FLOWER-TO-SEEDLING CONSEQUENCES OF DIFFERENT POLLINATION REGIMES IN AN INSECT-POLLINATED SHRUB

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Abstract. Insect pollinators of the Mediterranean shrub *Lavandula latifolia* (Labiatae) differ in components of pollinating effectiveness that may influence seed production and progeny quality. This study tests the prediction that *L. latifolia* flowers exposed to pollination regimes differing in pollinator composition should differ in their expected contribution to population recruitment in the field, as measured in terms of established seedlings, because of differences in seed production, progeny quality, or both. Using a randomized complete block design, two pollination regimes were induced during a 3-wk period on each of 15 *L. latifolia* plants by alternately exposing flowers in the two halves of each shrub to natural pollinators during two nonoverlapping diurnal periods (0930–1630, MD regime; dawn to 0930 and 1630 to dusk, R regime). The two pollination regimes differed in pollinator composition. The importance as flower visitors of butterflies and small bees was greater during the MD regime, which was also characterized by higher flower visitation rate and higher probability of visitation per time unit of individual flowers. MD flowers set proportionally more fruits, and each fruit contained more seeds, than R flowers. Seed production per inflorescence was also greater in MD than in R half-plants. Seeds from the MD and R regimes did not differ significantly in either mean mass, probability of germination under laboratory conditions, or probability of producing a seedling when planted in a greenhouse. When planted in the field, seeds originating from MD flowers had a greater probability of producing a seedling than those originating from R flowers, the difference being greatest where soil characteristics were most adverse for early seedling growth. In the field, MD and R seedlings did not differ in post-emergence survival rate and size of eventual survivors. The estimated average probability (± 1 SE) of one ovule eventually producing one third-year, established seedling in the home environment was significantly greater for MD (0.0192 $\pm$ 0.0041) than for R (0.0085 $\pm$ 0.0020) flowers. This demographic advantage of MD ovules was mainly accounted for by the greater seedling emergence rate of MD seeds, and only secondarily by the greater fruit- and seed-set of MD flowers. Observed patterns may be explained by a combination of increased outcrossing derived from the greater relative importance of small bees and butterflies as pollinators, and increased gametophytic competition resulting from increased overall visitation rates to individual flowers during the MD pollination regime. This study suggests that even relatively minor differences in pollinator composition and abundance may have a measurable demographic impact on plant populations.

Key words: generalized linear mixed models; insect pollination; Labiatae; Lavandula latifolia; *Mediterranean habitats; pollination consequences; pollinator composition; seed production; seedling recruitment.*

INTRODUCTION

The adaptation of flowering plants to animal pollinators has been a recurrent theme in evolutionary biology ever since Darwin's pioneering contributions (Darwin 1862, Baker 1983, Herrera 1996). A prerequisite for plant specialization is that differences among pollinators in components of pollinating quality eventually translate into differential plant reproductive success (Stebbins 1970, Waser 1983, Schemske and Horvitz 1984). By logical extension, pollination regimes differing in the composition and abundance of pollinators will lead to differential reproduction. To date, numerous studies have assessed the reproductive success associated with different pollinators or pollination regimes in terms of differential pollen deposition, pollen export, fertilization, and/or fruit and seed production rates (e.g., Bertin and Willson 1980, Motten et al. 1981, Schemske and Horvitz 1984, Davis 1987, Herrera 1987*a*, Nilsson 1988, Ramsey 1988, Wilson and Thomson 1991, Dieringer 1992, Thompson and Pellmyr 1992, Conner et al. 1995, Fishbein and Venable 1996, Stone 1996, Thøstesen and Olesen 1996, Olsen 1997). These magnitudes, however, only indirectly or partially estimate fitness, and differences between pol-

linators may lead to selection on plants only if they actually result in differential progeny production under natural field conditions. This aspect does not seem to have been directly addressed so far by any investigation.

Lavandula latifolia Med. (Labiatae) is a summer-flowering, Mediterranean shrub whose flowers are diurnally pollinated by nearly 80 species of bees, butterflies, and flies. These pollinators differ in flower visitation rate, proportion of visits resulting in pollen delivery to the stigma, amount of pollen deposited and removed per floral visit, flight distance between consecutive flower visits, and the relative importance of within vs. between-plant interfloral flights (Herrera 1987*a*, *b*, 1989; and C. M. Herrera, *unpublished data*). It was previously hypothesized that these differences should lead to flowers pollinated by different pollinators to differ in average seed production rate and/or progeny quality (Herrera 1987*a*, *b*). The objective of the present study was to test one specific prediction emanating from this hypothesis, namely that flowers of *L. latifolia* exposed to natural pollination regimes differing in pollinator composition should differ in their eventual contribution to population recruitment. Population recruitment was measured in terms of established seedlings or maternal fitness, because of differences in seed production, progeny quality, or both.

The relative importance of the major pollinator groups of *L. latifolia* changes predictably over daytime: butterflies and small bees are most abundant around the middle of the day, while large bees and flies tend to visit most flowers in early morning and late afternoon (Herrera 1990*a*). Furthermore, major pollinator groups differ consistently in aspects of pollinating effectiveness. Butterflies, flies and small bees pollinate flowers less frequently, and deposit smaller pollen loads, than large bees. However, they tend to fly longer distances between consecutive flower visits than large bees, and a considerably greater proportion of their foraging flights are between flowers on different shrubs. These differences may result in differential seed production and progeny quality (Herrera 1987*a*, *b*, 1990*a*). To test the prediction above, I took advantage of these circumstances and experimentally induced two different pollination regimes on *L. latifolia* plants in the field by exposing flowers to natural pollinators at two nonoverlapping daytime periods. The effects on reproductive success were subsequently assessed from the flower through the established seedling stage.

Methods

Plant natural history

L. latifolia is a low evergreen shrub (up to 35 cm high) producing long-stalked (up to 1.25 m high) inflorescences in early summer. It is common in the understory of open mixed woodlands at middle elevations in the eastern and southeastern Iberian Peninsula. The composition, abundance, and foraging behavior of pollinators, and most aspects of *L. latifolia* reproductive ecology, have been described elsewhere (Herrera 1987*a*, *b*, 1988, 1989, 1990*a*, *b*, 1991, 1995, 1997). Flowering takes place from early to mid July to late September. Flowers are produced over a terminal portion of the stalks, have pale blue tubular corollas (tube length 6–8 mm), and are hermaphroditic and protandrous. New flowers open continuously throughout the day, and each flower lasts for 1.5–2.5 d. The proportion of open flowers in the receptive female stage tends to remain roughly constant. Flowers are self-compatible, but spontaneous autogamy occurs very infrequently due to protandry and to the spatial separation of anthers and stigma. In the absence of pollinators, <4% of flowers set fruit. As blossoms are asynchronous within one inflorescence, the male and female stages of different flowers are not in phase with each other. Thus, geitonogamous pollinations can readily take place. Relative to self-pollination, outcross pollination results in greater proportion of flowers setting fruit, and greater proportion of ovules yielding seeds per fruit. In most sites and years, the proportion of flowers setting fruit is not limited by pollen supply for the majority of *L. latifolia* individuals. The number of seeds per fruit, however, seem to be limited by the number of pollen tubes reaching the ovary, which in turn depends on the number and quality of pollen grains received by stigmas during the flowers' female stage. As individual plants in any given population vary significantly in the mean number of pollen tubes reaching the ovary, the opportunity exists for selection to operate on any pollination-related trait(s) consistently enhancing the quantity or quality of pollen deposited on stigmas (C. M. Herrera, *unpublished data*). Seed maturation takes place 4–6 wk after anthesis. After maturation, seeds become loose within the calyx and independently fall to the ground. Most seeds germinate in April or May of the first spring after dispersal, and seedling mortality is concentrated on the first few weeks after emergence. Seedlings ordinarily occur at moderate to low densities and early mortality is largely density independent (C. M. Herrera, *unpublished data*).

Study site

This study was conducted in the Reserva de Navahondona-Guadahornillos, Parque Natural de Cazorla-Segura-Las Villas, Jaén province, southeastern Spain (see Luque [1995] for descriptions of the vegetation). The study population was located around the intersection of Arroyo Aguaderillos and the track joining Roblehondo and Hoyos de Muñoz, at 1160 m elevation. This is the ''Aguaderillos-1'' site of earlier studies (Herrera 1988, 1991, where further details may be found).

