of biomass and the increase in RGR ($P < 0.0001$, $r^2 = 0.78$). Numbers indicate the rank order of families by biomass at ambient CO_2.

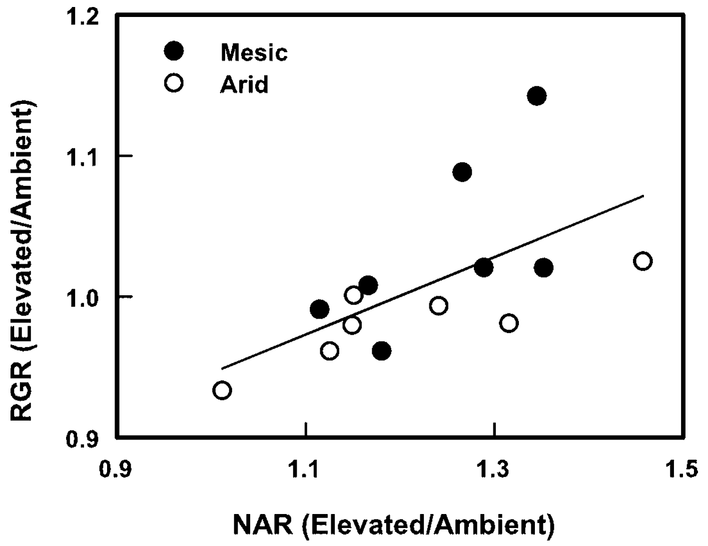

Figure 3. The relationship between the proportional increase in relative growth rate (RGR) of seedlings from 14 families of honey mesquite (7 families each from arid and mesic rangelands) at elevated CO_2 and the relative increase in net assimilation rate (NAR) of seedlings at elevated CO_2. RGR and NAR were calculated over days 10 to 30 following emergence. The line is a linear regression between the stimulation of RGR and the increase in NAR ($P = 0.02$, $r^2 = 0.37$).

response of biomass to CO_2 enrichment (1.03 to 1.74). Significantly, however, the proportional response of biomass to CO_2 enrichment was not well-correlated across families, with variation in either seedling biomass or RGR at ambient CO_2. Although CO_2 enrichment exaggerated size inequities among families, elevated CO_2 did not consistently favor the families with the largest or fastest-growing seedlings at ambient CO_2. These results are contrary to both our hypothesis and to the pattern that has been observed in interspecific comparisons (Poorter 1993; Bunce 1997; Atkin et al. 1999). Biomass stimulation at elevated CO_2 may have differed among families because of RGR differences that existed at ambient CO_2 and because of differences in the responses of RGR to CO_2 (Poorter 1993). Families with the most rapidly growing seedlings at ambient CO_2 will exhibit the greatest proportional increases in biomass at elevated CO_2 if CO_2 enrichment stimulates RGR similarly among families (Bunce 1997). The greater the differences in RGR among families at ambient CO_2, the more consistent will be the relationship between stimulation in biomass at elevated CO_2 and RGR at ambient CO_2. By contrast, variation in biomass responses to CO_2 must be determined primarily by differences in the stimulation of RGR when, as here, RGR at ambient CO_2 is similar among the plant groups investigated.

