

Comparing the Consequences of Induced and Constitutive Plant Resistance for Herbivore Population Dynamics

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ABSTRACT: Although it has been suggested that induced and constitutive plant resistance should have different effects on insect herbivore population dynamics, there is little experimental evidence that plant resistance can influence herbivore populations longer than one season. We used a density-manipulation experiment and model fitting to examine the effects of constitutive and induced resistance on herbivore dynamics over both the short and long term. We used likelihood methods to fit population dynamic models to recruitment data for populations of Mexican bean beetles on soybean varieties with no resistance, constitutive resistance, or induced resistance. We compared model configurations that fit parameters for resistance types separately to models that did not account for resistance type. Models representing the hypothesis that the three resistance types differed in their effects on beetle dynamics received the most support. Induced resistance resulted in lower population growth rates and stronger density dependence than no resistance. Constitutive resistance resulted in lower population growth rates and stronger density dependence than induced resistance. Constitutive resistance had a stronger effect on both short-term beetle recruitment and predicted beetle population dynamics than induced resistance. The results of this study suggest that induced and constitutive resistance can differ in their effects on herbivore populations even in a relatively complex system.

Keywords: plant-insect interactions, induced resistance, population dynamics, *Epilachna verivestis*, *Glycine max*, density-manipulation experiment.

An extensive body of basic and applied agricultural research on plant resistance to insect herbivores clearly dem-

onstrates that plant resistance traits affect individual insect behavior and performance (Fritz and Simms 1992). Because plant resistance can affect herbivore fecundity and survival, it is expected that plant resistance may also affect the long-term population dynamics of insect herbivores (Rhoades 1983, 1985; Schultz 1988; Hunter and Price 1992). However, relative to data on individual herbivore performance, data on population growth within one season are scant (but see, e.g., Webb and Moran 1978; Karban 1986), and experiments examining the effect of plant resistance types on long-term population dynamics are lacking. Thus, there is relatively little good experimental evidence that plant resistance traits can influence herbivore populations over longer than a few herbivore generations.

One aspect of plant resistance that has received a great deal of attention, with regard to herbivore dynamics, is induced resistance. Induced resistance can be most simply defined as a change in plant resistance resulting from herbivore damage. Because the strength of induced resistance produced by a single plant can depend on the density of attacking herbivores (Karbon and Baldwin 1997; Underwood 2000), it has been considered to be a potential source of density-dependent feedback to insect population sizes. Both verbal (Rhoades 1985; Schultz 1988) and mathematical (Edelstein-Keshet and Rausher 1989; Underwood 1999) theory predict that induced resistance should increase the density dependence acting on insect populations and thus contribute to the regulation of insect populations. For a population to be regulated (having a tendency to return to some mean size; Turchin 1995), net density dependence must be inverse to and stronger than density-independent factors affecting the populations. If induced resistance is strong and/or acts on a delay relative to herbivore generation time, theory also predicts that it can drive cycles in herbivore populations because delayed density-dependent factors tend to cause oscillations (Turchin and Taylor 1992; Lundberg et al. 1994). This is in contrast to constitutive plant resistance, which is the constant level of resistance a plant has, regardless of herbivore attack. Constitutive resistance cannot in itself be density dependent (by definition) and thus should not act to reg-

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ulate or drive cycles in herbivore populations. Constitutive resistance, however, might be expected to contribute to slower rates of population growth or lower equilibrium population sizes. Although these predicted differences in the effects of constitutive and induced resistance have been in the literature for at least 15 yr (Rhoades 1985), they have not been tested experimentally.

Predictions about the effects of plant resistance on long-term herbivore dynamics likely have not been tested because experiments involving long-term dynamics can be prohibitively time consuming in most systems. An alternative to directly observing long-term dynamics is to use density-manipulation experiments carried out in a single season to fit a model of population dynamics to the population of interest and then to use the model to estimate the longer-term behavior of the population. While using this method to predict the quantitative dynamics of specific populations requires numerous assumptions, it nevertheless offers a powerful tool for making qualitative comparisons of the effects of factors such as resistance types on long-term population dynamics (Belovsky and Joern 1995; Harrison and Cappuccino 1995; Underwood and Rausher 2000).

In this article, we present the results of a field experiment designed to determine how induced and constitutive plant resistance differ in their effects on herbivore population dynamics. We compared the dynamics of a specialist herbivore (the Mexican bean beetle) feeding on varieties of soybeans with three types of resistance: no resistance, constitutive resistance, and induced resistance. We fit population dynamic models to single-generation recruitment data for the beetles and used these models to examine the effects of resistance type on longer-term beetle population dynamics. In particular, we sought to determine the effects of induced and constitutive resistance on aspects of herbivore dynamics, including the strength of density dependence and rate of approach to equilibrium.

Methods

System

Soybeans (*Glycine max*, Fabaceae) are grown in North Carolina, where this research was conducted, from early May through October 1998. They are attacked by a wide range of pest insects, including the Mexican bean beetle (*Epilachna varivestis*, Coccinellidae), which is a specialist on legumes and an economic pest of soybeans in the Midwestern United States. The densities of Mexican bean beetles in soybean fields in North Carolina range from about 0.8 beetles/m² to 32 beetles/m², averaging 1.5 beetles/m² (Deitz et al. 1976). Beetles lay their eggs on soybean leaves, and adults and larvae feed on the leaves. Adults overwinter

under leaf litter. In North Carolina, Mexican bean beetles can complete three generations each year. Beetles for this study were reared on snap beans (*Phaseolus vulgaris*) in laboratory colonies at the New Jersey Department of Agriculture (West Trenton, N.J.) and Duke University. Soybean seed was provided by T. Carter, North Carolina State University, Raleigh, North Carolina. Soybean induced resistance to Mexican bean beetles is systemic, and both constitutive and induced resistance are known to affect Mexican bean beetle feeding preference, mortality, and growth in the lab and the field (Kogan and Fischer 1991; Underwood et al. 2000).

In a previous study, greenhouse bioassays were used to identify soybean varieties differing in their levels of constitutive and induced resistance to Mexican bean beetles (fig. 1; Underwood et al. 2000). In the bioassays, we measured induced resistance as the preference of adult beetles for undamaged leaves from an undamaged plant as opposed to undamaged leaves from a plant damaged by Mexican bean beetle larvae. We measured constitutive resistance as the preference of adult beetles for leaf tissue of undamaged plants of each variety as opposed to plants of all other varieties when plants were presented in pairs. All plants in the bioassays were of the same age, and leaves used in choice tests were of the same developmental stage.

