

Reproductive success in varying light environments: direct and indirect effects of light on plants and pollinators

Francis F. Kilkenny · Laura F. Galloway

Received: 8 March 2007 / Accepted: 17 October 2007 / Published online: 16 November 2007
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Abstract Plant populations often exist in spatially heterogeneous environments. Light level can directly affect plant reproductive success through resource availability or by altering pollinator behavior. It can also indirectly influence reproductive success by determining floral display size which may in turn influence pollinator attraction. We evaluated direct and indirect effects of light availability and measured phenotypic selection on phenological traits that may enhance pollen receipt in the insect-pollinated herb *Campanulastrum americanum*. In a natural population, plants in the sun had larger displays and received 7 times more visits than plants in the shade. Using experimental arrays to separate the direct effects of irradiance on insects from their response to display size, we found more visits to plants in the sun than in the shade, but no association between number of visits each flower received and display size. Plants in the sun were not pollen limited but pollen-augmented shade flowers produced 50% more seeds than open-pollinated flowers. Phenological traits, which may influence pollen receipt, were not under direct selection in the sun. However, earlier initiation and a longer duration of flowering were favored in the shade, which may enhance visitation in this pollen-limited habitat.

Keywords Light availability · Pollinator behavior · Pollen limitation · Phenotypic selection · Reproductive phenology

Introduction

Plant populations often exist in spatially heterogeneous environments. This environmental variation may affect fitness because abiotic factors determine resource availability (Lechowicz and Bell 1991). It may also influence fitness by affecting biotic factors, such as pollinator or herbivore abundance (Herrera 1995b; Rodriguez et al. 1994). Variation in abiotic and biotic conditions is likely to result in the environment-dependent selection that is commonly found in nature (e.g., Kalisz 1986; Stewart and Schoen 1987; Farris 1988; Widén 1991; Kelly 1992; Caruso 2000; Ashman and Morgan 2004; Moeller and Geber 2005; Vanhoenacker et al. 2006). Understanding adaptive evolution in populations that span environmental conditions requires dissecting environmental effects on plants into biotic and abiotic components, and investigating their combined influence on fitness.

Light environment has been shown to directly affect plant growth and resource allocation to structural components (McConnaughey and Coleman 1999). A low-light environment often reduces overall plant size but may also affect the allocation of resources to particular plant parts (Bloom et al. 1985; Gleeson and Tilman 1992; McConnaughey and Coleman 1999). This can influence reproduction because plants in low-light environments may shunt resources away from reproductive structures to parts which can increase light capture, such as leaves and stems. Therefore low light may directly reduce reproductive success because plants are smaller with reduced allocation to reproduction (Fig. 1).

The light environment can have additional effects on reproduction of outcrossing plants by directly influencing pollinator behavior (Fig. 1). Pollinator foraging may be influenced by forest canopy density (Liow et al. 2001) and light environment (Herrera 1995a, 1997). In forest environments

Communicated by Louis Pitelka.

F. F. Kilkenny (✉) · L. F. Galloway
Department of Biology, University of Virginia,
Charlottesville, VA 22904-4328, USA
e-mail: flk5p@virginia.edu

increasing shade decreases temperature (Liow et al. 2001; Herrera 1997; Newmark 2001), and the number of foraging bouts in many ectothermic insect pollinators is positively correlated with temperature (Herrera 1995a, b; Comba 1999; Liow et al. 2001; Totland 2001). However, the effect of temperature on pollinator behavior may vary among insect species (Herrera 1997; Bishop and Armbruster 1999). For example, one study found several pollinator guilds were influenced by environmental heterogeneity while others foraged indiscriminately across environmental mosaics (Herrera 1995b).

Pollinator behavior may also be indirectly affected by light environment. With greater light availability, plants typically grow larger and may produce larger floral displays. Larger display sizes have been shown to attract pollinators and increase the number of visits (Fig. 1; Andersson 1991; Conner and Rush 1996; Galloway et al. 2002; Grindeland et al. 2005).

