

# The Ecology and Genetics of Fitness in Forest Plants. IV. Quantitative Genetics of Fitness Components in Impatiens pallida (Balsaminaceae)

Daniel J. Schoen; Graham Bell; Martin J. Lechowicz

American Journal of Botany, Vol. 81, No. 2. (Feb., 1994), pp. 232-239.

Stable URL:

http://links.jstor.org/sici?sici=0002-9122%28199402%2981%3A2%3C232%3ATEAGOF%3E2.0.CO%3B2-R

American Journal of Botany is currently published by Botanical Society of America.

Your use of the JSTOR archive indicates your acceptance of JSTOR's Terms and Conditions of Use, available at <a href="http://www.jstor.org/about/terms.html">http://www.jstor.org/about/terms.html</a>. JSTOR's Terms and Conditions of Use provides, in part, that unless you have obtained prior permission, you may not download an entire issue of a journal or multiple copies of articles, and you may use content in the JSTOR archive only for your personal, non-commercial use.

Please contact the publisher regarding any further use of this work. Publisher contact information may be obtained at <a href="http://www.jstor.org/journals/botsam.html">http://www.jstor.org/journals/botsam.html</a>.

Each copy of any part of a JSTOR transmission must contain the same copyright notice that appears on the screen or printed page of such transmission.

The JSTOR Archive is a trusted digital repository providing for long-term preservation and access to leading academic journals and scholarly literature from around the world. The Archive is supported by libraries, scholarly societies, publishers, and foundations. It is an initiative of JSTOR, a not-for-profit organization with a mission to help the scholarly community take advantage of advances in technology. For more information regarding JSTOR, please contact support@jstor.org.

# THE ECOLOGY AND GENETICS OF FITNESS IN FOREST PLANTS. IV. QUANTITATIVE GENETICS OF FITNESS COMPONENTS IN *IMPATIENS PALLIDA* (BALSAMINACEAE)<sup>1</sup>

# DANIEL J. SCHOEN,<sup>2</sup> GRAHAM BELL, AND MARTIN J. LECHOWICZ

Department of Biology, McGill University, 1205 Avenue Docteur Penfield, Montreal, Quebec H3A 1B1, Canada

Genetic and environmental variances were estimated for a number of characters in the annual plant *Impatiens pallida* by planting seed obtained through controlled crosses into their native field site or pots maintained in the greenhouse. Significant additive genetic variance was detected for three of 11 characters studied—germination date, cotyledon area, and date of first flower production. Significant dominance and/or maternal variance was found for seed weight, proportion of seeds germinating, cotyledon area, plant height, and number of leaves produced. Environmental variance was greater in the field compared with the greenhouse. Characters found to be under strong directional selection in a previous study showed no detectable additive genetic variance. While these populations exhibit conditions that in theory could contribute to the maintenance of genetic variation (limited pollen and seed dispersal distances and small-scale variation for edaphic characteristics influencing plant growth), levels of additive genetic variance for most characters were not significantly different from zero.

The existence of additive genetic variation is a prerequisite for evolutionary change. Classical population genetics theory predicts that natural selection will deplete additive genetic variance for characters closely associated with fitness (Fisher, 1930). But theory has also shown that additive genetic variance can be maintained by a number of mechanisms including heterozygote advantage, mutation, immigration, spatially varying selection, frequency-dependent selection, and antagonistic pleiotropy (Barton and Turelli, 1989). Indeed, many studies have revealed significant levels of additive genetic variance for quantitative traits (Roff and Mousseau, 1987). The vast majority of these studies, however, have been conducted in the artificial environment of the laboratory, garden, greenhouse, or agricultural field plot. The relationship of estimates of heritability obtained under such artificial conditions to heritability in natural environments (e.g., undisturbed woods, prairie grassland, etc.) may be problematical. Artificial growth conditions are likely to differ from those of the natural field site such that the relative values of different genotypes are altered (Kahn, Antonovics, and Bradshaw, 1976; Gupta and Lewontin, 1982; Istock, 1983; Shaw, 1986). Moreover, artificial growth conditions are usually intended to minimize environmental variance, and would thus lead to heritability estimates that are biased upward (Falconer, 1981).

<sup>1</sup> Received for publication 1 February 1993; revision accepted 27 August 1993.

The authors thank Ruth Shaw and Nick Tinker for help with data analysis; Tony Brown, Susan Mazer, Tom Mitchell-Olds, and Ruth Shaw for discussion and comment; Elizabeth Bush, Cremilda Dias, Ashley Evans, Marika Egyed, Dorothy Jones, and Ron Kara for technical assistance; and the Gault Estate Board of Directors for permission to conduct field research at Mont St. Hilaire. This work was supported by grants from the Natural Sciences and Engineering Research Council of Canada and the Fonds pour la Formation de Chercheurs de l'Aide a la Recherche (FCAR-Quebec).

<sup>2</sup> Author for correspondence.