Pollination regimes

Two different pollination regimes were induced on each of 15 *L. latifolia* shrubs at the study locality. Each

PLATE 1. Honeybee (*Apis mellifera*, Hymenoptera: Apidae; left) and silver-spotted skipper butterfly (*Hesperia comma*, Lepidoptera: Hesperiidae; right) feeding on nectar in flowers of *Lavandula latifolia*. Bees and butterflies are the main pollinators of this summer-flowering Mediterranean shrub in southeastern Spain. Photographs by Carlos M. Herrera.

experimental plant was divided into two parts as similar in size as possible by vertically inserting a sheet of black tulle into the shrub. The sheet was suspended from a horizontal wire stretched between two vertical, green-painted metal rods 1.5-m high. Each half-plant was randomly assigned to one of two pollination regimes. In each plant, one pollinator exclosure made of a large piece of 1-mm mesh black tulle was changed in position twice daily from 23 July to 12 August 1991, so as to alternately expose one half-plant (and always the same half-plant) to pollinators from 0930 to 1630 (Greenwich Mean Time; midday or MD pollination regime hereafter), and the other half-plant for the rest of the daytime (from dawn to 0930, and again from 1630 to dusk; rest, or R pollination regime). These two specific time intervals were expected to maximize the contrast in pollinator composition, and were determined by extensive dawn-to-dusk pollinator census data from the study locality prior to the initiation of the study by applying a *k*-means clustering algorithm (procedure FASTCLUS in the SAS package; SAS Institute 1990) (Herrera 1988, 1990*a*).

Pollinator censuses were carried out on all experi-mental plants from dawn to dusk throughout the experimental pollination period (Plate 1 shows two common pollinators). Each census lasted for 5 min, during which I closely watched the activity of pollinators at the currently exposed half of one of the shrubs. I identified visually all flower visitors to species, and counted the total number of flowers probed by each visiting insect. In each pollination regime, plants were censused in turn according to a random permutation scheme. When a round of censuses was completed on all the plants, a different random permutation was used. The total number of open flowers in each half-plant was counted daily over the whole census period to assess flower availability to pollinators.

Fruit-set and seediness

To estimate the proportion of flowers setting fruits, or fruit-set, 10 inflorescences were marked on each half-plant at the beginning of the study. Cumulative flower production over the 3-wk experimental pollination period, and total number of ripe fruits (the unit formed by the persistent calyx plus the enclosed ripe nutlets) eventually produced, were determined for each

TABLE 1. Summary of variables, sampling and measurement units, and generalized linear mixed models used in this study to test for the differences between, and effects of, experimentally induced pollination regimes on plants of *Lavandula latifolia*.

Analysis no.	Variable	Sampling unit (N_{MID}, N_R)†	Measurement units
1	Pollinator approach rate	5-min census (226, 211)	Number of insect individuals
2	Flower visitation rate	5-min census (226, 211)	Number of flowers visited
3	Flower visitation probability	5-min census (226, 211)	Proportion of open flowers visited
4	Fruit-set	Inflorescence (150, 150)	Proportion of flowers setting fruit
5	Fruit seediness	Fruit (590, 506)	Number of seeds per mature fruit
6	Total seed production	Half-plant (15, 15)	Number of mature seeds
7	Seed mass	Seed (750, 750)	Milligrams
8	Seed germinability	Petri dish (30, 30)	Proportion of seeds germinating
9	Greenhouse seedling emergence rate	Half-container (45, 45)	Proportion of seeds yielding seedlings
10	Field seedling emergence rate	Subplot (37, 37)	Proportion of seeds yielding seedlings
11	Field seedling survival	Subplot (37, 37)	Proportion of seedlings surviving
12	Field seedling size	Seedling (72, 52)	Number of leaves

† N_{MD} and N_R are the numbers of sampling units in the MD and R pollination treatments, respectively, all plants combined.

‡ Codes and levels (in parentheses) for effects in models are as follows: AGE = age of seeds (1st vs. 2nd year); PLANT = individual plant (N = 15); SITE = location of experimental plantings in the field (N = 3 sites); TREAT = pollination regime (MD vs. R).

¶ Flowers of *Lavandula latifolia* have a fixed complement of four ovules. Thus the number of filled seeds per mature fruit was modeled as a binomial response variable, i.e., the proportion of the four ovules developing into seeds.

§ To account for slight differences between experimental units (half plants) in number of inflorescences (and thus flower production), which could influence the total number of seeds produced, number of inflorescences (log-transformed) was included as an offset variable (roughly analogous to a covariate) in this analysis. A log-linear model was thus fitted to the ratio of seed production to inflorescence number.

‖ The age of seedlings when leaves were counted (days elapsed from the date the seedling was first recorded, log-transformed) was included in the model as an offset variable.

marked inflorescence using methods based on marking individual flowers, as described in Herrera (1991). Fruit counts were performed in mid-September. At that time, fruit seediness or the number of enclosed ripe nutlets was also determined for each ripe fruit.

After completion of the experimental pollination period, pollinators were definitively excluded from experimental plants for the rest of the flowering season by individually bagging all inflorescences with a fine-mesh cloth. The cloth prevented the loss of seeds shed from ripe fruits. After fruit ripening and seed dispersal in mid-September to mid-October, all the mature seeds produced by flowers that had been subjected to experimental pollination regimes were collected from inside the bags, counted to determine total seed production by each experimental half-plant, and stored in the dark in paper bags at room temperature. Seeds from different half-plants were kept as separate batches.

Seeds and seedlings

Mean mass of seeds produced by each half-plant was determined by individually weighing a random sample of N = 50 seeds to the nearest 0.01 mg. Germination of seeds was tested in the laboratory by placing them on moist cotton in Petri dishes at room temperature (N = 25 seeds per half-plant). The number of germinating seeds with a radicle protruding at least 1 mm from the seed coat was recorded daily for 45 d. Preliminary analyses comparing the shape of cumulative germination curves did not reveal any difference between seeds from the two pollination treatments. Thus, the proportion of seeds germinated by the end of that period was the variable considered in the analyses. To evaluate the possibility of a differential decline in seed viability between the MD and R treatments, germination tests were performed during the first (1992) and second (1993) spring after fruit ripening.

The effect of pollination regime on the probability of seedling emergence was investigated by means of seed-planting experiments conducted in the field and the greenhouse. In the greenhouse experiment, seed samples were sown the winter following fruit ripening (January 1992) in Rootrainer (Ronaash Limited, Kelso, England) plastic trays filled with a homogenized mixture of washed sand, standard peat moss, and natural organic fertilizer (ground sheep manure). Three trays (replicates) were used for each of the 15 mother plants. Each tray was divided into two halves corresponding

TABLE 1. Extended.

| | Generalized Linear Mixed Model parameters‡ | | |
Error distribution	Link function	Fixed effects	Random effects
Poisson	Log	TREAT	PLANT, PLANT × TREAT
Poisson	Log	TREAT	PLANT, PLANT × TREAT
Binomial	Probit	TREAT	PLANT, PLANT × TREAT
Binomial	Complementary log-log	TREAT	PLANT
Binomial ¶	Logit	TREAT	PLANT
Poisson §	Log	TREAT	PLANT
Normal	Identity	TREAT	PLANT
Binomial	Logit	TREAT, TREAT × AGE	PLANT
Binomial	Probit	TREAT	PLANT, PLANT × TREAT
Binomial	Probit	TREAT	SITE, PLANT (SITE)
Binomial	Complementary log-log	TREAT	SITE, PLANT (SITE)
Poisson ‖	Log	TREAT	SITE, PLANT (SITE)

to the MD and R treatment, and 16 seeds sown in each. Trays were watered as needed and periodically relocated inside the greenhouse. Emergence of seedlings was recorded weekly for 6 mo until the end of the experiment in late June 1992. Seedlings were considered emerged when any part of the seedling broke through the soil surface.