In insect-pollinated plants, pollinator visitation is important for female fitness because the level of pollen receipt by ovules limits seed set. Therefore, pollen limitation can lead to reproductive failure and can affect the persistence of plant populations (Bond 1994; Groom 1998; Kearns et al. 1998; Ashman et al. 2004). Pollen limitation has been detected in a wide variety of systems although it is still unclear how often it occurs (Ashman et al. 2004). The detection of pollen limitation should be rare because plants are expected to evolve mechanisms of reproductive assurance. For example, increased self-fertilization or attractiveness is expected if pollen receipt is chronically low (Haig and Westoby 1988; Lloyd 1992). In outcrossing pollen-limited plants we expect to find selection on traits that may enhance pollen receipt, such as floral display characteristics and flowering phenology (Ashman et al. 2004). Changes in phenological traits may be particularly important if resources are limited and producing more flowers is costly. In particular, selection may act on the timing of flower initiation and peak flowering if pollinator abundance changes throughout the season. Selection may also favor increased flowering duration if variation in pollinator abundance is high across the season.

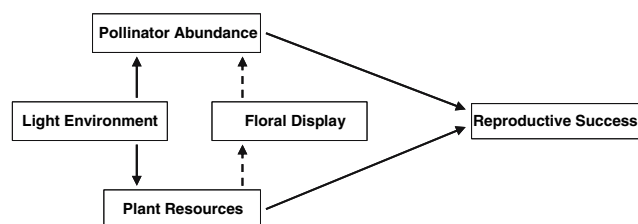


Fig. 1 Mechanistic depiction of the effect of light environment on plant reproductive success. Note that light environment can have both direct (solid arrows) and indirect (dashed arrows) effects on reproductive success

Here, we use a combination of observational and manipulative experiments to explore the direct and indirect effects of the light environment on plant fitness. We use an outcrossing monocarpic herb that inhabits divergent, but physically proximate, light environments to address the following questions:

1. Does light environment influence pollinator visitation?
2. If so, does the light environment influence pollinator behavior directly, and/or does it do so indirectly through the effect of resources on floral display size?
3. Do any differences in pollinator visitation between the light environments affect reproductive success?
4. If so, are there differences in selection on phenological characters between light environments that can be interpreted in the context of pollinator visitation patterns?

Materials and methods

Study system

Campanulastrum americanum Small (= *Campanula americana* L.) Campanulaceae is a protandrous, annual or biennial, insect-pollinated herb that flowers from mid July to late August. Flowers are located in compact indeterminate inflorescences at reproductive nodes on the main stem and, in larger plants, on side branches. Flowers are open in clusters along branches, composed of two to four adjacent nodes (Galloway et al. 2002). Each flower is typically male-phase for a single day and then female-phase for 2–3 days, but female phase can last 6–9 days if flowers are not pollinated (Evanhoe and Galloway 2002). *C. americanum* inhabits a range of light environments from full forest canopy to natural or human-made light gaps. Light availability varies considerably between environments and can be as much as 50 times greater in sun than in shade (Galloway 2005). The primary pollinators of *C. americanum* are bumblebees (*Bombus* spp., including *Bombus vagans*, *Bombus bimaculatus*, and *Bombus affinis*) foraging for nectar and halictids (Halictidae) collecting pollen (Galloway et al. 2002). In 2004 we studied a natural population of *C. americanum* located in southwestern Virginia, USA [Giles County, Salt Pond Mountain near Mountain Lake Biological Station (MLBS)]. The population straddled two distinct light environments: a large light gap created by a road cut, and an adjacent area under a deciduous canopy.

