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A garden study of the effects of ultraviolet-B radiation on pollination success and lifetime female fitness in *Brassica*

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Abstract While a large number of studies have examined the effects of increased ultraviolet-B radiation (UV-B) on growth and physiological function of plants, UV-B effects on pollination success and fitness are poorly understood. To examine this question, we measured growth, timing of flowering, pollination success, production of pollen, ovules, flowers, fruits, and seeds, and quality of offspring produced by *Brassica nigra* and *B. rapa* in a garden experiment. A total of 313 plants of the two species were randomly divided into two treatment groups. One group received only natural ambient levels of UV-B, while the other received an artificially enhanced UV-B dose. Fitness of *B. nigra* declined at the higher UV-B dose while *B. rapa* fitness did not change. One possible cause of this result was a shift in the relative attractiveness of the two species to pollinators: visitation to *B. nigra* declined at the high UV-B dose while *B. rapa* visitation increased.

Key words Ultraviolet radiation · Female fitness · *Brassica nigra* · *Brassica rapa* · Pollination

Introduction

Depletion of stratospheric ozone is increasing the level of ultraviolet-B radiation (UV-B) reaching the earth's surface. UV-B interferes with a variety of biological processes (Bornman 1989; Caldwell et al. 1989; Stapleton 1992). There have been numerous studies examining physiological and growth responses by plants to enhanced UV-B (see reviews by Teramura 1983; Caldwell et al. 1989; Krupa and Kickert 1989; Tevini and Teramura 1989; Bornman and Teramura 1993; Runeckles and Krupa 1994), but far less work has been done on the effects of UV-B on female fitness (i.e., seed number and quality). Fitness data are important in understanding how populations will respond to increases in UV-B (Caldwell et al. 1989; Teramura 1990; SCOPE 1992).

Increased UV-B may affect female fitness in several different ways. Stress caused by UV-B could affect pollination success by reducing the ability of the plant to produce floral attractants and rewards. Stress from UV-B could also cause reductions in the resources available for seed production. Finally, UV-B may cause deleterious mutations in the pollen, ovules, or developing embryos. None of these possibilities has received much study (Caldwell et al. 1989, 1995; SCOPE 1992).

A greenhouse study of two species of *Brassica* (Feldheim and Conner 1996) found *increases* in female fitness with increased UV-B in three of four cases, in spite of the fact that the family Brassicaceae is known to be sensitive to UV-B (Van et al. 1976; Hashimoto and Tajima 1980; Tevini et al. 1981; Wilson and Greenberg 1993). However, responses of plants in the greenhouse to UV-B may be quite different from responses in the field (Teramura 1986b; Teramura and Murali 1986). It may be that plants are more able to cope with UV-B when other stresses are minimized in the greenhouse.

The goal of this study was to determine the effects of enhanced UV-B on female fitness of these same species of *Brassica* in an outdoor garden experiment. To help determine the causes of fitness differences, a variety of measurements were made. Plant height, leaf production, and flower production were recorded weekly. Observations of pollinators were conducted to determine pollination success, and per-flower pollen and ovule production was measured. Total lifetime flower, fruit, and seed production were recorded, as well as several aspects of seed quality (seed size, germination success, and offspring growth). The results differ in important

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ways from the previous greenhouse study (Feldheim and Conner 1996).

Methods

Experimental design

Rapid-cycling *Brassica nigra* (black mustard, phenotype Rcb) and *B. rapa* (field mustard, canola, rape seed, phenotype Rcb) were used (Crucifer Genetics Cooperative, Madison, Wis., USA). Both of these species are common weeds and important agricultural crops. Ninety plants of each species were planted on 21 June and another 90 of each species were planted in a second block on 11 July for a total of 360 plants. Due to incomplete germination, final sample sizes were 162 *B. nigra* and 151 *B. rapa*. All plants were maintained in 7.5-cm pots in a 1:1:1 peat, perlite, soil mixture and watered twice daily. Fertilization to provide 237 ppm nitrogen and treatments of insecticidal soap were given weekly; the latter was to control aphid outbreaks. The UV-B absorbance of the soap is unknown, but the control and experimental plants were treated equally and the soap was applied in the late afternoon, so that the results should not be biased by the soap treatment. Plants were kept well-spaced to minimize shading. Both control and experimental plants were grown under outdoor UV light racks in an experimental garden at Plant Sciences Laboratory at the University of Illinois.

