

Spatial Variation in the Selective Scenarios of *Hormathophylla spinosa* (Cruciferae)

José M. Gómez* and Regino Zamora†

Grupo de Ecología Terrestre, Departamento de Biología Animal y Ecología, Facultad de Ciencias, Universidad de Granada, E-18071 Granada, Spain

Submitted July 20, 1999; Accepted December 9, 1999

ABSTRACT: The effects of multispecific systems containing both mutualistic and antagonistic interacting organisms on the evolution of plant traits have seldom been analyzed. We studied the selection exerted by several species of herbivores and pollinators in three populations of *Hormathophylla spinosa* (Cruciferae) in the Sierra Nevada (Spain) over 4 yr by using path analyses and structural equation modeling (SEM). The main selective pressures in our study sites were ungulates and pollinators. However, the importance of each kind of interacting organism differed among populations. Our results indicate a selection mosaic among populations of *H. spinosa* in the Sierra Nevada caused by the spatial variation in the relative importance of different interactions as selective pressures. We found two main selective scenarios, depending on the presence or absence of ungulates. In the populations with low ungulate pressure, there was positive phenotypic selection in flower number per plant and in flower density (mediated by nectarivorous pollinators). In the two populations with high ungulate pressure, there was a strong positive, ungulate-mediated selection in thorn density. Our results suggest that the application of SEM to several populations simultaneously monitored might help to isolate the major selection pressures on local populations and identify potential differences in selection among populations, becoming a useful exploratory approach to study the geographical variation of selection in complex systems.

Keywords: evolutionary ecology, multispecific systems, plant-animal interactions, spatial variation in selection, structural equation modeling, ungulate herbivory.

A central goal in evolutionary ecology is to discover the actual role of interspecific interactions (mutualistic and antagonistic) in the evolution of plant traits. Thus, many

studies have shown that some floral traits affect plant fitness by mediating pollinator visitation rate or effectiveness (e.g., Lloyd and Barrett 1996 for a recent review), whereas other traits, such as secondary metabolites or mechanical defenses, affect plant fitness by changing foraging behavior and feeding capacity of antagonistic organisms (Marquis 1992). Most studies focus on only one or a few temporally coincident organisms interacting with the plant. However, to understand the true overall effect of an organism on plant evolution, we need to consider that most plant species usually interact with a system composed of many species that influence the plant directly and indirectly through interaction with other organisms (Rausher and Simms 1989; Simms and Rausher 1989; Farris and Lechowicz 1990; Simms 1990; Swank and Oechel 1991; Meyer and Root 1993; Pilon 1996).

Multispecific systems inevitably contain both mutualistic (pollinators, symbionts, dispersers, etc.) and antagonistic (herbivores, seed predators, pathogens, etc.) species. The interaction between the host plant and each kind of interacting organism depends on the effect of the other kinds of organisms, and some plants reveal strong links between responses to mutualist and antagonist interacting organisms (Quesada et al. 1995; Niesenbaum 1996; Brody 1997; Delph et al. 1997; Strauss 1997). For example, pollinators can also be seed predators (Pellmyr 1989, 1997), floral herbivores (Galen 1983; Irwin and Brody 1998), vectors of plant disease (Jennersten 1988; Roy 1994), or prey of some carnivorous plants (Zamora 1999). Selection on plant traits related to pollination can sometimes be cancelled by the overwhelming influence of herbivory (Aarssen and Irwin 1991; Herrera 1993; Quesada et al. 1995; Niesenbaum 1996; Armbruster et al. 1997; Delph et al. 1997). Consequently, adaptations to avoid herbivory can constrain the evolution of plant-pollinator interactions and vice versa (Armbruster 1997; Armbruster et al. 1997; Fineblum and Rausher 1997; Shykoff et al. 1997). Nevertheless, the relative importance of balance between mutualism versus antagonism in the evolution of plant traits has seldom been studied (but see Schemske and Horvitz 1988; Zamora 1999).

* E-mail: jmgreyes@goliat.ugr.es.

† E-mail: rzamora@goliat.ugr.es.

An additional important but largely neglected factor is spatial variability in the relative importance of herbivores and pollinators as selective agents (Thompson 1988; Galen 1996; Travis 1996; Brody 1997). With very few and remarkable exceptions (e.g., Jarosz and Burdon 1991; McCauley 1991; van Tienderen and van der Toorn 1991a, 1991b; Mitchell 1994), most recent works have focused on natural selection acting in a single population (Thompson 1994, 1999a, 1999b). Nevertheless, the way that interacting organisms affect the relationship between phenotype and fitness can shift among populations (Thompson 1994, 1999a, 1999b). In this case, a selection mosaic among populations can appear, favoring different evolutionary trajectories to interactions in different populations (Thompson 1999b). Interaction-mediated natural selection would favor different adaptations in different environments as a result of spatial variation in the outcome of the interactions (Thompson 1994).

Path analysis and structural equation modeling (SEM) are techniques used to quantify causal pathways in networks of interactions (Mitchell 1992, 1993), representing one of the most powerful approaches available for analyzing complex multivariate relationships in nonexperimental systems (Grace and Pugesek 1998). Therefore, they can be used to test some evolutionary hypotheses, helping to identify the causal mechanisms of phenotypic selection in multispecific assemblages where experimental manipulations are highly constrained (Kingsolver and Schemske 1991; Mitchell 1992; Grace and Pugesek 1998). In previous work, we have shown intraspecific variation in the intensity of interaction among *Hormathophylla spinosa* and pollinator and herbivores as a prerequisite for selection by an interacting organism to occur (Gómez 1993; Gómez and Zamora 1999). In this article, by using path analysis and SEM, we examine the effect of the multispecific system, composed of pollinators, herbivorous insects, and vertebrate herbivores, on fitness, phenotype, and the relationship between fitness and phenotype of *H. spinosa* (Cruciferae). We seek to determine whether there is any interspecific interaction acting as selective pressure on this plant species. In addition, we consider the spatial inter-population differences in the selective regimes. Specifically, this article focuses on the main determinants of reproductive success through female function, addressing the following questions. First, what is the relative importance of pollination and herbivory on plant fitness both when occurring separately as well as together? Second, are there spatial differences in selective regimes? That is, is there any possibility for the existence of selection mosaic among populations? Third, is there a local fit between the plant phenotype and the main interacting organisms?