Pollination in the natural population

To determine whether pollinator visitation patterns differed between plants in light gap (sun) and canopy (shade) habi-

tats we observed pollinators in each light environment throughout the flowering season. On 7 days, separated by 5-day intervals, we observed from five to 20 haphazardly selected plants per environment, depending on availability. Each plant was observed for 10 min and the number and type of pollinators visiting the plant were recorded. Plants were observed in random order and display size was counted prior to all observations. Because the pollinator counts did not meet the assumption of normality, we analyzed these data with a generalized linear model using a Poisson distribution and χ^2 likelihood ratios to test for significance (PROC GENMOD; SAS Institute 2005). We tested the fixed effects of pollinator type (*Bombus*, halictid, syrphid), environment, date and the pollinator \times environment interaction on the number of visits. Display size was included as a covariate. Contrasts were used to determine whether visitation varied among pollinator groups and whether pollinator groups showed differences in visitation based on the environment. Means are presented \pm SE throughout.

To test pollen export in the sun and shade we measured pollen removal for 15 plants in each environment on a single day during the peak flowering period. We tagged two unopened flowers per plant and recorded the time of opening and the amount of pollen remaining at hourly intervals from 1200 hours, when the first tagged flower opened, to 1900 hours. Only flowers that opened during the observation period, 23 in sun and 26 in shade, were included in the data set. In *C. americanum*, pollen is presented on the surface of the style and can easily be visually assessed. Proportion of pollen removed per hour was measured using methods developed in Evanhoe and Galloway (2002). A *t*-test was performed to compare removal rates between sun and shade.

We evaluated whether pollen quantity limits seed set by comparing the number of seeds produced from flowers augmented by hand pollination to number of seeds from open-pollinated flowers. Fifteen plants were selected per environment. At 5-day intervals, a single flower on each plant was augmented with outcross pollen. The augmented flower and an unmanipulated control flower, selected from a similar spatial position on the same plant, were marked with colored fabric paint and collected as fruits. Pollen augmentation was performed for the majority of the flowering season in both environments. Seed number per fruit was counted and analyzed with a repeated measures ANOVA to determine whether pollen limitation differed between the two environments. The model contained three fixed effects, date, environment (sun, shade), and pollination treatment (augmented, control), and two interaction terms, pollination \times date and pollination \times environment. To assess whether particular light environments were pollen limited we performed paired *t*-tests separately for each

environment using all available paired observations. The dependent variable, seed number, was natural-log transformed to meet the assumptions of the model. One limitation of this design is that plants may reallocate resources away from control flowers to flowers that are pollen augmented (Zimmerman and Pyke 1988). To test this possibility we compared seed counts from open and augmented flowers to seed counts from flowers on plants that received no hand pollination. These control plants (14 shade, 26 sun) were chosen post hoc and had a comparable range of biomass and flower counts as the plants that received pollen. Flowers on these plants had been marked at similar time intervals as the experimental plants. *t*-tests were performed for each treatment pairing within each environment.

Pollination in experimental arrays

To separate the direct effects of light environment on pollinator visitation from the indirect effects of light, mediated through floral display size, we observed pollinators in experimental arrays. *C. americanum* were grown in a greenhouse at the University of Virginia and transported to the field just prior to initiation of flowering. Experimental arrays of five plants, spaced 1 m apart in a cross formation, were placed approximately 100 m from a natural population in either sun or shade and assigned a display size treatment. For the small display size treatment, flowers were removed until each plant had only three flowers in a single cluster on the main stem. Plants in the large display size treatment were not manipulated (21.5 ± 7.4 SD flowers), and included clusters on the main stem and branches. These displays are typical of natural plants growing in shade and sun environments (F. F. Kilkenny, personal observation).

Arrays were observed for pollinator visitation. Arrays were alternated between sun and shade and the order of the display size treatments was randomized to minimize the effect of changing pollinator abundances throughout the day. Once an array was set up, we waited 30 min before beginning observations to allow pollinators to acclimate to the new floral arrangement. Each array was observed for a single 20-min period. During this time we watched one randomly chosen plant and recorded the number of pollinators visiting and the number of flowers each pollinator visited. From these data we calculated the average number of visits received by each flower per hour. Prior to the observation period in each array, photosynthetically active radiation (PAR) and temperature were recorded. A total of 112 arrays were observed over a 19-day period. We tested the effects of light environment, date, patch display size and the environment \times display interaction on visits per plant and per flower using a generalized linear model (see above). Only visits by *Bombus* spp. were analyzed as other pollinator classes were rare during our observations.

