Oecologia (1994) 97:100–105
© Springer-Verlag 1994

ORIGINAL PAPER

Peter S. Curtis · Allison A. Snow · Amy S. Miller

Genotype-specific effects of elevated CO_2 on fecundity in wild radish (*Raphanus raphanistrum*)

Received: 16 June 1993 / Accepted: 8 November 1993

Abstract Rising atmospheric CO_2 may lead to natural selection for genotypes that exhibit greater fitness under these conditions. The potential for such evolutionary change will depend on the extent of within-population genetic variation in CO_2 responses of wild species. We tested for heritable variation in CO_2-dependent life history responses in a weedy, cosmopolitan annual, *Raphanus raphanistrum*. Progeny from five paternal families were grown at ambient and twice ambient CO_2 using outdoor open-top chambers (160 plants per CO_2 treatment). Elevated CO_2 stimulated net assimilation rates, especially in plants that had begun flowering. Across paternal families, elevated CO_2 led to significant increases in flower and seed production (by 22% and 13% respectively), but no effect was seen on time to bolting, leaf area at bolting, fruit set, or number of seeds per fruit. Paternal families differed in their response to the CO_2 treatment: in three families there were no significant CO_2 effects, while in one family lifetime fecundity increased by >50%. These genotype-specific effects altered fitness rankings among the five paternal families. Although we did not detect a significant genotype × CO_2 interaction, our results provide evidence for heritable responses to elevated CO_2. In a subset of plants, we found that the magnitude of CO_2 effects on fecundity was also influenced by soil fertility.

Introduction

Atmospheric carbon dioxide partial pressure has risen approximately 25% since the beginning of the industrial revolution and is now increasing at a rate of 1.5 μbar per year (Conway et al. 1988). With a continuation of current patterns of fossil fuel use, atmospheric CO_2 is expected to double within 60 to 80 years. Different species and cultivars vary widely in their response to elevated CO_2 (Hunt et al. 1991; Ziska and Teramura 1992), yet little is known about whether variation also occurs within natural populations. Documenting genetic variation in the effects of elevated CO_2 on growth and reproduction is an important step toward understanding the potential evolutionary consequences of global climate change (Geber and Dawson 1992). If some genotypes respond more than others due to genotype × CO_2 environment interactions, then higher CO_2 should favor a new set of genotypes. Rapid evolutionary change could thereby boost the average growth response to elevated CO_2 in natural populations. Of particular interest, then, is the extent of within-population genetic variation that could affect the course of natural selection as CO_2 continues to increase.

To date, only two studies have included explicit consideration of genotype-specific CO_2 responses in wild populations. Wulff and Alexander (1985) found significant differences in the germination and growth of progeny derived from 5 maternal families (clones) of *Plantago lanceolata* grown at 350 or 675 μbar CO_2. Some families responded much more to CO_2 and temperature than others, suggesting that there may be genetic variation in response to CO_2 during seed maturation and seedling development. Also working with *Plantago lanceolata*, Fajer et al. (1992) examined growth and leaf biochemistry in six clones, but they found no genotypic differences in response to CO_2.

We investigated genetic variation in responses to rising atmospheric CO_2 in a weedy annual, *Raphanus raphanistrum* (wild radish). This species has a cosmopolitan distribution; in northeastern North America, it occurs primarily in agricultural fields and on sheltered beaches. *Raphanus raphanistrum*, and its closely related congener *R. sativus*, have been the subject of several life history and quantitative genetic studies (e.g., Stanton

P. S. Curtis (✉) · A. A. Snow
Department of Plant Biology 1735 Neil Avenue,
Ohio State University,
Columbus, OH 43210–1293, USA

A. S. Miller
Department of Botany and Plant Pathology and University of Michigan Biological Station, Pellston, MI 49769, USA,
Oregon State University,
Corvallis, OR 97331, USA

1984a, b, 1985; Stanton and Preston 1988; Mazer 1987a, b; Mazer et al. 1986). Working with *R. sativus*, Mazer and Schick (1991) and Mazer and Wolfe (1992) found significant genotype × environment interactions in the expression of life history and floral traits. Both wild radish (*R. sativus* × *raphanistrum*, Chu et al. 1992) and commercial *R. sativus* cultivars (Overdieck et al. 1988; Idso and Kimball 1989; Barnes and Pfirrmann 1992) have been grown at elevated CO_2, and in each case a stimulation of growth was reported.

