J. Appl. Ent. 120, 369–373 (1996)

ISSN 0931-2048

Toxicological effects of selected insecticides on *Nephaspis oculatus* (Col., Coccinellidae), a predator of *Bemisia argentifolii* (Hom., Aleyrodidae)

T.-X. Liu and P. A. Stansly
Southwest Florida Research and Education Center, University of Florida, Immokalee, U.S.A.

Abstract: Toxicological effects of selected biorational insecticides were tested in the laboratory against all developmental stages of *Nephaspis oculatus* (Blatchley), a predator of whiteflies including silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring. Biorational insecticides included a mineral oil (Sunspray Ultra-Fine Oil), an insecticidal soap (M-Pede), and a mixture of sucrose and glucose esters (a detergent-like extract of *Nicotiana gossei* Domin). A pyrethroid (Capture 2EC) was used for comparison and water as control. *N. oculatus* has four distinct instars but the larvae were divided into two size categories (small and large) for bioassays. Pyrethroid was toxic to all developmental stages of *N. oculatus* and insecticide soap was toxic to larvae, but not to adults, eggs or pupae. Mineral oil was moderately toxic to eggs, but had no significant effect on other stages. Mixture of sucrose and glucose esters was innocuous to all life stages of the beetle. These results indicated that the use of pyrethroid would be incompatible with biological control of whitefly by *N. oculatus* and that considerable interference with predator activity could be expected from insecticidal soap, depending on the timing of application. On the other hand, mineral oil and especially *N. gossei* extract could be used against the whitefly with relative impunity to the coccinellids.

1 Introduction

Nephaspis oculatus (Blatchley), a native coccinellid of Central America, may have entered the West Indies and the United States through imported plants (GORDON, 1985). In addition to being widely established in Florida, *N. oculatus* has been recorded from Iowa, Louisiana, New Hampshire and Texas (GORDON, 1985). It has been observed on citrus to prey on Aleyrodidae species in Texas (MEYERDIRK et al., 1980) and Florida (MUMA et al., 1961; HOELMER et al., 1994). Under the name *N. amnicola* Wingo, it was successfully introduced into Hawaii from Central America (Honduras, Trinidad, and West Indies) in 1979 to control spiraling whitefly (*Aleurodicus dipersus* Russell) on vegetables (KUMASHIRO et al., 1983). Following the advent of silverleaf whitefly, *Bemisia argentifolii* Bellows & Perring as a pest of ornamental crops in Florida (HAMON and SALGUERO, 1987), the biological control potential of *N. oculatus* was recognized due to its capacity to consume large numbers of *B. argentifolii* eggs (HOELMER et al., 1994; LIU and STANSLY, unpublished data). A similar species, *Delphastus pusillus* (LeConte), has proven ability to control *B. argentifolii* on cotton and greenhouse-grown ornamental plants in California and Florida (HEINZ et al., 1994; HEINZ and PARRELLA 1994; HOELMER et al., 1993, 1994).

Integration of biological and chemical control requires knowledge of the effects of insecticides on natural enemies. HOELMER et al., (1990) found that adult *D. pusillus* were not affected by treatment with 0.3% azadirachtin for 2 weeks and that females feeding upon treated whiteflies for several days continued to lay eggs. We evaluated toxicological effects of a range of insecticide types to all developmental stages of *N. oculatus* with the objective of identifying materials useful for whitefly suppression but causing minimal interference to predation by this beetle.

2 Materials and methods

2.1 Insects and host plants

N. oculatus were maintained on an established greenhouse culture of *B. argentifolii* using a variety of plant hosts which included collard (*Brassica oleracea* L. var. *acephala*, 'Georgia LS'), hibiscus (*Hibiscus rosa-sinesis* L.), tomato (*Lycopersicum esculentum* Miller, 'Lana'), and sweet potato (*Impomoea batatas* L.). Plants grown in 15 cm plastic pots filled with Metro-Mix® 300 growing medium (Grace Sierra, Horticultural Products Company, Milpitas, CA), to which sufficient slow release fertilizer (N-P-K: 12-8-6) (Diamond R Fertilizer Company, Winter Garden, FL) was added as needed to maintain normal growth. Bioassays were conducted in a laboratory at 25±2 C, 14:10 (L:D) h photoperiod, and 65±5% RH. Voucher specimens of *N. oculatus* and *B. argentifolii* were deposited in the insect collection, Southwest Florida Research and Education Center, University of Florida, Immokalee, FL.