Plant Natural History

Hormathophylla spinosa (L.) Küpfer (Cruciferae) is a shrub of the Mediterranean high mountains that inhabits the Sierra Nevada (southeast Spain) from 1,600 m to 3,340 m elevation, mainly above the timberline. This crucifer is a mass-flowering species, with an average of over 8,600 flowers per reproductive season (482–74,343 flowers for three plant populations during 4 yr). Moreover, flower density is very high, with an average of 308.3 ± 23.9 flowers/100 cm² (Gómez and Zamora 1992). As a consequence, during the reproductive period, a typical individual is almost completely covered by flowers or fruits. Flowering begins at the end of June in the population situated at the lowest altitude and ends at the beginning of September for plants living at highest altitude (Gómez 1993). In the Sierra Nevada mountains, the flowers are pollinated by many insects, predominantly the ant *Proformica longiseta* (Gómez and Zamora 1992, 1999). It is also pollinated by the wind in some high-elevation populations (Gómez and Zamora 1996). Every flower harbors four ovules arranged in two locules, although most mature fruits have only two seeds (Gómez and Zamora 1992). These seeds are small (around 0.3–0.6 mg) and are dispersed by the wind. The secondary branches of *H. spinosa* form small rigid thorns, usually located at the basal portion of each inflorescence and associated with bracts. In addition, some stems of several-years-old infructescences become hard and rigid, forming additional thorny stems. The proportion of former to latter type of thorns is roughly 7 : 1. In our analyses, we pooled the two types of thorns. Detailed information on various aspects of *H. spinosa* reproductive ecology and interaction with pollinators and herbivores is provided in Gómez and Zamora (1992, 1994, 1996, 1999), Gómez (1993, 1994), and Zamora and Gómez (1993).

Methods

At the beginning of the study (1988), we tagged a total of 110 plants in three populations located at three different altitudes: 50 plants at 2,150 m (population A), 30 plants at 2,550 m (population B), and 30 plants at 3,130 m above sea level (population C). Populations were 8 km away from one another. However, populations A and B were in the same river valley (Rio San Juan valley), both on the northern slope of the mountain, whereas population C was in another river valley (Rio Veleta valley) on the southern slope.

The three studied populations were similar in plant density ($P > .1$, Kruskal-Wallis test), being 91.7 ± 20.6 ($\bar{X} \pm 1$ SD) individuals/100 m² in population A, 87.5 ± 13.6 in population B, and 74.2 ± 15.1 in population C. Plants were haphazardly selected in each population

among the reproductive individuals, the size distribution of tagged plants reflecting the natural distribution of plant size (Gómez 1993). In addition, plant size (expressed as shrub surface in cm^2) did not differ among populations ($F = 0.55$, $df = 2, 108$, $P = .57$), being $1,603 \pm 145$ ($\bar{X} \pm 1$ SD) square centimeters in population A, $1,225 \pm 351$ in population B and $1,739 \pm 188$ cm^2 in population C. There is a significant relationship between plant size and age ($\text{age} = 4.926 + 0.018 \times \text{size}$, $r^2 = 0.91$, $P = .0001$, $n = 20$), indicating that plant age was also probably similar among populations.

Traits Measured

Each phenotypic trait was measured for all tagged plants every year of the study (1988–1991). Plant size, estimated as two diameters of the shrub, was measured at the very beginning of the study, and then at the beginning of each reproductive season. Two floral traits were measured per plant and per year: flower size (in millimeters, by a digital caliper with 0.1 mm error; 20 flowers per plant and per year), estimated as the distance between the outer border of two opposite petals, and number of stamens per flower (by a yearly count of 50 flowers per plant), counting both short and long filaments. In addition, we quantified flower density (by a yearly count of five 6×6 cm surface quadrats per plant) and total number of flowers per tagged plant. In addition to those flowering traits, fruit size (length in millimeters) of each tagged plant was measured yearly with binocular magnifying glasses (0.01 mm error), for a total of 50–100 fruits per plant per year. Finally, we estimated the thorn production of each marked plant by quantifying thorn density by a count of five 6×6 -cm surface quadrats per plant.

Pollinator and Herbivore Observations

Two main groups of putative biotic selective pressures, pollinators and herbivores, have been considered in this study. Pollinator abundance, expressed as number of insects per plant and per time unit, was quantified for each tagged plant, by censuses carried out from 1988 to 1991. Censuses consisted of counting all flower visitors at tagged plants during 1-min periods (see Gómez and Zamora 1999 for details). We considered the two main categories of pollinators: nectarivores (mainly the ant *Proformica longiseta* but also *Fannia scalaris* and *Ramphomyia tenuirostris*) and pollinivores (mainly *Colletes* sp., *Eristalis tenax*; Gómez and Zamora 1992, 1999).

Damage by inflorescence-feeding herbivores was also quantified for all tagged plants during 4 yr. We considered the main groups of herbivores: ungulates (domestic sheep and goats and *Capra pyrenaica*), linnet (*Carduelis canna-*

bina), floral herbivores (*Timarcha lugens*, *Oiketicoides febredda*, and *Pimelia monticola*) and seed-predator weevils (*Ceuthorhynchus* sp.), quantifying the average damage produced by each type of herbivore.

Correlation among Plant Traits, Interacting Organisms, and Plant Fitness

We correlated in each population all phenotypic traits by product moment correlations. Furthermore, we related the intensity of potential selective pressures, pollinators and herbivores, against each phenotypic plant trait by simple linear regressions, and all putative selective pressures and plant fitness by multiple regressions. For this, we correlated the magnitude of damage caused by each main herbivore or the abundance of pollinators against both absolute (realized fecundity or number of seed dispersed per plant) and relative fitness (the absolute fitness of an individual relative to the population mean; Falconer 1989).

Before all analyses, we tested whether the relationship among each phenotypic trait, interactive organism, and relative fitness varied between years by performing ANCOVAs with interaction term, introducing years as covariate in all analyses described above. A nonsignificant interaction term between year and each independent variable suggests that the slope of the regression between each independent variable and the response variable was similar across years (Edwards 1985). Since the interaction term was in all cases nonsignificant, we pooled the data from the 4 yr of study (1988–1991), averaging all phenotypic estimates for each tagged plant. The REG procedure in the SAS package was used to perform the regression analyses and the GLM procedure to perform the ANCOVAs (SAS Institute 1997).

Randomization tests (Manly 1991) were used to assess the significance of the regression coefficients to control for small sample size per population. Randomization tests were carried out with 5,000 iterations using program MULTREG (routines provided by P. Jordano, based on Manly 1991 and Press et al. 1992). We transformed the variables that departed significantly from normality (Zar 1996) in all previous analyses. Moreover, to control for experiment-wise Type I errors produced by multiple comparisons, we adjusted the probability of error to $\alpha = 0.05$ by using the sequential Bonferroni technique (Rice 1989).