Our objective was to test the hypothesis that genetic variation in growth responses to elevated CO_2 is present in a natural population. We describe general physiological responses and genotype-specific reproductive success of plants grown at ambient and twice ambient CO_2. We focused on life history responses, especially lifetime seed production, in order to estimate the relative fitness of genotypes grown at ambient vs. elevated CO_2.

Materials and methods

Breeding design

Seeds for controlled crosses were collected from a single beach population at Deer Isle, Maine, USA, and grown under greenhouse conditions at the Ohio State University. Five individuals from separate maternal plants were randomly selected to serve as pollen donors, and 20 randomly selected individuals, also from separate maternal plants, served as pollen recipients. Each pollen donor (male parent) was crossed with four recipients (female parents) in a nested design (females within males), yielding five paternal families.

With this design, significant paternal effects are interpreted as evidence for additive genetic variance among paternal genotypes (c.f., Mazer and Schick 1991). By measuring paternal effects rather than maternal or clonal variation, we minimized the possibility of detecting phenotypic variation that lacks a genetic basis.

Field experiment

The CO_2 exposure experiment was conducted at the University of Michigan Biological Station in northern Lower Michigan, USA. Four outdoor open top chambers, 3.1 m in diameter and 4 m in height (Heagle et al. 1979), were used to establish the CO_2 treatments. Two chambers received additional CO_2 by dispensing 100% CO_2 into the ventilation fans (Elevated treatment). Monitoring and control of chamber CO_2 partial pressure was as described by Curtis and Teeri (1992) except CO_2 measurements were recorded every 8 minutes and logged onto a personal computer. Two chambers received no additional CO_2 (Ambient treatment). Temperature inside and outside chambers was monitored using shaded thermocouples attached to an LI-1000 datalogger (LICOR Inc., Lincoln NB). Daytime (0700–1900) CO_2 partial pressure averaged 685 ± 40 μbar (mean ± s.d.) inside Elevated chambers and 338 ± 49 μbar inside Ambient chambers. Temperature was $1.0 \pm 0.6°$ C higher inside chambers than outside but there was no difference between Elevated and Ambient chambers.

Seeds were planted on 28 June, 1992, in 2.5 L pots filled with a mixture of locally derived Rubicon sand and Kalkaska series topsoil (one seed per pot). The bottom 1.5 L of each pot was filled with sand and the top 1.0 L was filled with a 1:1 by volume mixture of sand and topsoil. Roots were allowed to penetrate the mesh bottom of each pot and grow into the Rubicon sand subsurface soil. All pots were watered daily as needed, but received no nutrient additions.

Eight seeds from each of 20 female parents were randomly assigned to each of the two CO_2 treatments and weighed to the nearest 0.01 mg. Four pots per female were placed in each chamber, for a total of 160 pots per treatment (5 paternal families × 4 maternal families × 4 replicates per female × 2 chambers). All pots within a chamber were rotated to another chamber weekly for the first 6 weeks to minimize chamber effects. During these rotations, plants from a given chamber always stayed together and were designated as Groups 1–4 for statistical analysis. After week 6, most plants had bolted and were too large to move without risk of damage. Thus, for the last 7 weeks of the experiment, both chamber and soil conditions (where roots emerged from their pots) could contribute to variation among groups.

Flowering began about 30 days after planting and each flower was hand-pollinated 1–2 d after opening. Hand-pollination was necessary because this species is self-incompatible (Samson 1967). Pollinations were performed using tissue-covered forceps that were dusted with pollen from several randomly chosen plants. Carbon dioxide treatments and hand-pollinations were terminated 1 October (96 days after planting) and all plants were moved into a greenhouse where they were allowed to senesce.

