Selection of the common green lacewing for resistance to carbaryl

Elizabeth E. Grafton-Cardwell □ Marjorie A. Hoy

Lacewings, ladybird beetles, predatory mites, wasp parasites, and other natural enemies of pests are important biological control agents in many integrated pest management (IPM) programs throughout California. These natural enemies, however, are usually more sensitive to pesticides than are the pests they attack. As a result, when a pesticide must be applied for a key pest that has no biological agents controlling it, the natural enemies of other pests are killed and secondary pest outbreaks occur.

There are several hypotheses to explain why natural enemies may be more sensitive to pesticides than pest insects and mites. One is that they have different feeding habits. The natural defense systems of plants may stimulate production of detoxifying enzymes in pests that also break down pesticides. Since natural enemies generally do not feed on plants, they do not have these enzymes in sufficient quantities to detoxify pesticides. Availability of food after pesticide applications also gives pests an advantage over natural enemies. When pesticides are applied, most of the hosts and prey of the natural enemies are destroyed, which can cause the few survivors to starve. The pest survivors, however, simply feed on plants and rapidly rebuild their populations. Thus, the pest population recovers much more rapidly after treatment with pesticides and develops pesticide resistance more rapidly.

One method of improving biological control in agricultural IPM systems that use pesticides has been to select the natural enemy in the laboratory for resistance to one or more commonly used pesticides. Predatory mites in the family Phytoseiidae are currently the only biological control agents that have been successfully selected for levels of resistance useful in the field. In California almonds, strains of the predatory mite *Metaseiulus* (=*Typhlodromus*) *occidentalis* (Nesbitt) have developed resistance to organosphosphorus pesticides in the field and to carbamates and pyrethroids in the laboratory. The selected strains have been mass-reared and released to control Pacific spider mite, *Tetranychus pacificus* McGregor, and two-spotted spider mite, *T. urticae* Koch. The predatory mites have survived sprays applied for navel orangeworm control and kept spider mites under control so that fewer applications or lower rates of acaricides were needed.

The adult common green lacewing, *Chrysoperla carnea* Stephens, is a widely dispersing nectar-feeding insect that deposits eggs on both orchard and field crops. As a larva, it is a voracious predator of aphids, mites, small lepidopterous larvae, psylla, and eggs of many insects. The lacewing has been shown to be relatively tolerant of some chlorinated hydrocarbons, pyrethroids, and microbial insecticides. It is susceptible, however, to most of the commonly used organophosphorus and carbamate insecticides. Enhancement of the lacewing's tolerance of these pesticides would improve its effectiveness in many crops. We report here on the first case of laboratory selection of *C. carnea* that resulted in its ability to survive field rates of a pesticide.

TABLE 1. Pesticide concentrations at which 50 percent (LC_{50}) and 90 percent (LC_{90}) mortality occurred in first-stage common green lacewing larvae collected from four counties

Pesticide (field rate)* and colony tested	LC_{50}†		LC_{90}
Permethrin (0.2 lb)	*lb active ingredient/gal*		
San Joaquin	0.64	a	1.88
Fresno	2.48	c	20.33
Kern	0.23	a	11.20
Imperial	1.72	b	4.45
Fenvalerate (0.1 lb)			
San Joaquin	1.50	a	5.39
Fresno	1.40	a	2.86
Kern	1.63	ab	8.16
Imperial	2.22	b	17.39
Carbaryl (1.5 lb)			
San Joaquin	0.0013	a	0.0053
Fresno	0.0023	b	0.0068
Kern	0.0030	b	0.0063
Imperial	0.0032	b	0.0158
Methomyl (0.45 lb)			
San Joaquin	0.0009	ab	0.0023
Fresno	0.0012	b	0.0045
Kern	0.0004	a	0.0035
Imperial	0.0043	b	0.0042
Diazinon (0.5 lb)			
San Joaquin	0.0023	a	0.0085
Fresno	0.0024	a	0.0048
Kern	0.0011	a	0.0045
Imperial	0.0043	b	0.0101
Phosmet (1.0 lb)			
San Joaquin	10.0	a	20.3
Fresno	17.5	a	72.0
Kern	36.6	b	395.8
Imperial	158.5	c	3206.3

* Field rate proposed for alfalfa, mixed in 10 gallons of water.