Phenotypic selection analysis

We monitored a natural population of *C. americanum* to determine whether phenotypic selection on reproductive phenology differed between light environments. We randomly chose 90 plants from the population, 50 in the sun and 40 in the shade. We counted flowers at 5-day intervals on all individuals for the entire flowering season to ascertain flowering patterns. Three phenological measurements were derived from the flower counts: date of flowering initiation, date of peak flower production, and flowering duration. To determine seed production, a single flower was marked at each 5-day interval on each plant and seeds were counted when fruits had matured. Total seed production was estimated by multiplying seed counts at every time interval by flower production at that date and then summing across the entire season. Total fruit production was quantified at the end of the flowering season as a second measure of fitness. Plants were harvested when all fruit had matured and aboveground dry biomass was recorded.

We analyzed phenotypic selection on flowering phenology in light gap and canopy regions of the population (Lande and Arnold 1983). Relative fitness was calculated by dividing the fitness estimates for each individual by the mean for its environment. Biomass was highly variable among plants and between environments; it was included in the analyses so that the relationship of phenological traits with fitness could be evaluated independent of plant size. For each analysis, biomass (log transformed) and phenological traits (flowering initiation, peak, and duration) were standardized to a mean of 0 and a SD of 1. Standardized phenotypic selection differentials (S) were estimated as the covariance between relative fitness and measured traits. S values indicate the overall directional change in phenotype due to selection, but do not distinguish between change due to selection on the trait of interest and selection on correlated traits. Standardized directional selection gradients (β) were estimated from partial regression coefficients of linear multiple regression of relative fitness on all four traits. β -values represent phenotypic selection directly on each trait with the effects from correlated traits removed.

To test whether phenotypic selection differed between the two light environments we performed a heterogeneity of slopes analysis [analysis of covariance (ANCOVA)] for each fitness measure on the data pooled across environments. The model included the three phenological characters and biomass, light environment, as well as interaction terms of each trait with environment. Significant trait \times environment interaction terms indicate a difference in slope between the two environments, and therefore the magnitude of selection. Only significant interaction terms are reported.

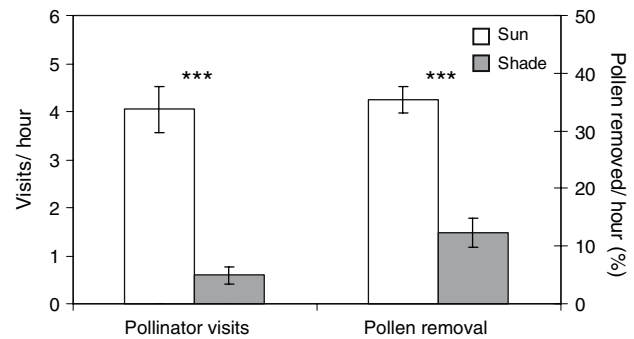


Fig. 2 Pollinator visitation to (\pm SE) and pollen removal from (\pm SE) *Campanulastrum americanum* plants growing in sun and shade areas of a natural population. *** $P < 0.001$, comparison between sun and shade habitats, see text for details

Results

Pollination in the natural population

The light environment influenced pollinator visitation. Plants in the sun received 7 times more visits than plants in the shade and visits increased with display size (Table 1, Fig. 2). Visit frequency varied among pollinator types and the relative proportion of visits from each pollinator type depended on light environment (pollinator \times environment). Contrasts demonstrate that *Bombus* spp. (5.41/h in sun; 0.84/h in shade; $\chi^2 = 6.55$, $P < 0.001$) and halictids (5.03/h in sun; 0.12/h in shade; $\chi^2 = 2.58$, $P < 0.001$) visited sun plants more often than shade plants, whereas syrphid flies visited plants in both environments with equal frequency (1.56/h in sun; 0.84/h in shade; $\chi^2 = 0.11$, $P = 0.767$). *Bombus* were the most frequent visitor with 16% more plant visits than halictids and 190% more than syrphids. Pollinator visitation was highest at the beginning of the flowering season for both light environments. In the sun, visits dropped from 6.26 visits/h early in the season to 2.41 visits/h near the end. Similarly in the shade, visits declined from 1.27 visits/h to 0.01 visits/h near the end of the season.