Leaf gas exchange

Net assimilation was determined on a set of 20 plants 57 days after planting. Two groups of plants within each CO_2 treatment were selected: those that had not yet bolted (vegetative), and those that had recently bolted and were maturing 3–5 fruits (reproductive). Five plants per group and CO_2 treatment were measured, and selection of plants was random with regard to family. Gas exchange was measured with an ADC LCA3 photosynthesis system using a water jacketed cuvette. All measurements were made at saturating light intensity (1800 μmol m^{-2} s^{-1}), the appropriate growth CO_2 (350 or 700 μbar), and at a leaf temperature between 25 and 27° C.

Life history traits

All seeds germinated within 4–6 days, so germination date will not be considered further. After producing a basal rosette, the plants bolted, flowered within a few days, and continued to produce flowers for two to three weeks. Early fruit set was near 100% on all plants, declining to zero for later flowers. Days to bolting, leaf area at bolting, total number of flowers produced, and total seed set were recorded for each plant. Total leaf area at bolting was estimated by measuring the length of each leaf (L) and relating this to individual leaf area (A) by the equation $A = 0.53 + 0.4(L)^2$, ($r^2 = 0.98$, $N = 31$). This equation was determined by destructive harvest of a separate set of plants.

Soil fertility experiment

The effect of soil fertility on the CO_2 response was investigated using progeny from a single maternal family, with eight plants per CO_2 and soil treatment. Plants were treated as described above but two levels of soil fertility were used. For the low fertility treatment, 100 ml topsoil was mixed with 900 ml sand in the upper 1 L of each pot. In contrast, 900 ml topsoil was mixed with 100 ml sand in the upper 1 L for the high fertility treatment. Soil fertility in the larger experiment that compared paternal families was intermediate (500 ml topsoil + 500 ml sand). To estimate relative nutrient availability in these soil mixtures, 5.0 g soil was extracted with 2M KCl and analysed for NO_3^- with a Technicon Autoanalyser 2. Extractable NO_3^- for combined subsamples (five per soil type) from the low, intermediate, and high fertility treatments was 5.1, 13.9, and 22.6 mg/kg respectively.

Statistical analysis

The effects of CO_2, father, mother, and group on various life history traits were analysed using the General Linear Models procedure of SAS (SAS Institute 1989), with individual plant as the unit of replication. The main ANOVA model for response (Y) across fathers was:

$$Y_{ijklm} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \gamma_{ik} + \delta_{jl} + \varepsilon_{ijklm}$$

where μ was the parametric mean, α was the effect of CO_2, β was the effect of father, $(\alpha\beta)$ was their interaction, γ was the effect of group nested within CO_2, δ was the effect of mother nested within father, and ε was random error. All effects were considered fixed. Response of progeny from individual fathers was evaluated using the model:

$$Y_{ijkl} = \mu + \alpha_i + \delta_j + (\alpha\delta)_{ij} + \gamma_{ik} + \varepsilon_{ijkl}$$

where symbols are as above and mother (δ) was considered a main effect.

Gas exchange data and responses to soil fertility were analysed as two-way ANOVAs with CO_2 and either reproductive state (vegetative vs reproductive) or soil fertility (low vs high) as main effects. Pairs of treatment means for net assimilation were contrasted by orthogonal comparisons (*a priori* tests); means from the soil fertility experiment were compared using the Minimum Significant Difference (*a posteriori* tests) (Sokal and Rohlf 1981).