Here we report the results of work conducted using materials from two large populations of the annual plant Impatiens pallida (yellow touch-me-not). Additive, dominance, maternal, and environmental variances were estimated for a number of plant characters using pedigreed seed planted into the native field site of the parents, and, in the case of one experiment, into both the natural site and the greenhouse. An earlier investigation (Stewart and Schoen, 1987) based on correlation of trait expression with components of fitness (Lande and Arnold, 1983; Arnold and Wade, 1984a, b) conducted in one of the same populations of *I. pallida* and including many of the same characters studied here showed strong and consistent directional phenotypic selection for some characters (e.g., plant height and number of leaves measured at the middle or end of the growing season), but for others (e.g., cotyledon area, germination date, date of initial cleistogamous flower production), phenotypic selection was weak, undetectable, or heterogeneous between either spatial positions or selection episodes (Stewart and Schoen, 1987). One aim of the present study, therefore, was to determine whether additive genetic variance is near zero for characters found to be under strong and consistent directional selection (Fisher, 1930; Falconer, 1981).

A second aspect of the present study pertains to earlier findings that showed significant environmental variation at spatial scales of 1 m or less in natural populations of *Impatiens pallida*. These included studies of the edaphic environment utilizing inbred lines of *Arabidopsis thaliana* and *Hordeum vulgare* as phytometers (Bell and Lechowicz, 1991) together with chemical analysis of soil pH, potassium, and nitrogen (Lechowicz and Bell, 1991). Such findings, coupled with evidence of restricted gene flow (Schmitt, Ehrhardt, and Swartz, 1985; Schoen and Latta, 1989) and heterogeneous spatial selection (Stewart and Schoen, 1987) suggest that small-scale environmental variation could play a role in maintaining genetic variation in these populations.



Fig. 1. Experimental design used to estimate quantitative genetic parameters in the Yellow Valley population of *Impatiens pallida*. Large squares represent the  $50 \text{-m} \times 50 \text{-m}$  study quadrat from which all parents of the crosses were obtained and into which all progeny were planted. This crossing design and the accompanying procedures were replicated 24 times.

### MATERIALS AND METHODS

**Populations and study site**—Impatiens pallida is a locally common, summer annual plant that grows in rich, mesic woods throughout much of eastern North America (Schemske, 1978). In Quebec the plants emerge as seedlings in late April, and flower and set seed from July through early October. Both cleistogamous (CL-closed, self-pollinating flowers) and chasmogamous (CH-open flowers) flowers are produced (Schemske, 1978). CH flowers are visited by honeybees and bumblebees. Mating system estimation conducted in the populations studied here and using the seeds of CH flowers indicates that these flowers are approximately 60% outcrossed (Schoen and Lloyd, 1992; S. C. Stewart, University of Guelph, personal communication), but as cleistogamy is the primary mode of reproduction in the populations studied (Stewart and Schoen, 1987; Bell, Lechowicz, and Schoen, 1991), the overall mating system is predominantly self-fertilization.

Two study populations, referred to hereafter as the Yellow Valley and Green Valley populations, were chosen for all field work reported below. These populations are located in separate drainage basins in beech-maple woods (*Fagus grandifolia-Acer saccharum*) at Mont St. Hilaire, Quebec, Canada (Maycock, 1961) and are separated from each other by 1 km of forested land. Both populations contain known polymorphisms for a number of genes, including flower color, flower spotting, and the enzyme phosphoglucomutase (PGM) (Schoen and Latta, 1989; Schoen, unpublished data).

**Experimental design**—In the Yellow Valley population, all work was conducted within a 50-m  $\times$  50-m study quadrat located in the middle of the population. To obtain materials for the estimation of variance components for quantitative characters, we replicated a single crossing design 24 times. The details of each replicate experiment are as follows. In April of 1986, four seedlings at the cotyledon stage were removed from the 50-m  $\times$  50-m study quadrat using random coordinates numbered from

1 m to 50 m along each dimension of the quadrat (one coordinate for each seedling)-thus no two seedlings were collected from closer together than 1 m, and, in general, the distance separating seedlings was much more than this (Fig. 1). These seedlings were transferred on the same day to an insect-free, screened enclosure located adjacent to the Mont St. Hilaire Research Center of McGill University where they were grown at low density and under optimal nutrient and watering conditions to promote large adult size. Two out of four plants so obtained were randomly designated as female parents and the other two as male parents. These plants were crossed factorially, i.e., in a Comstock and Robertson (1952) design-two cross to yield 80 seeds -20 seeds from each male  $\times$  female parent combination (Fig. 1). Seeds were stored on moist filter paper until October of 1986, at which time they were planted back into the 50-m  $\times$  50-m field site in four separate open-bottomed trays of 15 cm  $\times$  20 cm in size. Each of these four trays contained 20 seeds from the crossing design (five from each male  $\times$  female combination), and each tray was placed in a different randomly selected location (transplant plot) within the 50-m  $\times$  50-m study quadrat. The trays were filled with the native soil of the transplant plot which had previously been screened to remove any naturally dispersed seed (Fig. 1). This step was necessary so that the fate of pedigreed seed in the natural environment could be traced separately from those naturally dispersed seed present at the plots.

Transplantation of seeds into the natural environment is an important feature of our study. Experiments started with seedlings transplanted from the greenhouse or garden may yield different estimates of variance components, as both the phenotypes and survivorship of seedlings may be influenced by early environmental effects associated with artificial propagation conditions. Unfortunately, such a design also inevitably leads to high levels of mortality in the transplanted progeny, and consequent reduction in statistical power to detect significant levels of genetic variance.