Pollen deposition and removal, flower level surrogates of male and female fitness, differed between the light environments. The rate of pollen removal in the shade was only a third of that in the sun ($t = -6.79$, $P < 0.0001$; Fig. 2). Overall, pollen augmentation had a positive effect on seed production (Table 2). However, the degree of pollen limitation differed between light environments. In the shade, augmented flowers produced 50% more seeds than open-pollinated control flowers ($t = -2.65$, $P = 0.0121$), whereas seed production in the sun did not differ between treatments ($t = -0.89$, $P = 0.375$; Fig. 3). There is no evidence that increased seed production of augmented flowers is due to a relocation of resources. In the sun, there was no difference in seed number between flowers from control plants and

Table 1 Pollinator visitation to *Campanulastrum americanum* in a natural population and experimental arrays in sun and shade environments. Arrays were composed of plants with small or large displays, while display size in the natural population was recorded for every observation

Source	<i>df</i>	χ^2
Natural population		
Visits per plant		
Environment (Env)	1	26.40***
Pollinator (Poll)	2	6.55*
Poll × Env	2	13.32**
Display size	1	24.33***
Date	6	29.91***
Experimental arrays		
Visits per plant		
Env	1	56.05***
Display size	1	9.02**
Display × Env	1	0.00
Date	14	97.39***
Visits per flower		
Environment	1	23.15***
Display size	1	0.38
Display × Env	1	0.00
Date	14	59.90***

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

augmented or open-pollinated flowers (means, augmented 30.97 ± 1.79 , open 29.78 ± 1.69 , control 28.19 ± 1.33 ; control vs. augmented $t = 0.74$, $P = 0.461$; control vs. open $t = 1.25$, $P = 0.214$). In the shade, where plants were pollen limited, control plant flowers had fewer seeds than augmented flowers but similar seed numbers to open-pollinated flowers (means, augmented 11.11 ± 1.74 , open 7.4 ± 1.19 , control 4.54 ± 0.94 ; control vs. augmented $t = 3.02$, $P = 0.004$; control vs. open $t = 1.73$, $P = 0.089$). Seed production varied throughout the season, but the effect of pollen augmentation was consistent (date × pollination; Table 2).

Pollination in experimental arrays

Light environment influenced visitation of *Bombus* spp. in experimental plant arrays. Both PAR ($1,090.33 \pm 79.2 \mu\text{mol m}^{-2} \text{s}^{-1}$ sun; $16.76 \pm 2.05 \mu\text{mol m}^{-2} \text{s}^{-1}$ shade) and temperature ($30.95 \pm 0.49^\circ\text{C}$ sun; $23.96 \pm 0.29^\circ\text{C}$ shade) were greater in arrays located in a light gap than under the forest canopy. *Bombus* visits per plant and visits per flower were greater in the sun than in the shade (Table 1, Fig. 4). Pollinator visits per plant were greater for large displays than for small displays. However, the number of visits each flower received did not differ between

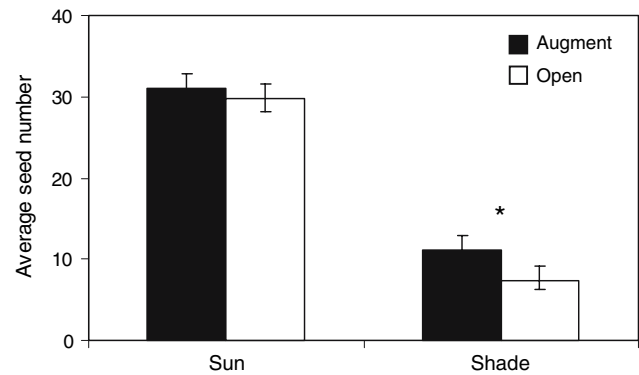


Fig. 3 The number of seeds (\pm SE) produced in open-pollinated and pollen-augmented flowers of *C. americanum* plants growing in sun and shade in a natural population. Differences between the pollination treatments indicate pollen limitation: * $P < 0.05$ see text for details

