Topical Toxicity of Imidacloprid, Fipronil, and Seven Conventional Insecticides to the Adult Convergent Lady Beetle (Coleoptera: Coccinellidae)[1]

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ABSTRACT The relative toxicities (comparing LD_{50} and LT_{50}s) of two synthetic pyrethroids (cypermethrin and fenvalerate), two organophosphorus insecticides (chlorpyrifos, diazinon), three carbamates (propoxur, carbaryl, bendiocarb), a phenylpyrazole representative (fipronil), and a heterocyclic nitromethylene representative (imidacloprid or NTN 33893) were assessed with topical bioassays in the laboratory against the convergent lady beetle, *Hippodamia convergens* Guérin Méneville. LD_{50} values decreased (i.e., toxicity increased) with an increased time after application of a specific insecticide. The differences between the LD_{50} values caused by various insecticides were significant. Among tested insecticides, cypermethrin and bendiocarb were the most toxic; fipronil was the least toxic. *H. convergens* responded differently to different insecticides within the same class. Beetles exhibited similar responses to both organophosphorothionates chlorpyrifos and diazinon 24 to 72 h after application. Of the carbamates, propoxur was 2.4 and 3.5 times less toxic than carbaryl and bendiocarb, respectively. Of the pyrethroids tested, cypermethrin was significantly more toxic than fenvalerate. At 800 ppm, cypermethrin and bendiocarb were the fastest in killing *H. convergens* among the tested insecticides. The ranking of insecticides in decreasing order of LT_{50} values was as follows: fipronil > diazinon > chlorpyrifos > propoxur > carbaryl > fenvalerate and imidacloprid.

KEY WORDS *Hippodamia convergens,* predator, topical toxicity

The convergent lady beetle, *Hippodamia convergens* Güerin-Méneville (Coleoptera: Coccinellidae), is an important cosmopolitan natural enemy species in many agricultural settings (Hagan 1962). The application of most insecticides produced a greater loss of beneficial arthropods than any other agricultural practice (van den Bosch 1966). From the standpoint of possible integrated chemical and biological control programs utilizing *H. convergens*, it is preferable to use insecticides that are less toxic to these predators. Most available

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organophosphorus, carbamate, and pyrethroid compounds used for insect control have an adverse effect on beneficial insects. There is, therefore, interest in the discovery of novel classes of insecticides with different biochemical targets that have toxic effect on the pest but not the beneficial insects. Toxicity of insecticides to *H. convergens* has been reported with laboratory research on peach (Moffitt et al. 1972), cotton (Burke 1959, Lingren and Ridgway 1967, Wilkinson et al. 1979), and pecan (Mizell and Schiffhauer 1990, Hurej and Dutcher 1994) and with field research (Dutcher 1983).

Toxicity of two new insecticide classes, heterocyclic nitromethylene and phenylpyrazole, are not known. These compounds have a mechanism of action that is dissimilar to currently-used insecticides (Moffat 1993). The toxicity of one nitromethylene, imidacloprid, to selected arthropod predators in the laboratory was recently described (Mizell and Sconyers 1992), but no quantitative data are available on the toxicity of either compounds to *H. convergens.* Imidacloprid, BAY NTN 33893, 1-[(Chloro-3-pyridinyl) methyl]-4,50dihydro-*N*-nitro-1*H*-imidazol-2-amine, was developed in the USA by Miles Corporation (Anonymous 1993). Imidacloprid is a broad-spectrum systemic insecticide that exhibits insecticidal activity against sucking insects such as Homoptera and some Coleoptera, Diptera, and Lepidoptera (Elbert et al. 1991). It has relatively low mammalian toxicity (Elbert et al. 1990). Imidacloprid interferes with the postsynaptic acetylcholine receptor in the insect nervous system and exhibits an agonist action at nicotinic acetylcholine receptors (Sattelle et al. 1989). Fipronil [(±)-5-amino-1-(2,6-dichloro-α,α,α-trifluoro-p-tolyl)-4-trifluoromethyl-sulfinylpyrazole-3-carbonitrile] is a phenylpyrazole insecticide discovered by Rhône-Poulenc Agro in 1987 at Ongar, UK (Colliot et al. 1992). Fipronil is being used against both piercing-sucking and chewing phytophagous insects. This compound is a nerve poison and blocks transmissions of signals by the inhibitory neurotransmitter gamma-amirobutyric acid (GABA) (Colliot et al. 1992, Cole et al. 1993). Both imidacloprid and fipronil can be delivered via soil, foliar, bait or seed treatment applications.