The position of each seed within the tray was used to keep track of its parental origin. In April of 1987, the emergent seedlings were identified and labeled to facilitate further monitoring during the growing season. Background plants were not removed, and every effort was made to minimize disturbance to the vegetation surrounding each transplant plot. The following characters were measured between April and October 1987: 1) seed weight (in mg at the time of seed production); 2) germination date (Julian day when at least one of the cotyledons exceeded 1 cm in length); 3) % germination (percentage of seeds out of the five from each of the male  $\times$  female parent combinations at each transplant plot that germinated); 4) cotyledon area (sum of the length in mm  $\times$ width in mm of each cotyledon); 5) May stem length (in cm from cotyledonary node to shoot apex after 1 mo of growth); 6) June stem length (in cm from cotyledonary node to shoot apex after 2 mo of growth); 7) May leaf total (number of leaves exceeding 1 cm in length counted after 1 mo of growth); 8) June leaf total (number of leaves exceeding 1 cm in length counted after 2 mo of growth); 9) CL flowering day (Julian day when first CL flower bud exceeded 1 mm in length); and 10) the lifetime total CL seed production (CH seed production accounted for less 
 TABLE 1. Quantitative traits of Impatiens pallida measured for estimation of variance components.

Trait (measurement units, transformation)	Symbol
Seed weight (mg, logarithm) <sup>a</sup>	SEEDWT
Germination date (Julian date) <sup>a</sup>	GDATE
Cotyledon area (mm <sup>2</sup> , logarithm)	COTAR
Stem height after 1 month (cm, logarithm) <sup>b</sup>	HT1
Stem height after 2 months (cm, logarithm)	HT2
Number of leaves after 1 month <sup>b</sup>	LVS1
Number of leaves after 2 month <sup>b</sup>	LVS2
Date of first cleistogamous flower <sup>b</sup> (Julian date)	CLDATE
Lifetime cleistogamous fruit production	
(logarithm) <sup>a,c</sup>	CLSUM
Biomass of dried plant (g, logarithm) <sup>a.c</sup>	BIOMASS
Proportion of germinating seeds in full	
sib family (arcsin $\sqrt{x}$ ) <sup>a,b</sup>	PGERM

<sup>a</sup> Not measured in Green Valley experiment.

<sup>b</sup> Not measured in Greenhouse experiment.

<sup>c</sup> Not measured in Yellow Valley experiment.

than 1% of the total seed output of this population). Table 1 lists the abbreviations used in the text and tables for each of these characters. In total, plants in 96 transplant plots were studied (i.e., 24 replicate sets of crosses  $\times$  four transplant plots for each replicate).

In the Green Valley population all work was conducted within a 100-m  $\times$  25-m quadrat. Seedlings were collected from the quadrat in April of 1987 using random coordinates and transplanted into the screened enclosure as described above. A single crossing design was replicated five times. This crossing design was intended to yield larger sample sizes than that used in the Yellow Valley study (Fig. 2). It entailed the crossing of four plants as male parents with each of three different female parents to yield a mixture of four paternal half sib families and 12 full sib families; i.e., a Comstock and Robertson (1952) design-one cross. The seeds from each of the five sets of crosses were stored as described above, and in October 1987 they were transplanted back into the field in up to ten different randomly located, 1-m  $\times$  1-m transplant plots within the original 25-m  $\times$  100-m study quadrat (some sets of crosses produced fewer seeds and correspondingly fewer transplant plots could be used). Eight seeds from each of the 12 full sib families produced in a cross were planted in each transplant plot, for a total of 96 seeds per transplant plot. Within the transplant plots the seeds were each planted in a random position in a hole 2 cm wide and 5 cm deep filled with a commercial soil mixture. Starting in April 1988, the germinating seeds and seedlings were monitored for most of the characters studied in the Yellow Valley experiment.

A third investigation was initiated in the greenhouse with parents collected from the Green Valley population in 1987. The experimental design was similar to the Green Valley experiment (i.e., a Comstock and Robertson design-one cross). In this case, however, seeds were available from crosses of 14 different male parents crossed with two different female parents each. Seeds from these crosses were stored in the refrigerator at 4 C on moist filter paper and planted into a greenhouse room in the McGill University Phytotron in March 1988. Progeny from each of the 28 full sib families were assigned at random to one

# February 1994]



Fig. 2. Experimental design used to estimate quantitative genetic parameters in the Green Valley population of *Impatiens pallida*. Large rectangles represent the 100-m  $\times$  25-m study quadrat from which all parents of the crosses were obtained and into which all progeny were planted. This crossing design and the accompanying procedures were replicated five times, but in some cases fewer than ten transplant sites were used.