Study of Phenotypic Selection

We used path analysis and SEM to investigate complex multivariate causal models of plant phenotypic traits and pollinators/herbivores on plant relative fitness values (Li 1981; Kingsolver and Schemske 1991; Mitchell 1992, 1993,

1994). Path analysis allows the partitioning of the simple correlations among a set of variables according to a particular working model, enabling an a priori hypothesis concerning the causal link between variables to be translated into a statistical model (Mitchell 1992; Shipley 1997). Structural equation modeling, as an extension of path analysis, tests whether a specific model, represented as a path diagram, is an acceptable description of the underlying causal mechanisms. This is accomplished by a goodness-of-fit test, by which the observed covariance matrix is compared with that expected if the model were true (see Mitchell 1992, 1993 for applications of SEM to evolutionary ecology). A significant goodness-of-fit test indicates that the model is a poor description of the observed covariance among the variables, while a nonsignificant value indicates that the pattern of covariance predicted by the model is not distinguishable from that observed (Mitchell 1994; Shipley 1997).

By using the information derived from our 4 yr of study, we hypothesized about the causal links among phenotypic traits, selective pressures and fitness of the plants, building an a priori path analytic basic model. We built the same basic model for the three populations. In this basic model, we introduced as response the relative fitness of the plants. As intermediate variables, directly related to the relative fitness, we introduced these interactive organisms obtaining significant regression coefficient in at least one plant population in the previous analyses. Finally, for independent variables we considered only those phenotypic traits that significantly related to these main interactive organisms.

After building the basic, most inclusive model, we considered a number of alternative, competing models (alternative hypotheses). All these alternatives were "nested" within the basic model; that is, they had the same causal structure but some paths were eliminated (Mitchell 1994). For this reason, in the decision to accept or reject them, the differences in goodness of fit between the models (both between the basic and alternatives and among different alternatives) were statistically compared, with degrees of freedom equal to the difference in degrees of freedom between the two models. A significant difference in goodness of fit indicates that the less inclusive model is significantly worse at describing the interaction. However, if the difference in goodness of fit is nonsignificant, both models are equally appropriate, and the simpler model is preferred on the principle of parsimony (Mitchell 1993). This means that when more than one alternative model was acceptable, we chose the one with the lowest degrees of freedom. When two alternative models with equal degrees of freedom were acceptable, we chose the one with the highest P value and the lowest χ^2 . We tried to maintain the ratio of number of independent samples to the number

of paths estimated per alternative model close to 10, the minimum acceptable threshold (Mitchell 1993).

The structural equation modeling was performed by using the CALIS procedure in the SAS package (SAS Institute 1997), using maximum-likelihood estimation on the covariance matrix to test the goodness of fit of the model to data.

Results

Correlation between Plant Traits

In population A, only the correlation between plant size versus number of flowers proved significant ($r = 0.865$, $P = .0001$). In population B, there were three significant correlations: floral diameter versus number of stamens ($r = 0.6091$, $P = .00001$), number of flowers versus floral density ($r = 0.673$, $P = .0001$), and plant size versus number of flowers ($r = 0.913$, $P = .0001$). In population C, only one correlation was significant: plant size versus number of flowers ($r = 0.694$, $P = .00001$).

Relationship between Plant Traits and Interacting Organisms

Number of flowers positively related to pollinator abundance in all populations, whether with nectarivorous pollinators in the populations A ($\beta = 0.805 \pm 0.25$, $F = 15.36$, $P = .001$, linear regression with significance tested by a randomization procedure) and B ($\beta = 0.29 \pm 0.16$, $F = 27.35$, $P = .002$) or with pollinivores in population C ($\beta = 0.32 \pm 0.15$, $F = 17.76$, $P = .002$). Flower density per plant also positively related with nectarivorous pollinators abundance in population B ($\beta = 0.69 \pm 0.24$, $F = 7.76$, $P = .006$). Finally, flower size correlated strongly with pollinator abundance in population C ($\beta = 0.38 \pm 0.64$, $F = 18.77$, $P = .002$) but not in the other populations ($P > .05$). No other plant traits were related with pollinator visitation rate.

Similarly, few traits correlated with herbivore pressure. Plants with more thorns were less damaged by ungulates both in population A ($\beta = -0.538 \pm 0.002$, $F = 6.78$, $P = .012$) and in population C ($\beta = -0.466 \pm 0.003$, $F = 4.98$, $P = .038$). Furthermore, ungulates fed preferentially on plants displaying many flowers in all populations, this trend being significant in population C ($\beta = 0.305 \pm 0.001$, $F = 17.76$, $P = .0001$). In this latter population, linnets fed in plants with large fruits ($\beta = 0.314 \pm 0.002$, $F = 6.06$, $P = .017$).

Relationship between Interacting Organisms and Plant Fitness

Only ungulates and pollinators significantly affected plant female fitness, and this effect varied between populations. There were negative relationships in populations A and C between damage by ungulate and relative plant fitness (table 1). In Population B, there was a positive relationship between pollinator visitation rate and relative fitness of the plants (table 1).

Path Analysis and Structural Equation Modeling

According to the results obtained in the previous analyses, the basic model was built introducing ungulate damage, pollinator visitation rate and number of flowers directly affecting plant female fitness, thorn density affecting to ungulate damage, and number of flowers, flower size and flower density affecting to pollinator visitation rate (fig. 1). In addition, seven alternative models were built in the three populations (table 2). These models were made by constraining to zero some paths from the basic model, according to the significant relationships found between phenotypic traits and herbivores/pollinators (table 2).

In population A the basic model was highly significant, suggesting that it was a very poor description of the observed data ($\chi^2 = 27.99$, $df = 8$, $P < .0001$, CALIS). According to this basic model, the main effect on female fitness was produced directly by ungulate damage (table 3). In addition, of the seven alternative models, model 7 (table 4) was the best description of the observations

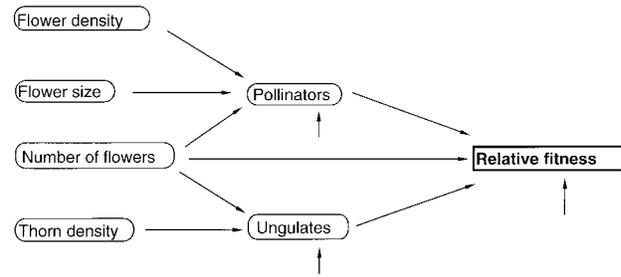


Figure 1: Hypothesized causal scheme for the relationships among phenotypic traits, pollinator visitation rate, ungulate damage, and female fitness for *Hormathophylla spinosa* (basic model). See table 3 for the actual values of the effects of each variable in each populations.

($\chi^2 = 0.109$, $df = 4$, $P = .950$, $\chi^2/df = 0.027$, lower than the suggested ratio of 5; see Jordano 1995 and references therein), since it was the simplest nested model obtaining a nonsignificant difference in goodness of fit when compared with the other more complex alternative models. This model explained 24% of the total variability of the between-plants differences in relative fitness ($r^2 = 0.243$). According to this model, the main negative influence on relative fitness was determined by the herbivory of ungulates (livestock; fig. 2). In addition, there was low positive direct effects of both number of flowers and pollinators on relative fitness. However, the total effect of number of flowers on fitness was diluted by its also positive effect on ungulate damage. Moreover, due to this strong effect of ungulates, there was a clear indirect effect of thorn density on female fitness (fig. 2).