† LC_{50} values in the same column for each pesticide followed by the same letter indicate that the responses of the colonies are not significantly different (p=0.05).

Geographical variability

The first stage of the laboratory selection program entailed collecting lacewings from widely separated areas of California. We then screened the lacewings with six pesticides to see if there was any geographical variability in the colonies' response to pesticides. If all of the lacewing populations showed a similar level of susceptibility to all pesticides, we would assume that selection was not occurring in the field and the genes necessary to develop resistance were rare. If so, laboratory selection could be slow and would require screening enough numbers to pick up such a rare gene. Variability in response to insecticides would imply that field selection for resistance had already begun.

We collected adult *C. carnea* from alfalfa in San Joaquin, Fresno, Kern, and Imperial counties during 1981 and 1982 and maintained them as four separate colonies. Adults, larvae, and eggs were screened with the organophosphates diazinon (50 percent WP) and phosmet (Imidan, 50 percent WP), the carbamates carbaryl (Sevin, 50 percent WP) and methomyl (Lannate, WS liquid), and the pyrethroids permethrin (Ambush, Pounce, EC) and fenvalerate (Pydrin, EC).

Adults were tested in petri dishes sprayed with a range of concentrations of formulated pesticide in water plus a wetting agent. Each dish containing five adult lacewings was closed with tissue paper, and adults were provided with food and water. Twenty adults were tested at each concentration, and mortality was assessed after 72 hours.

Adult lacewings were susceptible to field rates of the organophosphorus and carbamate insecticides. In contrast, they were tolerant of both pyrethroids, easily surviving field rates. This is a natural tolerance, not a field-selected resistance, because all colonies responded at a similar level and because other investigators have reported similar results. Two or more of the colonies differed significantly in their response to carbaryl, methomyl, and diazinon, indicating that field selection may be occurring. San Joaquin County lacewings consistently responded with the greatest susceptibility, which correlates with the lower use of pesticides in that county.

We tested lacewing eggs with the six pesticides using field rates proposed for alfalfa mixed in 10 gallons of water: permethrin, 0.2 pound; fenvalerate, 0.1 pound; carbaryl, 1.5 pounds; methomyl, 0.45 pound; diazinon, 0.5 pound; and phosmet, 1 pound. Female lacewings were al-

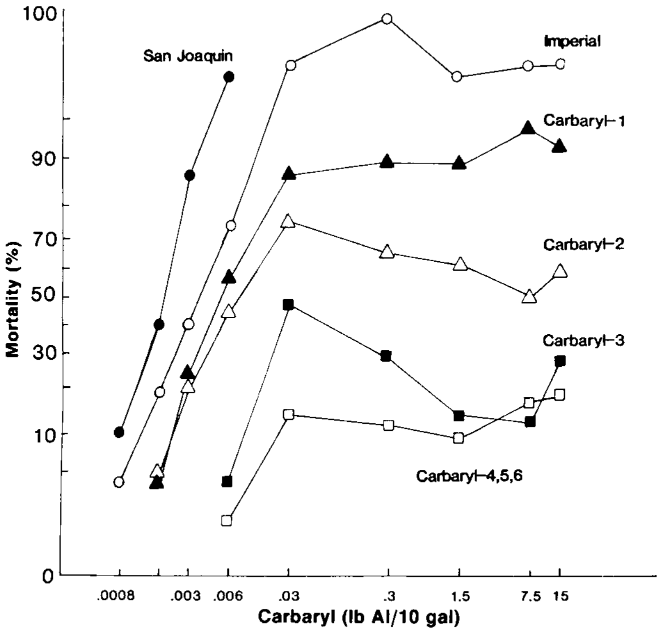


Fig. 1. Imperial County green lacewing larvae showed more than 80 percent survival after four generations of selection. All of highly susceptible San Joaquin larvae died at concentrations above .003.