Half of each species was randomly assigned to either a control or an experimentally enhanced dose of UV-B. UV-B treatments began before plants germinated and continued until all plants had senesced in early October. Control plants received only natural ambient UV-B. Experimental plants were given a daily supplemental UV-B dose of 11 kJ; this is the biologically effective dose based on the generalized plant damage spectrum of Caldwell (1971). This dose represents UV-B levels in central Illinois after approximately 45% destruction of the ozone layer (Green et al. 1980). This level of ozone destruction is more severe than the maximum expected according to most predictions (e.g., Stolarski et al. 1992). Ambient levels of UV-B at 40°N on a clear day vary between 6.5 and 7.5 kJ for the summer months according to a standard model (Green et al. 1980).

A rack of ten UV-B lamps (UV-B 313, Q-Panel Inc., Cleveland, Ohio, USA) was suspended over each of two adjacent garden plots. Before use, lamps were illuminated for at least 100 h to stabilize UV-B output. Control lamps were covered with mylar (0.0125 cm) that effectively absorbs all UV wavelengths (Middleton and Teramura 1993). Bulbs over experimental plants were covered with cellulose acetate (0.0075 cm), which absorbs UV-C while transmitting UV-B (Middleton and Teramura 1993). Because UV-B transmittance through cellulose acetate decreases with increasing UV-B exposure, both sets of filters (mylar and cellulose acetate) were changed every 10 days. All filters were exposed to sunlight for 8 h before use to stabilize transmittance.

Lamp height was adjusted daily so that experimental readings were 11 kJ in total darkness. The heights of the control and experimental racks were always kept equal, so that other effects of the light racks (e.g., shading) would not bias the results. Levels of UV-B reaching control plants were measured on 1–2 nights per week, and were always < 0.7 kJ. The UV-B dose was measured using a SED 240 radiometer (International Light Inc.) connected to a data logger (LI 1000, LiCor Inc.). The spectral response of the SED 240 closely approximates Caldwell's generalized plant-damage action spectrum (Caldwell 1971; DeLucia et al. 1991). All UV-B measurements were made at the average leaf height of the plants.

The UV-B lights were lit for 6 h centered around solar noon. The effects of UV-B on plants are greater when photosynthetically active radiation (PAR) levels are low (Bornman 1989), because visible and UV-A light are crucial cues for induction of a variety processes that help protect plants from UV-B (reviewed in Bornman and Teramura 1993). Therefore, PAR readings were taken several times a day on cloudy days. If PAR dropped below 200 μmol m^{-2}, the UV-B lamps were turned off, because the combination of high UV-B and low PAR does not occur naturally. This occurred nine times: on 3 days the UV lamps were never lit and on 6 days exposure time was reduced due to extremely cloudy conditions. Our cut-off of 200 μmol m^{-2} is fairly low, so that on some moderately cloudy days the UV-B/PAR ratio was high.

Treatment groups were not replicated because only two UV-B light racks were available. For this reason, a number of steps were taken to minimize the chance that factors other than UV-B would cause differences between treatment groups. The experimental and control plots were switched every 5 days by moving the plants from one plot to the other and switching the filters on the two light banks. Plants were rotated daily within plots, and the two species were interspersed, with each flat containing six of each species.

Even with these precautions, if there was some critical period in plant development that occurred mostly within a 5-day period during which the plants were not moved between plots, and there was some environmental difference between plots other than UV-B, then differences between sides might not be due to UV-B. The fact that plants were grown in two blocks separated in time means that critical periods were likely to happen in different plots and different environmental conditions in the two blocks. Since flowers are produced continuously over approximately 1 month, critical periods related to floral traits were spread across plots. To test for other environmental differences between UV-B treatment groups, PAR and temperature readings were taken at nine locations in each plot on 3 days spanning the experiment. There were no significant differences in PAR or temperature between the two treatment groups (average difference in PAR, control minus enhanced, $= -53$ mmol m^{-2}, paired $t = -0.9$, $P = 0.46$; mean difference in temperature $= -0.28$°C, paired $t = -0.7$, $P = 0.55$). Considering all these points together, it is very likely that differences between treatment groups were caused by the UV-B treatment itself.

Traits measured

Growth and flowering phenology

Growth and flower production was measured on all plants at weekly intervals from the development of the first true leaves to the cessation of flowering. The height from the soil surface to the tallest point of the plant was measured to the nearest 0.5 cm with a ruler and the number of true leaves and open flowers were recorded. The dates of first and last flowering were also recorded.