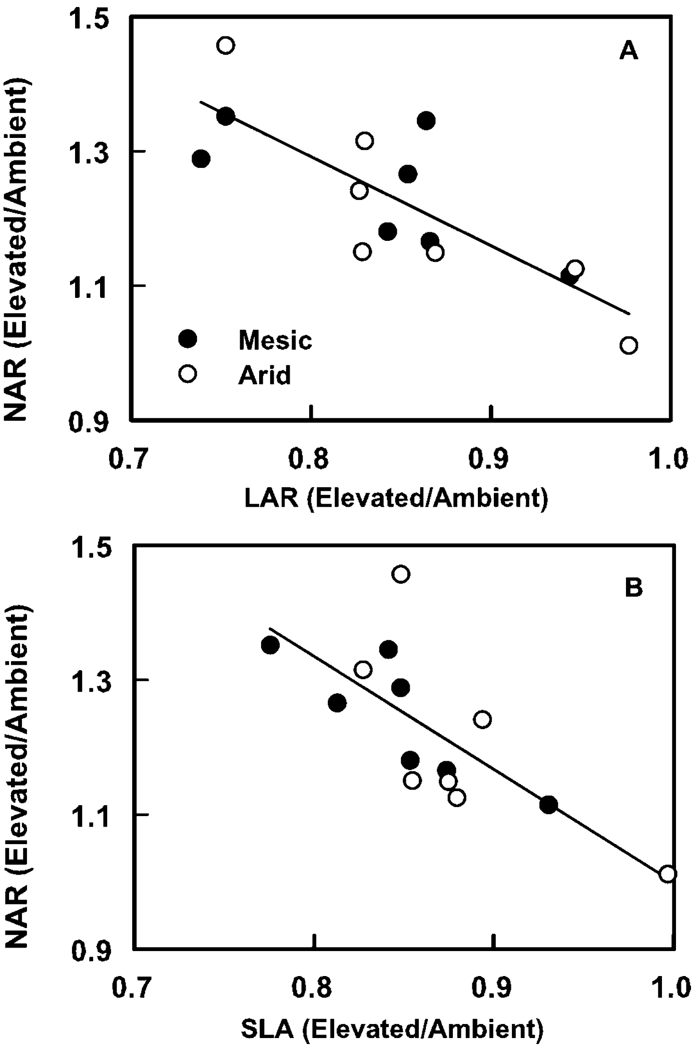

Figure 4. The relationship between the proportional increase in net assimilation rate (NAR) of seedlings from 14 families of honey mesquite (7 families each from arid and mesic rangelands) at elevated CO_2 and relative changes in (**A**) leaf area ratio (LAR) and (**B**) specific leaf area (SLA) of seedlings at elevated CO_2. Growth analysis parameters were calculated over days 10 to 30 following emergence. Lines are linear regressions between the stimulation of NAR and changes in LAR ($P = 0.0005$, $r^2 = 0.65$) and SLA ($P = 0.002$, $r^2 = 0.56$).

Differences in the responses of mesquite families to CO_2 enrichment were linked most directly to differences in the responses of NAR, rather than to changes in LAR. That is, CO_2 enrichment stimulated growth primarily by increasing rates of net carbon uptake (photosynthesis minus plant respiration) per unit of leaf area. This link between the response of RGR to CO_2 and CO_2 effects on NAR has been noted in studies which included several species (Bunce 1997; Tischler et al. 2000), but has not, to our knowledge, been reported for intraspecific comparisons. Contrary to the pattern reported by Bunce (1997), increases in NAR at elevated CO_2 were correlated with decreases in both LAR and SLA that lessened the sensitivity of RGR to CO_2 enrichment. A negative correlation between NAR and both LAR and SLA is common in studies in which the RGR of species or genotypes is similar (Konings 1990). Atkin et al. (1999) observed that declines in SLA partly offset the positive effects of CO_2 enrichment on NAR among 10 species of *Acacia*. Across families, CO_2 enrichment also increased the allometric coefficient of biomass partitioning to roots. Rarely has CO_2 treatment been found to alter the allometry of biomass partitioning (Farrar and Williams 1991).

At both ambient and elevated CO_2, greater values of NAR were associated with less negative values of $\delta^{13}C$. That is, NAR increased among mesquite families as leaf water use efficiency increased. Just the opposite relationship might be expected. Rapid rates of leaf gas exchange (presumably reflected in greater values of NAR) often are thought to be associated with reduced values of water use efficiency (more negative values of $\delta^{13}C$; Ehleringer et al. 1990). Poorter and Farquhar (1994), by contrast, found no correlation between leaf C isotope values and NAR among the 24 herbaceous species they studied. The positive correlation between NAR and leaf water use efficiency coupled with the negative relationship across families between change in NAR and change in LAR at elevated CO_2 implies that families that reduce LAR the most at elevated CO_2 will realize the largest gain in water use efficiency.

Seedlings derived from mesic environments generally were larger, grew more rapidly, and showed a greater response to CO_2 enrichment than did those from arid environments, but biomass and growth did not differ consistently between honey mesquite families from extremes of the precipitation gradient. Nevertheless, the inclusion of families from extremes of the species' distribution range increased variation in mean biomass of honey mesquite seedlings. Among-family variation in seedling biomass at ambient CO_2 was, in turn, highly correlated with mean seed mass. This correlation may have been strengthened by limiting variation in seed mass to ± 1 standard deviation of the mean for each family before planting. Contrary to the general pattern observed here of greater seed mass in mesquite families from mesic than from arid rangeland, mean seed mass has been found to decrease as average precipitation rises in some studies involving multiple species (Baker 1972; but see Westoby et al. 1992).

MANAGEMENT IMPLICATIONS

Atmospheric CO_2 enrichment increased seedling biomass in most families of honey mesquite and increased the inequity of seedling size among families under the favorable growth conditions employed. However, it remains to be established whether these intraspecific differences in seedling growth or in seedling response to CO_2 ultimately translate into differences in reproductive output (fitness) of mesquite. The most direct way in which CO_2 enrichment could change the relative abundances of genotypes is by differentially influencing seedling survivorship. Mesquite is most vulnerable to disturbances and to competition from neighboring plants as newly emerged seedlings (Brown and Archer 1989; Bond and Midgley 2000; Polley et al. 2002). In ungrazed grassland, seedling survivorship frequently is low because of competition from grasses for both light (Brown and Archer 1989; Bush and Van Auken 1990) and soil resources (Brown and Archer 1989; Van Auken and Bush 1997; Polley et al. 2002). By growing more rapidly, mesquite seedlings may more quickly become uncoupled from grass competition for essential resources (Brown and Archer 1989; Van Auken and Bush 1997). If survivorship during establishment in intact vegetation is positively correlated with seedling size or growth rate in mesquite, as it often is when multiple species are compared (Jakobsson and Eriksson 2000), CO_2 enrichment thus may enhance selection for the largest or most responsive genotypes. Larger seedlings also have a competitive advantage over smaller seedlings (Westoby et al. 1992; Turnbull et al. 1999), an advantage that may, in turn, affect fitness. Large size or rapid growth, therefore, may be especially advantageous to mesquite seedlings that emerge with seedlings of other rangeland plants. The extent to which rising CO_2 affects selection in mesquite will depend partly on whether neighboring plants are as sensitive to CO_2 enrichment as mesquite. Biomass of most plants increases at elevated CO_2, but growth generally is less responsive to CO_2 in C_4 plants, such as the dominant grasses in mesquite-invaded rangeland, than in C_3 species (Poorter 1993).