Results

General effects of CO_2 on wild radish

Elevated CO_2 stimulated leaf net CO_2 assimilation, as expected, and the magnitude of this effect depended on plant reproductive status (Fig. 1). In vegetative plants, net assimilation increased by 25% at elevated CO_2, while in reproductive plants net assimilation increased by 48%. Fecundity of plants grown at elevated CO_2 was significantly greater than that of plants grown at ambient CO_2 (Tables 1 and 2). The CO_2 treatment increased flower number by 22% and seed number by 13% but had no effect on days to bolting, total leaf area at bolting, percent fruit set, or number of seeds per fruit (Table 2). Regression analysis indicated that initial seed weight had no effect on flower or seed production (N = 150–155 per CO_2 treatment). Much of the variation in flower and seed production was explained by group nested within treatment (Table 1). This effect was stronger for seed production, and may be related to brief aphid infestations or growing conditions in the last chamber used in chamber rotations.

The magnitude of reproductive responses to elevated CO_2 was also influenced by soil fertility (Fig. 2). Effects of soil and CO_2 were significant ($P < 0.05$) for both flower and seed number; the soil × CO_2 interaction was significant ($P < 0.05$) for seeds but not flowers (two-way ANOVAs). At low fertility, elevated CO_2 increased seed set by 53%, while at high fertility seed set increased by more than 300%. Plants growing at high CO_2 responded much more to increased soil fertility, showing a 256% increase in seed set, while there was no significant change in ambient CO_2 grown plants. These results suggest that the plants from the five paternal families, which were grown at an intermediate soil fertility, would show greater CO_2 responses if grown in pure topsoil.

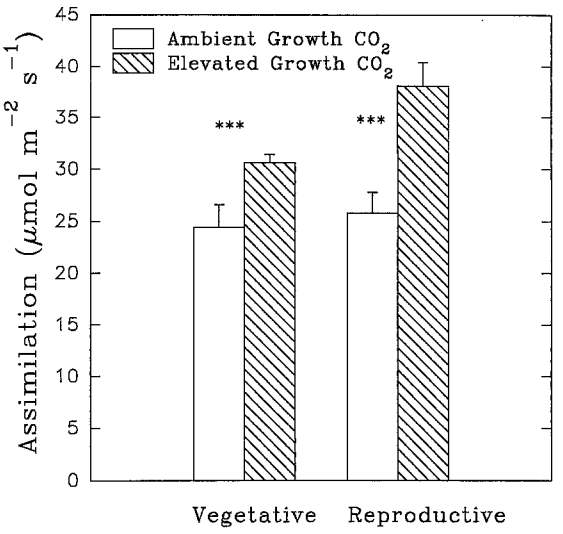

Fig. 1 Effects of CO_2 treatment and growth stage on net assimilation. Plants were grown at either elevated or ambient CO_2. Means ±1 SE; N = 5. CO_2 responses within each growth stage were significant at $P < 0.001$ based on orthogonal comparisons

Table 1 Effects of CO_2, paternal and maternal genotype, and group on life history characteristics. All effects were considered fixed for the analysis of variance

	F Values and Significance			
	Bolt Date[a]	Bolt Area[b]	Flowers	Seeds
CO_2	0.14	0.84	11.76***	5.38*
Father	44.55***	18.59***	7.36***	3.17*
CO_2 × Father	0.65	0.36	0.90	1.13
Mother (Father)	5.05***	7.51***	1.68	2.35**
Group (CO_2)	0.22	1.96	6.48**	11.72***

[a] Days to bolting
[b] Total leaf area at bolting
* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Table 2 Response of life history characteristics to elevated CO_2. Mean (±SE), N = 155–160. Significance tests as in Table 1

	CO_2 Treatment		
	Ambient	Elevated	$P <$
Days to bolting	36 (1)	37 (1)	ns
Leaf are at bolting (cm^2)	30 (2)	29 (2)	ns
Percent fruit set	43 (1)	42 (1)	ns
Seeds per fruit	2.7 (0.6)	2.7 (0.6)	ns
Flowers per plant	95 (4)	116 (5)	0.001
Seeds per plant	107 (6)	121 (7)	0.05

Comparisons among paternal families

Paternal family had a significant effect on several life history characteristics, irrespective of CO_2 treatment