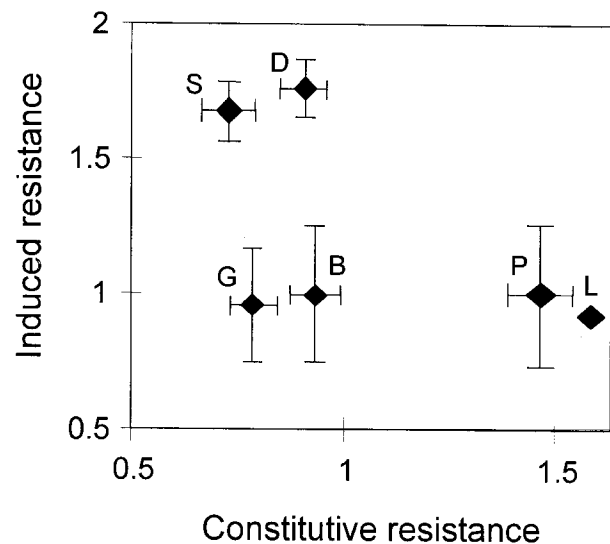


Figure 1: Levels of induced and constitutive resistance in six varieties of soybeans in greenhouse bioassays with adult Mexican bean beetles. Induced resistance is the relative preference of beetles for tissue from undamaged over damaged plants of the same variety. Constitutive resistance is the overall preference for undamaged plants of one variety over all other varieties. Sample sizes for each point range from 8 to 12, error bars indicate 1 SE (B, Bragg; D, Davis; G, Gasoy; L, Lamar; P, PI 416937; S, Stonewall). Data are from Underwood et al. (2000).

From these trials, we chose six varieties with three types of resistance: “no resistance” (low constitutive and no induced resistance; varieties Bragg and Gasoy), “induced resistance” (low constitutive and high induced resistance; varieties Davis and Stonewall), and “constitutive resistance” (high constitutive and no induced resistance; varieties Lamar and PI 416937). Lamar was not involved in the same set of greenhouse trials as the other varieties, but its high constitutive resistance is known from the agricultural literature, and it has never shown significant induced resistance in greenhouse trials (N. Underwood, personal observation). Measurements of cysteine proteinase inhibitor activity in plants during the experiment described here (results reported elsewhere; Underwood et al. 2002) support greenhouse measures of both induced and constitutive resistance characteristics. Proteinase inhibitors are a potential mechanism for resistance in plants (Ryan 1990), although little is known about how proteinase inhibitors in soybeans may affect beetles in the field.

Experimental Approach and Design

To determine how plant resistance type affects the population dynamics of Mexican bean beetles, we compared the dynamics of caged beetle populations in the field on these six soybean varieties. We characterized beetle dynamics by fitting population dynamic models to beetle recruitment data (see Hassell et al. 1976; Belovsky and Joern 1995; Underwood and Rausher 2000 for similar uses of recruitment curves). We obtained recruitment data by creating five initial densities of beetle populations on each soybean variety in the field and by following these populations for one full generation.

Using recruitment curves to predict specific long-term population dynamics requires assuming that the shape of the recruitment curve will not change over the time of the prediction. To our knowledge, data are not available to determine how recruitment curves may change over time in systems such as ours. Because the goal of this experiment was the qualitative comparison of beetle dynamics on plants with different types of resistance, rather than the prediction of the specific dynamics of particular populations, violations of this assumption should not affect our conclusions as long as any changes in curves affect all resistance types equally. Using recruitment curves for qualitative comparison of beetle dynamics also assumes that the curves reflect all of the important influences on dynamics that might differ between plant resistance types. This experiment was necessarily too short to include the influence of delayed density-dependent factors (such as maternal effects). Delayed density-dependent factors have been shown in other systems to have a potentially important influence on population dynamics (Prout and

McChesney 1985; Turchin 1990; Rossiter 1991). This experiment did, however, include the effects of predators and competitors and allowed for movement of herbivores both within and among plants. The recruitment curves thus reflect many of the ecological complexities that would be present in unmanipulated populations in the field. Any differences between recruitment curves detected in this relatively complex environment should then reflect meaningful differences in the field.

This experiment was carried out at the Duke University biology department's field station in Durham, North Carolina. Five densities of beetle populations (2, 6, 10, 14, and 20 per cage) were created on each of the six soybean varieties in three replicate spatial blocks. These densities span a range within what has been observed in the field in North Carolina (Deitz et al. 1976). This design was carried out twice during the summer of 1998 (i.e., in two temporal blocks), yielding a total of 160 experimental populations. Each population was located in a $1.5 \times 1.5 \times 2$ -m screen cage with a door opening to the outside above a 0.5-m sill (which helped to contain walking beetle larvae). Each block of 30 cages was surrounded by a 2-m-wide strip of soybean plants that served to attract beetle predators or parasites that might be attracted by the host plant. There is no natural population of Mexican bean beetles at this site, and the nearest agricultural fields that might have such populations are >10 km away.

Each cage was filled with 20 potted plants containing one of six varieties of soybeans. Soybeans were planted in 15-cm pots in Fafard P4 potting mix and treated with *Rhizobium*. Each plant had approximately five fully expanded trifoliolate leaves at the beginning of the experiment. All soybeans were fertilized with Osmocote time-release fertilizer when they had two fully expanded trifoliate leaves. Soybeans were subject to natural damage by insects other than Mexican bean beetles (primarily grasshoppers and lepidopteran larvae) throughout the experiment.

We initiated beetle populations by releasing equal numbers of male and female adult beetles into the cages. Adult beetles were marked with one or two spots of enamel paint on their elytra to identify the cages in which they belonged. We allowed the adults to mate and lay eggs for 1 wk before removing them and opening the doors of the cages to the surrounding soybean field to allow predators and competitors access to the cages. We allowed predators access to experimental beetle populations because it has been shown that genetic variation among host plants can affect the impact of predators (Fritz 1995) on herbivores in other systems, and we wanted to include both direct and indirect effects of host-plant resistance on beetle population dynamics. Cage doors remained open until larvae began to pupate. Soybean plants surrounding the cages were ex-

amed periodically to determine whether any larvae left the cages during this time. Each population was censused weekly for one generation (5–6 wk). Each week we counted clutches of eggs, eggs per clutch, larvae in each instar, pupae, and emerging adults. Egg masses and new adults were individually marked, so counts of these stages were likely to be very accurate, while counts of larvae in each instar were more approximate. Emerging adults were weighed. Analyses of data on beetle performance can be found in Underwood et al. (2002).