of two nutrient regimes involving regular watering with either: 1) 100% Hoagland's solution (nutrient rich regime); or 2) 10% Hoagland's solution (nutrient poor regime). This was done as part of a separate study designed to examine the effects of varying levels of nutrients on growth and photosynthesis of *Impatiens pallida* plants (Lechowicz, unpublished data). The research greenhouse room was suitably equipped and under computer control to allow environmental homogeneity over the growing area. Plants in this greenhouse population were monitored for some of the same characters studied in the two field experiments and were also dried and weighed at the end of the study (Table 2). Methods used to estimate quantitative genetic parameters—Estimates of additive  $(V_a)$ , dominance  $(V_d)$ , maternal  $(V_m)$ , and environmental  $(V_e)$  variances were obtained by pooling data from all transplant plots in the Yellow Valley experiment as one data set, and all transplant plots in the Green Valley experiment as a second data set. In the case of the greenhouse study, data were pooled from the two nutrient treatments. Hence, three separate analyses were conducted. The method of unconstrained restricted maximum likelihood (REML) estimation (Shaw, 1987) was used. Calculations were conducted using a program provided by Dr. Ruth Shaw (University of California, Riverside). REML estimation of variances has been shown to be a useful alternative approach to the conventional sums of squares method in evolutionary investigations which often suffer from highly unbalanced design (Harville, 1977; Shaw, 1987). The unconstrained analysis permits estimated variances to be negative. Simulations have shown that constraining variance components to be nonnegative leads to biased variance estimates (Shaw, 1987). Because of the nature of the crossing design (i.e., the lack of reciprocal crosses between parents) the maternal variance,  $V_m$ , may be due to cytoplasmic, nuclear maternal, or maternal direct environment effects (Cockerham and Weir, 1977). In a few instances it was found that  $V_d$  and  $V_m$  were confounded, and it was therefore possible to estimate only one of these two variance components ( $V_m$  was chosen arbitrarily).

The pooling of data from separate plots requires that a fixed effect associated with each plot be estimated in conjunction with the variance component estimates. This pooling assumes no genotype  $\times$  environment (i.e., genotype  $\times$  transplant plot) interaction for the characters measured. The magnitude of such genotype  $\times$  environment interaction was examined in detail in the Green Valley experiment by estimating variance components associated with paternal parents, maternal parents and sites, as well as all pairwise and the three-way interactions, using REML as implemented in the SAS VARCOMP procedure (SAS Institute, Inc., 1985). Parents and plots were considered to be random factors.

To test for the significance of the estimated variance components, the log-likelihood ratio,  $G = -2 (L_o - L_{max})$ , was calculated, where  $L_o$  is the log likelihood specified by the null hypothesis (e.g., with  $V_a$  constrained to zero), and  $L_{max}$  is the log likelihood associated with the REML estimates of all four variance components. The test statistic, G, is asymptotically distributed approximately as chisquare with r degrees of freedom, where r is the number of parameters specified under the null hypothesis (Shaw, 1987).

**Inbreeding coefficient of population** — When parents are inbred but unrelated (as in the above-described experiments), the covariance of paternal half-sibs is (1 + F)/4, where F is Wright's inbreeding coefficient (Cockerham, 1963; Mitchell-Olds and Rutledge, 1986). Ignoring the inbreeding coefficient of the parents and using the uncorrected paternal half-sib covariance will, therefore, lead to inflated estimates of  $V_a$ . Data on the inbreeding coefficient of the parental generation were unavailable, but an electrophoretic survey of genotypic variability at the *Pgm-1* locus (coding for the enzyme PGM) was conducted 2 yr after the crosses were made (i.e., in 1988), as part of a mating system analysis of the Yellow Valley population (S. C. Stewart, personal communication). This survey involved the collection for six seeds from CH fruits in each of 254 seed parents located along a 150-m transect directly adjacent to the 50-m  $\times$  50-m study quadrat in this population. Parental gene frequencies and inbreeding coefficients were estimated from arrays of progeny genotypes using an estimation procedure based on the correlated mating model described by Ritland (1988).

Assumptions – In obtaining estimates of variance components we assume that parents are randomly sampled

 
 TABLE 2.
 Means (standard deviation and range) of quantitative traits of *Impatiens pallida* measured for estimation of variance components.<sup>a</sup>

Trait	Mean	SD	Range	N				
Yellow Valley Population (1987)								
SEEDWT (mg)	114.1	26.7	40-251	1,837				
GDATE (Julian date)	109.4	4.1	101-143ء	1,008				
PGERM (%)	0.54	0.33	0-1.0	377				
COTAR (mm <sup>2</sup> )	303.6	131.6	20.0-828.1	689				
HT1 (cm)	9.8	3.8	0.3-26.6	695				
HT2 (cm)	17.4	7.3	3.1-50.1	440				
LVS1 (number)	3.0	1.0	1–6	683				
LVS2 (number)	4.8	1.6	1-10	440				
CLDATE (Julian date)	163.6	4.7	123-184	471				
CLSUM (number)	0.86	2.56	0-21	1,059				
Green Valley Population (1988)								
COTAR (mm <sup>2</sup> )	330.3	152.0	34.8-1,032.7	1,580				
HT1 (cm)	13.3	5.5	2.9-36.9	1,603				
HT2 (cm)	19.7	7.7	4.0-50.9	838				
LVS1 (number)	4.0	1.2	0-10	1,601				
LVS2 (number)	5.9	2.3	0-23	835				
CLDATE (Julian date)	189.8	11.7	179–242	721				
Greenhouse Population								
SEEDWT (mg)	178.0	55.3	45-349	625				
GDATE (Julian date)	84.2	7.6	65-127	625				
COTAR (mm <sup>2</sup> )	343.1	122.0	37.5-798.5	583				
HT2 (cm)	30.5	5.2	7.0-48.5	618				
BIOMASS (g)	1.0	0.3	1.1-7.4	604				