Table 2 ANOVA to test for pollen limitation on seed production in *C. americanum* growing in sun and shade environments in a natural population. Flowers were either open-pollinated (control) or pollen-augmented at regular intervals throughout the reproductive season

Source	<i>df</i>	MS	<i>F</i>
Pollination	1	1.88	6.79**
Environment	1	34.24	41.11***
Poll × Env	1	1.53	5.53*
Date	7	3.30	11.95***
Date × Poll	7	0.25	0.90
Plant (Env)	42	0.83	3.02***
Error	180	0.28	

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

plants with large displays and those with small displays (Table 1, Fig. 4).

Phenotypic selection analysis

On average, plants growing in the sun outperformed plants growing in the shade. Sun plants were 17 times larger (11.66 ± 1.07 g sun; 0.67 ± 0.09 g shade), initiated flowering 9 days earlier ($15 \text{ July} \pm 0.57$ days sun; $24 \text{ July} \pm 0.74$ days shade), flowered for nearly twice the duration (45.6 ± 1.2 days sun; 24.3 ± 1.8 days shade), produced 6 times more flowers at peak flowering (37.72 ± 3.35 sun; 6.20 ± 0.59 shade), and produced 9 times more flowers throughout the season (sum of flowers open at 5-day intervals 163.06 ± 13.31 sun; 17.58 ± 1.67 shade).

Phenotypic selection on flowering phenology was stronger in the shade than in the sun (Table 3). In the sun, the selection differentials for both fitness measures indicate that plants with earlier flowering initiation and longer flowering durations had greater fitness. However, the near-zero

Table 3 Phenotypic selection analyses^a on phenological traits in a natural population of *C. americanum* growing in sun ($n = 50$) and shade ($n = 40$)

	Seed production		Fruit production		Mass
	S	β	S	β	R
Sun					
Initiation	-0.237 [†]	-0.012	-0.260 [†]	-0.037	-0.329*
Duration	0.245 [†]	-0.083	0.304*	-0.004	0.328*
Peak	0.110	0.125*	-0.020	0.019	-0.068
Mass	0.885***	0.605***	0.931***	0.562***	
Shade					
Initiation	-0.415**	-0.596*	-0.471**	-0.301*	-0.343*
Duration	0.189	0.116	0.362*	0.223*	0.126
Peak	-0.020	0.258	-0.082	0.091	-0.137
Mass	0.533**	0.499**	0.722***	0.431***	

[†] $P < 0.1$, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

^a Separate analyses were performed for each light environment and each fitness estimate and selection differentials (S) and gradients (β) are given. Differences between S and β are primarily due to the correlation of phenological traits with biomass (R)