Pollen production

Pollen was collected from one flower on each of 56 plants (33 *B. nigra* and 23 *B. rapa*), approximately half from each UV-B dose. On these plants, one group of unopened flower buds was enclosed in a mesh bag to exclude pollinators until a flower opened (usually less than 24 h). All six anthers were then collected from this flower for later pollen counts. Pollen was counted using a Coulter counter; for details see Rush et al. (1995). The bridal-veil mesh used for the bags absorbs less than 5% of PAR and UV-B (unpub. data).

Pollinator attraction

To determine if enhanced UV-B could affect competition between the two plant species for the attraction of pollinators, plants were taken to the University of Illinois Phillips Tract natural area for pollinator observations. Small native bees were the most common visitors (57% of all visits), while syrphid flies made up most of the remainder (42%). For details of pollinators visiting *Brassica* at this site see Conner and Neumeier (1995). Observations began within a few days of first flowering. The plants taken to the observation site on a given day were a random sample (without replacement) of plants that were flowering at that time. Eighteen plants of each species from the same UV-B dose were interspersed in a 6×6 grid with 0.5 m spacing. Two observers simultaneously observed one

plant of each species for 10 min; this design controls for temporal variation in visitation. Before the observations began, the height of the plant and the number of open flowers were recorded. During the observations the number of visits, taxon of visitor, duration of the visit, and number of flowers visited were recorded. Plants were returned to the experimental garden the same day so that they could receive their UV-B dose. A total of 40 pairs of plants were observed from 13 July to 10 August; individual plants were observed only once. While one 10-min observation period is not enough to characterize the attractiveness of individual plants, the goal was to examine the effect of UV-B on attractiveness, not evaluate individual plants.

Pollen and ovule quality

The effects of UV-B on pollen and ovule quality were studied using hand pollinations. A pair of two pollen donor plants, one from each UV-B dose, was paired randomly with two pollen recipient plants, again one from each dose. Pollen from each donor was used to pollinate two flowers on each recipient. Each recipient flower received pollen from only one of the donors, so a total of four flowers were pollinated on each recipient. This entire procedure was repeated 13 times for each species, each time with a unique set of two donor and two recipient plants, for a total of 104 plants and 208 flowers pollinated. Recipient flowers were bagged as buds and kept in a pollinator-free laboratory for 4 h after hand-pollination to prevent natural pollination. The number of seeds produced by each hand-pollinated flower was then counted.

This experimental design allowed us to make paired comparisons (using paired *t*-tests) of the ability of pollen from control vs. enhanced UV-B plants to fertilize seeds on both control and enhanced UV-B recipient plants. Similarly, we made paired comparisons of the number of seeds produced by control and enhanced UV-B plants when they were pollinated by pollen from both control and enhanced fathers.

Fitness components

Plants were harvested upon senescence, air dried, and the following counts were made: total lifetime number of flowers produced (by counting pedicels), number of fruits, and total number of seeds. The entire above-ground part of the plant, all fruits, and all seeds were weighed. From these data, the following fitness components were calculated: number of flowers, number of fruits/flower, number of seeds/fruit (these multiply to equal total lifetime seed production), and reproductive effort (fruit mass/total above-ground biomass). Then 32 seeds were randomly sampled for offspring growth measurements.

Since the two UV-B treatment groups were grown in adjacent plots (to minimize other environmental differences), an unknown amount of cross-pollination occurred between treatments. The plants were taken to the Phillips Tract for pollination (see above) because visitation rates were very low in the experimental garden. At the Phillips Tract, where visitation rates were much higher, plants from only one UV-B dose were present at any one time. Therefore, it seems likely that most pollination was within treatments. Any differences in reproduction that we observed between treatments would be underestimated to the extent to which cross-pollination occurred, and the extent to which UV-B induced pollen-quality differences affect seed production. Cross-pollination from natural *Brassica* plants is unlikely, because no natural populations were found either on campus (where the experimental garden was located) or at the Phillips Tract.

Offspring measurements

To determine effects of UV-B exposure on offspring quality without confounding results with the direct effects of UV-B on offspring, offspring were grown in a greenhouse in the absence of UV-B. The randomly sampled seeds were planted in two 7.5-cm pots (16 seeds/pot) in MetroMix 360 potting soil. To minimize microenvironmental effects, seeds from different UV-B dose treatments and species were interspersed within flats.