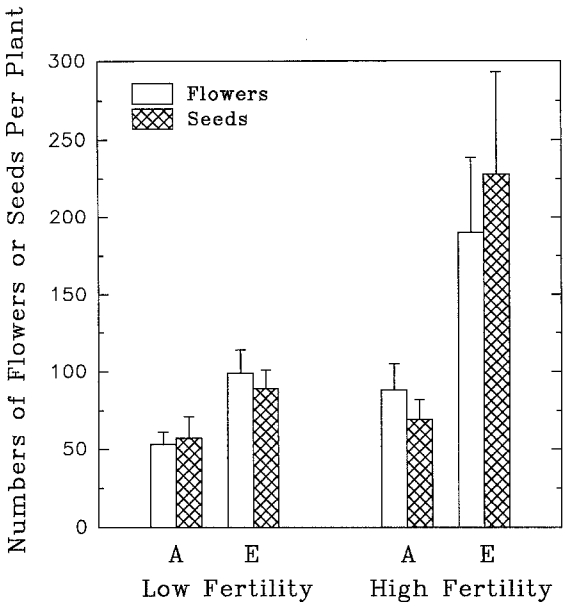

Fig. 2 Effects of CO_2 and soil fertility on lifetime fecundity. Means ± 1 SE, N = 7–8: *A* = ambient CO_2, *E* = elevated CO_2. A two-way ANOVA showed that effects of CO_2 and soil type were significant at $P < 0.05$ for both flowers and seeds; their interaction was significant at $P < 0.05$ for seeds only

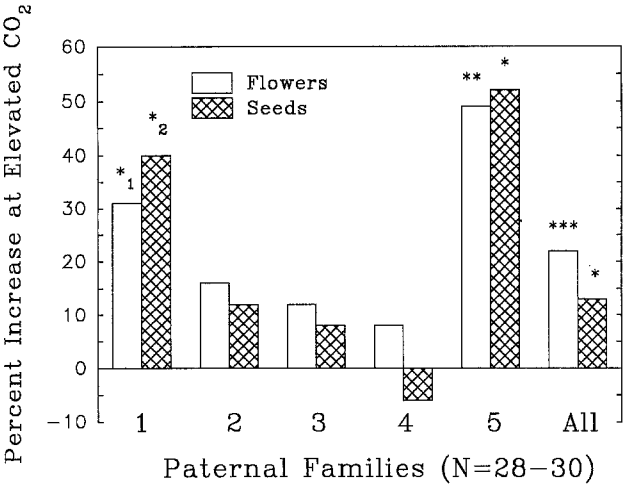

Fig. 3 Differences among paternal families in percent increase in flower and seed production when grown in elevated as compared to ambient CO_2. Significance levels are for effect of CO_2 from two-way ANOVAs within paternal families and from Table 1 for all families combined; *1 $P < 0.06$, *2 $P < 0.08$, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

(Tables 1 and 3). In comparison to Families 1–3, Families 4 and 5 had smaller seeds initially, bolted later, attained a greater size at bolting, and produced fewer flowers and seeds (presumably because they bolted near the end of the growing season). Other investigators have also found evidence for genetic variation in life history traits in *Raphanus* spp. (Mazer 1987a, b; Mazer and Schick 1991; Mazer and Wolfe 1992).

We did not detect significant father × CO_2 interactions in the overall ANOVA (Table 1), but paternal families clearly differed in their response to CO_2 (Table 3; Figure 3). For number of flowers, the response to elevated CO_2 varied between an 8% increase in Family 4 and a 50% increase in Family 5 (Fig 3). Seed number showed a similar pattern. Only two paternal families (1 and 5) showed significant positive response to elevated CO_2, based on N = 28 – 32 (Table 3).