Analysis

To determine how plant resistance affected beetle dynamics, we fit a series of alternative models to the recruitment data at different densities for all resistance types simultaneously using likelihood methods (Edwards 1992). Before constructing and comparing models differing in the effects of variety and resistance type on population parameters, we performed model fits to all our data to identify the best overall model form and observation error distribution. We compared the fits of the Ricker ($N_{t+1} = N_t r e^{-bN_t}$) and discrete logistic models ($N_{t+1} = N_t \lambda (1 - [N_t/K])$; Edelman-Keshet 1988) using either Poisson or lognormal observation errors. In these models, N represents beetle population size, λ and r represent maximum per capita population growth rate, b represents the strength of density dependence in the system, K represents carrying capacity, and t indicates time step. We used Akaike weights (Burnham and Anderson 1998), which account for differences in the number of parameters between models and express the relative support for each model in a group, to select an appropriate population dynamics model and error structure. Akaike weights sum to 1 for the models being compared, and higher values indicate greater support for a given model. The combination of the Ricker model with lognormal error received the overwhelming majority of the support, so this combination was used for subsequent analyses (table 1).

Using the Ricker/lognormal configuration, we compared a set of seven models to determine the effects of plants on the two parameters (r and b) that determine beetle dynamics (table 2). The “saturated” model (model 1) represents the hypothesis that individual varieties differ in their effects on beetle dynamics (both r and b). Model 2 represents the hypothesis that resistance types differ in their effects on both r and b . Model 3 represents the hypothesis that resistance types differ in their effects on r but not on b . Likewise, model 4 represents the alternative that resistance classes differ in b but not r . Models 5 and 6 represent the hypotheses that varieties differ in their effects on either r (model 5) or b (model 6) but not both. Finally, we fit the minimal model (model 7) of no resis-

Table 1: Relative support for two population dynamics models and two error structures when fit to recruitment data

Model/error structure (using “saturated” configuration)	AIC	AIC weight
Ricker/lognormal	543.2089	1
Ricker/Poisson	2,307.501	0
Discrete logistic/Poisson	2,330.809	0

Note: AIC (Akaike Information Criterion) could not be estimated for the discrete logistic with lognormal error because the minimization routine was unable to converge.

tance class nor variety effects on beetle dynamics. For each model, a minimization routine (S-Plus 4.5; MathSoft 1998) was used to find parameter values that minimized the negative log likelihood of the data given the model. Relative support for each model was determined by calculating Akaike weights.

We generated 95% confidence limits around the best-fit parameters for each resistance type using bootstrapping (Manly 1991). Predicted equilibrium sizes of beetle populations on soybeans of each resistance type (N^*) were determined by setting $N_t = N_{t+1} = N^*$ and solving the best-fit Ricker equation for each resistance type for N^* .

We also used ANOVA (PROC GLM; SAS Institute 2000) to ask how temporal block, spatial block, resistance class, variety nested within resistance class, and initial population density affected per capita recruitment in each beetle population. Because temporal block and variety were never significant in preliminary analyses, final analyses were done with only spatial block, resistance type, and initial density. For this analysis, recruitment was log transformed to meet assumptions of normality and run, block, resistance class, initial density, and variety were considered classification variables.

Results

Per Capita Recruitment

Plant resistance type affected per capita recruitment of beetles (number of adult beetles produced by each beetle in the first generation in each experimental population; $F = 3.68$, $df = 2, 153$, $P = .03$; fig. 2). Beetle populations on plants with constitutive resistance had lower recruitment than those on plants with inducible resistance (post hoc contrast: $F = 5.34$, $df = 1, 153$, $P = .02$) and no resistance ($F = 5.76$, $df = 1, 153$, $P = .02$). There was clear evidence of density dependence in recruitment: slopes of linear regressions of \ln (per capita recruitment) on each resistance type differed significantly from 0 (no resistance: slope = -0.05 , $F = 4.19$, $df = 1, 55$, $r^2 = 0.07$, $P =$

.05; induced resistance: slope = -0.08 , $F = 8.79$, $df = 1, 57$, $r^2 = 0.14$, $P = .004$; constitutive resistance: slope = -0.1 , $F = 18.75$, $df = 1, 55$, $r^2 = 0.26$, $P < .0001$; fig. 3). Estimates of the slope of density dependence suggest that density dependence was weakest for no resistance (shallowest slope) and progressively stronger for induced resistance and constitutive resistance. Although there was no statistical support for the slopes differing among resistance types (no significant interaction between resistance type and initial density: $F = 1.27$, $df = 8, 153$, $P = .26$), the power to detect this interaction was relatively low (power $[1 - \beta] \approx 0.6$, $\Phi = 1.06$, $\alpha = 0.05$, $\nu_1 = 8$, $\nu_2 = 153$). There was no significant effect of variety nested within resistance on per capita recruitment.

Recruitment Curves

Despite substantial noise in the raw data, there were consistent differences in the recruitment curves for beetles on different host-plant types (fig. 4). Although no one model received an overwhelming share of support (table 3), the three models that estimate separate parameter values for each resistance type (models 2, 5, and 6) received between 1.8 and 746 times more support than models that estimate separate values for each variety. In total, the probability that the best model was one incorporating separate parameters for different resistance types was 0.89. It should be noted that differences among varieties are a combination of differences due to resistance class and differences due to additional (unmeasured) factors. We might therefore expect that models estimating separate parameters for each variety (models 1, 3, and 4) should provide a better fit to the data than models including only resistance classes. The Akaike Information Criterion (AIC) corrects for the fact that models with more parameters always provide better fits, giving a measure of whether additional parameters provide meaningful improvement in model fit. The fact that models that include separate parameters for each variety do not receive more support than models that include separate parameters for each resistance type indicates that including parameters for each variety does not provide a