In the field experiment, seed samples were also sown the winter following fruit ripening (February 1992) at three different locations: the Aguaderillos home site and two nearby localities, Agracea (2 km away, 1200 m elevation) and Correhuelas (4 km away, 1600 m elevation). These two additional sites were included to broaden the range of field conditions faced by planted seeds, and were chosen because of their evident contrast with Aguaderillos in soil characteristics (see *Results: Reproductive consequences: Seeds to seedlings*). At each location, the planting array replicated the design used to induce the experimental pollination regimes on plants, the only exception being that two plants did not have their seeds sown at all sites due to an insufficient number of seeds. At each site, rectangular plots (25 × 50 cm) scattered over the planting area (~10 m²) were randomly assigned to mother plants. Each of these plots was in turn subdivided into two contiguous 25 × 25 cm subplots, which were randomly assigned to receive seeds from either the MD or R treatment half-plants ($N = 25$ seeds sown per subplot). Seeds were individually placed ~5 mm deep in 1.5-mL microcentrifuge tubes with the bottom cut off, embedded in the soil, and filled with sieved and homogenized local topsoil. Emerging seedlings were monitored weekly during the spring germination season at all sites until early summer of 1994. The criterion

for scoring an emerged seedling was the same as in the greenhouse experiment. The monitoring period encompassed three consecutive spring seed germination periods (1992, 1993, 1994). Of all seedlings emerging, most did during the first (74.5% of total) and second (24.2%) years, while the number of emergences was negligible during the third year (1.3%). I am thus confident that the cumulative number of seedlings emerging between 1992 and 1994 adequately reflects the actual emergence prospects of seeds sown under field conditions. Seedling survival rate was estimated as the proportion of emerged seedlings remaining alive by the end of the monitoring period (summer 1994). The number of leaves of surviving seedlings was used as a size index. Comparisons of seedling size accounted statistically for differences in the age of seedlings by including this later variable as a covariate.

Five soil samples (top 20 cm soil, excluding gross surface litter) were collected at each of the three planting sites (nearest sampling points were 1.25–1.75 m apart). Texture (percent coarse and fine sand, silt, and clay content) and chemical (pH, organic matter, nitrogen, potassium, and phosphorus) characteristics were determined on these samples using standard soil analytical procedures.

Statistical analyses

Interspersion of experimental units was ensured, and pseudoreplication avoided, by consistently using a randomized complete block design (Hurlbert 1984). This applies not only to the initial induction of the two pollination regimes on plants, but also to subsequent field and greenhouse experiments. Individual plants were in all cases treated as blocks and plant halves as experi-

TABLE 2. Percentage of total flower visits recorded in censuses contributed by major pollinator groups and species in the two pollination regimes. N = total number of flowers visited, all species combined.

Major group and species	Pollination regime†		Partial χ^2‡
	MD (N = 2440)	R (N = 1676)	
Large bees (body length >11 mm)			
Apis mellifera	52.4	50.1	
Bombus terrestris	10.8	27.4	
Anthophora ochroleuca	2.4	2.8	
Total (6 species)	69.5	81.9	30.12
Small bees (body length <9 mm)			
Anthidium breviusculum	8.5	4.6	
Ceratina cyanea + mocsaryi	8.7	2.4	
Megachile pilidens	1.1	0.0	
Total (5 species)	19.5	7.5	90.96
Butterflies			
Argynnis paphia	2.0	1.3	
Argynnis adippe	2.4	0.0	
Thymelicus acteon	2.3	0.4	
Total (12 species)	8.9	3.7	35.90
Flies (6 species)	1.1	2.5	12.65

† In each column, percentages do not sum to 100 because flower visitation data for two species of sphecid wasps and one species of day-flying hawk moth were omitted from the table.

‡ Contribution of each major pollinator group to the overall χ^2 of the Group $\times$ Pollination Regime two-way table (χ^2 = 169.6, df = 3, P < 0.0001). For every major group, the proportions of flowers visited in the MD and R regimes differed significantly (P < 0.001; significance was tested by comparing the partial χ^2 for each group with the critical value from the χ^2 distribution for df = 1).

mental units, each of which corresponded to one of the two pollination regimes.

The effect of pollination regime on the variables describing reproductive performance from the flower through the established seedling stage was tested by fitting generalized linear mixed models to the data. The application of generalized linear models (McCullagh and Nelder 1989) is justified by the nature of dependent variables under consideration. Only seed mass is expected to follow a normal distribution. All the other parameters examined here to assess reproductive performance are not expected to be normally distributed, as they represent either counts or proportions (Table 1). Nevertheless, conventional generalized linear model estimation treats all effects in a model as fixed. This represents a serious limitation in the context of the present study, where the randomized block design that underlies all analyses requires the treatment of plant as a random effect. Generalized linear *mixed* models represent a particularly useful extension of ordinary generalized linear models, as they combine the desirable properties of relaxing normality assumptions and at the same time allow for a distinction between fixed and random effects in the model (for further details, see Littell et al. 1996). Computations were performed with SAS Macro program GLIMMIX (Version 19 May 1997; available on the World Wide Web at http://www.sas.com/techsup/download/stat), which iteratively calls SAS Procedure MIXED (SAS Institute 1996*a*) until convergence, in the context of Wolfinger and O'Connell's (1993) method of reweighted likelihoods.

Model parameter estimates were fitted using the restricted pseudolikelihood method (Littell et al. 1996). The MIXED procedure implements a generalization of the standard linear model that allows for proper incorporation of random effects. For further details on the MIXED procedure, see Littell et al. (1996) and SAS Institute (1996*a*, *b*).

A summary of all the models fitted to the data is shown in Table 1. Rather than automatically applying in each analysis the canonical link function corresponding to the presumed error distribution of the data (McCullagh and Nelder 1989), I first fitted each model using all applicable link functions, and then selected the one that minimized the deviance of the model. Pollination regime was in all cases treated as a fixed effect, and Plant and the interaction Plant $\times$ Treatment (in those analyses where true replication within experimental units allowed for estimation of this effect) as random effects. As this study focuses on the consequences of the different pollination regimes, I do not consider covariance parameter estimates and significance tests associated with random effects in models. It must be noted, however, that covariance components associated with the Plant effect were almost always different from zero, thus denoting the existence of significant individual variation in most reproductive parameters considered.

RESULTS

The two pollination regimes

The pollinator assemblage.—A total of 32 insect taxa were recorded visiting *L. latifolia* flowers during

TABLE 3. Summary of characteristics of the two pollination regimes experimentally induced on *Lavandula latifolia* plants.

Analysis[†] no.	Variable	Treatment means (95% confidence limits)[‡]		Wald χ^2	P
		MD	R		
1	Pollinator approach rate (individuals/census)				
	Large bees	0.48 (0.32–0.74)	0.43 (0.28–0.67)	0.57	0.50
	Small bees	**0.26** (0.14–0.46)	**0.06** (0.03–0.13)	10.03	**0.006**
	Butterflies	**0.21** (0.11–0.38)	**0.06** (0.03–0.13)	10.39	**0.001**
	All species	**1.00** (0.66–1.52)	**0.62** (0.40–0.95)	6.02	**0.014**
2	Flower visitation rate (flowers/census)				
	Large bees	5.33 (2.99–9.50)	4.59 (2.55–8.27)	0.41	0.52
	Small bees	**1.87** (1.08–3.21)	**0.49** (0.24–1.04)	9.39	**0.002**
	Butterflies	**0.64** (0.32–1.31)	**0.18** (0.08–0.44)	10.68	**0.001**
	All species	8.16 (4.92–13.49)	5.90 (3.49–9.84)	2.48	0.11
3	Single-flower visitation probability (per census)				
	Large bees	0.094 (0.065–0.131)	0.062 (0.042–0.090)	3.61	0.057
	Small bees	**0.031** (0.020–0.045)	**0.006** (0.003–0.012)	18.41	**<0.0001**
	Butterflies	**0.011** (0.007–0.019)	**0.002** (0.001–0.005)	20.99	**<0.0001**
	All species	**0.144** (0.110–0.184)	**0.080** (0.058–0.107)	10.74	**0.001**

Note: Treatment means and *P* values corresponding to statistically significant differences are shown in bold type.

† Numbers correspond to Analysis column in Table 1.

‡ Model-adjusted treatment means. In GLIMMIX, model-adjusted treatment means are expressed in terms of the link function used in the model (see Table 1). Values presented here are expressed in the original measurement scale after back-transforming using the inverse link. Confidence intervals were also obtained by back-converting to the original scale the parametric (*t*-based) confidence limits expressed in terms of the link function; hence their asymmetry around the mean.

this study. All of them should be considered as legitimate pollinators of this species on the basis of previous investigations (Herrera 1987*a*) or their behavior at flowers recorded during this study. Large bees (body length >11 mm; 6 species, 74.5% of flowers visited), small bees (body length <9 mm; 5 species, 14.6% of flowers visited), and butterflies (12 species, 6.8% flowers visited) were the most important pollinator groups, accounting collectively for 96% of all flowers visited during pollinator censuses ($N = 4116$ flowers). The importance of flies (6 species, 1.7% of flowers visited) was negligible. Considering individual species, 9 taxa (listed in Table 2) were responsible for 90% of total flower visits. *Apis mellifera* was responsible for 51.5% of all flowers visited, followed by *Bombus terrestris* (17.5%), *Anthidium breviusculum* (6.9%), and *Ceratina cyanea + mocsaryi* (6.1%).