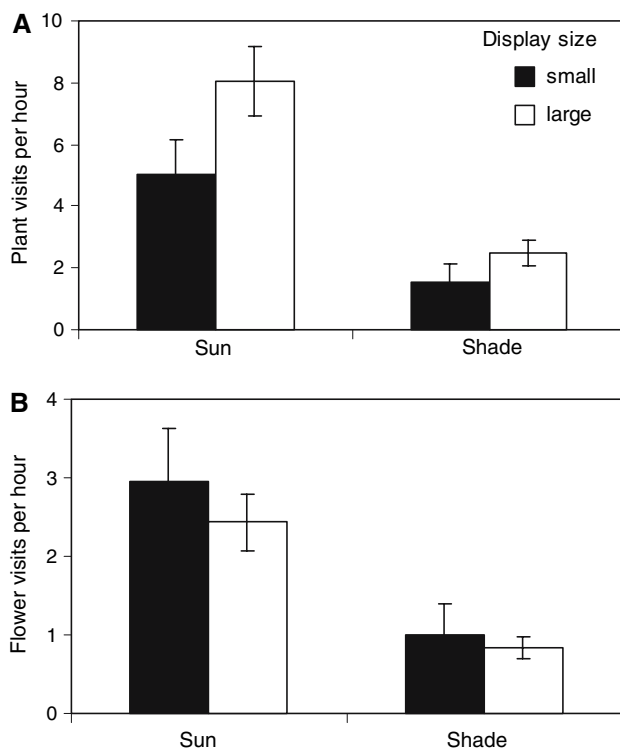


Fig. 4 *Bombus* spp. visit number (\pm SE) per plant (a) and per flower (b) to experimental arrays of *C. americanum* in sun and shade. Each array consisted of plants with either large or small display sizes

selection gradients reveal that these traits were not under direct selection, but rather that phenotypic selection was acting on these traits through their correlation with plant size. In the shade, plants that flowered earlier had higher seed and fruit production and plants that flowered longer had higher fruit production. Significant β indicate that these

traits were under direct selection. For seed production, phenotypic selection on flowering initiation was stronger in the shade (ANCOVA, environment \times initiation $F_{1,88} = 5.67$, $P < 0.05$). For fruit production both initiation of flowering (environment \times initiation $F_{1,88} = 7.64$, $P < 0.01$) and duration of flowering (environment \times duration $F_{1,88} = 4.13$, $P < 0.05$) also experience stronger phenotypic selection in the shade. In general, there was almost no selection on date of peak flowering and, not surprisingly, strong positive selection on biomass (Table 3).

Discussion

Light environment directly influenced pollinator behavior. Pollinators visited plants in the light gap more often than plants under the canopy, indicating that conditions associated with these habitats determined pollinator behavior. Activity in flying insects is typically dependent on body temperature, which is influenced by ambient air temperature and direct sunlight (Bishop and Armbruster 1999). Both temperature and irradiance levels were higher in the sun than in the shade, and so these factors likely influenced visitation rates in the two light environments. Although *Bombus* species have the ability to heat their own bodies (Heinrich 1993), and thus can be active at lower temperatures than other insects, they still preferred the sun environment. Pollinator response to environmental conditions can differ among taxa (Herrera 1997; Bishop and Armbruster 1999). Indeed, we found greater visitation by *Bombus* and halictids in the sun, but no difference in syrphid visits between light environments. Interestingly, another study found no difference in pollinator visitation between

meadow and forest environments by bumblebees or flies, although the irradiance levels ($977.4 \mu\text{mol m}^{-2} \text{s}^{-1}$ in meadow, and $49.4 \mu\text{mol m}^{-2} \text{s}^{-1}$ in forest) and the plant species (*Campanula persicifolia*) were similar to our study (Hansen and Totland 2006). This suggests that pollinator preferences may arise from local environmental conditions.

Differences in pollinator abundance between light gap and canopy habitats influenced plant reproductive success. Pollen augmentation increased seed production for shade plants but not sun plants, and there was no evidence of reallocation of resources to pollinated flowers, indicating that the shade environment was pollen limited. Also, pollen removal rates were higher in the sun than the shade, corroborating our conclusions that pollinator visits limit fitness under the forest canopy. Pollen limitation can occur when plant populations are small and have a low total number of flowers to attract pollinators (Groom 1998). This was true of plants under the canopy, where density was lower and display size smaller. Populations can also become pollen limited if pollinator behavior is influenced by the environment (e.g., Totland 2001). The experimental arrays enabled us to distinguish the effect of reduced density and display size from the effects of light level on pollinator behavior. We found arrays in the sun and with larger displays received more visits per plant. However, there was no difference in visits per flower between display size treatments indicating a proportional increase in pollinator visits with increasing display size. This is an important point, because although reproductive success is expected to increase with the number of open flowers, we found no evidence of increased pollination success in plants with larger displays. In contrast, both visits per plant and per flower were greater in the sun. Therefore light environment had direct pollinator-mediated effects on plant reproductive success, but no indirect effects mediated through pollinator response to display size (Fig. 1).