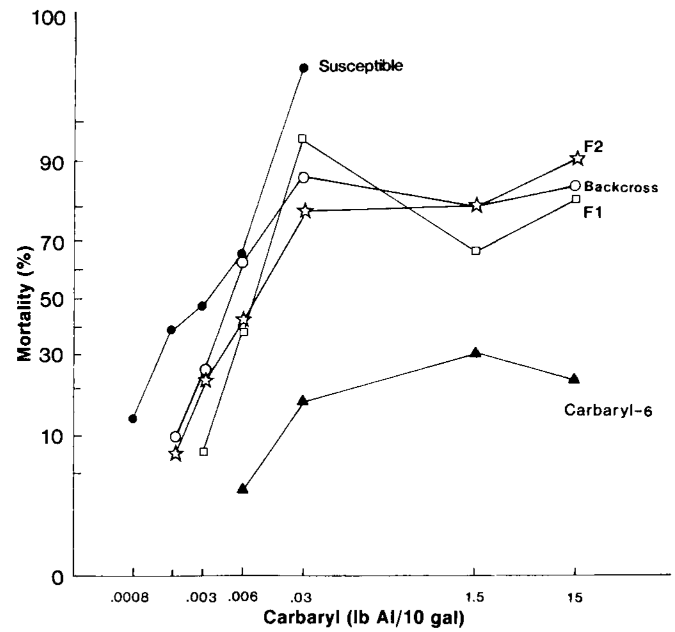


Fig. 2. Genetic analysis of purified susceptible strain crossed with the carbaryl-resistant strain (after six rounds of selection) and their offspring indicated no single dominant gene was involved.

lowed to deposit eggs on paper toweling. Each sheet of eggs was cut in two; one half was sprayed with formulated pesticide in water and the other half was sprayed with water alone. None of the pesticides tested affected egg hatch.

We tested lacewing larvae individually (to avoid cannibalism) in small wells of plastic microtiter plates. Each plate was treated with a concentration of pesticide mixed with distilled water plus a wetting agent, then air-dried; the tops of the wells were ringed with stickem. Grain moth eggs, *Anagasta kuehniella* Zeller, were provided as food for the larvae. Mortality was assessed after 48 hours. We tested 30 to 200 first-stage (instar) larvae at each concentration.

Like the adults, larvae tolerated field rates of the two pyrethroids, as shown by the fact that the LC_{50} values (the concentrations at which 50 percent mortality occurs) were higher than the field rates (table 1). The larvae were also highly tolerant of the organophosphate phosmet. Since no susceptible colonies were found, these are most likely natural tolerances. The larvae were susceptible to field rates of carbaryl, methomyl, and diazinon. In all cases, even for the three pesticides that the lacewings tolerated, there were significant differences in the responses of the four lacewing colonies, confirming the adult data suggesting that field selection is occurring. San Joaquin County lacewing larvae showed the greatest susceptibility (lowest LC_{50} values) and Imperial County larvae generally showed the greatest tolerance of the pesticides tested.

The greater tolerance to pesticides exhibited by the Imperial County lacewing larvae correlates well with the greater use of pesticides in that county. It is most likely that resistance has not reached field spray levels because lacewing adults fly to many different crops to deposit their eggs and so may be exposed to many pesticides during their lifetime. Because different pesticides are likely to have different modes of action, selection for any one is slow when an insect is exposed to several at the same time.

Although field-selected green lacewings did not have levels of resistance useful against field rates, the existence of more tolerant strains prompted us to attempt laboratory selection.

Carbaryl resistance

The Imperial County lacewing larvae showed the greatest overall tolerance of pesticides and so were chosen for laboratory selection with carbaryl. We screened larvae because they are the predatory stage of the lacewing and because they are most directly and continuously exposed to the pesticide residues on plants.