Table 1: Results of multiple regression of damage by each herbivore and pollination visitation rate on relative fitness per population

	A	B	C
	$\beta \pm SE$	$\beta \pm SE$	$\beta \pm SE$
Ungulates ^a	-.448 ± .650 ^{***}	-.222 ± .811 ^{ns}	-.880 ± .459 ^{***}
Granivorous birds ^b	-.183 ± 1.832 ^{ns}	.011 ± 1.012 ^{ns}	-.001 ± .102 ^{ns}
Floral herbivores ^c	-.024 ± 1.799 ^{ns}	-.551 ± 2.284 ^{ns}	-.308 ± 2.404 ^{ns}
Seed predators ^d	.400 ± 6.717 ^{ns}	-.150 ± 3.091 ^{ns}	...
Pollinators ^e	.021 ± .086 ^{ns}	.352 ± .117 [*]	.870 ± .109 ^{ns}

Note: Figures are standard coefficients ± standard error. The significance of each regression coefficient was tested using a randomization procedure. Multivariate analyses against absolute fitness offered similar results but are not shown.

^a Livestock in population A and *Capra pyrenaica* in populations B and C.

^b Linnets (*Carduelis cannabina*) in all populations.

^c *Pimelia monticola* in population A, *Oiketicoides febretea* and *Timarcha lugens* in population B, and *T. lugens* in population C.

^d *Ceutorhynchus* sp. nov.

^e Nectarivorous ants (mainly *Proformica longiseta*) and flies in populations A and B and solitary bees and hoverflies in population C.

* $P < .05$.

*** $P < .001$.

^{ns} Not significant.

Table 2: Description of the alternative models considered in each population

Model	Model description (paths constrained to zero from basic model)
1	1. Flower size to pollinator visitation rate 2. Number of flowers to ungulate damage
2	1. Flower size to pollinator visitation rate 2. Number of flowers to pollinator visitation rate
3	1. Flower density to pollinator visitation rate 2. Number of flowers to pollinator visitation rate 3. Number of flowers to ungulate damage
4	1. Flower size to pollinator visitation rate 2. Number of flowers to pollinator visitation rate 3. Number of flowers to ungulate damage
5	1. Flower density to pollinator visitation rate
6	1. Flower size to pollinator visitation rate 2. Number of flowers to ungulate damage 3. Thorn density to ungulate damage
7	1. Flower density to pollinator visitation rate 2. Flower size to pollinator visitation rate 3. Number of flowers to pollinator visitation rate

Note: The description refers to the paths appearing in the basic model (fig. 1) that were constrained to zero in each alternative model.

In population B, the basic model was nonsignificant ($\chi^2 = 4.14$, $df = 8$, $P = .84$). This model suggests that in this population the effect of ungulates was negligible, most effect on female fitness being produced directly by number of flowers (table 3). There was also an important positive effect of flower density on nectarivorous pollinators visitation rate. There were six simpler alternative models also adequately describing the observed data (table 4), all of which showed a nonsignificant goodness of fit when compared to the basic model. However, when compared to the alternative model among them, model 6 (fig. 3) provided the best description of the relationship among our variables since it was the simplest model (5 paths, $df = 4$) giving the lowest goodness of fit ($\chi^2 = 0.116$, $P = .946$, $\chi^2/df = 0.029$) and explaining about 70% of the variability in the relative fitness ($r^2 = 0.696$). According to this model, the effect of ungulate herbivory on female fitness of the plants was negligible, whereas the number of flowers was the main factor affecting, positively, the difference between plants in relative fitness. In addition, flower density significantly affected nectarivore abundance, although its indirect effect on female fitness was highly negated by the small direct effects of nectarivores on plant fitness (fig. 3).

Finally, the basic model was also nonsignificant in pop-

ulation C ($\chi^2 = 7.08$, $df = 8$, $P = .63$), showing that ungulate damage was the main negative effect on female fitness in this population (table 3). This basic model also suggests that flower size, number, and density were related to pollinator visitation rate, whereas thorn density negatively affected ungulate damage (table 3). Moreover, all alternative nested models proved nonsignificant in this population (table 4), with the three simplest models (models 5, 6, and 7) providing adequate description of the relationships among our variables. Nevertheless, model 7 was the simplest model (5 paths, $df = 4$) giving the lowest goodness of fit ($\chi^2 = 1.57$, $P = .456$, $\chi^2/df = 0.39$). According to this model, the effect of Spanish ibex herbivory on female fitness was very strong and negative (fig. 4). For this reason, the positive effect of thorn density was even greater in this population than in population A. Finally, the effect of flower number on female fitness was direct positive but indirect, via ungulate damage, negative. As a consequence, the total effect of flower number on female fitness was very low (fig. 4).

Table 3: Direct (DE) and total (TE) effects for basic path analyses in the three populations

Independents	Pollinators	Ungulates	Relative fitness	
	DE	DE	DE	TE
Population A:				
Pollinators195*	.195
Ungulates	-.420***	-.420
Flower size	.001001
Flower density	.083016
No. of flowers	-.212*	.060	.227*	.186
Thorns	...	-.148*062
r^2	.025	.040	.235	...
Population B:				
Pollinators079	.079
Ungulates	-.136*	-.136
Flower size	.097007
Flower density	.798***062
No. of flowers	.130	-.265*	.601***	.625
Thorns	...	-.106002
r^2	.484	.062	.465	...
Population C:				
Pollinators075	.075
Ungulates	-.506***	-.506
Flower size	.167*012
Flower density	-.234**	-.18
No. of flowers	.321**	.231**	.181*	.087
Thorns	...	-.199*101
r^2	.173	.125	.239	...

* $P < .05$.

** $P < .01$.

*** $P < .0001$.

Table 4: Summary of the goodness of fit for each alternative model in each plant population in the Sierra Nevada

Model	df	Population A		Population B		Population C	
		χ^2	P value	χ^2	P value	χ^2	P value
1	6	27.84	.0001	3.15	.789	4.82	.567
2	6	30.09	.0001	1.17	.978	8.03	.236
3	7	31.15	.0001	12.02	.099	9.35	.229
4	7	29.78	.0001	3.49	.837	10.80	.148
5	5	30.10	.0001	20.76	.005	3.87	.568
6	4	28.74	.0001	.116	.946	1.79	.408
7	4	.109	.950	1.263	.745	1.57	.456

Note: See table 2 for description of the alternative models.