The following study was undertaken to determine the relative toxicity of materials in these two new classes of insecticides in comparison with two widely used synthetic pyrethroids (cypermethrin, fenvalerate), two organophosphorothionates (chlorpyrifos, diazinon), three carbomates (propoxur, carbaryl, bendiocarb), a phenylpyrazole representative (fipronil), and a chloronicotinyl representative (imidacloprid) on *H. convergens.*

Materials and Methods

Hippodamia convergens were obtained from A-1 Unique Insect Control (Citrus Height, CA). Beetles were collected as adults (sex ratio was approximately 1:1) from wild populations in Northern Sierra Foothills, CA. After the arrival of the adults in the laboratory, they were placed in plastic boxes (30 × 24 × 10 cm) with 25-ml cotton-stoppered water vials and fed daily on BugPro® (protein diet) obtained from Gardens Alive (Lawerenceburg, IN). Adults were maintained in the laboratory (24 ± 1°C and 55-58% RH) for 1 wk prior to test initiation.

Technical grade insecticides were used in all tests and provided by the following manufacturers: diazinon (88% A. I.; Ciba-Geigy, Greensboro, NC); chlorpyrifos (99%; DowElanco, Indianapolis, IN), bendiocarb (97.7%; NOR-AM Chemical

Company, Wilmington, DE), fenvalerate (80%; MGK, Minneapolis, MN), carbaryl and fipronil (96.7%; Rhone-Poulenc, Research Triangle Park, NC), cypermethrin (92.3%; Zeneca Ag Products, Richmond, CA), imidacloprid (95%; code name: NTN 33893), and propoxur (99%; Bayer, Kansas City, MO).

Insecticides were dissolved in acetone into a series of concentrations (10, 50, 100, 200, 300, and 800 ppm) and applied topically by microapplicator (Arnold Automatic Microapplicators, Burkhard Manufacturing) in 1 µl droplets to intercoxal regions of the meso- and metathoracic segments of beetles anesthetized by CO_2. A range of percentage mortality (0-100%) for all tested insecticides were determined on adults in preliminary tests. Each insecticide treatment was administered to 40 adults per concentration. Acetone-treated controls were included for each treatment. Immediately after treatment, adults were grouped (10 adults per replication) in 100 × 20-mm plastic Petri dishes in the top of which a 15-mm hole had been cut and covered with a nylon screen. Adults were supplied with moist cotton wicks to furnish water; no food was provided. Dishes containing treated adults were held at 23 ± 1°C, 55% RH, and 16:8 (L:D) photoperiod. Mortality was measured at 6, 24, 48, and 72h after application. Adults not able to walk normally when prodded were counted as dead.

Concentrations (µg/ml) were corrected for insect body weights to describe the dose (µg/g) each beetle received on treatment. Median lethal doses (LD_{50}s) for all insecticides were estimated by regression analysis of probit mortality to the adult stage versus log10 of dose. Separate LD_{50} analysis was conducted for each time period after application. The speed of action (LT_{50}) for each insecticide tested at 800 ppm (i.e., the highest concentration) was estimated by regression of the cumulative daily probit mortality versus log10 of time (h). SAS software (SAS Institute 1990) was used for both estimation procedures. Estimates of LD_{50}s and LT_{50}s were considered significantly different when the 95% fiducial limits did not overlap.

Results and Discussion

No mortality was detected in any of the acetone-treated controls. The LD_{50}s (± 95% confidence limits), chi-square, and slopes of the probit lines for all treatments are presented in Table 1. LD_{50}s decreased (i.e., toxicity increased) with an increased time after application of a specific insecticide. *H. convergens* varied widely in susceptibility to toxicants. At 72 h after application, LD_{50} values ranged from 0.007µg/g for cypermethrin to 2.6 µg/g for fipronil. The differences between LD_{50}s caused by various insecticides were significant (i.e., no overlap in 95% CL values). Cypermethrin and bendiocarb were the most toxic to *H. convergens* with LD_{50}s of 0.007 and 0.2 µg/g at 72 h after application, respectively. Fipronil was 2.6, 4.3, 3.7, 1.5, 13.0, 371.4, 5.2, and 6.5 times less toxic than chlorpyrifos, diazinon, carbaryl, propoxur, bendiocarb, cypermethrin, fenvalerate, and imidacloprid, respectively, 72 h after application. Similar toxicity levels were obtained at 24 and 48 h after application. Imidacloprid was approximately 2.6, 2.5, 5.0, and 6.5 times more toxic than fipronil at 6, 24, 48, and 72 h after application.