Table 3 Life history characteristics of paternal families grown at Ambient and Elevated CO_2. N = 28–32. Within CO_2 treatments, means of families sharing the same superscript were not significantly different ($P < 0.05$, Tukey tests). Within two families, CO_2 had significant effects on flowers per plant ($P < 0.06$ in Family 1, $P < 0.001$ in Family 5) and on seeds per plant ($P < 0.08$ in Family1, $P < 0.05$ in Family 5); other comparisons between means were not significant (two-way ANOVAs)

Paternal Family		Initial Seed Wt. (mg)		Bolt Date[1]		Bolt Area[2] (cm^2)		Flowers Per Plant		Seeds Per Plant	
		CO_2 Treatment									
		Amb.	Elev.	Amb.	Elev.	Amb.	Elev.	Amb.	Elev.	Amb.	Elev.
1	Mean	6.7^{d}	6.2^{d}	32^{a}	34^{a}	27^{ab}	25^{ab}	108^{b}	141^{b}	97^{ab}	135^{a}
	(SE)	(0.2)	(0.2)	(1)	(1)	(2.3)	(2.2)	(11)	(15)	(14)	(21)
2	Mean	5.7^{c}	5.8^{cd}	29^{a}	28^{a}	20^{ab}	18^{a}	107^{b}	125^{b}	131^{b}	147^{a}
	(SE)	(0.4)	(0.4)	(1)	(1)	(0.9)	(0.8)	(10)	(13)	(15)	(16)
3	Mean	5.7^{c}	5.3^{bc}	33^{a}	33^{a}	23^{ab}	21^{a}	102^{ab}	114^{b}	120^{ab}	130^{a}
	(SE)	(0.2)	(0.3)	(1)	(1)	(1.1)	(2.0)	(10)	(9)	(14)	(17)
4	Mean	4.7^{ab}	4.8^{ab}	41^{b}	42^{b}	35^{b}	37^{bc}	77^{ab}	83^{a}	99^{ab}	93^{a}
	(SE)	(0.2)	(0.2)	(2)	(2)	(4.0)	(6.1)	(7)	(8)	(11)	(12)
5	Mean	4.4^{a}	4.3^{a}	45^{b}	43^{b}	49^{c}	40^{bc}	72^{a}	107^{b}	81^{a}	123^{a}
	(SE)	(0.2)	(0.2)	(2)	(2)	(8.4)	(5.6)	(7)	(10)	(11)	(14)

[1] Days after planting
[2] Leaf area at time of bolting

Discussion

Across the five paternal families, flower and seed production were stimulated by 22% and 13% respectively due to elevated CO_2 (Table 2). Considered separately, however, only two paternal families showed significant positive effects (30–50% increase; Table 3, Fig. 3). Moreover, Families 4 and 5 were similar in size at bolting and time of bolting, yet only Family 5 responded to elevated CO_2. This is the first time that variation among paternal lines has been demonstrated for CO_2 response characteristics. Previous studies have considered vegetative clones (Wulff and Alexander 1985; Fajer et al. 1992) or inbred lines (Ziska and Teramura 1992).

Although we examined only five paternal genotypes, our results suggest that within natural populations of *R. raphanistrum* there is heritable variation in life history responses to elevated CO_2. Since seed set is directly related to lifetime fitness in an annual species, those genotypes that show a greater response to elevated CO_2 should be favored evolutionarily in a future high CO_2 atmosphere. Natural selection would only result, however, if the relative fitness rankings of different genotypes changed with increasing CO_2. Our data suggest this could be the case as the rank order of flower and seed production for two paternal families (Families 1 and 5) changed relative to the others between ambient and elevated CO_2 conditions (see Table 3). We cannot draw a firm conclusion regarding shifts in relative performance, however, given nonsignificant father × CO_2 interactions. Based on the results reported here, we expect that father × CO_2 interactions may be detectable with more genotypes, larger sample sizes, and higher levels of soil fertility. The high soil fertility treatment used with one maternal family led to a much larger response to elevated CO_2 than did low or intermediate topsoil mixtures.