First-stage lacewing larvae were exposed to carbaryl at a rate that allowed, at most, 10 percent of the lacewings to survive. Larvae were placed in the wells of the treated plates (77°F) for 48 hours. Survivors were placed individually in untreated 1-ounce plastic cups with lids and reared to adulthood at room temperature (75° to 81°F). Their offspring were then used for the next selection. We tested 24 to 48 larvae from the San Joaquin and the original Imperial colonies as controls. Six generations of selection were performed using 0.003 and 0.3 pound active ingredient carbaryl per 10 gallons of water for the first two selections, and 7.5 pounds for the last four selections.

Selection for carbaryl resistance occurred very rapidly in this assay (fig. 1). All San Joaquin County larvae died at rates above 0.003 pound active ingredient per 10 gallons. Of the unselected Imperial County larvae, 97 to 99 percent died at rates above 0.03 pound, but after only one selection (carbaryl-1), mortality decreased to approximately 90 percent at these rates. Mortality decreased to 50 to 70 percent after the second round of selection (carbaryl-2) and to less than 20 percent after four rounds of selection. Up to 80 percent of the selected lacewing larvae survived on formulated material at the field rate recommended for alfalfa (approximately 1.5 pounds active ingredient per 10 gallons), compared with only 3 percent of the unselected strain.

An analysis of the mode by which the resistance was inherited indicated that no single, completely dominant or recessive gene was involved (fig. 2). The F_1 offspring (produced by crossing the susceptible and resistant parents), F_2 offspring (produced by crossing F_1 individuals), and the backcross offspring (produced by crossing F_1 individuals with susceptible individuals) all responded in an intermediate manner compared with the purified susceptible and resistant strains. Reciprocal crosses responded similarly, indicating no sex linkage of the resistance traits. The lack of complete dominance suggests that, in the field, interbreeding with susceptible lacewings would result in offspring with lower levels of resistance than the resistant parent. The lack of complete return to susceptibility in the F_1 offspring indicates that, if carbaryl sprays were applied, carbaryl resistance could be maintained by field selection.

Agricultural Experiment Station
UNIVERSITY OF CALIFORNIA
Berkeley, California 94720
Lowell N. Lewis, Director

Publication
PENALTY FOR PRIVATE USE, $300

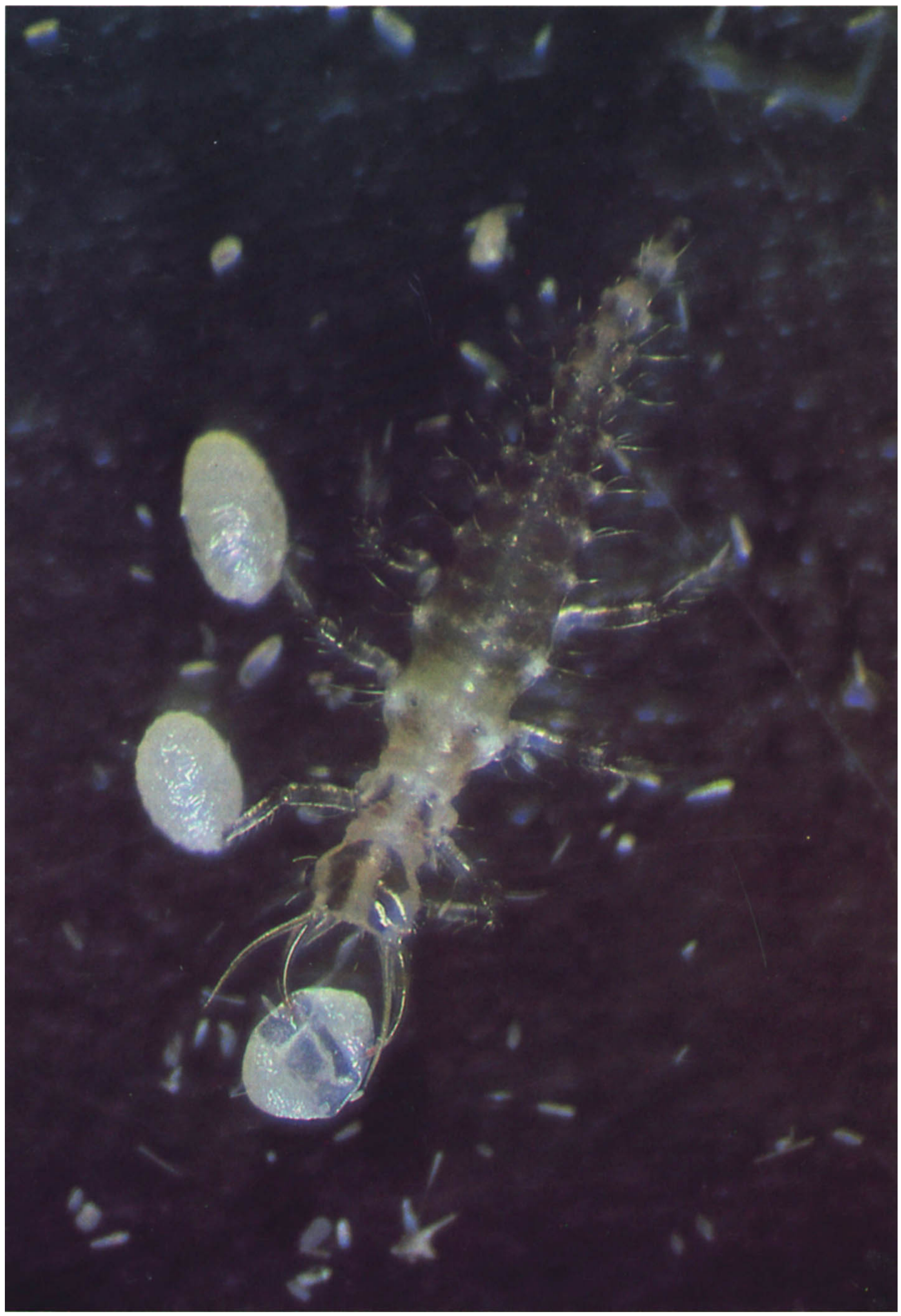
BULK RATE
POSTAGE & FEES PAID
USDA
PERMIT No. G269