The variation in seedling responses to elevated CO_2 we documented may not translate directly to field settings because responsiveness to CO_2 depends on environmental conditions and on the intensity of plant–plant interactions (Schmid et al. 1996). Biomass partitioning patterns change with plant size, so our results with seedlings also may not directly translate to larger mesquite plants. Ultimately, however, traits that underlie differences in plant performance, rather than composite metrics like size or growth rate, likely will be the targets of selection at elevated CO_2. The response of NAR to CO_2 varied considerably among the mesquite families we studied, indicating that

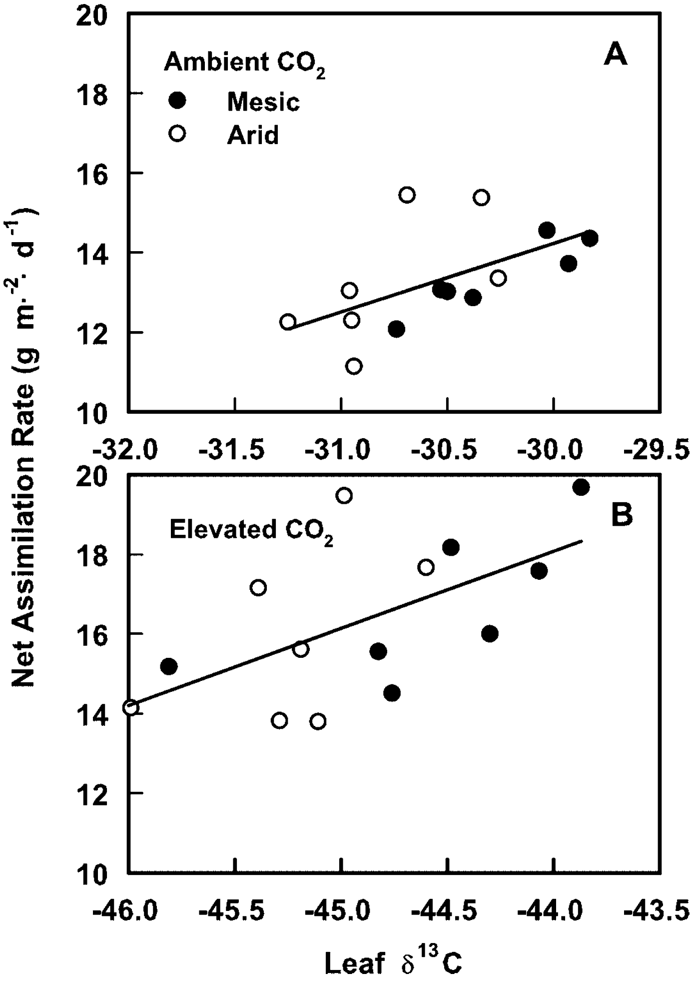

Figure 5. Relationships between mean values of net assimilation rate (NAR) and leaf $\delta^{13}C$ for seedlings from 14 families of honey mesquite (7 families each from arid and mesic rangelands) at (**A**) ambient and (**B**) elevated CO_2 concentrations. NAR was calculated over days 10 to 30 following emergence. The $\delta^{13}C$ was measured on leaves collected on day 30. Standard errors for $\delta^{13}C$ values ranged from 0.080 to 0.235 at ambient CO_2 and from 0.189 to 0.369 at elevated CO_2. Lines are linear regressions between NAR and leaf $\delta^{13}C$ values ($P = 0.03$, $r^2 = 0.34$ at ambient CO_2 and $P = 0.02$, $r^2 = 0.36$ at elevated CO_2). Note that leaf $\delta^{13}C$ values were more negative at elevated than at ambient CO_2 because ^{13}C-depleted CO_2 was used for enrichment.

variation in physiological processes might be large enough for directional selection to occur. Selection for greater NAR could augment the mean growth response of honey mesquite to CO_2 and exacerbate the challenge faced by rangeland managers to minimize mesquite encroachment. Even in the absence of selection, the ongoing increase in atmospheric CO_2 concentration could enhance the establishment or survival of honey mesquite by enhancing seedling growth and thereby accelerate woody encroachment on both arid and mesic rangelands.

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