Our study indicated that *H. convergens* responded differently to different insecticides within the same class. Adults exhibited similar responses to both

Table 1. Topical toxicity of acetone solutions of several insecticides against *Hippodamia convergens* to determine the median lethal dose (LD) at selected time after application; the rates evaluated were 10, 50, 100, 200, 400, and 800 ppm.

Class	Insecticide	% A.I.	Time, h	n	X^{2}*	Slope ± SE	LD_{50}**	95% confidence limits of LD_{50}
Organophosphate	Chlorpyrifos	99.0	6	240	7.7	0.7 ± 0.2	16.5	6.6 - 190.1
			24	240	1.0	1.2 ± 0.2	2.4	1.8 - 3.6
			48	240	2.6	1.6 ± 0.2	1.2	0.9 - 1.6
			72	240	5.2	1.5 ± 0.2	1.0	0.7 - 1.3
	Diazinon	88.0	6	240	1.8	0.4 ± 0.2	456.9	27.9 - >4000
			24	240	4.9	1.0 ± 0.2	2.0	1.3 - 3.1
			48	240	10.6 a	1.0 ± 0.3	1.2	0.4 - 3.6
			72	200	15.3 a	1.4 ± 0.3	0.6	0.1 - 1.5
Carbamate	Carbaryl	99.5	6	240	10.0 a	1.8 ± 0.5	8.2	4.2 - 511.6
			24	240	6.2	1.9 ± 0.2	2.1	1.6 - 2.7
			48	240	18.4 a	1.8 ± 0.4	1.0	0.3 - 2.7
			72	240	19.6 a	1.6 ± 0.4	0.7	0.1 - 1.7
	Propoxur	99.0	6	240	0.5	3.4 ± 0.7	9.3	7.3 - 14.7
			24	240	2.2	1.4 ± 0.2	4.2	3.1 - 6.5
			48	240	3.3	1.3 ± 0.2	2.8	2.1 - 4.1
			72	240	12.8*	1.9 ± 0.4	1.7	0.9 - 3.6
	Bendiocarb	97.7	6	200	5.2	1.9 ± 0.2	0.3	0.2 - 0.4
			24	200	5.2	1.9 ± 0.2	0.3	0.2 - 0.4
			48	200	3.6	2.0 ± 0.3	0.3	0.2 - 0.3
			72	200	4.2	2.1 ± 0.3	0.2	0.2 - 0.3
Pyrethroid	Cypermethrin	92.3	6	200	6.5 a	1.8 ± 0.3	0.5	0.2 - 0.9
			24	200	3.4	1.5 ± 0.2	0.2	0.1 - 0.3
			48	200	2.0	1.2 ± 0.2	0.1	0.04 - 0.2
			72	200	3.2	0.8 ± 0.3	0.007	0.0 - 0.04

Table 1. Continued.

Class	Insecticide	% A.I.	Time, h	n	X^{2}*	Slope ± SE	LD_{50}**	95% confidence limits of LD_{50}
	Fenvalerate	80.0	6	240	8.8 a	1.2 ± 0.3	3.4	1.7 - 13.4
			24	240	14.0 a	1.4 ± 0.3	1.6	0.6 - 4.3
			48	240	16.9 a	1.8 ± 0.4	0.7	0.2 - 1.6
			72	240	7.7	2.0 ± 0.2	0.5	0.3 - 0.6
Nitromethylene	Imidacloprid	95.0	6	240	13.8 a	2.2 ± 0.6	4.1	2.2 - 15.2
			24	240	11.1 a	2.8 ± 0.5	1.8	1.0 - 2.8
			48	240	8.7 a	2.1 ± 0.3	0.7	0.4 - 1.1
			72	240	19.2	1.9 ± 0.5	0.4	0.1 - 1.0
Phenylpyrazole	Fipronil	96.7	6	240	9.3 a	2.4 ± 0.8	10.8	5.9 - >4000
			24	240	8.6 a	3.3 ± 0.7	4.5	3.1 - 7.6
			48	240	21.5 a	1.4 ± 0.5	3.5	1.2 - 472.7
			72	240	35.2 a	1.5 ± 0.6	2.6	†

* Pearson's Goodness-of-Fit (the measure of how well data fit the assumption of the model). Since the chi-square is mall ($P < 0.1$), fiducial limits were calculated using a t value of 1.96. The large chi-square ($P > 0.1$), value with an "a" is not caused by systematic departure from the model; a t value of ≥ 2.11 was used in computing fiducial limits.