<sup>a</sup> Back-transformed to original measurement units (see Table 1).

from the population (the random sampling of parental material should ensure this); there is no inbreeding, migration, or assortative mating (the crossing design should ensure this); inheritance is disomic and there is no linkage disequilibrium among the genes controlling the quantitative traits studied (Impatiens pallida has a diploid chromosome number of 20; Clevenger, 1971); and there is no genotype-environment correlation (the randomizing of planting locations of progeny should ensure this). We also assumed that parents used are equally inbred (violation of this assumption is unlikely to be serious in inbreeding species as noted by Mitchell-Olds [1986]), that competition among relatives does not exceed random expectations (this assumption may be violated as relatives may have been planted closer together in plots than they may normally occur in the field), and that there is no additive  $\times$  additive epistatic variance (the presence of this will inflate estimates of  $V_a$ ; Bulmer, 1980).

#### RESULTS

Overwinter mortality rates were high, resulting in low germination rates (PGERM = 54% in the Yellow Valley, 73% in the Green Valley, and 65% in the greenhouse). Thus the results reported below are based on fewer seeds than were originally planted. Mortality of established seedlings in the first 2 mo of the experiments (the period during which most of the traits were measured) was about 20% in the populations. Mean values of the characters measured are given in Table 2. Sample sizes listed in this table also correspond to those used for variance component estimation. TABLE 3. Variance components of quantitative traits measured in Impatiens pallida with plots treated as fixed effects.

	Variance component					Hypothesis test (G statistic) <sup>a</sup>		
Trait	V <sub>a</sub>	V <sub>d</sub>	V <sub>m</sub>	V <sub>e</sub>	h² (%)	$V_a = 0$	$V_d = 0$	$V_m = 0$
			Yellow	Valley (1987)		· · · · · · · · · · · · · · · · · · ·		
SEEDWT	0.012	-0.025	0.009	0.151	8.1	b	_	18.58***
GDATE	-0.21	1.39	0.64	10.22	0	_	0.72	1.12
PGERM	-165.4	536.2	26.9	-48.3	0	_	16.6***	0.80
COTAR	-0.004	0.056	0.009	0.128	0	_	1.10	0.74
HTI	-0.007	0.021	0.016	0.104	0	_	1.54	4.60*
HT2	0.019	-0.008	0.007	0.153	11.1	0.30	_	0.28
LVS1	0.112	0.087	0.035	0.559	14.1	0.54	0.24	0.54
LVS2	-0.037	0.512	0.141	1.441	0	_	0.78	0.72
CLDATE	5.08	-6.90	0.05	20.86	26.6	5.86*	_	0
CLSUM	0.022	-0.029	0	0.366	6.1	2.20	_	_
			Green V	/alley (1988)				
COTAR	0.017	-0.265	0.068	0.393	7.8	4.58*	_	6.02*
HT1	0.010	-0.326	0.080	0.417	5.5	2.04	_	2.88
HT2	-0.001	_	0.005	0.147	0	_	_	1.44
LVS1	0.044	-2.212	0.566	2.881	3.4	1.16	_	4.68*
LVS2	-0.075	-0.535	0.303	5.436	0	_	_	0.54
CLDATE	11.27	_	-1.08	116.38	8.9	1.58	_	_
			Gre	enhouse				
GDATE	65.44	_	-12.07	13.90	97.3	6.62**	_	_
COTAR	0.029	_	0.007	0.107	20.0	0.90	—	0.34
HT2	0.009	_	-0.001	0.029	24.3	2.88	_	
BIOMASS	0.007	_	-0.001	0.049	12.0	1.46	—	_

<sup>a</sup> See text.

<sup>b</sup> G test not performed when variance component was <0.

\* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.005.

Few of the individual estimates of  $V_a$  for the traits measured were significant (Table 3). Only CLDATE in the Yellow Valley, COTAR in the Green Valley, and GDATE in the Greenhouse population were significantly different from 0. This is without accounting for the fact that with multiple significance tests the chi-square threshold should be adjusted upward (e.g., in the Yellow Valley experiment where ten comparisons were made, the Bonferoni correction gives a protected 5% chi-square threshold of ca. 7.9). Heritability estimates were low in both field populations and ranged from 0% to 27%. Heritability

 TABLE 4.
 Variance components of quantitative traits measured in Impatiens pallida including the transplant plot effect (Green Valley data only).

	Variance component (% of total)							
Trait	Male parent	Female parent	Trans- plant plot	Male × female	Male × Trans- plant plot	Female × Trans- plant plot	Error	
COTAR	0.003	0.006	0.006	0	0.005	0	0.201	
	(1.3)	(2.7)	(2.7)		(2.3)		(91.0)	
HT1	0.002	0.002	0.013	0	0.001	0	0.178	
	(1.0)	(1.0)	(6.6)		(0.5)		(90.8)	
HT2	0	0.005	0.024	0	0	0	0.146	
		(2.9)	(13.7)				(83.4)	
LVS1	0.016	0.032	0.092	0	0	0	1.237	
	(1.2)-	(2.3)	(6.7)				(89.8)	
LVS2	0.004	0.150	0.261	0.003	0	0	4.933	
	(0)	2.8)	(4.9)	(0)			(92.2)	
CLDATE	2.96	0.62	12.05	0.62	0	1.44	121.24	
	(2.1)	(0.5)	(8.7)	(0.5)		(1.0)	(87.3)	

estimates in the greenhouse population were higher and ranged from 12% to 97%. Several traits exhibited significant nonadditive genetic variation (Table 3). Significant  $V_d$  or  $V_m$  were detected for SEEDWT, PGERM, and HT1 in the Yellow Valley, and for COTAR and LVS1 in the Green Valley population (Table 3). Genotype × environment interaction accounted for none or a very small percentage (i.e., 0–2%) of the overall phenotypic variance of most traits in the Green Valley population (Table 4).