All germinating seeds were counted, and then seedlings were randomly thinned to one per pot. Two weeks (*B. rapa*) or three weeks (*B. nigra*) after planting, the number of true leaves and plant height were recorded. These measurements were made again on the day the plant first flowered.

Analysis

All statistical tests were conducted using JMP on Macintosh computers (SAS Institute 1994). For all data except hand-pollination results, related variables were grouped together and multivariate analyses of variance (MANOVA) were run to test for differences caused by UV-B for the group of traits as a whole. If a multivariate test was found to be significant at $P < 0.10$, univariate ANOVAs were conducted on each variable separately to determine which was affected most by UV-B. Flower number, flowering duration, and weekly leaf counts were ln-transformed to reduce heteroscedasticity. Because plants were grown in two blocks separated in time, all analyses included a block fixed effect and all possible interactions with block. Since the block effect was not of primary interest, it was not presented in the tables and figures for simplicity.

Weekly growth measurements

Repeated measures MANOVA and ANOVA were used to test for effects of week, dose, and the week by dose interaction on the three traits (height, leaf, and flower number).

Single measurements

Traits that were measured one time on all plants were split into three categories for MANOVAs: flowering phenology (number of days to first flower, flowering duration), fitness components (number of flowers, number of fruits/flower, number of seeds/fruit, number of seeds, average mass/seed, total above-ground biomass, reproductive effort), and offspring growth (percent germination, first and second measurements of height and leaf number, and number of days until first flower). MANOVAs and ANOVAs were performed with dose and species as fixed effects.

Pollinator visitation

Pollination data were split into two categories: plant traits (height and flower number) and visitation by pollinators (number of visits, number of flowers/visit, and time/flower) for MANOVAs.

Results

Weekly growth measurements

The weekly growth measurements are presented in Fig. 1. A strong week main effect occurred in all cases, indicating that plants were growing over time. MANOVA results for both species show significant dose main effects and significant week × dose interactions for the three traits as a group, indicating an effect of UV-B on growth that varied over time.

Increased UV-B caused significant reductions in height in both species (significant dose main effect). In *B. rapa* this reduction occurred mostly in week three

(significant week × dose interaction). Leaf and flower number were largely unaffected by UV-B except for a significant week × dose interaction for leaf number in *B. nigra*.

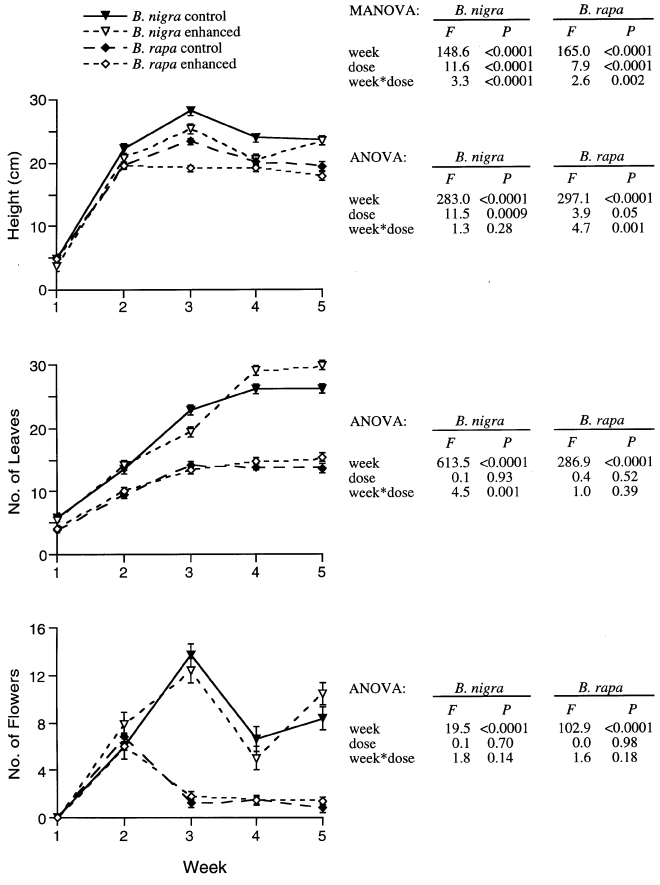

Fig. 1 Weekly growth and flower production results (means ± 1 SEM). MANOVA table for all three traits simultaneously is at the *top*; ANOVA results for each trait are located to the *right* of their respective graphs. Analysis of leaf number was on ln-transformed values, but untransformed data were used in the graph for ease of interpretation. Total $n = 313$