The two pollination regimes differed significantly in the proportion of flowers visited by the four major groups of pollinators (Table 2; $\chi^2 = 169.6$, df = 3, *P* < 0.0001). The proportional importance of large bees and flies was significantly greater during the R regime, and that of small bees and butterflies significantly greater in the MD regime (Table 2). Partial χ^2 figures indicate that the main compositional differences between the MD and R regimes were due to the variations exhibited by small bees and, to a lesser extent, butterflies and large bees (Table 2). For this reason, and because of its negligible importance during the two pollination regimes, flies will not be considered again in the remainder of this paper.

Flower visitation.—This section compares the MD and R pollination regimes with regard to three different quantitative aspects of flower visitation, namely pollinator approach rate (number of different insect individuals visiting a half-plant per census), flower visitation rate (number of flowers visited per census), and flower visitation probability (the probability of single flowers in half-plants being visited at least once during the census period), and correspond to Analyses 1, 2, and 3 in Table 1, respectively.

Considering all pollinator species combined, the MD regime was characterized by significantly greater pollinator approach rate (Table 3). Analyzing the data for large bees, small bees, and butterflies separately, however, only the difference for small bees and butterflies reached statistical significance. Average flower visitation rates were greater in MD than in R for both small bees and butterflies, but not for large bees or for all species combined. Finally, the mean estimated probability of individual flowers being visited during a 5-min period was significantly greater in MD than in R, for all species combined and for both small bees and butterflies analyzed separately (Table 3). The probability of visitation by large bees was also greater during MD, but the difference was only marginally significant. In summary, the MD regime was mainly characterized by (1) higher activity of pollinators at plants, as measured by approach rates, and higher probability of individual flowers being visited per time unit period; and (2) greater importance of small bees and butterflies as pollinators.

TABLE 4. Effects of experimentally induced pollination regimes on the reproductive success of *Lavandula latifolia* from the flower through the established seedling stages.

Analysis no.[†]	Variable	Treatment means (95% confidence limit)[‡]		Wald χ^2	P
		MD	R		
4	Fruit-set	**0.58** (0.51–0.64)	**0.53** (0.47–0.59)	6.61	**0.010**
5	Fruit seediness (seeds/fruits)	**1.55** (1.44–1.67)	**1.40** (1.29–1.52)	16.25	**<0.0001**
6	Total seed production§	**5.91** (3.75–9.31)	**5.07** (3.22–8.00)	6.35	**0.012**
7	Seed mass (mg)	1.35 (1.26–1.45)	1.34 (1.24–1.43)	1.85	0.17
8	Seed germinability‖	0.90 (0.85–0.94)	0.93 (0.88–0.96)	1.31	0.25
9	Greenhouse seedling emergence rate	0.057 (0.028–0.105)	0.054 (0.026–0.101)	0.04	0.85
10	Field seedling emergence rate	**0.19** (0.13–0.26)	**0.13** (0.09–0.19)	7.30	**0.007**
11	Field seedling survival	0.10 (0.0–1.0)	0.12 (0.0–1.0)	0.30	0.58
12	Field seedling size¶	0.15 (0.13–0.18)	0.15 (0.12–0.18)	0.17	0.68

Note: Treatment means and *P* values corresponding to statistically significant differences are shown in bold type.

† Numbers correspond to Analysis column in Table 1.

‡ Model-adjusted treatment means. See note in Table 3.

§ Figures for treatment means represent estimates of total seed production on a per-inflorescence basis, as the total number of inflorescences per half-plant was included in the model as an offset variable (Table 1).

‖ The TREAT × AGE effect was also included as a further fixed effect in this analysis (Table 1) and was not found to be significant (Wald $\chi^2 = 5.20$, $P = 0.074$).

¶ Figures for treatment means represent estimates of number of leaves per week of age, as seedling age was included in the model as an offset variable (Table 1).
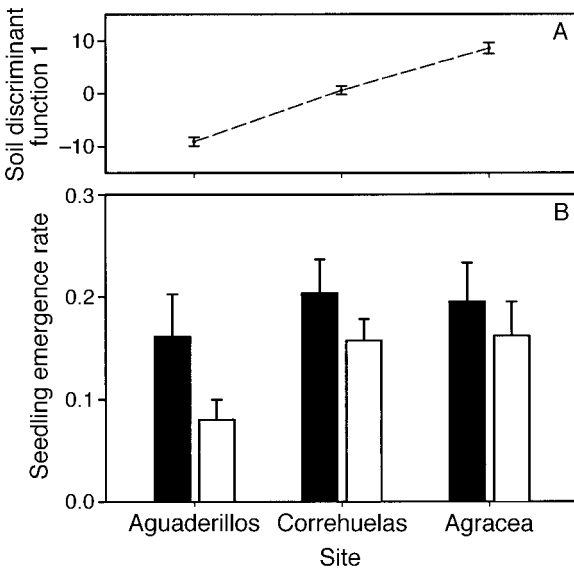

FIG. 1. Variation among field planting sites in soil characteristics and seedling emergence rate of seeds from the two pollination regimes. (A) Mean values (±2 SE) of the first canonical discriminant function for soil characteristics. See Table 5 for correlations between soil parameters and this discriminant function. (B) Seedling emergence rate (probability of producing a seedling) of seeds from the MD (black bars) and R (open bars) pollination regimes. Vertical bars extend over 1 SE. Means were obtained by back-transforming model-adjusted values obtained on the probit scale. Standard error estimates were obtained by bootstrapping.

Reproductive consequences

Flowers to seeds.—MD flowers exhibited a significant fecundity advantage over R flowers, because they set proportionally more fruits and each fruit contained more seeds (Table 4). This fecundity differential in favor of MD flowers was corroborated by independent estimates based on counts of the actual number of seeds produced by half-plants. After statistically accounting for differences between half-plants in the number of inflorescences, mean seed production per inflorescence was ~17% higher in MD than in R experimental units (Table 4).

Seeds from the MD and R regimes were statistically indistinguishable with regard to either mean mass or probability of germination when tested in Petri dishes in the laboratory (Table 4). Laboratory germination rate was very high (≥90%) for both MD and R seeds, and no evidence was found that storage of seeds at ambient temperature for 1 yr had any differential effect on the germination of MD and R seeds (Table 4).

Seeds to seedlings.—When planted in the greenhouse, individual seeds from the two pollination regimes did not differ significantly in seedling emergence rate (Table 4). When seeds were planted in the field, in contrast, seedling emergence of seeds from MD flowers was significantly greater than that of seeds from the R regime (Table 4).

When seedling emergence data were analyzed separately for the three planting sites, MD seeds were still more likely to produce a seedling than R seeds at all sites, yet the magnitude of the treatment effect varied

TABLE 5. Variation in soil characteristics among planting sites.

Soil parameter	Site			Difference between sites		Correlation with canonical variable	
	Aguaderillos	Correhuelas	Agracea	F	P	First	Second
Texture							
Coarse sand (%)	6.1 ± 2.3	16.3 ± 2.0	31.6 ± 6.3	50.2	<0.001	0.939	−0.166
Find sand (%)	4.4 ± 1.2	5.6 ± 1.2	5.9 ± 0.9	2.9	0.096	0.550	0.166
Silt (%)	28.3 ± 2.3	22.3 ± 2.6	28.1 ± 3.7	6.7	0.011	−0.071	−0.748
Clay (%)	61.2 ± 1.5	55.7 ± 2.5	34.4 ± 5.9	68.6	<0.001	−0.896	0.373
pH	7.6 ± 0.3	7.7 ± 0.1	7.7 ± 0.1	1.5	0.26	0.379	0.249
Organic matter (%)	2.9 ± 0.6	6.1 ± 0.4	6.0 ± 0.7	46.6	<0.001	0.844	0.444
Nitrogen (%)	0.17 ± 0.04	0.22 ± 0.02	0.26 ± 0.04	9.0	0.004	0.781	0.006
Phosphorus (μg/g)	0.96 ± 0.75	2.96 ± 2.58	6.56 ± 1.70	12.0	0.001	0.802	−0.184
Potassium (mg/g)	0.42 ± 0.03	0.49 ± 0.02	0.28 ± 0.06	31.7	<0.001	−0.568	0.750