2.2 Insecticides

Three biorational insecticides were used: M-Pede (an insecticidal soap, 49% potassium salt of a naturally derived fatty

 0931-2048/96/2006-0369 $ 11.50/0

acid; Mycogen, San Diego, CA) at 0.5% (vol : vol), Sunspray Ultra-Fine Spray Oil (a mineral oil, 68 s viscosity, 212.8°C 50% distillation point, 94% unsulfonated residue; Safer, Newton, MA) at 0.20% (vol : vol of water), and an extract of *Nicotiana gossei* Domin (0.2 g [AI]/l of water), containing surfactant-like sucrose and glucose esters (BUTA et al., 1993; NEAL et al., 1994) obtained from the Phytochemistry Research Laboratory, USDA-ARS, Athens, GA, and prepared as described in LIU and STANSLY (1995a). A pyrethroid, capture (2EC, FMC, Middleport, NY) was used at 0.048 g (AI)/l for comparison and purified tap water (reverse osmosis, 7 ppm dissolved solids) was used as control. Diagnostic insecticide concentrations were chosen based on known effects to *B. argentifolii* (LIU and STANSLY, 1995a,b).

Adults, eggs, small larvae (first and second instars), large larvae (3rd and 4th instars), and pupae were used for bioassay. Instars were determined by head capsule measurements (LIU and STANSLY, 1996).

2.3 Bioassays

2.3.1 Adults

Collard leaf disks (8 cm diameter) with 200–300 whitefly eggs and nymphs were dipped in appropriate insecticide solutions for 5 s, or sprayed with 2 ml insecticide solution using the Potter spray tower (Burkard Manufacturing, Rickmansworth, Hertfordshire, England) operating at 0.70 kg/cm^2 (10 psi) pressure. After air-drying for 4 h, leaf disks were placed in plastic petri dishes (9 cm diameter) with lower leaf surfaces facing upwards. A 9 cm diameter piece of Whatman No. 1 filter paper was placed at the bottom of each petri dish, to which a few drops of water were applied periodically for moisture. Eight to 11 adult beetles (total 485) previously collected with a small soft paint brush (#0) from the greenhouse culture were introduced into each petri dish. The experiment had 10 replicates and was repeated three times. Mortality was evaluated twice, once after 24 h and then again after 48 h.

2.3.2 Eggs

Sweet potato leaves cultured in root-cubes as described in LIU and STANSLY (1995b) were individually confined to 0.9 l clear, plastic cup-cages, each provided with a 9 cm screened opening on top and a corked access hole (1.2 cm diameter) on the side. More than 100 whitefly adults were introduced into each cage and allowed to oviposit. Whitefly adults were removed with a vacuum at the end of 3 days and the number of eggs verified to exceed 500. Six *N. oculatus* adults were then introduced onto the leaf to oviposit for 3 days. *N. oculatus* eggs were individually marked using an india ink pen and coded before insecticide treatments. Leaves bearing whitefly eggs and nymphs and *N. oculatus* eggs were dipped in appropriate insecticide solutions for 5 s, were air-dried on paper towels for 2–3 h. The rooted leaves were recaged and examined daily under a stereoscopic microscope until all live eggs had hatched. Nineteen to 23 eggs were tested with each insecticide; in total, 131 eggs tested.

2.3.3 Larvae

Larvae were tested in two groups, small (1st and 2nd instars) and large (3rd and 4th instars). Larvae from the greenhouse culture were carefully placed with a brush on collard leaf disks (7–8 cm diameter) containing 200–300 whitefly eggs face up on a 9 cm piece of moistened filter paper in a plastic petri dish. Two ml of insecticide solution was sprayed onto the leaf disks using the Potter Spray Tower at 0.70 kg/cm^2 air pressure. Leaf disks were allowed to dry for 2 h, then held in petri dishes and examined at 24 and 72 h to determine mortality. Each experiment had 8 replicates (leaf disks) and was repeated twice with a total of 336 and 251 small and large larvae, respectively.

2.3.4 Pupae

Collard leaf disks (9 cm diameter) bearing pupae were collected directly from the greenhouse to avoid direct handling and treated with insecticides as described for larvae. The number of dead or live pupae was recorded daily until beetles emerged from all live pupae. The experiment had eight replicates (leaf disks) and was repeated twice with a total of 346 pupae.

2.4 Data analysis

Percentage mortalities (%) of adults, eggs and larvae were transformed to the arc sine square root [arc sine (percent mortality/100)$^{1/2}$] before analysis to stabilize error variance (GOMEZ and GOMEZ, 1984). Treatment effects were analyzed using analysis of variance (ANOVA), and means were separated using the least significant difference (LSD) test following a significant *F*-test (SAS INSTITUTE, 1988). Although all tests of significance were based on the transformed data, we report the untransformed percent mortality (%mean $\pm$ SE).