Flowering phenology and fitness components

The MANOVA results (Table 1) show that UV-B had no significant effect on flowering phenology but did have a highly significant effect on the fitness components (dose and dose × species interactions). Increased UV-B caused a slight decrease in lifetime flower production in both species (Table 1, dose main effect; Fig. 2A) and caused a reduction in fruits per flower in *B. nigra* only (Table 1, Fig. 2B). The number of seeds per fruit increased in *B. rapa* and decreased in *B. nigra* with increased UV-B (Table 1, dose × species interaction, Fig. 2C). The declines in all three of these multiplicative fitness components in *B. nigra* resulted in an approximately 30% decrease in total seed production at the high UV-B dose, while there was little overall change in seed production by *B. rapa* (Table 1, dose main effect and dose × species interaction; Fig. 2D). The increase in seeds per fruit compensated for the decrease in flower number in *B. rapa*, with the end result being little change in seed number. However, the increased number of seeds per fruit in *B. rapa* at the enhanced UV-B dose apparently came at the expense of decreased seed mass (Table 1 and Fig. 2E). Above-ground biomass showed sharp declines with increased UV-B in both species, but the proportion of biomass allocated to reproduction (reproductive effort) did not change (Table 1, dose main effects, Fig. 2F). Thus, female reproduction, in terms of total mass of fruit produced, was closely related to overall plant size as measured by above-ground biomass.

Pollination success

Increased UV-B did not significantly affect per flower pollen production by plants in either species (dose main effect: $F = 0.76$, $P = 0.39$; dose × species interaction: $F = 0.39$, $P = 0.55$). Similarly, no significant differences between treatments occurred in the hand-pollination study, suggesting that UV-B did not affect pollen or ovule quality in our study.

Table 1 Flowering phenology and fitness components result summary. In each group of variables, the first line presents the results of a MANOVA including all dependent variables in that group. If either the dose or dose × species tests in the MANOVA were significant at $P < 0.10$, the results of individual ANOVAs for each dependent variable are presented separately ($n = 307$ for mass/seed; $n = 312$ for number of seeds/fruit, $n = 313$ for all other traits)

	Dose		Species		Dose × Species	
	F	*P*	*F*	*P*	*F*	*P*
MANOVA (Flowering phenology)	0.95	0.39	61.6	<0.0001	0.94	0.39
MANOVA (Fitness components)	6.66	<0.0001	65.7	<0.0001	2.91	0.006
ln no. of flowers	5.78	0.02	215.4	<0.0001	0.05	0.82
No. fruits/flower	3.05	0.08	12.6	0.0004	2.13	0.15
No. seeds/fruit	0.01	0.93	52.8	<0.0001	4.22	0.04
No. seeds	16.26	0.0001	37.5	0.0001	12.38	<0.0001
Average mass/seed	8.02	0.005	147.8	<0.0001	1.80	0.18
Total above-ground biomass	28.83	<0.0001	0.8	0.36	1.29	0.25
Reproductive effort	0.63	0.43	4.1	0.04	1.67	0.20

The MANOVA results for the height and flower number of plants used in pollinator observations show a highly significant dose main effect and dose × species interaction (Table 2). Increased UV-B caused a dramatic reduction in both height and flower number in *B. nigra,* but little or no declines in these traits occurred in *B. rapa* (Table 2; Fig. 3A, B).

The MANOVA for the visitation variables showed no dose main effect, but a strong dose × species interaction (Table 2). For both the number of visits and the number of flowers probed per visit, increased UV-B caused strong decreases in *B. nigra* and slight increases in *B. rapa* (Table 2; Fig. 3C, D).