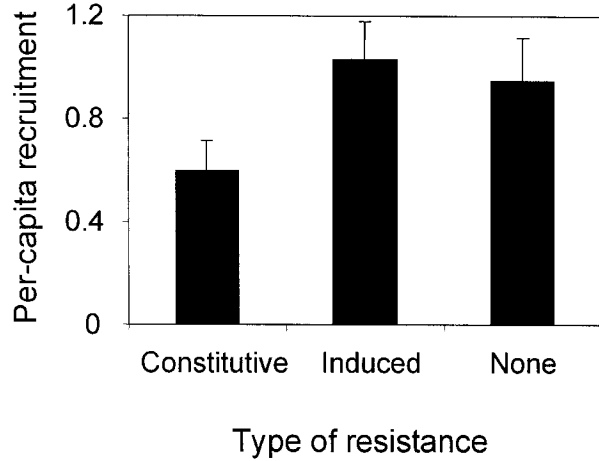


Figure 2: Per capita recruitment for beetles on plants with no resistance, constitutive resistance, and induced resistance. Error bars indicate 1 SE. $N = 60$ for each bar.

meaningful improvement over models including only resistance classes. This suggests that resistance classes account for the bulk of differences among varieties in their effects on herbivore dynamics.

Parameters for the three best-fit Ricker models and their 95% confidence intervals are shown in figure 5. We have presented the parameter estimates for all three models because models 5 and 6 fit fairly equally well but examine resistance effects on two different parameters. Presenting a combined model estimate (Burnham and Anderson 1998) could therefore be misleading. The two best-supported models show that either the rankings for b (model 6) are constitutive \gg inducible $>$ no resistance or that the rankings for r (model 5) are no resistance \gg inducible $>$ constitutive resistance. The model that includes resistance effects on both r and b (model 2, not as well supported) agrees with models 5 and 6 on the rankings for b but not for r (inducible $>$ no resistance $>$ constitutive resistance). Taken together, these results suggest that induced resistance tends to be intermediate in its effects on

Table 2: Models for examining effects of plant variety and resistance on beetle population dynamics

Model	Parameters for r	Parameters for b
1. Varieties affect r and b (saturated)	$r_{\text{Bragg}}, r_{\text{Davis}}, r_{\text{Gasoy}}, r_{\text{Lamar}}, r_{\text{PI}}, r_{\text{Stonewall}}$	$b_{\text{Bragg}}, b_{\text{Davis}}, b_{\text{Gasoy}}, b_{\text{Lamar}}, b_{\text{PI}}, b_{\text{Stonewall}}$
2. Resistance affects r and b	$r_{\text{constitutive}}, r_{\text{inducible}}, r_{\text{no resistance}}$	$b_{\text{constitutive}}, b_{\text{inducible}}, b_{\text{no resistance}}$
3. Varieties affect r	$r_{\text{Bragg}}, r_{\text{Davis}}, r_{\text{Gasoy}}, r_{\text{Lamar}}, r_{\text{PI}}, r_{\text{Stonewall}}$	b
4. Varieties affect b	R	$b_{\text{Bragg}}, b_{\text{Davis}}, b_{\text{Gasoy}}, b_{\text{Lamar}}, b_{\text{PI}}, b_{\text{Stonewall}}$
5. Resistance affects r	$r_{\text{constitutive}}, r_{\text{inducible}}, r_{\text{no resistance}}$	b
6. Resistance affects b	R	$b_{\text{constitutive}}, b_{\text{inducible}}, b_{\text{no resistance}}$
7. No effect of resistance or variety (minimal)	r	b

Note: All models are based on the Ricker model.

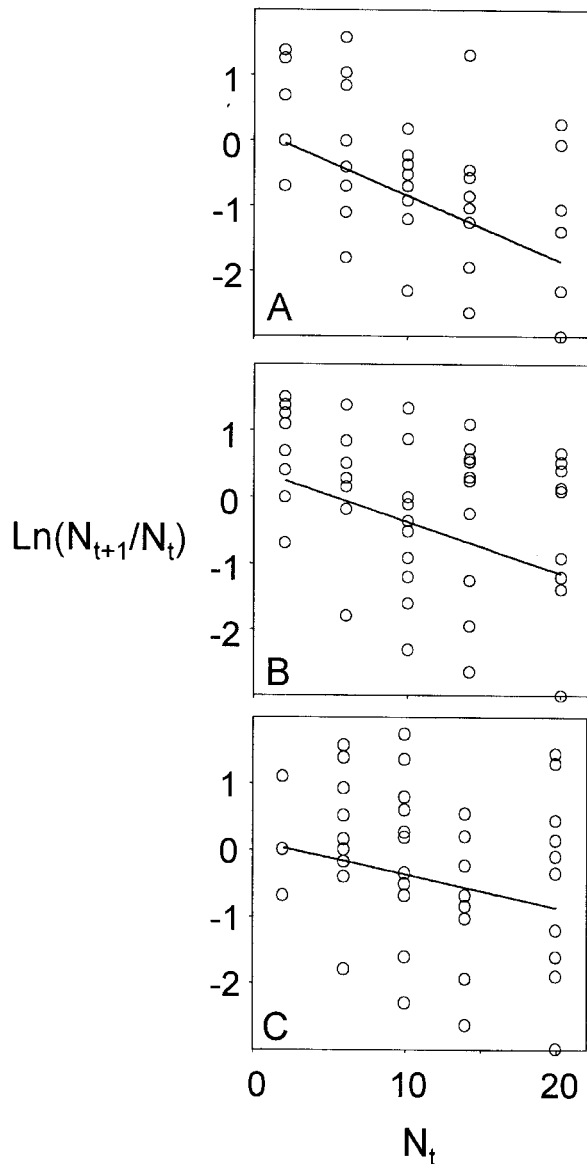


Figure 3: Effect of initial population density on $\ln(\text{recruitment})$ for beetle populations on plants with three types of resistance (A, constitutive; B, inducible; C, no resistance). Circles indicate individual beetle populations; lines indicate least squares regressions (constitutive: slope = -0.1 , intercept = 0.16 ; inducible: slope = -0.08 , intercept = 0.38 ; no resistance: slope = -0.05 , intercept = 0.13). Slopes are all significantly different from 0 ($P < .05$); intercepts are not significantly different from 0.

population dynamic parameters between constitutive resistance and no resistance. The 95% confidence intervals around the best-fit parameters for beetle populations on the three plant types do not overlap, further suggesting that resistance types have distinctly different parameters. Estimated values of r and b differ among resistance types

by up to 50% (table 3). Parameters for constitutive resistance differ more from those for no resistance than do parameters for induced resistance. The best-fit recruitment curves predict that populations on all resistance types will be stable, though for many resistance type/model combinations populations are predicted to go extinct (i.e., be trivially stable). Resistance type affects the rate of approach to extinction (constitutive < inducible < no resistance; fig. 6).