3 Results

3.1 Adults

Significant differences were found in mortality responses of *N. oculatus* adults to residues of insecticides applied as leaf dips ($F = 377.0$: df = 4, 9; P = 0.0001) or as sprays ($F = 407.7$; df = 4, 9; P = 0.0001, table 1). However, only Capture caused high levels of mortality (76.3–97.6% We observed beetles dying within 2–3 h after treatment with Capture, while others attempted to leave the treated leaf disk, apparently to avoid contact with the insecticide. Responses to other insecticides tested were not significantly different from water.

3.2 Eggs

Hatch rates were significantly different among insecticides ($F = 71.8$; df = 4, 9; P = 0.0001), with lowest hatch rate (4.4%) for eggs treated with Capture (table 2). Capture-killed eggs lost normal coloration (shiny yellowish-green) and turgor, collapsing within 24 h of treatment. Some appeared to remain viable, but never hatched. Eggs treated with Sunspray oil also hatched at a lower rate (69.6%) than the water control, but significant effects on hatch rate were not seen with other insecticides tested.

3.3 Larvae

Larvae were the most susceptible stage to insecticides, especially small larvae to Capture (tables 3, 4). Larvae arched the middle of the body upward with only the head and caudal end touching leaf surfaces, as if to avoid contact with residues of Capture. Dying larvae shrank, became yellowish or brownish, and collapsed.

Table 1. *Susceptibility of* N. oculatus *adults on collard leaf disks to insecticide residues applied at discriminating doses*

Insecticide	Rate	Mortality (%) ± SE				
		Leaf dip			Spray	
		N	24 h	48 h	N	24 h	48 h
Capture (g [AI]/l)	0.048	83	76.3 ± 7.8*a	97.8 ± 4.1*a	78	81.2 ± 6.9*a	97.6 ± 7.8*a
Sunspray oil (vol : vol)	0.20%	63	3.3 ± 6.2b	8.6 ± 9.7b	62	1.6 ± 4.4b	3.0 ± 5.5b
M-Pede (vol : vol)	0.50%	65	1.4 ± 3.9b	1.4 ± 3.9b	61	3.3 ± 6.2b	3.3 ± 6.2b
N. gossei extract (g [AI]/l)	0.2	73	5.8 ± 4.2b	5.3 ± 7.8b	64	7.3 ± 10.8b	7.3 ± 10.8b
Water (Control)		73	1.2 ± 3.5b	1.3 ± 3.5b	63	1.6 ± 4.4b	1.6 ± 4.4b

Means in the same column followed by different letters, and means between 24 and 48 h followed by an * are significantly different ($P < 0.05$, LSD [SAS INSTITUTE, 1988]).

Table 2. *Hatch rate of* N. oculatus *eggs on sweet potato leaves treated with insecticide dips*

Insecticide	Rate	N	Hatch rate (%)		Larvae died (%) after hatching
			4 d	7 d	
Capture (g [AI]/liter)	0.048	22	0.0c	4.4c	100.0a
Sunspray oil (vol : vol)	0.20%	23	43.5b	69.6b	17.6b
M-Pede (vol : vol)	0.50%	21	71.4a	85.6ab	36.8b
N. gossei extract (g [AI]/l)	0.2	28	71.4a	92.5a	20.0b
Water (Control)		19	73.6a	94.9a	11.1b

Means in the same column followed by different letters are significantly different ($P < 0.05$, LSD [SAS INSTITUTE, 1988]).

Table 3. *Mortality of small larvae (first and second instars) of* N. oculatus *on collard leaf disks 24 and 72 h after treatment with insecticide sprays*

Insecticide	Rate	N	Mortality (%) ± SE	
			24 h	72 h
Capture (g [AI]/l)	0.048	73	80.9 ± 0.4a	97.1 ± 2.9a
Sunspray oil (vol : vol)	0.20%	64	10.9 ± 2.9c	12.7 ± 5.0b
M-Pede (vol : vol)	0.50%	71	47.2 ± 2.8b	85.2 ± 14.8a
N. gossei extract (g [AI]/l)	0.2	64	8.9 ± 8.9c	13.7 ± 7.4b
Water (Control)		64	8.1 ± 2.6c	12.4 ± 1.9b

Means in the same column followed by different letters are significantly different ($P < 0.05$, LSD [SAS INSTITUTE, 1988]).