Note: Mean values ± 1 SD are shown for each parameter considered ($N = 5$ soil samples per site). The table also reports univariate tests of among-site heterogeneity and the total sample correlations between the original and the two canonical variables obtained from canonical discriminant analysis (see also Fig. 1A).

among sites (Fig. 1B). The advantage of MD over R seeds was greatest when seeds were planted in the Aguaderillos home environment, and declined when planted in Correhuelas and Agracea. It must be emphasized that it is the absolute magnitude of the difference, not its direction, that varied among sites, as illustrated by the nonsignificance of the Treatment × Site interaction (Wald $\chi^2 = 1.02$, df = 2, $P = 0.60$; a variant of Analysis 10 in Table 1 having Site, Treatment and their interaction as fixed effects and individual plants as replicates). This variation across planting sites was mainly due to differences in seedling emergence
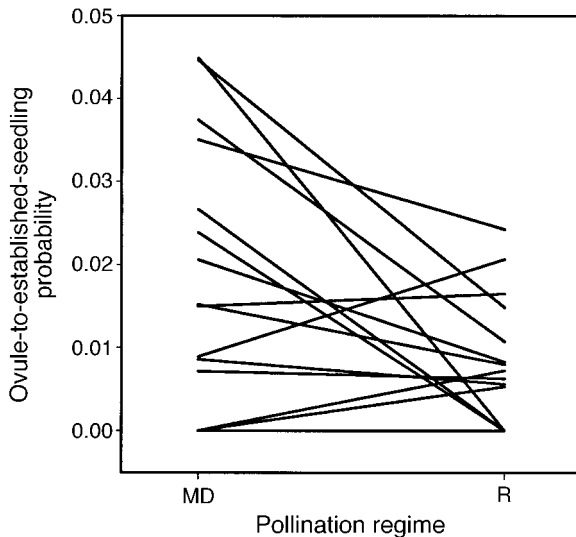

FIG. 2. Effect of pollination regime on the estimated probability of an individual ovule eventually producing a 3-yr-old established seedling in the Aguaderillos home environment. Probabilities were estimated separately for each half-plant by multiplicatively combining transition probabilities from the ovule through the seed, newly emerged seedling, and established seedling stages. In the graph, each line corresponds to a different plant, and it connects the values obtained for the two experimental halves.
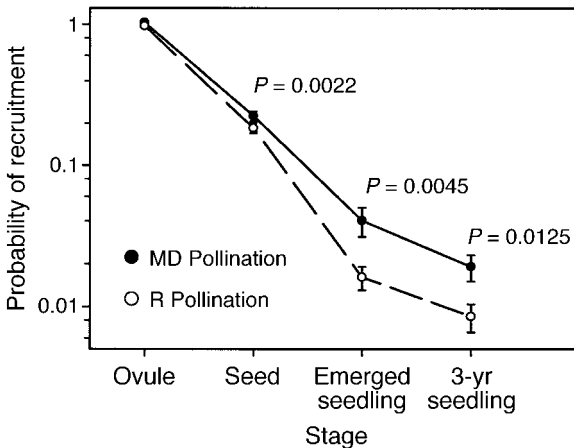
rate of R seeds, which increased from Aguaderillos through Correhuelas to Agracea (Fig. 1B).

As anticipated, the three planting sites differed significantly in mean soil characteristics. They differed in all the soil parameters considered excepting pH and proportion of fine sand (Table 5). In comparison with the other two sites, Aguaderillos soil stood out because of its high clay content, and low organic matter, nitrogen, and phosphorus content (Table 5). A canonical discriminant analysis of soil analytical data revealed significant heterogeneity among sites in the position of centroids in the multivariate space defined by soil parameters (F statistic for Wilk's $\lambda = 13.27$, df = 18, 8, $P < 0.001$). The first canonical discriminant function, accounting for 82.7% of variance, reflected a gradient defined by organic matter, nitrogen, and phosphorus on the positive extreme, and clay and potassium content on the negative one (Table 5). Average values for the three planting sites were rather evenly spaced along this gradient (Fig. 1A).

Seedlings that emerged from MD and R seeds did not differ significantly in either percent survival up to the end of the study, or final size, measured by the number of leaves (after statistically accounting for age differences) (Table 4).

Recruitment prospects in the home environment

The effect of pollination regime has so far been tested by separately considering different stages in the reproductive process. Significant effects have been found for some stages but not for others. Do observed differences in fruit-set and seediness, and in seedling emergence rate in the field, effectively result in differential seedling recruitment prospects for MD and R flowers? If they do, what reproductive components are more directly responsible for the difference? I will address these questions in this section, referring specifically to the conditions of the Aguaderillos home environment.

FIG. 3. Mean probability (±1 SE) of recruitment from the ovule through the established (3-yr-old) seedling stage at the Aguaderillos study site for flowers exposed to the MD (filled dots) and R (open dots) pollination regimes. Transition probabilities were separately obtained for each half-plant by multiplicatively combining figures for fruit-set and seediness, seedling emergence rate, and seedling survival, and then averaged across plants. Computations took into consideration that *L. latifolia* flowers have a fixed complement of four ovules per ovary. *P* values denote significance levels of the difference between MD and R regimes up to a given recruitment stage, assessed using a one-tailed *t* test for paired data and randomization.

For each half-plant I estimated the probability of an individual ovule contributing a 3-yr-old, established seedling to the home population. This was done by multiplicatively combining for each half-plant the figures for fruit-set and seediness, and seedling emergence rate and survival for seeds planted in the Aguaderillos site. These computations took into consideration that *L. latifolia* flowers have a fixed complement of four ovules per ovary. Results are summarized in Fig. 2. On average, the probability of an ovule eventually contributing an established, 3-yr-old seedling to the Aguaderillos population was significantly greater for MD (mean ± 1 SD = 0.0192 ± 0.0157; N = 15 halfplants) than for R (0.0085 ± 0.0076; N = 15 halfplants) flowers (P = 0.012; tested using one-tailed Student's *t* test for paired data; significance assessed by means of randomization with 5000 repetitions).

To evaluate the relative importance of the various reproductive stages in determining the differential recruitment prospects between MD and R ovules, I also estimated for each half-plant the transition probabilities from ovule to seed and from ovule to newly emerged seedling. Mean recruitment probabilities from the ovule through the established seedling stages are separately plotted in Fig. 3 for MD and R flowers. The difference between MD and R seeds in transition probability from the seed to the newly emerged seedling stage was the critical determinant of overall difference in ovule-to-established-seedling recruitment prospects.

The combined influence of differences in fruit-set and seediness was negligible.

DISCUSSION

The experimental design: rationale and potential limitations

The use of half-plants, rather than whole shrubs, as experimental units was justified by practical and statistical considerations. Differences between experimental pollination regimes in pollinator composition were a priori expected to be small, which should also lead to small treatment effects on plant reproduction. I expected variance in reproductive parameters induced by the pollination treatment to be much smaller than naturally occurring between-plant variance in these parameters, an expectation confirmed in this study (Fig. 2). To achieve sufficient statistical power while using whole plants as experimental units, the number of individuals sampled should have been considerably greater than that used here. This was impractical, mainly because of the impossibility of conducting sufficient pollinator censuses on many individual plants. The randomized complete block design adopted provided a convenient way of increasing statistical power without inordinately increasing sampling effort. Assuming that individual plants (blocks) are internally homogeneous units, blocking allowed a more precise estimation of treatment effects through the elimination from the comparison of treatments of large differences between experimental units in different blocks (Mead 1988). This explains why this study was able to detect small treatment effects in spite of broad confidence intervals around treatment means (Tables 3 and 4). Fig. 2 exemplifies a statistically significant treatment effect despite broad within-treatment variability due to individual variation.

The block design used here has one possible limitation that must be explicitly acknowledged, namely that the two experimental units (half-plants) recognized in each block might be not fully independent in their response to the treatment. This would happen, for instance, if decreased fruit-set and seediness in one half-plant (caused, e.g., by reduced pollination) enhanced reproductive output in the adjacent halfplant by relaxing among-inflorescence competition for resources and inducing resource translocation between contiguous experimental units. Although there is no information on patterns of within-plant resource allocation in *L. latifolia*, relevant data on the congeneric *Lavandula stoechas* (quite similar to *L. latifolia* in general architecture and inflorescence structure) suggest that experimental units were largely independent from the viewpoint of resource allocation. Inflorescences of *L. stoechas* have been experimentally shown to behave as isolated modules of resource allocation, and reallocation among inflorescences of the

same plant is either negligible or does not occur at all (J. Herrera 1991).