Discussion

Results of this experiment suggest that induced and constitutive resistance have the potential to differ in their effects on the short- and long-term population dynamics of Mexican bean beetles. Per capita recruitment and aspects of individual beetle performance, including number of eggs laid, final adult weight, and development time from first instar to adult differed significantly among plants with no resistance, induced resistance, and constitutive resistance (fig. 2; Underwood et al. 2002). These effects of resistance type on performance and recruitment indicate that plant resistance can influence herbivores in a single season. Recruitment curves summarize the expected dynamics of beetle populations on plants with different types of resistance over the long term. The three models of recruitment with the most support indicate that induced and constitutive resistance differed in their effects on the two parameters (r and b) determining beetle dynamics (table 3; fig. 5). The 95% confidence limits around the parameter values estimated for each resistance type do not overlap, further suggesting that plant resistance types may have distinct effects on population dynamics parameters. Thus, plant resistance may not only influence within-season herbivore numbers but may also influence across-season population trajectories.

In this system (Underwood 2000) and others (Karban and Baldwin 1997), the level of induced resistance expressed by a plant can be affected by herbivore density. This leads to the prediction that beetle populations on plants with induced resistance should experience increased density dependence relative to beetle populations on plants with no resistance or constitutive resistance. In the Ricker model, the strength of density dependence is controlled by the parameter b . While the best-fit values of b for beetle populations on plants with induced resistance were indeed larger than the values of b for plants with no resistance (table 3; fig. 5), the values for plants with constitutive resistance were even higher. Density dependence can also be measured as the relationship between per capita recruitment and initial density (fig. 3). Although ANOVA suggested that this measure of density dependence did not differ among resistance types, the power of this test to

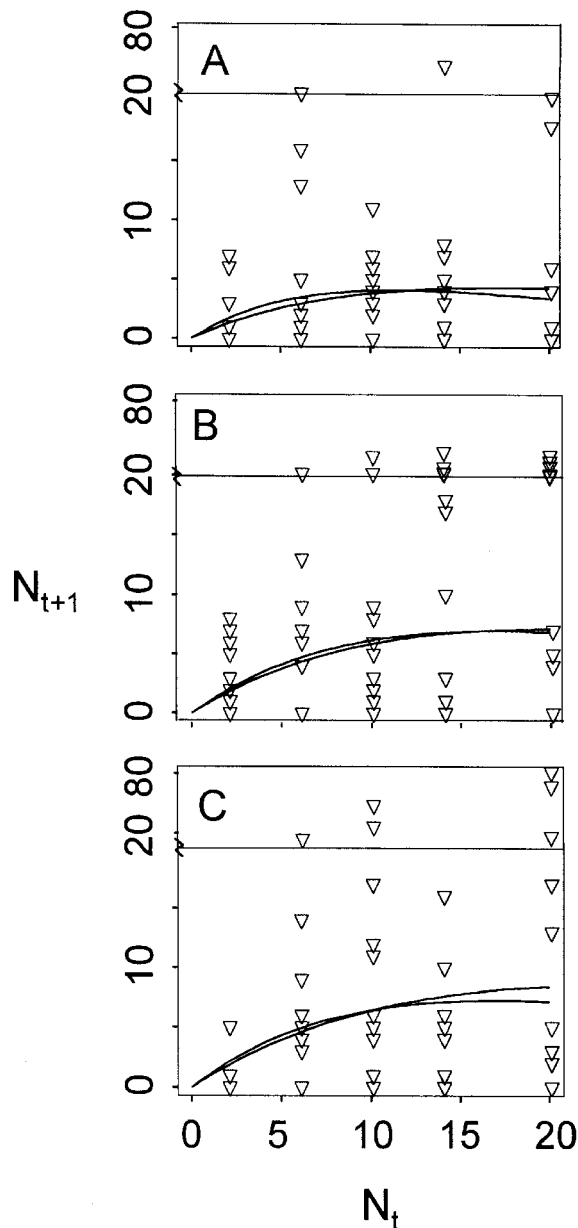


Figure 4: Numbers of adult beetles emerging from populations of beetles on plants with three types of resistance, initiated at five different beetle densities. Each triangle indicates a single experimental beetle population. Curved indicate best-fit recruitment curves, which are derived from the configurations of the Ricker model with the most support (model 5: resistance affects b ; model 6: resistance affects r). A, Constitutive. B, Inducible. C, No resistance.

detect such an interaction was only 0.6—a slightly better-than-even chance of detecting the interaction if it exists. The regression slopes agreed with the best-fit b parameters of the Ricker model, with density dependence (slope) in-

creasing from no resistance through induced resistance to constitutive resistance. By both measures of density dependence (slope of recruitment with density or value of b), plants with induced resistance added to the density dependence acting on beetle populations relative to plants with no resistance, and plants with constitutive resistance exhibited the strongest density dependence.

In theory, no resistance and constitutive resistance should not impose density dependence on herbivore populations. The fact that beetle populations on plants with no resistance exhibited significant density dependence suggests that there were sources of density dependence other than plant resistance acting in this experiment. Plants were not completely defoliated during the experiment, so beetle populations should not have been limited by total food quantity. However, there could have been behavioral competition for higher-quality plant parts, density-dependent predation, or other nonplant sources of density dependence acting on beetle populations. The strong density dependence exhibited by plants with constitutive resistance could be the result of an interaction between resistance type and other sources of density dependence. For example, lower overall plant quality could exacerbate competition for high-quality plant parts. It is also possible that soybean varieties with constitutive resistance have induced resistance not detected by beetles in greenhouse-feeding trials. Proteinase inhibitors are a candidate mechanism for induced resistance in this system. Plants with constitutive resistance changed PI levels, as damage accumulated over the experiment, less than plants with induced resistance, which suggests that induced resistance involving this mechanism was not occurring (Underwood et al. 2002). However, proteinase inhibitors are unlikely to represent the induced response in this system adequately, so it is not possible to determine conclusively whether induced resistance exists in the constitutively resistant varieties of soybean used in this experiment.