Table 4. *Mortality of large larvae (third and fourth instars) of* N. oculatus *on collard leaf disks 24 and 72 h after treatment with insecticide sprays*

Insecticide	Rate	N	Mortality (%) ± SE	
			24 h	72 h
Capture (g [AI]/l)	0.048	52	50.2 ± 9.8a	95.0 ± 5.0a
Sunspray oil (vol : vol)	0.20%	46	11.4 ± 8.6b	26.9 ± 13.1c
M-Pede (vol : vol)	0.50%	52	50.4 ± 4.1a	66.0 ± 2.3b
N. gossei extract (g [AI]/l)	0.2	49	0.0 ± 0.0b	7.6 ± 2.4c
Water (Control)		52	0.0 ± 0.0b	5.8 ± 3.3c

Means followed by different letters are significantly different ($P < 0.05$, LSD [SAS INSTITUTE, 1988]).

Table 5. Eclosion rates from pupae of N. oculatus *on collard leaf disks treated with insecticide sprays*

Insecticide	Rate	N	Eclosion (%)±SE
Capture (g [AI]/l)	0.048	56	34.9±6.9b
Sunspray oil (vol : vol)	0.20%	82	88.7±6.9a
M-Pede (vol : vol)	0.50%	61	88.7±4.2a
N. gossei extract (g [AI]/l)	0.2	83	81.5±3.5a
Water (Control)		64	92.3±3.4a

Means followed by different letters are significantly different ($P < 0.05$, LSD [SAS INSTITUTE, 1988]).

M-Pede was as toxic as Capture to small larvae after 72 h (table 3) and large larvae after 24 h (table 4). Responses to *N. gossei* extract and Sunspray oil were not significantly different from the water control.

3.4 Pupae

Significant effects ($F = 11.4$; df = 4, 14; $P = 0.0001$) of insecticides on pupae were observed as indicated by eclosion rates (table 5). However, only the response from capture was significantly different from the water control. Dead pupae lost their creamy coloration within 24 h, and later shrunk, curled and desiccated.

4 Discussion

Capture was clearly the most detrimental insecticide tested against *N. oculatus*. Similar effects could probably be expected from many broad-spectrum insecticides. Insecticidal soap was quite toxic to larvae and should only be used when most beetles are not in larval stages in systems where *N. oculatus* is an important predator. Mineral oil and *N. gossei* extract were relatively innocuous to *N. oculatus*, except for moderate toxicity of oil to eggs. Sunspray oil is quite toxic to adults of the parasitoid *Encarsia pergandiella* Howard (Hym., Aphelinidae) and should be used with caution if parasitism is to be an important component of whitefly mortality (LIU and STANSLY, unpubl.). We have yet to detect any deleterious effects to either *N. oculatus* or *E. pergandiella* of sucrose and glucose ester-containing extracts of *N. gossei*, in spite of the activity of this material to *B. argentifolii* (LIU and STANSLY, 1995a,b, unpubl.; BUTA et al., 1993; NEAL et al., 1994). This sugar ester insecticide appears to be 'biorational' in the sense of compatibility with biological control (STANSLY et al., 1995) and could become a useful component in integrated pest management of *B. argentifolii* and similar pests where hymenopterous parasitoids and coccinellids make significant contributions to pest mortality.

Acknowledgements

We would like to thank D. J. SCHUSTER and D. DEAN (University of Florida, Institute of Food and Agricultural Sciences, Bradenton), and K. A. HOELMER (USDA-APHIS, Brawley) for review of this manuscript, O. J. CHORTYK (Phytochemistry Research Laboratory, USDA-ARS, Athens) for providing *Nicotiana gossei* extract and J. CONNER, T. YOST, and Y. M. ZHANG for technical assistance. This study was funded by USDA Competitive Grant #93-34103-8433. Florida Agricultural Experiment Station Journal Series R-04396.

References

BUTA, J. G.; LUSBY, W. R.; NEAL, J. W., JR.; WATERS, R. M.; PITTARELLI, G. W., 1993; Sucrose esters from *Nicotiana gossei* active against the greenhouse whitefly, *Trialeurodes vaporariorum*. Phtyochem. **22,** 859–864.

CHERRY, R.; DOWELL, R. V., 1979: Predators of citrus blackfly (Hom.: Aleyrodidae). Entomophaga **24,** 385–391.

GOMEZ, K. A.; GOMEZ, A. A., 1984: Statistical procedures for agricultural research, 2nd edn. John Wiley & Sons, New York.