The inbreeding coefficient estimated for the Yellow Valley population in 1988 was 0.64, indicating that the population is highly inbred, as expected given the predominance of cleistogamy in this species. If such an inbreeding level were representative of the parents of the crosses (collected from the Yellow and Green Valley populations in 1986 and 1987) the  $V_a$ s reported above, which assume noninbred parents, would have to be adjusted downward; i.e., ignoring the inbreeding coefficient of the parents and using the uncorrected paternal half-sib covariance would have inflated the estimates of  $V_a$ . Due to the lack of significant  $V_a$  for most characters as

Due to the lack of significant  $V_a$  for most characters as well as limitations on available computer memory, genetic correlations between characters were not calculated.

## DISCUSSION

In light of previous studies of selection in this species (Stewart and Schoen, 1987; Brassard and Schoen, 1990), it is informative to consider the few characters for which significant levels of additive genetic variance were detected, namely cotyledon area (COTAR), germination date (GDATE), and date of first cleistogamous flower production (CLDATE). Phenotypic directional selection of these characters was generally weak or nondetectable compared with size-related characters such as numbers of leaves and plant height measured in the middle or end of the growing season (e.g., LVS2, HT2), for which directional selection was strong and no significant additive genetic variance was detected. In general, these findings are consistent with theory that predicts the depletion of additive genetic variance for traits closely related to fitness (Fisher, 1930), but we regard them as tentative, as such results may be attributable in part to the low statistical power associated with the reduced number of surviving

plants later in the season. Nevertheless, it is notable that in the greenhouse study where sample sizes remained large and fairly constant throughout the course of the growth period (Table 2), and where environmental variance was smaller than in the field (Table 3), significant  $V_a$  for height, leaf number, or biomass was also not detected.

Other studies of additive genetic variance for quantitative characters in plants grown in natural habitats have yielded a variety of results. Mitchell-Olds (1986), in a study of quantitative genetic variation of 16 different fitness components in the related species Impatiens capensis, was able to uncover significant narrow-sense heritabilities for only three traits in one population-germination date, June stem diameter, and final dry weight. A second population showed no significant narrow-sense heritabilities for any of these traits. In a follow-up study, analysis of additional families together with jackknife methods of resampling revealed significant additive genetic variation for germination date alone (Mitchell-Olds and Bergelson, 1990). Schmitt and Antonovics (1986) and Shaw (1986), studying old field perennial herbs in North Carolina, both reported significant differences among genotypes for a number of traits related to fitness. In Phlox drummondii, Schwaegerle and Levin (1991) reported significant levels of additive genetic variation for approximately one-third of the characters they studied. These characters were associated with both plant growth and floral morphology. Stratton (1992) found low but significant broad sense heritabilities for date of emergence, size, survivorship, and fecundity in Erigeron annuus.

Studies of heritable variation in natural populations of insects include *Drosophila*, where significant levels of heritability of a number of morphological characters were found, and were shown to be lower under natural compared with laboratory conditions (Coyne and Beecham, 1987; Prout and Barker, 1989). Among birds studied in natural habitats, significant heritabilities of a number of life history traits were reported by Boag and Grant (1978) and Boag (1983) in Darwin's finches, and by Smith and Dhondt (1980) in song sparrows. Gibbs (1988), however, was unable to demonstrate significant heritability for clutch sizes in Darwin's finches.

It is not clear whether differences among the abovecited studies in the amount of additive genetic variance detected under natural conditions reflect real biological variation among species (and characters studied) or are due to differences in statistical power from study to study. In the present case it was not feasible to conduct a power analysis for each estimate, as sample sizes were unbalanced and differed from character to character. It is informative, however, to examine the results of simulations

conducted by Shaw (1987), who investigated the empirical power of REML to detect additive genetic variation under a variety of situations. Empirical power was examined for an experimental design involving ten sires mated to three different dams each, and with two or six progeny per sire-dam combination (yielding 60 or 180 progeny in total). These simulated experiments involve smaller experimental designs than the 24 pooled 2  $\times$  2 factorial crosses used in the Yellow Valley, or the 15 sire  $\times$  3 dam design used in the Green Valley experiment, where typically ten or more progeny per sire-dam combination were measured for a total of at least 400 progeny per character. With 60 and 180 progeny in total, and a ratio of  $V_a$  to the total phenotypic variance  $(V_p)$  of 0.2, Shaw (1987) found that the statistical power of REML to detect  $V_a$  was roughly 8% and 20%, respectively. With 60 and 180 progeny and a ratio of  $V_a$  to  $V_p$  of 0.5, the statistical power was approximately 16% and 60%, respectively. Using these simulation results as a conservative guide to interpreting the findings of our study it seems likely that the heritabilities of most characters investigated in our study were much less than 0.5.