** µg / g expressions for all dosage estimators.

† In some instances, the use of very large t value caused a non-realistic confidence interval to be calculated.

the organophosphorothionates, chlorpyrifos, and diazinon, 24 to 72 h after application. Of the carbamates, propoxur was 2.4 and 3.5 times less toxic than carbaryl and bendiocarb, respectively. Of the pyrethroids tested, cypermethrin was significantly more toxic than fenvalerate at each time after application. Similar results were obtained by Coats et al. (1979) who reported an LD_{50}(±95% CL) of 0.042 (0.017-0.066) for cypermethrin and 1.5 (0.70-2.8) µg/g for fenvalerate after 24 h of topical application.

Cypermethrin and bendiocarb, evaluated at 800 ppm, killed *H. convergens* faster than the other insecticides tested (Table 2). LT_{50}s could not be estimated because both compounds killed all the beetles prior to the first response time (i.e., at 6 h after application). The ranking of insecticides in decreasing order of LT_{50}s was as follows: fipronil > diazinon > chlorpyrifos and propoxur > carbaryl > imidacloprid and fenvalerate > bendiocarb and cypermethrin. Wilkinson et al. (1975) found carbaryl (50% WP) highly toxic to *H. convergens* in a contact bioassay with an LT_{50} of 8.8 h. In our test, an LT_{50} of 8.1 h was obtained.

Table 2. Topical toxicity of acetone solutions of several insecticides against *Hippodamia convergens* to determine the lethal time (LT_{50}) at 800 ppm concentration; mortality was recorded 6, 24, 48, and 72h after application.

Class	Insecticide	χ^{2*}	Slope ± SE	LT_{50}**	95% Confidence Limits of LT_{50}
Organophosphate	Chlorpyrifos	0.6	2.1 ± 0.3	10	6-13
	Diazinon	2.3	2.3 ± 0.3	10	7-13
Carbamate	Carbaryl	0.1	2.0 ± 0.3	8	5-11
	Propoxur	9.2a	1.4 ± 0.6	10	†
	Bendiocarb‡	160.0a	–	–	–
Pyrethroid	Cypermethrin‡	160.0a	–	–	–
	Fenvalerate	1.2	1.4 ± 0.3	4	1- 8
Nitromethylene	Imidacloprid	0.6	2.0 ± 0.4	5	2- 7
Phenylpyrazole	Fipronil	1.4	1.4 ± 0.3	13	7-18

* Pearson's Goodness-of-Fit. Since the chi-square is small ($P < 0.100$), fiducial limits were calculated using a t value of 1.96. The large chi-square ($P > 0.1$), values with an "a" is not caused by systematic departure from the model and a t value of 4.3 was used in computing confidence limits; n = 160 observations.

** Number of hours before death following exposure to the insecticide.

† In some instances, the use of a very large t value caused a non-realistic confidence interval to be calculated.

‡ There was a single response value; no lethal time estimate was done.

The relative toxicity of insecticides to *H. convergens* in this study is in agreement with results reported by various researchers. Bartlett (1963) and Moffitt et al. (1972) rated diazinon and carbaryl residues to be highly toxic to adults. Stern et al. (1960) reported carbaryl to be highly toxic to adults and larvae under field conditions. Hurej and Dutcher (1994) found carbaryl to cause 100% mortality of adults 48 h after feeding on insecticide-treated pecan aphids. Exposure of adult *H. convergens* to full registered rates of diazinon AG 500, chlorpyrifos 4E, fenvalerate 4E, cypermethrin 3E, and carbaryl 80S caused 91 to 100% adult mortality, 72 h after treatment (Mizell and Schiffhauer 1992).

Mizell and Sconyers (1992) reported imidacloprid to be toxic to most predators tested. Imidacloprid caused 78% mortality to adult *H. convergens* after exposure to residues at 127.4 ppm for 48-72 h. In our topical tests, 77.5 and 95.0% mortalities were recorded 72 h after application of 100 and 200 ppm, respectively. It has been suggested that seed treatment or application as a systemic soil drench may reduce the harmful effects of imidacloprid to some biocontrol agents (Mizell and Sconyers 1992).

This study suggested that each of the five different classes tested generated a different response on adult *H. convergens*. Characterization of the toxicity of each insecticide class to natural enemies can be useful in an integrated pest management program that seeks to conserve natural enemies. Although we have been able to determine the relative toxicities of the insecticides to *H. convergens*, it is important to emphasize that this was a laboratory study. As such, high rates of mortality may not be expected from similar doses under field conditions. Confirmation of our results from field experiments is needed.