The estimated maximum growth rates (r) of beetle populations varied among resistance types by up to 40%, and many were below one (table 3; fig. 5). Growth rates below one result in the prediction that beetle populations will become extinct (fig. 6). For populations that do become extinct, the rates of extinction differ among resistance types. Beetle populations on plants with no resistance had the highest maximum growth rates in the best-supported models and thus the slowest rate of extinction, while beetle populations on plants with constitutive resistance had the lowest growth rates and fastest predicted extinction time. This result is intuitively appealing because plants with induced resistance should only have high levels of resistance when high densities of beetles are high, while constitutive resistance is expected to impose its negative effects on beetles at all times.

Table 3: Maximum likelihood estimates of the parameters (r and b) from seven configurations of the Ricker model fit to recruitment data from populations of beetles on varieties of soybeans with three types of resistance

Model	r			b			AIC	AIC weight	N^*
1. Saturated	.816 _{Lamar} [‡]	1.08 _{PI} [‡]	.874 _{Davis} [‡]	.074 _{Lamar} [‡]	.096 _{PI} [‡]	.037 _{Davis} [‡]	543.2089	.0008	...
	1.4 _{Stonewall} [‡]	.68 _{Bragg} [‡]		.081 _{Stonewall} [‡]	.008 _{Bragg} [‡]				
	1.05 _{Gasoy} [‡]			.057 _{Gasoy} [‡]					
2. Resistance r and b	1.115 _{inducible} [‡]	.849 _{no resistance} [‡]		.060 _{inducible} [‡]	.033 _{no resistance} [‡]		533.5886	.0945	1.83 _{inducible} [‡] , 0 _{no resistance} [‡]
	.940 _{constitutive}			.085 _{constitutive}					0 _{constitutive}
3. Varieties r	.679 _{Lamar} [‡]	.692 _{PI} [‡]	1.122 _{Davis} [‡]	.059			537.6261	.0125	...
	1.085 _{Stonewall} [‡]	1.241 _{Bragg} [‡]							
	1.072 _{Gasoy} [‡]								
4. Varieties b	.051			.967 _{Lamar} [‡]	.085 _{PI} [‡]	.088 _{Davis} [‡]	534.8398	.0505	...
				.044 _{Stonewall} [‡]	.056 _{Bragg} [‡]				
				.031 _{Gasoy} [‡]					
5. Resistance r	1.102 _{inducible} [‡]	1.149 _{no resistance} [‡]		.059			531.8799	.2220	1.66 _{inducible} [‡] , 2.37 _{no resistance} [‡]
	.685 _{constitutive}								0 _{constitutive}
6. Resistance b	.965			.050 _{inducible} [‡]	.041 _{no resistance} [‡]		529.9771	.5749	0 _{inducible} [‡] , 0 _{no resistance} [‡]
				.087 _{constitutive}					0 _{constitutive}
7. Minimal	.965			.059			535.0882	.0446	...

Note: Models estimate parameters separately for each variety (Lamar, PI, Davis, Stonewall, Bragg, and Gasoy) or for each resistance type (no resistance, induced resistance, or constitutive resistance); r = maximum population growth rate; b = strength of density dependence; AIC (Akaike Information Criterion) weights = the relative support for each model; N^* = the predicted equilibrium population size (calculated only for models including parameters for separate resistance types).

The low maximum growth rates estimated in this experiment are likely an artifact of limiting beetle egg laying to a single week, which was necessary to complete two temporal blocks of the experiment and achieve necessary replication. If beetles in this experiment had been allowed to lay eggs for their full adult lifetimes (approximately 3 wk), recruitment would likely have been much higher and yielded more nonzero equilibrium populations. Likewise, the specific values of b estimated here depend on the range of densities observed in the experiment. Estimates of r and b were also likely affected by other aspects of the experiment, such as the presence of competitors and predators. Thus, the specific parameter estimates reported here should not be taken as “realistic,” in the sense of being accurate for any particular “natural” population. However, differences in parameters among treatments are valid measures of the effects of different types of resistance. Because aspects of the natural system, such as competition and predation, were included in this experiment, these estimates give us some idea of the importance of induced and constitutive resistance in a somewhat realistic field setting. However, they should not be taken as a measure of the minimum or maximum possible effects of induced or constitutive resistance, which would require isolating the effects of resistance from other effects on beetle population dynamics.

There has been considerable interest in the possibility that induced resistance might be responsible for driving fluctuations in herbivore populations (e.g., Rhoades 1985). Theory predicts that for induced resistance alone to drive

fluctuations in herbivore populations there must either be lags in the production of induced resistance longer than a single herbivore generation, or induced resistance must have very strong effects (Underwood 1999). Very high population growth rates can also drive population fluctuations in the Ricker model, but this source of fluctuation is distinct from effects of induced resistance. The time course of the induced resistance produced by a single bout of Mexican bean beetle damage indicates that induced resistance is produced by 3 d and has decayed by 20 d following damage (Underwood 1998)—much too fast to produce lags that are likely to drive population fluctuations. However, the timing of induced responses to repeated damage is unknown. The experiment described here followed beetle populations for only one generation and thus could not directly detect longer-delayed effects of induced resistance. In a wide variety of insects, adult weight is related to fecundity (Engelmann 1970), and thus adult weight might indicate an effect of resistance on the next generation. However, while resistance type did affect the weight of emerging adults in this experiment (Underwood et al. 2002), there was no interaction of resistance type with density, suggesting that the density-dependent effect of resistance on fecundity did not differ. Thus, for fecundity at least, delayed effects are perhaps unlikely. The strength of density dependence produced by all resistance types was relatively low (the best-fit values of b were all small; table 3), suggesting that under the conditions of this experiment, induced resistance in the soybeans was not strong enough to drive cycles in Mexican bean beetle