Light environment also directly influenced plant reproductive success (Fig. 1). Sun plants were larger and produced more fruits than shade plants. Resource differences between sun and shade habitats likely contributed to differences in phenotypic selection on phenological characters. We found direct selection for earlier flowering initiation (seed production and fruit production) and longer duration (fruit production) in the shade, but no direct selection for these traits in the sun. Plants in the sun flowered earlier and for a greater length of time than plants in the shade, so phenotypic selection measured in the shade habitat favored individuals with phenotypes similar to those found in the sun. Because selection gradients adjust for variation in biomass, significant gradients are evidence for selection pressures due to pollinators and not just resource availability. Whether selection pressure from pollinators will cause the evolution of phenological characters in the shade depends

on the degree of genetic control for those characters. Studies of this and nearby populations of *C. americanum* show that flowering initiation and duration are differentiated when grown in a common field environment (Haggerty 2006) and flowering initiation is heritable (Burgess et al. 2007). This suggests that variation in these traits is genetically based and can evolve in response to selection.

Differences in pollen removal and receipt between sun and shade are the major selection pressures on flowering phenology. High visitation rates early in the flowering season coupled with a later initiation of flowering of shade plants likely caused phenotypic selection for earlier flowering in the shade. Early visits may have been especially critical in the shade because plants were generally pollen limited. In the sun, selection did not favor earlier flowering initiation. This is likely because sun plants already had the phenological pattern favored by selection and received sufficient pollinator visits. Greater fruit production resulted from longer flowering durations in the shade. There was no correlation between initiation of flowering and flowering duration ($R = -0.068$, $P = 0.675$). Therefore, plants that flowered for a longer duration were not any more likely to capitalize on high early season visitation than plants that flowered for a shorter duration. Positive selection on duration may be better explained by pollen limitation. In the shade, plants that spread flower production over a longer time period would likely have more opportunities to be pollinated by rare insect visitors than plants with a compact flowering schedule. Increased flowering duration may be important in low-resource conditions in which plants cannot support large floral displays, but can support production of a small number of flowers over time.

Pollinator-mediated phenotypic selection on flower morphology has been shown to differ between environments with different levels of pollen receipt (Caruso 2000; Totland 2001; Vanhoenacker et al. 2006), but not between environments with similar levels of pollen receipt (Hansen and Totland 2006). These studies highlight the importance of environmental differences in pollen receipt on phenotypic selection for reproductive characters. However, our study is one of the few that has demonstrated environment-specific selection on flowering phenology due to differences in pollen receipt (e.g., Widén 1991). This lack of data is surprising given the importance of phenological traits to life history evolution, the abundance of studies that have considered effects of seasonal changes in pollinator abundance on phenotypic selection of plant phenology (e.g., Gross and Werner 1983; Brody 1997; Abe 2001; Wright and Meagher 2003), and findings that these selective effects vary temporally (Domínguez and Dirzo 1995).

Our results indicate that light environment has a direct resource-mediated effect and a direct pollinator-mediated effect on *C. americanum* fitness, but lacks an indirect effect

on fitness through the influence of display size on pollinators. Under this scenario, traits that increase resource capture or the number of pollinator visits would increase plant fitness. The importance of resource-mediated versus pollinator-mediated effects on plant fitness may vary depending on light environment. In particular, if pollinator abundance is low then traits that increase pollinator visitation may be at a premium in comparison to traits that increase resource capture. Indeed, phenological traits that increased pollinator visits were under selection in the pollen-limited shade environment but not in the pollen-saturated sun environment. Phenological traits that increase visits by adjusting plants to pollinator schedules may be less costly than morphological traits that increase pollinator attraction. In such a case, phenological traits should be under selection any time that pollinator-mediated effects limiting reproduction are present. Whereas, morphologically based attractive characters should only to be selected when their benefit is disproportionate to the cost of their maintenance.

Acknowledgements Thanks to Tim Kugler for work conducting array experiments, MLBS for logistical support, and NSF DBI-0453380 REU-sites for support to MLBS and DEB-0316298 to L. F. G. All experiments comply with the current laws of the United States of America.

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