Discussion

Our results have shown that although *Hormathophylla spinosa* interacts simultaneously with many mutualistic and antagonistic species in the Sierra Nevada, only ungulates and pollinators significantly affected plant fitness. This functional specialization increases far more if we consider that the relative effect of each main interacting organism on plant fitness differed between populations. In populations A and C, where ungulate pressure was most intense, ungulates influenced *H. spinosa* female fitness much more than did either the other antagonistic or the mutualistic organisms. By contrast, in population B, a population with low ungulate damage, the female fitness of the host plant was related mainly to pollinators. Consequently, *H. spinosa* must actually cope in each Sierra Nevada population with only a small local subsample of the whole assemblage of interacting organisms living on it, which presumably are the organisms exerting significant selective pressures on the plant (Thompson 1994, 1998).

Pollinators and herbivores respond locally to different phenotypic traits of *H. spinosa*; ungulates are significantly related to thorn density; whereas flower size, number, and density significantly affected to pollinator visitation rate. The path analyses and SEM confirmed the existence of a geographic variation in the selection affecting the *H. spinosa* phenotypic traits related to herbivory and pollination. In fact, the highest path coefficients both in the basic and the alternative models were obtained by different variables in each population.

Low-Herbivory Selective Scenario

In population B, a population with low ungulate herbivory pressure and pollinated almost exclusively by nectarivorous pollinators, the path analyses, both the basic and the fittest alternative, showed a strong relationship between flower density and pollinator visitation rate. Nectarivorous ants and flies move among flowers of *H. spinosa* by crawl-

ing (Gómez and Zamora 1992, 1999), which means that they would benefit from abundant, densely grouped flowers. This may explain why these insects were found on plants with high flower density, irrespective of the total number of flowers displayed, suggesting that these pollinators may be the actual selective agent provoking the positive correlation between flower density and female fitness in this population.

The path analysis and SEM also demonstrated a very strong relationship between number of flowers and plant female fitness. Plants with greater flower number produced more seeds than did conspecifics with fewer flowers. Since nectarivorous pollinators seemed to be attracted to plants with more flowers, according to the significant positive correlations we found between pollinator visitation rate and flower number in this population, this benefit of flower number could be mediated by pollinators. Flower number is a trait that affects the visitation rate of many pollinators (e.g., Conner and Neumeier 1995; Conner et al. 1995; Conner and Rush 1996; Ohashi and Yahara 1998; Rademaker and De Jong 1998; Vaughton and Ramsey 1998) and therefore affects the fitness of many plant species (e.g., Klinkhamer et al. 1992; Lawrence 1993; De Jong and Klinkhamer 1994; Conner et al. 1996; Vaughton and Ramsey 1998). However, in the case of *H. spinosa*, as shown in figure 3, the number of flowers per plant affects plant fitness not only indirectly via pollinator visitation rate but mainly directly by increasing potential reproduction (total number of ovules per plant), irrespective of the pollinator effect (see e.g., Herrera 1993; Jordano 1995; Conner and Rush 1997). These two nonmutually exclusive causes may act synergically to benefit many-flowered plants in this population.

High-Herbivory Selective Scenario

A second selective scenario occurred in Sierra Nevada populations in which ungulate damage was intense and the

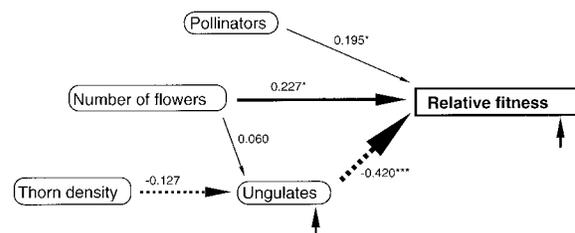


Figure 2: Representation of the path diagram corresponding to the best alternative model for population A (model 7). Positive effects are indicated by solid lines and negative effects by broken lines. Pollinators are nectarivorous ants (*Proformica longiseta*) and flies. Ungulates are domestic sheep and goats. * $P < .05$, ** $P < .01$, *** $P < .0001$.

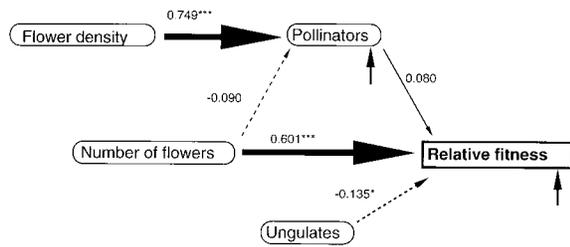


Figure 3: Representation of the path diagram corresponding to the best alternative model for population B (model 6). Positive effects are indicated by solid lines and negative effects by broken lines. Pollinators are nectarivorous ants (*Proformica longiseta*) and flies. Ungulates are mainly Spanish ibex (*Capra pyrenaica*). * $P < .05$, ** $P < .01$, *** $P < .0001$.

effect of any other antagonistic and mutualistic organisms was not detected (e.g., populations A and C). In these two populations, irrespective of the model considered, we found a strong relationship between plant fitness and ungulate damage, indicating that ungulates were the major selective agent. The main phenotypic trait affecting to ungulate damage was thorn density. Thorn presence is currently beneficial due to ungulate herbivory, given that we also found a high negative correlation between thorn density and ungulate damage, with herbivores consistently avoiding plants with many thorns. Spines and thorns are assumed to be defensive traits against mammal herbivores, mainly in nutrient-poor habitats (Campbell 1986; Cooper and Owen-Smith 1986; Gowda 1997; Obeso 1997; Cooper and Ginnett 1998), although a negative relationship between mammalian herbivory and thorn density has been shown for only some woody species (e.g., Belovsky et al. 1991; Milton 1991; Gowda 1996). In this study, we found a significant relationship of the defensive trait not only with the herbivore but also with the fitness of the plant, of which it explained a high intraspecific variance (up to 18%; J. M. Gómez and R. Zamora, unpublished data). As a consequence, thorns can be considered in these populations as a functional defense against mammal herbivory (*sensu* Zamora et al. 1999).

A major consequence of the high ungulate pressure in these populations was the cancellation of the positive effect of number of flowers, all plants having the same low fitness irrespective of initial flower production. More striking was that Spanish ibex and pollinators exerted opposite selection on flower number in population C; the first selective agent conferred reproductive advantage to low-flowered plants, whereas the latter conferred an advantage to many-flowered plants. As a consequence, flower number rendered a very low total effect in the SEM. As demonstrated for other ungulate-herbivorized plant species (e.g., Ehrlén

1997), this result points to a conflict between maximizing the probability of finding a mate and minimizing the risk of herbivory by ungulates in plants from highly herbivorized populations.

However, we have found some evidence that plants respond simultaneously to multiple selective agents in the same population, if those agents select for different, independent, and noncorrelated traits. This appears to occur with flower size, which was not affected by ungulate herbivory, and it did not correlate with any other plant traits related to ungulate herbivory. Thus, in population C, local pollinators (primarily *Colletes* sp., *Eristalis tenax*, and other species of bees and hoverflies) more frequently visited plants with larger flowers (see also Galen 1989, 1996; Eckhart 1991; Conner and Rush 1996). In contrast, flower size did not relate to pollinator visitation rate in the remaining, nectarivore-visited populations. This suggests spatial variation also in the relationship between flower size and fitness mediated by pollinator activity, with selection in flower size largely depending on the local composition of pollinator assemblage (see, e.g., Galen 1996 for similar results). This difference in strength and direction of selection exerted by different kinds of floral-visitor assemblage have also been reported for other plant species (Galen 1996) and is necessary for the maintenance of geographical variation in floral attributes.