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References Cited

Anonymous. 1993. BAY NTN 33893 Technical and safety information sheets. Miles Corporation Agricultural Chemicals Division. Kansas city, MO 64120. 11 pp.

Bartlett, B. R. 1963. The contact toxicity of some pesticide residues to hymenopterous parasites and coccinellid predators. J. Econ. Entomol. 56: 694-698.

Burke, H. R. 1959. Toxicity of several insecticides to two species of beneficial insects on cotton. J. Econ. Entomol. 52: 616-618.

Coats, S. R., J. R. Coats and C. R. Ellis. 1979. Selective toxicity of three synthetic pyrethroids to eight coccinellids, a eulophid parasitoid, and two pest chrysomelids. Environ. Entomol. 8: 720-722.

Cole, L. M., R. A. Nicholson and J. E. Casida. 1993. Action of phenylpyrazole insecticides at the GABA-gated chloride channel. Pestic. Biochem. Physiol. 46: 47-54.

Colliot, F., K. A. Kukorowski, D. W. Hawkins and D. A. Roberts. 1992. A new soil and foliar broad spectrum insecticide. Proc. Brighton Crop. Prot. Conf. Pests and Diseases, 1992, pp. 29-34.

Dutcher, J. D. 1983. Carbaryl and aphid resurgence in pecan orchards. J. Georgia Entomol. Soc. 18: 492-495.

Elbert, A., B. Becker, J. Hartwig and C. Erdelen. 1991. Imidacloprid- a new systemic insecticide. Pflanzenschutz-Nachrichten Bayer 44: 113-136.

Elbert, A., H. Overbeck, K. Iwaya and S. Tsuboi. 1990. Imidacloprid, a novel systemic nitromethylene analogue insecticide for crop protection. Proc. Brighton Crop. Prot. Conf., Pest and Diseases, 1990, pp. 21-28.

Hagen, K. S. 1962. Biology and ecology of predacious Coccinellidae. Annu. Rev. Entomol. 7: 289-326.

Hurej, M. and J. D. Dutcher. 1994. Indirect effect of insecticides on convergent lady beetle (Coleoptera: Coccinellidae) in pecan orchards. J. Econ. Entomol. 87: 1632-1635.

Lingren, P. D. and R. L. Ridgway. 1967. Toxicity of five insecticides to several insect predators. J. Econ. Entomol. 60: 1639-1641.

Mizell, R. F. and D. E. Schiffhauer. 1990. Effects of pesticides on pecan aphid predators *Chrysoperla rufilabris* (Neuroptera: Chrysopidae), *Hippodamia convergens, Cycloneda sanguinea* (L.), *Olla v-nigrum* (Coleoptera: Coccinellidae), and *Aphelinus perpallidus* (Hymenoptera: Encyrtidae). J. Econ. Entomol. 83: 1806-1812.

Mizell, R. F. and M. C. Sconyers. 1992. Toxicity of imidacloprid to selected arthropod predators in the laboratory. Florida Entomol. 75: 277-280.

Moffat, A. S. 1993. New chemicals seek to outwit insect pests. Science 261: 550-551.

Moffitt, H. R., E. W. Anthon and L. O. Smith. 1972. Toxicity of several commonly used orchard pesticides to adult *Hippodamia convergens.* Environ. Entomol. 1: 20-23.

SAS Institute. 1990. *SAS/STAT User's Guide,* Vol. 1 and 2. SAS Institute. Cary, NC.

Sattelle, D. B., S. D. Buckingham, K. A. Wafford, S. M. Sherby, N. M. Bakry, A. T. Eldefrawi, M. E. Eldefrawi and T. E. May. 1989. Actions of the insecticide 2 (nitromethylene) tetrahydro-1,3-thiazine on insect and vertebrate nicotinic acetylcholine receptors. Proc. R. Soc. Land. B. 237: 501-514.

van den Bosch, R. 1966. The role of parasites and predators on integrated pest control. Proc. FAO Symp. Integrated Pest Control. 2: 143-147.

Wilkinson, J. D., K. D. Biever and C. M. Ignoffo. 1975. Contact toxicity of some chemical and biological pesticides to several insect parasitoids and predators. Entomophaga 20: 113-120.

Wilkinson, J. D., K. D. Biever and C. M. Ignoffo. 1979. Synthetic pyrethroid and organophosphate insecticides against the parasitoid *Apanteles marginiventris* and the predators *Geocoris punctipes, Hippodamia convergens,* and *Podisus maculiventris.* J. Econ. Entomol. 72: 473-475.