Offspring quality

There was no evidence for detrimental effects of UV-B on offspring germination and growth. The MANOVA for offspring growth shows both a highly significant dose main effect and a highly significant dose × species interaction (Table 3). The UV-B treatment had no effect on germination success. Both leaf number and height at the first measurement were greater in offspring of *B. nigra* plants grown under enhanced UV-B, while there were no differences in *B. rapa* offspring (Fig. 4A, B). These effects disappeared by the second measurement (Table 3). A

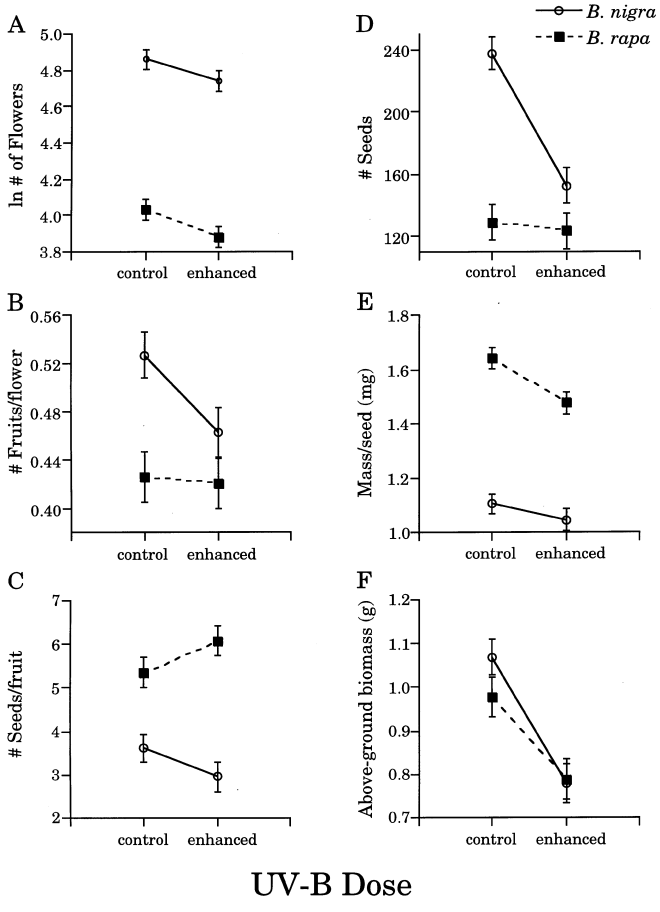

Fig. 2A–F Flowering phenology and fitness component results (means ± 1 SEM); $n = 307$ for mass/seed; $n = 312$ for number of seeds/fruit, $n = 313$ for all other traits

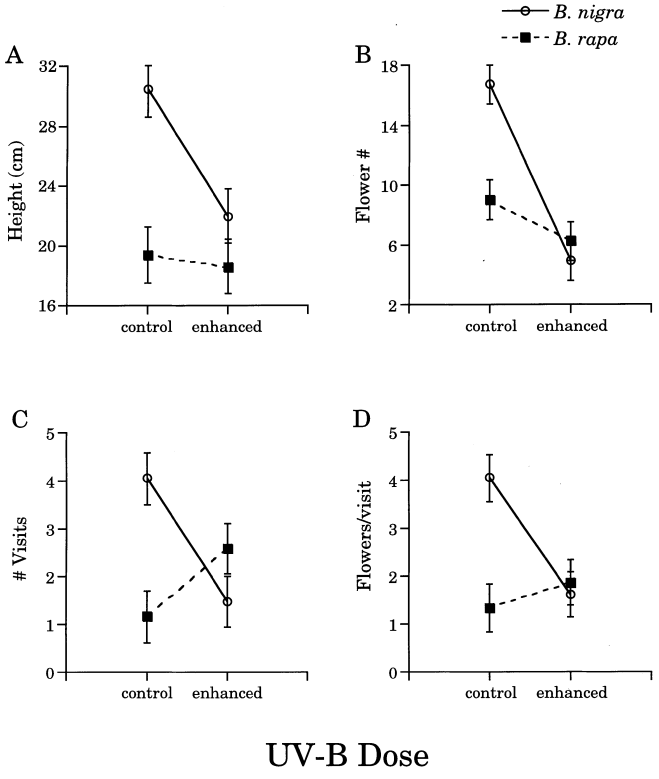

Fig. 3A–D Pollinator visitation trial results (means ± 1 SEM). See Table 2 for statistical tests; $n = 74$

Table 2 Pollinator visitation MANOVA and ANOVA summary ($n = 74$; see Table 1 for details)