Pollination regimes and their effects

Previous investigations have taken advantage of differences among pollinators in activity rhythms to generate artificial nighttime vs. daytime pollination regimes on plants pollinated by both diurnal and nocturnal animals (Bertin and Willson 1980, Jennersten and Morse 1991, Guitián et al. 1993, Sahley 1996). In this study, two diurnal pollination regimes differing in pollinator abundance and composition were induced on experimental *L. latifolia* shrubs. Pollinator approaches per census period (all species combined) were more frequent, and individual flowers had a higher probability of being visited at least once per time unit period, during the MD regime. The MD regime was also characterized by reduced importance of large bees and, principally, a concomitant increase in the importance of small bees and butterflies (measured as either number of approaches, number of flowers visited, or probability of visitation of individual flowers by time unit). These major differences between the MD and R regimes are consistent with the findings of earlier studies on *L. latifolia* pollinators at the same locality, and will not be discussed further here. Species-specific daily activity rhythms exhibited by *L. latifolia* pollinators reflect temperature- and irradiance-based microhabitat preferences, which are in turn related to interspecific differences in body size, thermal tolerance, and thermoregulation method (Herrera 1990*a*, 1995, 1997).

Although biologically and statistically significant, the magnitude of the difference between the MD and R regimes in pollinator composition and abundance was rather limited, particularly in comparison with geographical, seasonal, or annual variabilities in pollinator composition and abundance commonly experienced by plant species (e.g., Horvitz and Schemske 1990, Pettersson 1991, Eckhart 1992, Thompson and Pellmyr 1992, Kearns and Inouye 1994, Fishbein and Venable 1996, Guitián et al. 1996). It was thus not surprising that treatment effects on plant reproduction were of a small magnitude, but rather that such modest differences translated into measurable, statistically significant differences in reproductive parameters. This finding should warn us on the risks of a priori disregarding relatively minor variations in pollinator composition as ecologically or evolutionarily unimportant.

Results of this study support the prediction that flowers of *L. latifolia* exposed to different pollination regimes should differ in their contribution to population seedling recruitment because of differential seed production or progeny quality. MD flowers produced more seeds (because of greater fruit-set and seediness), and their seeds had a greater probability of yielding a seedling in the field (presumably because of improved progeny quality, since the mean mass of MD and R seeds did not differ). As a consequence, ovules in MD flowers

had a significantly greater probability of eventually contributing an established, 3rd-yr seedling to the *L. latifolia* study population under natural field conditions. In the study area, mortality of *L. latifolia* seedlings is greatest within 4 mo of emergence, and becomes negligible afterwards (C. M. Herrera, *personal observation*). Differences in seedling recruitment prospects found in this study can thus confidently be interpreted as actual evidence of differential population recruitment up to the age of first reproduction (5–8 yr).

Were observed differences in reproductive success between MD and R flowers actually attributable to differences between treatments in pollinator composition and abundance, or did they originate from factors unrelated to pollinator differences? Two concurrent lines of reasoning suggest that factors unrelated to pollinator composition and abundance were probably unimportant as determinants of the differential reproductive consequences of the MD and R regimes. On one side, consideration of differences between major pollinator groups in foraging behavior and pollinating effectiveness will provide a plausible, parsimonious explanation of observed patterns (see *Discussion: Potential mechanisms*). On the other side, there is no support for alternative explanations attributing a significant role to factors unrelated to pollinators.

While exposed to pollinators, flowers from the two pollination regimes experienced different ambient temperature and solar irradiance levels. The greater fruit and seed-set of MD flowers, and the greater seedling emergence rate of MD seeds, might thus have been caused by some unknown, advantageous effect of higher floral temperatures at the time of pollination, rather than from pollinator-related differences. This possibility could be properly tested, because the thermal and radiant microenvironment of study plants, and their daily variation, had been characterized for other studies (Herrera 1995, 1997). If the contrasting thermal environments of MD and R regimes were a major cause of observed differences in fruit-set, seed-set, and seedling emergence rate, I would expect the magnitude of the reproductive advantage of MD over R flowers to be positively correlated across plants to the mean difference in solar irradiance and ambient temperature between the MD and R half-plants. Study plants varied broadly in the differential between MD and R periods in both ambient temperature (2.4°–9.1°C range) and solar irradiance (-65–777 W/m^2 range), yet these between-treatment differentials in abiotic conditions were unrelated to the corresponding differentials in fruit-set ($r_s = -0.011$ and -0.343 for temperature and radiation, respectively; $P > 0.20$), fruit seediness ($r_s = 0.132$ and -0.193, $P > 0.45$), and seedling emergence rate ($r_s = -0.283$ and -0.193, $P > 0.30$).

The smaller fruit and seed-set of R flowers might be seen as an incidental consequence of reduced resource availability for seed and fruit maturation. In R half-plants, black tulle exclosures were covering plant fo-

liage during the time of day at which incident radiation was highest; hence the proportion of total daily photosynthetically active radiation (PAR) intercepted by exclosures was greater for R than for MD half-plants. Nevertheless, the influence of this effect on differential fruit and seed-set seems very unlikely, because (1) percent reduction of PAR (measured using a quantum sensor) by black tulle exclosures was only around 15%; (2) for Mediterranean plants in summer (including *L. latifolia*; Herrera 1991), water availability, rather than light, is the resource ordinarily limiting photosynthesis; and (3) if greater resource availability for fruit and seed maturation were actually responsible for the advantage of MD over R flowers in fruit and seed-set, increased resources would have also led to a concomitant increase in the size of the seeds produced by MD half-plants, yet no significant difference in mean seed mass was found between treatments. The positive effect of increased resource availability on seed size has been demonstrated experimentally for both *L. latifolia* (Herrera 1990*b*) and *L. stoechas* (J. Herrera 1991).

Potential mechanisms

The central objective of this investigation was to test the hypothesis that different pollination regimes had measurable demographic consequences for *L. latifolia*, rather than elucidating the actual mechanisms involved. There is evidence, however, suggesting that increased outcrossing levels (derived from the increased participation of small bees and butterflies as pollinators) and increased pollen deposition on stigmas (resulting from increased overall visitation rates to individual flowers), most likely account for the observed reproductive advantage of MD flowers.

Of the three major pollinator groups considered here, butterflies fly the longest distances between consecutively visited flowers, large bees the shortest, and small bees are intermediate (Herrera 1987*b*; see also Schmitt 1980, Waser 1982, Olesen and Warncke 1989). As a consequence, average flight distance of pollinators between flower visits is longest during the daytime period corresponding to the MD regime (Herrera 1990*a*: Fig. 6). The three major pollinator groups also differ in the proportion of total interfloral flights that take place between flowers on different plants. In this study, that proportion was greatest for butterflies (mean $\pm$ 1 SD $= 29.6 \pm 13.5\%$, $N = 81$ foraging bouts), smallest for large bees ($15.4 \pm 12.8\%$, $N = 235$), and intermediate for small bees ($22.0 \pm 14.0\%$, $N = 91$), and between-group heterogeneity was statistically significant ($\chi^2 = 71.0$, df $= 2$, $P < 0.0001$, Kruskal-Wallis ANOVA). These observations suggest that butterflies and small bees produced geitonogamous pollinations less often than large bees, and that outcross pollination thus occurred more often during the MD period. As outcrossing is known to increase fruit-set and fruit seediness relative to self-pollination in *L. latifolia* (Herrera 1987*a*), the greater fruit-set and seediness of MD flow-

ers may be partly due to increased outcrossing during that period due to a greater participation of butterflies and small bees as pollinators.

Increased outcrossing may also account for the greater inherent ability of MD seeds to produce seedlings under field conditions. In self-compatible species, larger environmental sensitivity of inbred individuals to both biotic and abiotic stresses very often leads to increased inbreeding depression (including decreased seedling emergence rate) with increasing severity of conditions (Schemske 1983, Mitchell-Olds and Waller 1985, Dudash 1990, Schmitt and Ehrhardt 1990, Biere 1991, Wolfe 1993, Belaoussoff and Shore 1995, Hauser and Loeschcke 1996, Ramsey and Vaughton 1998). The finding of this study that the advantage of MD over R seeds manifested itself only under field conditions and, within these, was greater under the relatively most severe ones, points to the occurrence of inbreeding depression in *L. latifolia* at the seed and early seedling stages, and is consistent with the interpretation that MD seeds originated more frequently from outcross pollination (due to increased participation of butterflies and small bees).