Consequences of the Spatial Heterogeneity in Selective Scenarios

Overall, it appears that the between-population differences in the occurrence and abundance of main selective pressures, ungulates and pollinators, provokes a spatial variation in the selection on *H. spinosa* phenotypic traits, and thus the ecological and evolutionary consequences of associations between this crucifer and pollinators and herbivores are molded by local conditions that shift between populations. This spatial variation can potentially produce

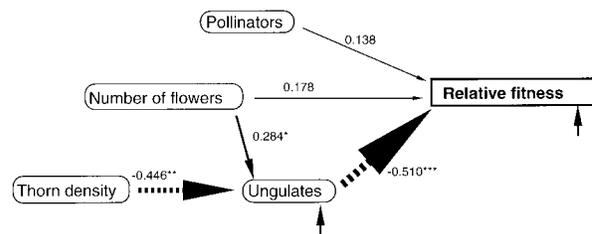


Figure 4: Representation of the path diagram corresponding to the best alternative model for population C (model 7). Positive effects are indicated by solid lines, and negative effects by broken lines. Pollinators are solitary bees and hoverflies. Ungulates are mainly Spanish ibex (*Capra pyrenaica*). * $P < .05$, ** $P < .01$, *** $P < .0001$.

Table 5: Between-population differences in plant traits

	Population A	Population B	Population C	F-ratio
No. flowers/plant	6,739 ± 585 ^a	11,621 ± 2,693 ^b	7,745 ± 635 ^a	2.81
Flower density	16.23 ± .68 ^a	27.95 ± 1.70 ^b	16.29 ± .75 ^a	36.26
Thorn density	33.20 ± 2.65 ^a	39.00 ± 2.16 ^a	51.36 ± 2.88 ^b	12.84
Flower diameter	6.02 ± .15 ^{a,b}	5.64 ± .21 ^a	6.72 ± .15 ^b	59.95

Note: F-ratio after Bonferroni correction. Means followed by different superscript letters are significantly different at $P < .05$ according to Bonferroni-Dunn post hoc test. Source for number of flowers per plant and flower diameters, Gómez and Zamora (1999).

local specialization and spatial divergence of plant traits as a consequence of the resulting divergent overall selective pressures (Bell et al. 1991; Campbell 1991; Van Tienderen and Van der Toorn 1991a; Arnqvist 1992; Thompson 1994; Galen 1996; Wilson and Thomson 1996). This idea is reinforced if we take into account that the selective regimes, although spatially variable, were temporally predictable in each populations during the years of study, according to the similar relationship found between fitness, phenotypic traits and interactive organisms according to the ANCOVA analyses. A possibility exists that the spatial variation in selection had produced local fitting, since we found significant interpopulational differences in the value of those *H. spinosa* phenotypic traits selected by pollinators and herbivores (see table 5). For example, plants from population B had many more flowers despite being as big as plants from other populations and had significantly more densely grouped flowers than did plants from each of the other two populations (table 5), which correlates with the foraging behavior of nectarivorous pollinators. Furthermore, plants from population C bore larger flowers than did plants from the other populations (table 5), a fact that agrees with the selection pattern of the local pollinating fauna, and had many more thorns than did plants from the other populations, in accord also with the heaviest ungulate herbivory.

A Final Consideration: Limitations and Advantages of SEM to Study Phenotypic Selection

Selection measurements based exclusively on observational phenotypic correlations between characters and fitness may be biased by the effect of unmeasured environmental or external correlations (Rausher 1992). The most appropriate way to test selective causal claims is, thus, to perform randomized experiments with genetically controlled individuals (Shiple 1999). However, for several reasons, this approach becomes highly impractical when studying many natural systems. First, working with long-lived species makes difficult the growth of families with different character expression for use in randomized experiments. Second, in the study of a complex system composed of many

ecologically different herbivores and pollinators, the use of experimental manipulations of each ostensibly important selective factor becomes unfeasible in practice. Third, the analysis of the system directly in nature and the quantification of the effect of how the major pressures varied among populations is a final limitation that makes experimental control almost impossible. In the cases in which the relevant variables cannot be physically controlled, we must rely on statistical control (Shiple 1999). Despite its limitations, SEM has proven to be a useful tool for performing inferential tests in these type of nonexperimental complex systems (Grace and Pugsek 1998; Shiple 1999). Our results suggest that the application of this analytical technique to several simultaneously monitored populations might help to isolate the major selection pressures on local populations and to identify potential selection differences among populations under field conditions. Consequently, SEM provides a practical exploratory approach to study the geographical variation of selection in complex systems.

Acknowledgments

We are most grateful to D. García and J. A. Hódar for invaluable help in the field work. Pedro Jordano advised us on the use of randomization tests. T. E. Miller, M. Rausher, J. Travis, and one anonymous reviewer improved early versions of the manuscript. We thank D. Nesbitt for linguistic advice. J.M.G. was supported by a grant from Plan de Formación del Personal Investigador (PFPI—Junta de Andalucía). This work was partially supported by Dirección General Interministerial de Ciencia y Tecnología (DGICYT) projects PB90-0852 and AMB95-0479.

Literature Cited

- Aarssen, L. W., and D. L. Irwin. 1991. What selection: herbivory or competition? *Oikos* 60:261–262.
- Armbruster, W. S. 1997. Exaptations link evolution of plant-herbivore and plant-pollinator interactions: a phylogenetic inquiry. *Ecology* 78:1661–1672.
- Armbruster, W. S., J. J. Howard, T. P. Clausen, E. M. Debevec, J. C. Loquvam, M. Matsuki, B. Cerendolo, and