The general stimulation of flower and seed number by elevated CO_2 in *R. raphanistrum* is consistent with observations on a number of crop (Kimball 1983) and native species (Garbutt and Bazzaz 1984). In the annual species *Phlox drummondii*, there were significant differences among populations in reproductive responses to CO_2 (Garbutt and Bazzaz 1984). We found that maternal and paternal genotype, as well as the CO_2 environment, contributed to variation in reproductive output, but CO_2 had no effect on time to bolting or leaf area at bolting. Species appear to be quite variable in flowering time responses to elevated CO_2; both earlier and later flowering have been reported (Reekie and Bazzaz 1991). In our study, paternal and maternal genotypes had significant effects on age and size at reproduction. Unusually cool temperatures during the summer of 1992 generally slowed growth and delayed flowering in our experiment, and this may have obscured CO_2 effects on flowering phenology.

Net assimilation increased at elevated CO_2 in both vegetative and reproductive plants and this stimulation was more pronounced in reproductive plants. The presence of strong carbon sinks, such as developing fruits, may play an important role in determining the timing and degree of stimulation of assimilation by CO_2 (Clough et al. 1981; Peet et al. 1986). In *R. raphanistrum*, greater sink strength resulted in a 92% increase in the degree of CO_2 stimulation of assimilation. Assimilation in ambient CO_2 grown plants was not affected by reproductive status, suggesting greater potential for sink regulation of photosynthesis at elevated CO_2. If a plant becomes source limited during rapid seed fill this assimilatory response to elevated CO_2 could provide carbon for continued reproduction. This would be of considerable ecological importance to indeterminate annuals with high reproductive capacity such as *R. raphanistrum*.

In conclusion, we have shown that lifetime reproduction in *R. raphanistrum* is sensitive to changing CO_2, and that genetically based variation in this response occurs within a single wild population. If such variation is widespread in populations of this species and others, rising atmospheric CO_2 could act as a selective agent, favoring certain genotypes over others. The potential for rapid evolutionary responses to CO_2 and related climatic variables should be considered in evaluating the ecological consequences of global climate change.

Acknowledgements We thank Susan Wadkowski, Jennifer Bashore, Scott Smith, and Chris Vogel for field assistance, Richard Spray for logistical help, and Susan Kalisz, Monica Geber and Susan Mazer for comments on the manuscript. James Teeri and the University of Michigan Biological Station provided important institutional support. This research was funded by grants from The Ohio State University, the National Institute for Global Environmental Change, and NSF's Research Experience for Undergraduates Program.

References

Barnes JD, Pfirrmann T (1992) The influence of CO_2 and O_3, singly and in combination, on gas exchange, growth and nutrient status of radish (*Raphanus sativus* L.). New Phytol 121:403–412

Chu CC, Coleman JS, Mooney HA (1992) Controls of biomass partitioning between roots and shoots: atmospheric CO_2 enrichment and the acquisition and allocation of carbon and nitrogen in wild radish. Oecologia 89:580–587

Clough JM, Peet MM, Kramer PJ (1981) Effect of high atmospheric CO_2 and sink size on rates of photosynthesis of a soybean cultivar. Plant Phys 67:1007–1010

Conway TJ, Tans P, Waterman LS, Thoning KW, Maserie KA, Gammon RM (1988) Atmospheric carbon dioxide measurements in the remote global troposphere. Tellus 40B: 81–115

Curtis PS, Teeri JA (1992) Seasonal responses of leaf gas exchange to elevated carbon dioxide in *Populus grandidentata*. Canadian J For Res 22:1320–1325

Fajer ED, Bowers MD, Bazzaz FA (1992) The effect of nutrients and enriched CO_2 environments on production of carbon-based allelochemicals in *Plantago*: A test of the carbon/nutrient balance hypothesis. Am Nat 140:707–723

Garbutt D, Bazzaz FA (1984) The effects of elevated CO_2 on plants III. Flower, fruit and seed production and abortion. New Phytol 98:433–446

Geber MA, Dawson TE (1992) Evolutionary responses of plants to global change, pp 179–197 in: Karieva PM, Kingsolver JG,