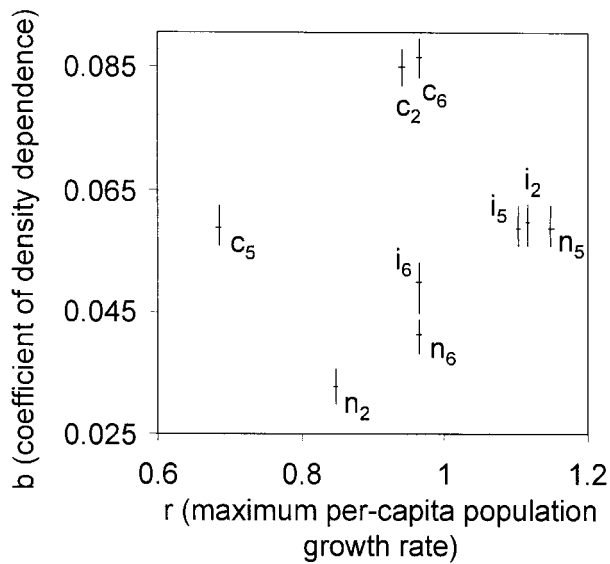


Figure 5: Best-fit parameters of three configurations of the Ricker model fit to beetle populations on plants with three types of resistance. *C*, Constitutive. *I*, Inducible. *N*, No resistance. Error bars indicate bootstrapped 95% confidence intervals.

populations. The population dynamics predicted by the best-fit recruitment curves for all resistance types and models in this experiment are in fact both stable and non-fluctuating (fig. 6), although this stability is mostly a trivial result since most of the populations are predicted to become extinct.

It is not clear what specific plant traits are responsible for induced and constitutive resistance in soybeans. The varieties of soybeans used in this experiment were assigned to resistance classes based on beetle-feeding preference. While this provides a clear measure of the effect of the plant on the herbivore, it does not tell us what aspect(s) of plant physiology are responsible for those effects. Cystein proteinase inhibitors were measured at the beginning and end of this experiment in all soybean populations (Underwood et al. 2002). Cystein proteinase inhibitors are a candidate for causing induced resistance to beetles in soybeans because they have been shown to affect beetle digestion in the lab and are increased by wounding. The constitutive and induced levels of proteinase inhibitors in each variety agree with beetle-feeding preferences (i.e., support the ratings of resistance based on bioassays) but do not predict performance in the field. Thus, while proteinase inhibitors are correlated with and may be involved in induced and constitutive resistance in this system, they are unlikely to account for the bulk of the response. Previous studies have found many changes in soybeans resulting from beetle injury, including increases in

isoflavones (Glyceolin), lipoxygenases, peroxidases, and proteinase inhibitors (e.g., Kogan and Fischer 1991; Felton et al. 1994).

In this study, we have used two different statistical approaches to examining the effects of resistance on herbivore dynamics: ANCOVA and the AIC. These two approaches yielded similar results, with both approaches suggesting differences among resistance classes in effects on recruitment and both approaches producing the same rankings of resistance classes in terms of recruitment (rate of increase) and strength of density dependence. We have used both approaches in this article because ANCOVA is

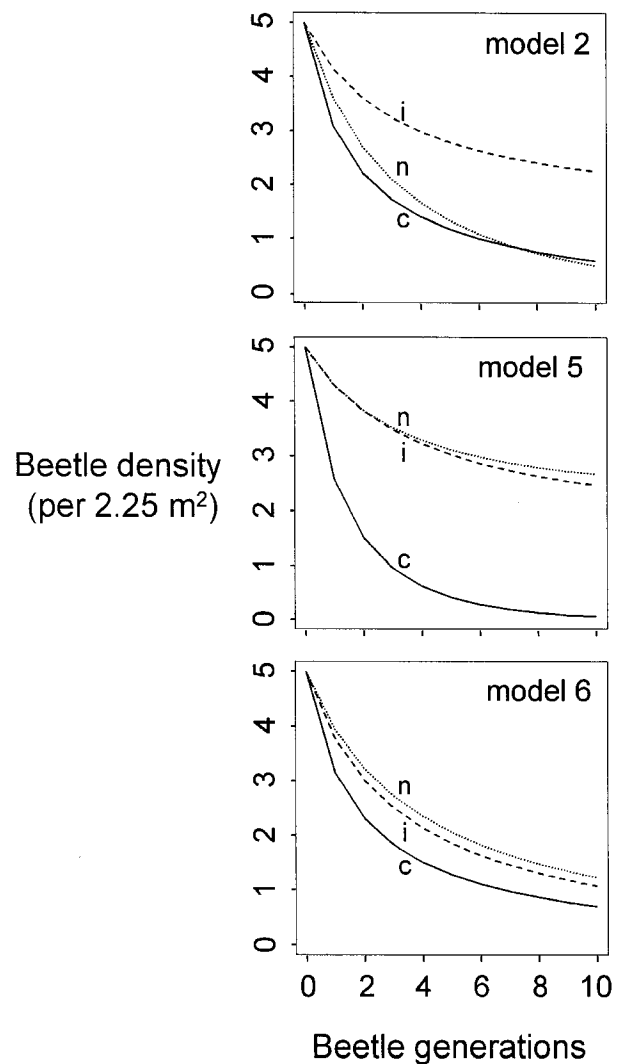


Figure 6: Dynamics of beetle populations predicted by three configurations (models 2, 5, and 6) of the Ricker model for beetle populations on plants with three types of resistance. *C*, Constitutive. *I*, Inducible. *N*, No resistance.

more familiar to most ecologists, while the AIC allows us to ask questions that the ANCOVA approach does not. For example, the AIC allows determination and use of the most appropriate error structure, while ANCOVA is limited to a smaller range of possible error structures. The AIC allows us to fit a large variety of models to the data, while ANCOVA only allows examination of models that can be linearized. Using AIC weights, we also can compare the relative support for many models at once rather than having to examine each individually.

Conclusions

Using a density-manipulation experiment and model fitting, we examined the effects of constitutive and induced resistance on herbivore population dynamics over both the short and long term. By all measures in this study, constitutive and induced resistance differed in their effects on beetle populations, and constitutive resistance had a stronger effect than induced resistance. This experiment included the presence of competitors and predators and was done at a spatial scale that allowed beetles to move among numerous plants. Much of the previous experimental work on effects of induced resistance on herbivores has occurred in tightly controlled environments and at the scale of single plants. However, this experiment necessarily sacrificed some precision in exchange for some realism. More controlled experiments and larger-scale and longer-running experiments are needed to isolate the effects of induced resistance on herbivore populations and to determine the importance of induced resistance relative to other factors influencing herbivore populations. The results of this study suggest that induced and constitutive resistance can differ in their effects on herbivore populations even in a relatively complex system. Characterizing herbivore dynamics with model fitting is a valuable tool for examining these population-level questions experimentally.