- F. Andel. 1997. Do biochemical exaptations link evolution of plant defense and pollination systems? historical hypotheses and experimental test with *Dalechampia* vines. *American Naturalist* 149:461–484.
- Arnqvist, G. 1992. Spatial variation in selective regimes: sexual selection in the water strider, *Gerris odontogaster*. *Evolution* 46:914–929.
- Bell, G., M. J. Lechowicz, and C. J. Schoen. 1991. The ecology and genetics of fitness in forest plants. III. Environmental variance in natural populations of *Impatiens pallida*. *Journal of Ecology* 79:697–713.
- Belovsky, G. E., O. J. Schmitz, J. B. Slade, and T. J. Dawson. 1991. Effects of spines and thorns on Australian arid zone herbivores of different body masses. *Oecologia* (Berlin) 88:521–528.
- Brody, A. K. 1997. Effects of pollinators, herbivores, and seed predators on flowering phenology. *Ecology* 78:1624–1631.
- Campbell, B. M. 1986. Plant spinescence and herbivory in a nutrient poor ecosystem. *Oikos* 47:168–172.
- Campbell, D. R. 1991. Effects of floral traits on sequential components of fitness in *Ipomopsis aggregata*. *American Naturalist* 137:713–737.
- Conner, J. K., and R. Neumeier. 1995. Effects of black mustard population size on the taxonomic composition of pollinators. *Oecologia* (Berlin) 104:218–224.
- Conner, J. K., and S. Rush. 1996. Effects of flower size and number on pollinator visitation to wild radish, *Raphanus raphanistrum*. *Oecologia* (Berlin) 105:509–516.
- . 1997. Measurements of selection on floral traits in black mustard, *Brassica nigra*. *Journal of Evolutionary Biology* 10:327–335.
- 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.
- Conner, J. K., S. Rush, and P. Jennetten. 1996. Measurements of natural selection on floral traits in wild radish (*Raphanus raphanistrum*). I. Selection through lifetime female fitness. *Evolution* 50:1127–1136.
- Cooper, S. M., and T. F. Ginnett. 1998. Spines protect plants against browsing by small climbing mammals. *Oecologia* (Berlin) 113:219–221.
- Cooper, S. M., and N. Owen-Smith. 1986. Effects of plant spinescence on large mammalian herbivores. *Oecologia* (Berlin) 68:446–455.
- De Jong, T. J., and P. G. L. Klinkhamer. 1994. Plant size and reproductive success through female and male function. *Journal of Ecology* 82:399–402.
- Delph, L. F., M. H. Johannsson, and A. G. Stephenson. 1997. How environmental factors affect pollen performance: ecological and evolutionary perspectives. *Ecology* 78:1632–1639.
- Eckhart, V. M. 1991. The effects of floral display on pollination visitation vary among populations of *Phacelia linearis* (Hydrophyllaceae). *Evolutionary Ecology* 5:370–384.
- Edwards, A. L. 1985. Multiple regression and the analysis of variance and covariance. 2d ed. W. H. Freeman, New York.
- Ehrlén, J. 1997. Risk of grazing and flower number in a perennial plant. *Oikos* 80:428–434.
- Falconer, D. S. 1989. Introduction to quantitative genetics. 3d ed. Longman Scientific, New York.
- Farris, M. A., and M. J. Lechowicz. 1990. Functional interactions among traits that determine reproductive success in a native annual plant. *Ecology* 71:548–557.
- Fineblum, W. L., and M. D. Rausher. 1997. Do floral pigmentation genes also influence resistance to enemies? the W locus in *Ipomoea purpurea*. *Ecology* 78:1646–1654.
- Galen, C. 1983. The effects of nectar-thieving ants on seed set in floral scent morphs of *Polemonium viscosum*. *Oikos* 41:245–249.
- . 1989. Measuring pollinator-mediated selection on morphometric floral trait: bumblebees and the Alpine Sky Pilot, *Polemonium viscosum*. *Evolution* 43:882–890.
- . 1996. Rates of floral evolution: adaptation to bumblebee pollination in an alpine wildflower, *Polemonium viscosum*. *Evolution* 50:120–125.
- Gómez, J. M. 1993. Phenotypic selection on flowering synchrony in a high mountain plant, *Hormathophylla spinosa* (Cruciferae). *Journal of Ecology* 81:605–613.
- . 1994. Importance of direct and indirect effects in the interaction between a parasitic angiosperm (*Cuscuta epithimum*) and its host plant (*Hormathophylla spinosa*). *Oikos* 71:97–106.
- Gómez, J. M., and R. Zamora. 1992. Pollination by ants: consequences of the quantitative effects on a mutualistic system. *Oecologia* (Berlin) 91:410–418.
- . 1994. Top-down effects in a tritrophic system: parasitoids enhance plant fitness. *Ecology* 75:1023–1030.
- . 1996. Wind pollination in high-mountain populations of *Hormathophylla spinosa* (Cruciferae). *American Journal of Botany* 83:580–585.
- . 1999. Generalization vs. specialization in the pollination system of *Hormathophylla spinosa*. *Ecology* 80:796–805.
- Gowda, J. H. 1996. Spines of *Acacia tortilis*: what do they defend and how? *Oikos* 77:279–284.
- . 1997. Physical and chemical response of juvenile *Acacia tortilis* trees to browsing: experimental evidence. *Functional Ecology* 11:106–111.
- Grace, J. B., and B. H. Pugesek. 1998. On the use of path