GORDON, R. D., 1985: The Coccinellidae (Coleoptera) of America north of Mexico. J. New York Entomol. Soc. **93,** 1–912.

HAMON A. B.: SALGUERO, V., 1987: *Bemisia tabaci*, sweetpotato whitefly, in Florida (Homoptera: Aleyrodidae: Aleyrodinae). Florida Dept. Agric. and consumer Serv., Division of Plant Industry, Entomol. Cir. No. **292,** 1–2.

HEINZ, K. M.; PARRELLA, M. P., 1994: Biological control of *Bemisia argentifolii* (Homoptera: Aleyrodidae) infesting *Euphorbia pulcherrima*: Evaluations of releases of *Encarsia luteola* (Hymenoptera: Aphelinidae) and *Delphastus pusillus* (Coleoptera: Coccinellidae). Environ. Entomol. **23,** 1346–1353.

HEINZ, K. M.; BRAZZLE, J. R.; PICKETT, C. H.; NATWICK, E. T.; NELSON, J. M.; PARRELLA, M. P., 1994: *Delphatus pusillus* as a potential biological control agent for sweetpotato (silverleaf) whitefly. Calif. Agric. **48**(2), 35–40.

HOELMER, K. A.; OSBOURNE, L. S.; YOKOMI, R. K., 1990: Effects of neem extracts on beneficial insects in greenhouse culture. In: Neem's potential in pest management programs. Proc. of the USDA Workshop, Ed. by LOCKE, J. C., LAWSON, R. H. USDA/ARS. ARS-**86,** 100–106.

—; —; —, 1993: Reproduction and feeding behaviour of *Delphastus pusillus* (Coleoptera: Coccinellidae), a predator of *Bemisia tabaci* (Homoptera: Aleyrodidae). J. Econ. Entomol. **86,** 322–329.

HOELMER, K. A.; OSBOURNE, L. S.; BENNETT, F. D.; YOKOMI, R. K., 1994: Biological control of sweetpotato whitefly in Florida, pp. 101–103. In: Pest Management in the Subtropics: Biological Control—a Florida Perspective. Ed. by ROSEN, D., BENNETT, R. D., CAPINERA, J. I. Intercept Press, Andover, UK.

KUMASHIRO, B. R.; LAI, P. Y.; FUNASAKI, G. Y. TERAMOTO, K. K., 1983: Efficacy of *Nephaspis amnicola* and *Encarsia haitiensis* in controlling *Aleurodicus dipersus* in Hawaii. Proc. Hawaii Entomol. Soc. **24,** 261–269.

LIU, T.-X.; STANSLY, P. A., 1995a: Toxicity and repellency of biorational inseciticides to *Bemisia argentifolii* on tomato plants. Entomol. Exp. Appl. **74,** 137–143.

—; —, 1995b: Toxicity of biorational insecticides to *Bemisia argentifolii* (Homoptera: Aleyrodidae) on tomato leaves. J. Econ. Entomol. **88,** 564–568.

—; —, 1996: Morphology of *Nephaspis oculatus* and *Delphastus pusillus* (Coleoptera: Coccinellidae), predators of *Bemisia argentifolii* (Homoptera: Aleyrodidae). Proc. Entomol. Soc. Wash. **98,** 292–300.

MEYERDIRK, D. E.; KREASKY, J. B.; HART, W. G., 1980: Whiteflies (Aleyrodidae) attacking citrus in southern Texas with notes on natural enemies. Can. Entomol. **112,** 1253–1258.

MUMA, M. H.; SELHIME, A. G.; DENMARK , H. A., 1961: An annotated list of predators and parasites associated with insects and mites on Florida citrus. University of Florida, Agric.. Exp. Sta., Tech. Bull. **634.**

NEAL, JR., J. N., ; BUTA, J. G.; PITTARELLI, G. W.; LUSBY, W. R.; BENTZ, L.-A., 1994: Novel sucrose esters from *Nicotiana gossei*: Effective biorational against selected horticultural pests. J. Econ. Entomol. **87,** 1600–1607.

SAS INSTITUTE, 1988: SAS/STAT user's guide. SAS Institute, Cary, NC.

STANSLY, P. A.; LIU, T.-X.; SHUSTER, D. J.; DEAN, D. E. 1995: Role of biorational insecticides in management of *Bemisia* species. Phytoparasitica **24,** 350–351.

Authors' address: Dr TONG-XIAN LIU (corresponding author), PHILIP A. STANSLY, University of Florida PO Drawer 5127, Immokalee, FL 33934, USA