Huey RB (eds), Biotic interactions and global change. Sinauer Associates, Inc, Sunderland, MA, USA
Heagle AS, Philbeck RB, Rogers HH, Letchworth MB (1979) Dispensing and monitoring ozone in open-top field chambers for plant effects studies. Phytopathology 69:15–20
Hunt R, Hand DW, Hannah MA, Neal AM (1991) Response to CO_2 enrichment in 27 herbaceous species. Funct Ecol 5:410
Idso SB, Kimball BA (1989) Growth response of carrot and radish to atmospheric CO_2 enrichment. Env Exp Bot 29:135–139
Kimball BA (1983) Carbon dioxide and agricultural yield: an assemblage and analysis of 430 prior observations. Agron J 75:779–788
Mazer SJ (1987a) Parental effects on seed development and seed yield in *Raphanus raphanistrum*: Implications for natural and sexual selection. Evolution 41:355–371
Mazer SJ (1987b) The quantitative genetics of life history and fitness components in *Raphanus raphanistrum* L. (Brassicaceae): Ecological and evolutionary consequences of seed-weight variation. Am Nat 130:891–914
Mazer SJ, Schick CT (1991) Constancy of population parameters for life history and floral traits in *Raphanus sativus* L. I. Norms of reaction and the nature of genotype by environment interactions. Heredity 67:143–156
Mazer SJ, Wolfe LM (1992) Planting density influences the expression of genetic variation in seed mass in wild radish (*Raphanus sativus* L. Brassicaceae). Am J Bot 79:1185–1193
Mazer SJ, Snow AA, Stanton ML (1986) Fertilization dynamics and parental effects upon fruit development in *Raphanus raphanistrum*: Consequences for seed size variation. Am J Bot 73:500–511
Overdieck D, Reid CH, Strain BR (1988) The effects of preindustrial and future CO_2 concentrations on growth, dry matter production and the C/N relationship in plants at low nutrient supply: *Vigna unguiculata* (Cowpea), *Abelmoschus esculentus* (Okra) and *Raphanus sativus* (Radish). Angew Botanik 62:119–134
Peet MM, Huber SC, Patterson DT (1986) Acclimation to high CO_2 in monoecious cucumbers. II. Carbon exchange rates, enzyme activities, and starch and nutrient concentrations. Plant Physiol 80:63–67
Reekie EG, Bazzaz FA (1991) Phenology and growth in four annual species grown in ambient and elevated CO_2. Can J Bot 69:2475–2481
Samson DR (1967) Frequency and distribution of self-incompatibility alleles in *Raphanus raphanistrum*. Genetics 56:241–251
SAS Institute (1989) SAS/STAT user's guide, version 6, fourth edition, volume 2. Cary, NC, USA, SAS Institute Inc
Sokal RR, Rohlf FJ (1981) Biometry. W H Freeman and Co, New York
Stanton ML (1984a) Developmental and genetic sources of seed weight variation in *Raphanus raphanistrum* L. (Brassicaceae). Am J Bot 71:1090–1098
Stanton ML (1984b) Seed variation in wild radish: effects of seed size on components of seedling and adult fitness. Ecology 65:1105–1112
Stanton ML (1985) Seed size and emergence time within a stand of wild radish (*Raphanus raphanistrum* L.): the establishment of a fitness hierarchy. Oecologia 67:524–531
Stanton ML, Preston RE (1988) Ecological consequences and phenotypic correlates of petal size variation in wild radish, *Raphanus sativus* (Brassicaceae). Am J Bot 75:528–539
Wulff RD, Alexander HM (1985) Intraspecific variation in the response to CO_2 enrichment in seeds and seedlings of *Plantago lanceolata* L. Oecologia 66:458–460
Ziska LH, Teramura AH (1992) Intraspecific variation in the response of rice (*Oryza sativa*) to increased CO_2 – photosynthetic, biomass and reproductive characteristics. Physiol Plant 84:269–276