The detection of  $V_m$  for characters expressed early in the life cycle of *Impatiens pallida* is compatible with the expectation that maternal effects are important early in the life cycle (Roach and Wulff, 1987). Moreover, the presence of significant  $V_d$  is compatible with reports of inbreeding depression for fitness components in the related species *I. capensis*, as dominance in genes controlling quantitative traits may be necessary for inbreeding depression to be expressed (Falconer, 1981; Mitchell-Olds and Bergelson, 1990). Mitchell-Olds and Bergelson (1990) have also found evidence of significant dominance and/ or maternal variance for fitness components expressed early in the life cycle in *Impatiens capensis*.

Studies of the beech-maple forest habitats occupied by Impatiens pallida at Mont St. Hilaire have shown considerable small-scale environmental heterogeneity for edaphic features (soil pH and minerals) affecting plant growth (Bell and Lechowicz, 1991; Lechowicz and Bell, 1991). These findings are consistent with the discovery of larger levels of environmental variance in the field- vs. greenhouse-based estimates of variance components (Table 3). In addition to the large amount of environmental heterogeneity suggested by these studies, it is known that gene flow distances in I. pallida are restricted; e.g., due to self-fertilization, short-distance pollinator movement (Dubé, 1988), and limited seed dispersal distances (Schmitt, Ehrhardt, and Swartz, 1985). The combination of small-scale environmental variation and restricted gene flow can, in theory, lead to local adaptation to specific microsites within the larger habitat area (Hedrick, Ginewan, and Ewing, 1976) and the maintenance of  $V_a$ . Yet results from the present study suggest that the large amount of phenotypic variation often observed in *I. pallida* in nature is primarily environmental in origin or due to nonadditive genetic effects. Such findings are difficult to reconcile with the discovery of local adaptation and differentiation in adjacent populations of *I. pallida* (Schemske, 1984; Schoen et al., 1986). It appears, therefore, that even when conducting rather large experiments such as those reported here, the investigator has only limited ability to detect levels of quantitative genetics in

February 1994]

plants grown under natural conditions. These levels, as pointed out by Mitchell-Olds and Bergelson (1990) are, however, sufficient for natural selection to act.

#### LITERATURE CITED

- ARNOLD, S. J., AND M. J. WADE. 1984a. On the measurement of natural and sexual selection: theory. *Evolution* 38: 709–719.
- ------, AND -------. 1984b. On the measurement of natural and sexual selection: applications. *Evolution* 38: 720–734.
- BARTON, N. H., AND M. TURELLI. 1989. Evolutionary quantitative genetics: how little do we know. Annual Review of Genetics 23: 337–370.
- BELL, G., AND M. J. LECHOWICZ. 1991. The ecology and genetics of fitness in forest plants. I. Environmental heterogeneity as measured by transplant trials. *Journal of Ecology* 79: 663–685.
  - , —, AND D. J. SCHOEN. 1991. The ecology and genetics of fitness in forest plants. III. Environmental variance in native populations. *Journal of Ecology* 79: 697–713.
- BOAG, P. T. 1983. The heritability of external morphology in Darwin's Ground Finches (*Geospiza*) on Isle Daphne Major, Galapagos. *Evolution* 37: 877–894.
  - ——, AND P. R. GRANT. 1978. Heritability of external morphology in Darwin finches. *Nature* 274: 793–794.
- BRASSARD, J. T., AND D. J. SCHOEN. 1990. Analysis of phenotypic selection among locations in *Impatiens pallida* and *Impatiens ca*pensis. Canadian Journal of Botany 68: 1098–1105.
- BULMER, M. G. 1980. The mathematical theory of quantitative genetics. Oxford University Press, Oxford.
- CLEVENGER, S. 1971. Anthocyanides in some Impatiens species. Evolution 25: 669–677.
- COCKERHAM, C. C. 1963. Estimation of genetic variances. In W. D. Hanson and H. F. Robertson [eds.], Statistical genetics and plant breeding. NAS-NRC Publication #982, 53-94. NAS-NRC, Washington, D.C.
- , AND B. S. WEIR. 1977. Quadratic analyses of reciprocal crosses. Biometrics 33: 187–203.
- COMSTOCK, R. E., AND H. F. ROBERTSON. 1952. Estimation of average dominance of genes. *In J. W. Gowen* [ed.], Heterosis, 494–516. Iowa State College Press, Ames, IA.
- COYNE, J. A., AND E. BEECHAM. 1987. Heritability of two morphological characters within and among natural populations of *Drosophila melanogaster*. *Genetics* 117: 727–737.
- DUBÉ, D. 1988. Flight path of pollinators foraging on *Impatiens*. Decision rules and their implications for gene flow. M.Sc. thesis, McGill University. Montreal, Quebec.
- FALCONER, D. S. 1981. Introduction to quantitative genetics, 2d ed. Longman, London.
- FISHER, R. A. 1930. The genetical theory of natural selection. Clarendon Press, Oxford, U.K.
- GIBBS, H. L. 1988. Heritability and selection on clutch size in Darwin's Medium Ground Finches (*Geospiza fortis*). Evolution 42: 750–762.
- GUPTA, A. P., AND R. C. LEWONTIN. 1982. A study of reaction norms in natural populations of *Drosophila pseudoobscura*. Evolution 36: 934–948.
- HARVILLE, D. 1977. Maximum likelihood approaches to variance component estimation and to related problems. *Journal of the American Statistical Association* 72: 320–340.
- HEDRICK, P. W., M. E. GINEWAN, AND E. P. EWING. 1976. Genetic polymorphism in heterogeneous environments. Annual Review of Ecology and Systematics 7: 1–32.
- ISTOCK, C. A. 1983. The extent and consequences of heritable variation for fitness characters. In C. R. King and P. S. Dawson [eds.], Population biology: retrospect and prospect, 61–96. Oregon State Colloquium in Biology, Columbia University Press, New York, NY.
- KAHN, M. A., J. ANTONOVICS, AND A. D. BRADSHAW. 1976. Adaptation