Conclusions

Our studies showed that common green lacewing larvae collected from widely separated areas of California had different levels of tolerance to six pesticides. The most tolerant larvae, collected from Imperial County, were selected in the laboratory for resistance to carbaryl. Carbaryl-resistant larvae exhibited more than 80 percent survival after four generations of selection, in contrast to 3 percent survival of the unselected strain. This rapid selection response demonstrates the feasibility of selection for pesticide resistance in predatory insects. The mode of inheritance of carbaryl resistance in the selected strain of lacewing is not due to a single completely dominant or recessive gene. Thus, the level of resistance is lowered when the resistant strain interbreeds with susceptible lacewings.

Although field tests of the carbaryl-resistant strain have not been conducted, it is interesting to speculate where this strain might be used for biological control. The purpose of selecting a resistant strain is to help it survive carbaryl sprays used to control pests that are not its prey. Because carbaryl resistance is not dominant, interbreeding with susceptible lacewings must be avoided. This can be accomplished by using the selected strain where susceptible natives do not exist, such as in greenhouses. Where susceptible lacewings cannot be eliminated, the selected strain could be used only in inundative single-generation, short-term control. The economics and mechanics of rearing and releasing adequate numbers of resistant lacewings for either type of program must be studied before the strain could be commercially available.

Elizabeth E. Grafton-Cardwell is Postdoctoral Researcher, Department of Entomology, University of California, Davis, and Marjorie A. Hoy is Professor, Department of Entomology and Parasitology, UC Berkeley. *The authors thank Ciba-Geigy, Dupont, Shell, Stauffer, and Union Carbide Chemical companies for providing the pesticides used in screening and selection. Partial support for this project was provided by the California Department of Food and Agriculture, Environmental Monitoring and Pest Management and Biological Control Services Program; and by the Western Regional Research Project W-84, "Establish, improve, and evaluate biological control in pest management systems."*



The green lacewing, shown here feeding on the egg of a grain moth, is a valued predator of pests in many crops. Its effectiveness may be enhanced by improving its tolerance to pesticides.