	Dose		Species		Dose × Species	
	F	*P*	*F*	*P*	*F*	*P*
MANOVA (Plant traits)	15.65	< 0.0001	8.41	0.0006	6.55	0.003
Height	6.34	0.01	15.27	0.0002	4.33	0.04
Flower no.	31.22	< 0.0001	6.04	0.01	12.10	0.0009
MANOVA (Pollinator visitation)	1.75	0.17	2.95	0.04	4.78	0.004
No. visits	1.19	0.28	2.78	0.10	13.92	0.0004
Flowers/visit	3.84	0.05	6.46	0.01	9.25	0.003
Time/flower	3.50	0.07	6.50	0.01	1.20	0.28

Table 3 Offspring growth MANOVA and ANOVA summary. Leaf 1, leaf 2, height 1, and height 2 refer to the first and second measurements of leaf number and overall plant height respectively ($n = 298$ for days to 1st flower; $n = 299$ for all other traits)

	Dose		Species		Dose × Species	
	F	*P*	*F*	*P*	*F*	*P*
MANOVA	4.75	0.0001	167.3	< 0.0001	6.54	< 0.0001
% Germination	0.97	0.33	4.3	0.04	0.62	0.43
Leaf 1	7.52	0.007	445.7	< 0.0001	12.98	0.0004
Height 1	9.11	0.003	209.2	< 0.0001	14.57	0.0002
Leaf 2	0.36	0.55	872.3	< 0.0001	0.85	0.36
Height 2	3.09	0.08	157.9	< 0.0001	1.55	0.21
Days to 1st flower	2.16	0.14	171.8	< 0.0001	6.81	0.01

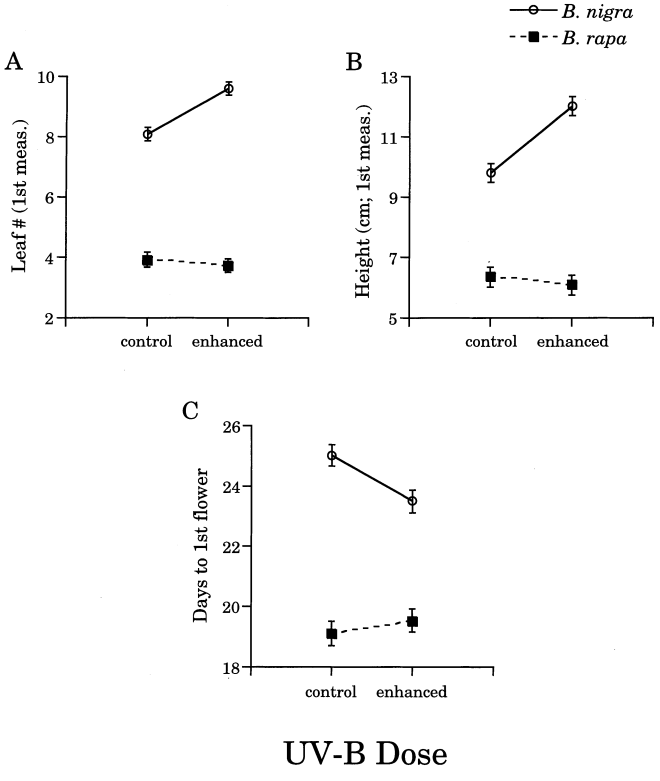

Fig. 4A–C Offspring growth results (means ± 1 SEM). Only traits for which dose or dose × species were significant at $P < 0.05$ (see Table 3) are presented ($n = 298$ for days to 1st flower; $n = 299$ for all other traits)

similar pattern occurred in the time to first flower: offspring of *B. nigra* plants from the enhanced UV-B flowered earlier, while *B. rapa* offspring flowering time did not change (Fig. 4C). These early growth patterns were not predictable based on seed size: the seeds of both species were smaller in the enhanced UV-B treatment (note that this was only significant for *B. rapa*; Fig. 2).

Discussion

Our results suggest that increased UV-B will have detrimental effects on reproduction in *Brassica nigra* but not *B. rapa*. Seed production by *B. nigra* decreased by over 30% with increased UV-B while *B. rapa* seed production remained essentially constant. The offspring of *B. nigra* grown at high UV-B had higher early growth, but this is not likely to make up for the 30% decrease in seed number, particularly since germination success was not affected by UV-B in either species. With increasing UV-B in the future, populations of tolerant plant species like *B. rapa* may increase at the expense of sensitive plants like *B. nigra*.