to heterogeneous environments. III. The inheritance of response to spacing in Flax and Linseed (*Linum usitatissimum*). Australian Journal of Agricultural Research 27: 649–659.

- LANDE, R., AND S. J. ARNOLD. 1983. The measurement of selection on correlated characters. *Evolution* 37: 1210–1226.
- LECHOWICZ, M. J., AND G. BELL. 1991. The ecology and genetics of fitness in forest herbs. II. Microspatial scale of the edaphic environment. *Journal of Ecology* 79: 687–696.
- MAYCOCK, P. F. 1961. Botanical studies on Mont St. Hilaire, Rouville County, Quebec. General descriptions of the area and floristic survey. *Canadian Journal of Botany* 39: 1293–1325.
- MITCHELL-OLDS, T. 1986. Quantitative genetics of survival and growth in *Impatiens capensis*. Evolution 40: 107–117.
- ——, AND J. BERGELSON. 1990. Statistical genetics of an annual plant, Impatiens capensis. I. Genetic basis of quantitative variation. Genetics 124: 407–415.
- ——, AND J. J. RUTLEDGE. 1986. Quantitative genetics in natural plant populations: a review of the theory. *American Naturalist* 127: 379–402.
- PROUT, T., AND J. S. F. BARKER. 1989. Ecological aspects of the heritability of body size in *Drosophila buzzatii*. *Genetics* 123: 803-813.
- RITLAND, K. 1988. The genetic structure of subdivided populations. II. Correlated mating model. *Theoretical Population Biology* 34: 320–346.
- ROACH, D. A., AND R. D. WULFF. 1987. Maternal effects in plants. Annual Review of Ecology and Systematics 17: 667–693.
- ROFF, D. A., AND T. A. MOUSSEAU. 1987. Quantitative genetics and fitness: lessons from *Drosophila*. *Heredity* 58: 103–118.
- SAS INSTITUTE, INC. 1985. SAS user's guide: statistics, version 5 ed. SAS Institute, Inc., Cary, NC.
- SCHEMSKE, D. W. 1978. Evolution of reproductive characteristics in *Impatiens* (Balsaminaceae): the significance of cleistogamy and chasmogamy. *Ecology* 59: 596–613.
- ——. 1984. Population structure and local selection in *Impatiens* pallida (Balsaminaceae): a selfing annual. Evolution 38: 817–832.
- SCHMITT, J., AND J. ANTONOVICS. 1986. Experimental studies of the evolutionary significance of sexual reproduction. III. Maternal and paternal effects during seedling establishment. *Evolution* 40: 817– 829.
- ——, D. EHRHARDT, AND D. SWARTZ. 1985. Differential dispersal of self-fertilized and outcrossed progeny in jewelweed (*Impatiens capensis*). American Naturalist 126: 570–575.
- SCHOEN, D. J., AND R. G. LATTA. 1989. Spatial autocorrelation of genotypes in populations of *Impatiens pallida* and *Impatiens ca*pensis. Heredity 63: 181–190.
- ——, AND D. G. LLOYD. 1992. Self- and cross-fertilization in plants. III. Methods for studying modes and functional aspects of selffertilization. *International Journal of Plant Sciences* 153: 381–393.
- ——, S. C. STEWART, M. J. LECHOWICZ, AND G. BELL. 1986. Partitioning the transplant site effect in reciprocal transplants experiments with *Impatiens capensis* and *I. pallida. Oecologia* 70: 149– 154.
- SCHWAEGERLE, K. E., AND D. A. LEVIN. 1991. Quantitative genetics of fitness traits in *Phlox. Evolution* 45: 169–177.
- SHAW, R. G. 1986. Response to density in a wild population of the perennial herb Salvia lyrata: variation among families. Evolution 40: 492-505.
- 1987. Maximum-likelihood approaches applied to quantitative genetics of natural populations. *Evolution* 41: 812–826.
- SMITH, J. N. M., AND A. A. DHONDT. 1980. Experimental confirmation of heritable morphological variation in a natural population of song sparrows. *Evolution* 34: 1155–1160.
- STEWART, S. C., AND D. J. SCHOEN. 1987. Pattern of phenotypic viability and fecundity selection in a natural population of *Impatiens* pallida. Evolution 41: 1290–1301.
- STRATTON, D. A. 1992. Life-cycle components of selection in *Erigeron* annuus. II. Genetic variation. *Evolution* 46: 107–120.