Differences between pollination regimes in flower visitation rates may also have contributed to differences in fruit-set, fruit seediness, and seedling emergence rate. Estimated probabilities of individual flowers being visited at least once during a 5-min period were 0.144 and 0.080 for the MD and R regimes, respectively (Table 3). This leads to binomial expectations of 9.8 and 5.1 pollinator visits received, on average, by individual MD and R flowers over the 6-h period of daily exposure to pollinators. This nearly twofold difference presumably originated larger pollen loads in MD flowers. At the study locality, the stigmas of flowers of *L. latifolia* receive an average (± 1 SD) of 13.8 ± 22.8 pollen grains ($N = 153$ flowers), yet the number of pollen tubes eventually reaching the base of the style is only 8.9 ± 7.1 tubes ($N = 384$ flowers) (C. M. Herrera, *unpublished data*), and the number of ovules available for fertilization is even smaller (four). These figures suggest that gametophytic competition (Lee 1984, Winsor et al. 1987) probably occurs, and that it should be most intense during the MD regime because of increased pollen loads. This might lead to increased fruit-set, fruit seediness, and progeny vigor via the same mechanisms known from other species (e.g., Bertin 1982, 1990, Schemske and Fenster 1983, Winsor et al. 1987, Richardson and Stephenson 1992, Niesenbaum and Casper 1994, Mitchell 1997*a*, *b*).

Correlations across experimental units ($N = 30$ half-plants) between reproductive and pollinator visitation parameters provide strong support for the preceding interpretations. Fruit-set ($r = 0.499$, $P = 0.005$), fruit seediness ($r = 0.406$, $P = 0.026$), and seedling emergence rate ($r = 0.349$, $P = 0.056$), were all positively correlated with single-flower visitation probability. Seedling emergence rate was positively correlated with

the mean number of flowers visited by small bees and butterflies combined per census ($r = 0.471$, $P = 0.008$), but not with the number of flowers visited by large bees ($r = 0.109$, $P = 0.56$). Finally, the estimated probability of an individual ovule eventually producing a 3-yr-old seedling was directly related to single-flower visitation probability ($r = 0.434$, $P = 0.016$) and to the number of flowers visited by small bees and butterflies combined ($r = 0.441$, $P = 0.014$), but not to the number of flowers visited by large bees ($r = 0.178$, $P = 0.35$).

Context dependence of pollination effects

This study has revealed that the reproductive consequences of different pollination regimes may be strongly context dependent. In the Aguaderillos home environment, differential seedling emergence rate was the main determinant of the difference between MD and R flowers in seedling recruitment prospects. The combined influence of differences in fruit-set and seediness, although statistically significant, was quantitatively negligible. Differences in fruit-set and seediness will, however, become proportionally more important at sites where differences in seedling emergence rate are smaller, as found in Agracea and Correhuelas. This leads to the important practical conclusion that, in *L. latifolia,* fruit-set and seediness parameters may be reliable descriptors of the reproductive consequences of different pollination regimes in some populations but not in others.

Planting conditions had a decisive influence on whether MD and R seeds differed in their ability to produce seedlings. While no significant difference existed in the greenhouse, a significant advantage of MD seeds was revealed in the field. Among field-planted seeds, in turn, the contrast between MD and R seeds increased with increasing clay content, and decreasing nitrogen, phosphorus, and organic matter, of the soil. *Lavandula latifolia* seeds are small (mean mass of 1.3 mg) and radicle growth and hypocotyl elongation are seriously hampered by very clay-rich soils, such as those occurring in the Aguaderillos site and elsewhere in the Sierra de Cazorla study region (C. M. Herrera, *unpublished data*). The low nitrogen, phosphorus, and organic matter content of these soils may also have adversely affected early seedling growth. Since MD and R seeds did not differ in either size or germinability in the laboratory, their difference in ability to produce seedlings in the field should be attributed exclusively to intrinsic differences in vigor or ability to grow shortly after germinating and before emerging above the soil surface. This cryptic difference seems to express itself only when conditions for the earliest underground life of seedlings become most severe, tending to remain concealed otherwise. This result highlights the need, when testing for the effects of contrasting experimental or natural conditions on seed performance, of conducting such tests under a broad range of environmental conditions encompassing those actually found by plants in the field. In the only other study known to me where the consequences of different pollination regimes were studied from flowers through seedlings (Jennersten and Morse 1991), the failure to detect seed or seedling performance differences between pollination regimes could thus have been due to the tests being conducted in a greenhouse.

Context dependence of the consequences of different pollinator regimes may have important implications for the relationship between *L. latifolia* and its pollinators. The most obvious one is that the possibilities of pollinators exerting selection on floral or flowering characteristics of *L. latifolia* will most likely depend on the degree to which local adversity for seedling emergence allows for the expression of cryptic heterogeneity in progeny quality. All else being equal, pollinators would be most likely to select on floral characteristics (e.g., favoring traits enhancing visitation by small bees and butterflies) in populations established on the most unfavorable soil types. Studies are currently underway to test this prediction.

Literature Cited

Baker, H. G. 1983. An outline of the history of anthecology, or pollination biology. Pages 7–28 *in* L. Real, editor. Pollination biology. Academic Press, Orlando, Florida, USA.

Belaoussoff, S., and J. S. Shore. 1995. Floral correlates and fitness consequences of mating-system variation in *Turnera ulmifolia.* Evolution **49**:545–556.

Bertin, R. I. 1982. Paternity and fruit production in the trumpet creeper (*Campsis radicans*). American Naturalist **119**: 694–709.

Bertin, R. I. 1990. Effects of pollination intensity in *Campsis radicans.* American Journal of Botany **77**:178–187.

Bertin, R. I., and M. F. Willson. 1980. Effectiveness of diurnal and nocturnal pollination of two milkweeds. Canadian Journal of Botany **58**:1744–1746.

Biere, A. 1991. Parental effects in *Lychnis flos-cuculi.* II: Selection on time of emergence and seedling performance in the field. Journal of Evolutionary Biology **4**:467–486.

Conner, J. K., R. Davis, and S. Rush. 1995. The effect of wild radish floral morphology on pollination efficiency by four taxa of pollinators. Oecologia (Berlin) **104**:234–245.

Darwin, C. 1862. On the various contrivances by which Brit-

ish and foreign orchids are fertilised by insects. Murray, London, England.

Davis, M. A. 1987. The role of flower visitors in the explosive pollination of *Thalia geniculata* (Marantaceae), a Costa Rican marsh plant. Bulletin of the Torrey Botanical Club **114**:134–138.

Dieringer, G. 1992. Pollinator effectiveness and seed-set in populations of *Agalinis strictifolia* (Scrophulariaceae). American Journal of Botany **79**:1018–1023.

Dudash, M. R. 1990. Relative fitness of selfed and outcrossed progeny in a self-compatible, protandrous species, *Sabatia angularis* L. (Gentianaceae): a comparison in three environments. Evolution **44**:1129–1139.

Eckhart, V. M. 1992. Spatio-temporal variation in abundance and variation in foraging behavior of the pollinators of gynodioecious *Phacelia linearis* (Hydrophyllaceae). Oikos **64**:573–586.

Fishbein, M., and D. L. Venable. 1996. Diversity and temporal change in the effective pollinators of *Asclepias tuberosa.* Ecology **77**:1961–1073.

Guitián, P., J. Guitián, and L. Navarro. 1993. Pollen transfer and diurnal versus nocturnal pollination in *Lonicera etrusca.* Acta Oecologica **14**:219–227.

Guitián, J., P. Guitián, and L. Navarro. 1996. Spatio-temporal variation in the interactions between *Cornus sanguinea* and its pollinators. Acta Oecologica **17**:285–295.

Hauser, T. P., and V. Loeschcke. 1996. Drought stress and inbreeding depression in *Lychnis flos-cuculi* (Caryophyllaceae). Evolution **50**:1119–1126.

Herrera, C. M. 1987a. Components of pollinator ''quality'': comparative analysis of a diverse insect assemblage. Oikos **50**:79–90.

Herrera, C. M. 1987b. Componentes del flujo génico en *Lavandula latifolia* Medicus: polinización y dispersión de semillas. Anales del Jardín Botánico de Madrid **44**:49–61.

Herrera, C. M. 1988. Variation in mutualisms: the spatio-temporal mosaic of a pollinator assemblage. Biological Journal of the Linnean Society **35**:95–125.