These conclusions are opposite to those reached from similar UV-B doses in a greenhouse study (Feldheim and Conner 1996). In the greenhouse, exposure to UV-B 11 kJ above control levels increased seed production in *B. nigra* but decreased seed production in *B. rapa*. These differences suggest that other environmental factors affect the response of these species to UV-B (see also Bornman and Teramura 1993). The greenhouse was a less stressful environment than the outdoor garden: average seed production per plant ranged from about 260 to 320 in the greenhouse at similar UV-B doses (Feldheim and Conner 1996) but was only 120–240 in the garden. In a study of soybeans, responses to UV-B were similar in the greenhouse and field for vegetative traits, but seed yield responses varied greatly across the two environments for the six cultivars tested (Teramura and Murali 1986). Therefore, caution is needed when attempting to extrapolate greenhouse results to the field, particularly for traits closely related to fitness.

While there have been few other studies that have focused on the effects of UV-B on plant fitness, a number of papers have reported seed or fruit yield data for several agricultural species. The majority of these studies have reported decreases or no change in yield with increasing UV-B, similar to our results, but some have found significant increases in yield (reviewed by Teramura 1986a, 1990; Krupa and Kickert 1989; Teramura et al. 1990; Feldheim and Conner 1996). This variability in results, even when the same cultivars are tested in multiple years, indicates that multiple-year studies provide the best overall picture of the effects of UV-B (Teramura et al. 1990). In a recent greenhouse study of wild plants, seed number decreased in three of four species of dicots but increased in three of four species of monocots (Musil 1995).

The only detrimental effect of UV-B on seed quality that we found was a decrease in seed mass in *B. rapa* (Fig. 2). However, this did not result in a decrease in germination success or growth of the resulting offspring (Fig. 4). Musil (1995) found increases in seed mass with

increased UV-B in four species, but this translated to increased germination success in only one species, similar to our results. Note that the offspring in both these studies were grown in the greenhouse; therefore, it is possible that detrimental effects on offspring of *B. rapa* grown at high UV-B might have become apparent if they were grown under more stressful field conditions (see above). Also, while little detrimental effect on offspring was found in our single-generation study, UV-B may cause the accumulation of deleterious mutations across multiple generations; this is an important topic for further work (e.g., Musil 1996).

In contrast to our results for fitness, we found few detrimental effects of UV-B on growth and flowering phenology. The only significant effect was that increased UV-B caused decreased height in both species. This decrease in height could cause additional decreases in fitness under more crowded field conditions, if *Brassica* was competing for light with a species whose height was not affected by increased UV-B (e.g., Barnes et al. 1988). In our experiment, pots were kept well-spaced, minimizing light competition.

We found that increased UV-B strongly affected pollination success, in contrast to the results of our previous greenhouse study (Feldheim and Conner 1996). Height and flower number were significantly reduced in *B. nigra* but not *B. rapa* at the time of pollination trials. The result of this was a reversal in the outcome of competition for pollinators in these species. In plants raised at ambient levels of UV-B, *B. nigra* attracted more pollinators than *B. rapa*. At enhanced UV-B, *B. rapa* attracted more (Fig. 3C). Visitation to *B. rapa* increased at high UV-B relative to the ambient dose in spite of a slight reduction in flower number. This may be due to the much greater decrease in flower number in *B. nigra*, making *B. rapa* relatively more attractive at the high UV-B dose. More studies of the effects of UV-B on pollinator attraction are necessary to determine the generality of these results.

While pollinator attraction was affected by UV-B, neither pollen production per flower nor pollen quality were affected in either species. This result contrasts with a greenhouse study of *B. rapa*, which found evidence for decreased pollen production per flower and pollen quality with increased UV-B (Demchik and Day 1996); interestingly, this study used lower UV-B doses than our study.

As noted in Methods, the enhanced UV-B dose used here is quite high, higher than what is predicted to occur in the next century according to some predictions (e.g., Stolarski et al. 1992). The actual extent of future ozone destruction is unknown, and depends on many factors ranging from worldwide CFC production and use patterns to complex atmospheric physics and chemistry (Madronich 1993). Therefore, it seems prudent to examine the biological effects of a wide range of UV-B levels.

In summary, our results suggest that reproduction in *B. nigra* may decline with increased UV-B, but that *B. rapa* is more resistant. One of the causes of this difference between species may be the higher relative attractiveness to pollinators of *B. rapa* at higher UV-B. These results contrast sharply with a very similar study of these species conducted in the greenhouse (Feldheim and Conner 1996). Future work should include multiyear fitness studies on a variety of species to enable us to predict the possible consequences of increased UV-B radiation on plant populations.

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