- analysis and related procedures for the investigation of ecological problems. *American Naturalist* 152:151–159.
- Herrera, C. M. 1993. Selection on floral morphology and environmental determinants of fecundity in a hawk moth–pollinated violet. *Ecological Monographs* 63: 251–275.
- Irwin, R. E., and A. K. Brody. 1998. Nectar robbing in *Ipomopsis aggregata*: effects on pollinator behavior and plant fitness. *Oecologia* (Berlin) 116:519–527.
- Jarosz, A. M., and J. J. Burdon. 1991. Host-pathogen interactions in natural populations of *Linum marginale* and *Melampsora lini*. II. Local and regional variation in patterns of resistance and racial structure. *Evolution* 45: 1618–1627.
- Jennersten, O. 1988. Insect dispersal of fungal disease: effects of *Ustilago* infection pollinator attraction in *Viscaria vulgaris*. *Oikos* 51:163–170.
- Jordano, P. 1995. Frugivore-mediated selection on fruit and seed size: birds and St. Lucie's cherry, *Prunus mahaleb*. *Ecology* 76:2627–2639.
- Kingsolver, J. G., and D. W. Schemske. 1991. Path analyses of selection. *Trends in Ecology & Evolution* 6:276–280.
- Klinkhamer, P. G. L., E. Meelis, T. J. de Jong, and J. Weiner. 1992. On the analysis of size-dependent reproductive output in plants. *Functional Ecology* 6:308–316.
- Lawrence, W. S. 1993. Resource and pollen limitation: plant size-dependent reproductive patterns in *Physalis longifolia*. *American Naturalist* 141:296–313.
- Li, C. C. 1981. Path analysis, a primer. 3d ed. Boxwood, Pacific Grove, Calif.
- Lloyd, D. G., and S. C. H. Barrett. 1996. *Floral biology: studies on floral evolution in animal-pollinated plants*. Chapman & Hall, New York.
- Manly, B. F. J. 1991. *Randomization and Monte Carlo methods in biology*. Chapman & Hall, London.
- Marquis, R. J. 1992. The selective impact of herbivores. Pages 301–325 in R. S. Fritz and E. L. Simms, eds. *Plant resistance to herbivores and pathogens*. University of Chicago Press, Chicago.
- McCauley, D. E. 1991. The effect of host plant patch size variation on the population structure of a specialist herbivore insect, *Tetraopes tetraophthalmus*. *Evolution* 45: 1675–1684.
- Meyer, G. A., and R. B. Root. 1993. Effects of herbivorous insects and soil fertility on reproduction of goldenrod. *Ecology* 74:1117–1128.
- Milton, S. J. 1991. Plant spinescence in arid southern Africa: does moisture mediate selection by mammals. *Oecologia* (Berlin) 87:279–287.
- Mitchell, R. J. 1992. Testing evolutionary and ecological hypotheses using path analysis and structural equation modeling. *Functional Ecology* 6:123–129.
- . 1993. Path analysis: pollination. Pages 211–231 in S. M. Scheiner and J. Gurevitch, eds. *Design and analysis of ecological experiments*. Chapman & Hall, New York.
- . 1994. Effects of floral traits, pollinator visitation, and plant size on *Ipomopsis aggregata* fruit production. *American Naturalist* 143:870–889.
- Niesenbaum, R. A. 1996. Linking herbivory and pollination: defoliation and selective fruit abortion in *Lindera benzoin*. *Ecology* 77:2324–2332.
- Obeso, J. R. 1997. The induction of spinescence in European holly leaves by browsing ungulates. *Plant Ecology* 129:149–156.
- Ohashi, K., and T. Yahara. 1998. Effects of variation in flower number on pollinator visits in *Cirsium purpuratum* (Asteraceae). *American Journal of Botany* 85: 219–224.
- Pellmyr, O. 1989. The cost of mutualism: interactions between *Trollius europaeus* and its pollinating parasites. *Oecologia* (Berlin) 78:53–59.
- . 1997. Pollinating seed eaters: why is active pollination so rare? *Ecology* 78:1655–1660.
- Pilson, D. 1996. Two herbivores and constraints on selection for resistance in *Brassica rapa*. *Evolution* 50: 1492–1500.
- Press, W. H., S. A. Teukolski, W. T. Vetterling, and B. P. Flannery. 1992. *Numerical recipes in FORTRAN*. Cambridge University Press, Cambridge.
- Quesada, M., K. Bollman, and A. G. Stephenson. 1995. Leaf damage decreases pollen production and hinders pollen performance in *Cucurbita texana*. *Ecology* 76: 437–443.
- Rademaker, M. C. J., and T. J. De Jong. 1998. Effects of flower number on estimated pollen transfer in natural populations of three hermaphroditic species: an experiment with fluorescent dye. *Journal of Evolutionary Biology* 11:623–641.
- Rausher, M. D. 1992. The measurement of selection on quantitative traits: biases due to environmental covariance between traits and fitness. *Evolution* 46:616–626.
- Rausher, M. D., and E. L. Simms. 1989. The evolution of resistance to herbivory in *Ipomoea purpurea*. I. Attempts to detect selection. *Evolution* 43:563–572.
- Rice, W. R. 1989. Analyzing tables of statistical tests. *Evolution* 43:223–225.
- Roy, B. A. 1994. The effects of pathogen-induced pseudo-flowers and buttercups on each otheas insect visitation. *Ecology* 75:352–358.
- SAS Institute. 1997. *SAS/STAT software: changes and enhancements through release 6.12*. SAS Institute, Cary, N.C.
- Schemske, D. W., and C. C. Horvitz. 1988. Plant-animal interactions and fruit production in a neotropical herb: a path analysis. *Ecology* 69:1128–1137.

- Shiple, B. 1997. Exploratory path analysis with applications in ecology and evolution. *American Naturalist* 149: 1113–1138.
- . 1999. Testing causal explanations in organismal biology: causation, correlation and structural equation modeling. *Oikos* 86:374–382.
- Shykoff, J. A., E. Buchell, and O. Kaltz. 1997. Anther smut disease in *Dianthus silvester* (Caryophyllaceae): natural selection on floral traits. *Evolution* 51:383–392.
- Simms, E. L. 1990. Examining selection on the multivariate phenotype: plant resistance to herbivores. *Evolution* 44: 1177–1188.
- Simms, E. L., and M. D. Rausher. 1989. The evolution of resistance to herbivory in *Ipomoea purpurea*. II. Natural selection by insects and costs of resistance. *Evolution* 43:573–585.
- Strauss, S. Y. 1997. Floral characters link herbivores, pollinators, and plant fitness. *Ecology* 78:1640–1645.
- Swank, S. E., and W. C. Oechel. 1991. Interactions among the effects of herbivory, competition, and resource limitation on chaparral herbs. *Ecology* 72:104–115.
- Thompson, J. N. 1988. Variation in interspecific interactions. *Annual Review of Ecology and Systematics* 19: 65–87.
- . 1994. *The coevolutionary process*. University of Chicago Press, Chicago.
- . 1998. Coping with multiple enemies: 10 years of attacks on *Lomatium dissectum* plants. *Ecology* 79: 2550–2554.
- . 1999a. The raw material of coevolution. *Oikos* 84:5–16.
- . 1999b. Specific hypotheses on the geographic mosaic of coevolution. *American Naturalist* 153(suppl.): S1–S14.
- Travis, J. 1996. The significance of geographical variation in species interactions. *American Naturalist* 148(suppl.): S1–S8.
- van Tienderen, P. H., and J. van der Toorn. 1991a. Genetic differentiation between populations of *Plantago lanceolata*. I. Local adaptation in three contrasting habitats. *Journal of Ecology* 79:27–42.
- . 1991b. Genetic differentiation between populations of *Plantago lanceolata*. II. Phenotypic selection in a transplant experiment in three contrasting habitats. *Journal of Ecology* 79:43–59.
- Vaughton, G., and M. Ramsey. 1998. Floral display, pollinator visitation and reproductive success in the dioecious perennial herb *Wurmbea dioica* (Liliaceae). *Oecologia* (Berlin) 115:93–101.
- Wilson, P., and J. D. Thomson. 1996. How do flowers diverge? Pages 88–112 in D. G. Lloyd and S. C. H. Barrett, eds. *Floral biology, studies on floral evolution in animal-pollinated plants*. Chapman & Hall, New York.
- Zamora, R. 1999. Conditional outcomes of interactions: the pollinator-prey conflict of an insectivorous plant. *Ecology* 80:786–795.
- Zamora, R., and J. M. Gómez. 1993. Vertebrate herbivores as predator of insect herbivores: an asymmetrical interaction mediated by size differences. *Oikos* 66: 223–228.
- Zamora, R., J. A. Hódar, and J. M. Gómez. 1999. Plant-herbivore interaction: beyond a binary vision. Pages 677–718 in F. Pugnaire and F. Valladares, eds. *Handbook of functional plant ecology*. Dekker, New York.
- Zar, J. H. 1996. *Biostatistical analysis*. 3d ed. Prentice-Hall, Upper Saddle River, N.J.