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Literature Cited

- Belovsky, G. E., and A. Joern. 1995. The dominance of different regulating factors for rangeland grasshoppers. Pages 359–386 in N. Cappuccino and P. W. Price, eds. *Population dynamics: new approaches and synthesis*. Academic Press, New York.
- Burnham, K., and D. Anderson. 1998. *Model selection and inference: a practical information-theoretic approach*. Springer, New York.
- Deitz, L. L., R. L. Rabb, J. W. Van Duyn, W. M. Brooks, J. R. J. Bradley, and R. E. Stinner. 1976. A guide to the identification and biology of soybean arthropods in North Carolina. North Carolina Agricultural Experiment Station Technical Bulletin no. 238. Raleigh, N.C.
- Edelstein-Keshet, L. 1988. *Mathematical models in biology*. Random House, New York.
- Edelstein-Keshet, L., and M. D. Rausher. 1989. The effects of inducible plant defenses on herbivore populations. 1. Mobile herbivores in continuous time. *American Naturalist* 133:787–810.
- Edwards, A. W. F. 1992. *Likelihood*. Johns Hopkins University Press, Baltimore.
- Engelmann, F. 1970. *The physiology of insect reproduction*. Pergamon, New York.
- Felton, G., C. B. Summers, and A. J. Mueller. 1994. Oxidative responses in soybean foliage to herbivory by bean leaf beetle and three-cornered alfalfa hopper. *Journal of Chemical Ecology* 20:639–650.
- Fritz, R. S. 1995. Direct and indirect effects of plant genetic variation on enemy impact. *Ecological Entomology* 20: 18–26.
- Fritz, R. S., and E. L. Simms. 1992. *Plant resistance to herbivores and pathogens: ecology, evolution, and genetics*. University of Chicago Press, Chicago.
- Harrison, S., and N. Cappuccino. 1995. Using density-manipulation experiments to study population regulation. Pages 131–147 in N. Cappuccino and P. W. Price, eds. *Population dynamics: new approaches and synthesis*. Academic Press, San Diego, Calif.
- Hassell, M., J. Lawton, and R. May. 1976. Patterns of dynamical behaviour in single-species populations. *Journal of Animal Ecology* 45:471–486.
- Hunter, M. D., and P. W. Price. 1992. Playing chutes and ladders: heterogeneity and the relative roles of bottom-up and top-down forces in natural communities. *Ecology* 73:724–732.
- Karban, R. 1986. Induced resistance against spider mites in cotton: field verification. *Entomologia Experimentalis et Applicata* 42:239–242.
- Karban, R., and I. T. Baldwin. 1997. *Induced responses to herbivory*. University of Chicago Press, Chicago.
- Kogan, M., and D. C. Fischer. 1991. Inducible defenses in

- soybean against herbivorous insects. Pages 347–378 in D. W. Tallamy and M. J. Raupp, eds. *Phytochemical induction by herbivores*. Wiley, New York.
- Lundberg, S., J. Jaremo, and P. Nilsson. 1994. Herbivory, inducible defence and population oscillations: a preliminary theoretical analysis. *Oikos* 71:537–539.
- Manly, B. F. J. 1991. *Randomization, bootstrap and Monte Carlo methods in biology*. Chapman & Hall, London.
- MathSoft. 1998. *S-Plus 4.5*. MathSoft, Seattle.
- Prout, T., and F. McChesney. 1985. Competition among immatures affects their adult fertility: population dynamics. *American Naturalist* 126:521–558.
- Rhoades, D. F. 1983. Herbivore population dynamics and plant chemistry. Pages 155–220 in R. F. Denno and M. S. McClure, eds. *Variable plants and herbivores in natural and managed ecosystems*. Academic Press, San Diego, Calif.
- . 1985. Offensive-defensive interactions between herbivores and plants: their relevance in herbivore population dynamics and ecological theory. *American Naturalist* 125:205–238.
- Rossiter, M. C. 1991. Environmentally-based maternal effects: a hidden force in insect population dynamics? *Oecologia (Berlin)* 87:288–294.
- Ryan, C. A. 1990. Protease inhibitors in plants: genes for improving defenses against insects and pathogens. *Annual Review of Phytopathology* 28:425–449.
- SAS Institute. 2000. *The SAS system*. SAS Institute, Cary, N.C.
- Schultz, J. C. 1988. Plant responses induced by herbivores. *Trends in Ecology & Evolution* 3:45–49.
- Turchin, P. 1990. Rarity of density dependence or population regulation with lags? *Nature* 344:660–663.
- . 1995. Population regulation: old arguments and a new synthesis. Pages 19–40 in N. Cappuccino and P. W. Price, eds. *Population dynamics: new approaches and synthesis*. Academic Press, New York.
- Turchin, P., and A. D. Taylor. 1992. Complex dynamics in ecological time series. *Ecology* 73:289–305.
- Underwood, N. 1999. The influence of plant and herbivore characteristics on the interactions between induced resistance and herbivore population dynamics. *American Naturalist* 153:282–294.
- . 2000. Density dependence in induced plant resistance to herbivore damage: threshold, strength and genetic variation. *Oikos* 89:295–300.
- Underwood, N., and M. D. Rausher. 2000. The effects of host-plant genotype on herbivore population dynamics in a model system. *Ecology* 81:1565–1576.
- Underwood, N., W. Morris, K. Gross, and J. R. I. Lockwood. 2000. Induced resistance to Mexican bean beetles in soybeans: variation among genotypes and lack of correlation with constitutive resistance. *Oecologia (Berlin)* 122:83–89.
- Underwood, N., M. D. Rausher, and W. Cook. 2002. Bioassay versus chemical assay: measuring the impact of induced and constitutive resistance on herbivores in the field. *Oecologia (Berlin)* 131:211–219.
- Underwood, N. C. 1998. The timing of induced resistance and induced susceptibility in the soybean Mexican bean beetle system. *Oecologia (Berlin)* 114:376–381.
- Webb, J. W., and V. C. Moran. 1978. The influence of the host plant on the population dynamics of *Acizzia russallae* (Homoptera: Psyllidae). *Ecological Entomology* 3: 313–321.