Herrera, C. M. 1989. Pollinator abundance, morphology, and flower visitation rate: analysis of the ''quantity'' component in a plant-pollinator system. Oecologia (Berlin) **80**: 241–248.

Herrera, C. M.. 1990a. Daily patterns of pollinator activity, differential pollinating effectiveness, and floral resource availability, in a summer-flowering Mediterranean shrub. Oikos **58**:277–288.

Herrera, C. M. 1990b. Brood size reduction in *Lavandula latifolia* (Labiatae): a test of alternative hypotheses. Evolutionary Trends in Plants **4**:99–105.

Herrera, C. M. 1991. Dissecting factors responsible for individual variation in plant fecundity. Ecology **72**:1439–1448.

Herrera, C. M. 1995. Microclimate and individual variation in pollinators: flowering plants are more than their flowers. Ecology **76**:1516–1524.

Herrera, C. M. 1996. Floral traits and plant adaptation to insect pollinators: a devil's advocate approach. Pages 65–87 *in* D. G. Lloyd and S. C. H. Barret, editors. Floral biology: studies on floral evolution in animal-pollinated plants. Chapman and Hall, New York, New York, USA.

Herrera, C. M. 1997. Thermal biology and foraging responses of insect pollinators to the forest floor irradiance mosaic. Oikos **78**:601–611.

Herrera, J. 1991. Allocation of reproductive resources within and among inflorescences of *Lavandula stoechas* (Lamiaceae). American Journal of Botany **78**:789–794.

Horvitz, C. C., and D. W. Schemske. 1990. Spatiotemporal variation in insect mutualists of a neotropical herb. Ecology **71**:1085–1097.

Hurlbert, S. H. 1984. Pseudoreplication and the design of ecological field experiments. Ecological Monographs **54**: 187–211.

Jennersten, O., and D. H. Morse. 1991. The quality of pollination by diurnal and nocturnal insects visiting common milkweed, *Asclepias syriaca.* American Midland Naturalist **125**:18–28.

Kearns, C. A., and D. W. Inouye. 1994. Fly pollination of *Linum lewisii* (Linaceae). American Journal of Botany **81**: 1091–1095.

Lee, T. D. 1984. Patterns of fruit maturation: a gametophyte competition hypothesis. American Naturalist **123**:427–432.

Littell, R. C., G. A. Milliken, W. W. Stroup, and R. D. Wolfinger. 1996. SAS System for mixed models. SAS Institute, Cary, North Carolina, USA.

Luque, P. 1995. Mapa de vegetación del Parque Natural de las Sierras de Cazorla, Segura y Las Villas. Agencia de Medio Ambiente, Junta de Andalucía, Sevilla, Spain.

McCullagh, P., and J. A. Nelder. 1989. Generalized linear models. Second edition. Chapman and Hall, London, England.

Mead, R. 1988. The design of experiments. Statistical principles for practical application. Cambridge University Press, Cambridge, England.

Mitchell, R. J. 1997a. Effects of pollination intensity on *Lesquerella fendleri* seed-set: variation among plants. Oecologia (Berlin) **109**:382–388.

Mitchell, R. J. 1997b. Effects of pollen quantity on progeny vigor: evidence from the desert mustard *Lesquerella fendleri.* Evolution **51**:1679–1684.

Mitchell-Olds, T., and D. M. Waller. 1985. Relative performance of selfed and outcrossed progeny in *Impatiens capensis.* Evolution **39**:33–544.

Motten, A. F., D. R. Campbell, D. E. Alexander, and H. L. Miller. 1981. Pollination effectiveness of specialist and generalist visitors to a North Carolina population of *Claytonia virginica.* Ecology **62**:1278–1287.

Niesenbaum, R. A., and B. B. Casper. 1994. Pollen tube numbers and selective fruit maturation in *Lindera benzoin.* American Naturalist **144**:184–191.

Nilsson, L. A. 1988. The evolution of flowers with deep corolla tubes. Nature **334**:147–149.

Olesen, J. M., and E. Warncke. 1989. Temporal changes in pollen flow and neighbourhood structure in a population of *Saxifraga hirculus* L. Oecologia (Berlin) **79**:205–211.

Olsen, K. M. 1997. Pollination effectiveness and pollinator importance in a population of *Heterotheca subaxillaris* (Asteraceae). Oecologia (Berlin) **109**:114–121.

Pettersson, M. W. 1991. Pollination by a guild of fluctuating moth populations: option for unspecialization in *Silene vulgaris.* Journal of Ecology **79**:591–604.

Ramsey, M. W. 1988. Differences in pollinator effectiveness of birds and insects visiting *Banksia menziesii* (Proteaceae). Oecologia (Berlin) **76**:119–124.

Ramsey, M., and G. Vaughton. 1998. Effect of environment on the magnitude of inbreeding depression in seed germination in a partially self-fertile perennial herb (*Blandfordia grandiflora,* Liliaceae). International Journal of Plant Science **159**:98–104.

Richardson, T. E., and A. G. Stephenson. 1992. Effects of parentage and size of pollen load on progeny performance in *Campanula americana.* Evolution **46**:1731–1739.

Sahley, C. T. 1996. Bat and hummingbird pollination of an autotetraploid columnar cactus, *Weberbauerocereus weberbaueri* (Cactaceae). American Journal of Botany **83**: 1329–1336.

SAS Institute. 1990. SAS/STAT user's guide. Version 6. Fourth edition. SAS Institute, Cary, North Carolina, USA.

SAS Institute. 1996a. SAS/STAT software: changes and enhancements through Release 6.11. SAS Institute, Cary, North Carolina, USA.

SAS Institute. 1996*b*. SAS/STAT software: changes and enhancements for Release 6.12. SAS Institute, Cary, North Carolina, USA.

Schemske, D. W. 1983. Breeding system and habitat effects on fitness components in three Neotropical *Costus* (Zingiberaceae). Evolution **37**:523–539.

Schemske, D. W., and C. Fenster. 1983. Pollen grain interactions in a Neotropical *Costus*: effects of clump size and competitors. Pages 405–410 *in* D. L. Mulcahy and E. Ottaviano, editors. Pollen: biology and implications for plant breeding. Elsevier, New York, New York, USA.

Schemske, D. W., and C. C. Horvitz. 1984. Variation among floral visitors in pollination ability: a precondition for mutualism specialization. Science **225**:519–521.

Schmitt, J. 1980. Pollinator foraging behavior and gene dispersal in *Senecio* (Compositae). Evolution **34**:934–943.

Schmitt, J., and D. W. Ehrhardt. 1990. Enhancement of inbreeding depression by dominance and suppression in *Impatiens capensis*. Evolution **44**:269–278.

Stebbins, G. L. 1970. Adaptive radiation of reproductive characteristics in angiosperms, I: Pollination mechanisms. Annual Review of Ecology and Systematics **1**:307–326.

Stone, J. L. 1996. Components of pollination effectiveness in *Psychotria suerrensis*, a tropical distylous shrub. Oecologia (Berlin) **107**:504–512.

Thompson, J. N., and O. Pellmyr. 1992. Mutualism with pollinating seed parasites amid co-pollinators: constraints on specialization. Ecology **73**:1780–1791.

Thøstesen, A. M., and J. M. Olesen. 1996. Pollen removal and deposition by specialist and generalist bumblebees in *Aconitum septentrionale*. Oikos **77**:77–84.

Waser, N. M. 1982. A comparison of distances flown by different visitors to flowers of the same species. Oecologia (Berlin) **55**:251–257.

Waser, N. M. 1983. The adaptive nature of floral traits: ideas and evidence. Pages 241–285 *in* L. A. Real, editor. Pollination biology. Academic Press, Orlando, Florida, USA.

Wilson, P., and J. D. Thomson. 1991. Heterogeneity among floral visitors leads to discordance between removal and deposition of pollen. Ecology **72**:1503–1507.

Winsor, J. A., L. E. Davis, and A. G. Stephenson. 1987. The relationship between pollen load and fruit maturation and the effect of pollen load on offspring vigor in *Cucurbita pepo*. American Naturalist **129**:643–656.

Wolfe, L. M. 1993. Inbreeding depression in *Hydrophyllum appendiculatum*: role of maternal effects, crowding, and parental mating history. Evolution **47**:374–386.

Wolfinger, R., and M. O'Connell. 1993. Generalized linear models: a pseudo-likelihood approach. Journal of Statistical Computation and Simulation **48**:233–243.