

Impacts of Selected Insecticides on the Predatory Mite, *Neoseiulus californicus* (Acari: Phytoseiidae)¹

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Abstract Impacts of field rates of selected insecticides on the predatory mite, *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae), were evaluated in laboratory bioassays. In topical treatments with chlorantraniliprole, flonicamid, flubendiamide, indoxacarb, metaflumizone, pymetrozine, spinetoram, and thiacloprid, 86–92% of *N. californicus* adult females survived 168 h after exposure. Females exposed to the insecticides produced 86–98% as many eggs as did the females in the controls, and eclosion of eggs was not affected. Moreover, the percentage of eggs that hatched, and larval survival following direct exposure to the insecticides, were not reduced. Immature *N. californicus* survived on leaf discs with insecticidal residues, with 94–98% reaching adulthood. Based on these results, these insecticides evaluated in these laboratory bioassays are promising candidates for use in integrated pest management programs where *N. californicus* is a natural enemy.

Key Words *Neoseiulus californicus*, predatory mite, field rates, insecticides, integrated pest management

The twospotted spider mite, *Tetranychus urticae* Koch, is an important arthropod pest affecting various fruit trees and greenhouse crops in Korea (Cheon et al. 2007, Cho 2000). Outbreaks of this pest are often a consequence of repeated applications of nonselective pesticides, which enhance development of pesticide resistance in the tetranychid mite and decimate its natural enemies (Castagnoli et al. 2005, Kim and Yoo 2002). Using biological control agents represents a tool for slowing or avoiding development of pesticide resistance, especially in view of growing interest in environmentally friendly approaches to the control of pests (Cheon et al. 2007). Phytoseiid predatory mites play a central role in many agricultural systems as biological control agents of tetranychid phytophagous mites (Argolo et al. 2014, Liburd et al. 2007, Nguyen et al. 2015). *Neoseiulus californicus* (McGregor) is a phytoseiid mite known for its efficiency in the control of phytophagous mites and for its ability to feed on alternative foods (El Taj and Jung 2012, Rhodes and Liburd 2006, Sato et al. 2007). *Neoseiulus californicus* is produced commercially and sold in agricultural production areas worldwide (Yorulmaz Salman and Ay 2014). It is

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essential to assess methods and strategies for conserving naturally occurring predator populations, such as the natural population of *N. californicus* in Korea found on Jeju Island in a citrus orchard (Jung et al. 2006), and to develop guidelines for inoculative or augmentative releases in various cropping systems. The impacts of pesticides routinely used in cropping systems on these natural enemies is a logical first step in this process of developing effective integrated pest management (IPM) programs, as many may have deleterious effects on predatory mites (Sato et al. 2000, Yorulmaz Salman et al. 2015) and other natural enemies. Therefore, the objective of this study was to evaluate the impacts of eight insecticides, generally used to control aphids, whiteflies, thrips, and caterpillars in greenhouses and outdoor vegetable crops in Korea, on the survival and reproduction of *N. californicus* adult females and immatures.

Materials and Methods

Insecticides. All insecticides used in this study were commercial formulations and were selected on the basis of their current use for control of key greenhouse arthropod pests. The products were chlorantraniliprole (Altacor® 5WG, Farm Hannong, Seoul, Korea), flonicamid (Setis® 10WG, Farm Hannong, Seoul, Korea), flubendiamide (Anychung® 20SC, Hankook Samgong, Seoul, Korea), indoxacarb (Stewardgold® 5SC, Farm Hannong, Seoul, Korea), metaflumizone (Alverde® 20SC, Enbio, Seoul, Korea), pymetrozine (Chess® 25WP, Syngenta, Seoul, Korea), spinetoram (Delegate® 5WG, Farm Hannong, Seoul, Korea), and thiacloprid (Calyso® 10SC, Bayer, Seoul, Korea). The rates tested were recommended field rates in Korean crops.

Colony sources and experimental conditions. The *N. californicus* colony was established with mites obtained from Rural Development Administration of Korea in 2007 and has since been reared in the laboratory on kidney bean plants, *Phaseolus vulgaris* var. *humilis* Alefeld infested with *T. urticae*. The *N. californicus* colony was originally collected from a citrus orchard on Jeju Island in June 2006 (Jung et al., 2006). The *T. urticae* colony was collected from pear, *Pyrus* sp., trees and maintained on kidney bean plants in a greenhouse. All tests were conducted at 24–26°C at 50–60% relative humidity on an 18-h photophase. An individual test arena was a bean leaf disc (3-cm diameter) placed bottom-side up on moistened cotton in a plastic Petri dish (9-cm diameter) with a 1-cm diameter opening in the center of the top of the Petri dish. Each dish was placed in a plastic container (14-cm diameter, 5-cm height) containing water with a 1-cm diameter opening in its lid. A cotton wick was fitted through the center hole of the Petri dish and the plastic container for maintaining moisture in the cotton. Two holes (each 3-cm diameter) were drilled in the upper part of the side wall of the larger container to replenish the water as needed.

Insecticides were applied to runoff with a 1-L hand-operated sprayer (Komax Co., Seoul, Korea) held 23 cm away from the leaf disc. The leaf discs were bordered with a barrier of wet cotton (0.3–0.4 cm height) on the moistened cotton in each plastic Petri dish to prevent the escape of mites (Kim and Yoo 2002).

Impacts of insecticides on *N. californicus*. Topical toxicity of insecticides on the survival and reproduction of adult females of *N. californicus* and eclosion of

eggs deposited by treated females were evaluated in trials with 50 adult females (5 replicates with 10 mites per leaf disc). For each insecticide, *N. californicus* adult females were transferred from the source colony to leaf discs with the aid of a fine brush. Some twospotted spider mites were added to each disc to keep the adult female predators on the leaf discs. The leaf discs with adult female predators were sprayed with aqueous solution of each insecticide or distilled water (control) and then allowed to air-dry for 1 h. A surplus of all stages of *T. urticae* was added to each disc daily to insure an abundance of food. The survival of adult female predators was recorded at 1, 3, 5, and 7 d after treatment. Predatory mites were considered dead when they did not respond to touches by a fine brush. The eggs on each leaf disc were counted daily and transferred to a separate untreated disc to assess eclosion rates for each treatment.

The ovicidal effects of insecticides were evaluated with 50 eggs (10 eggs per leaf disc). Adult females of *N. californicus* were placed on leaf discs, allowed to deposit eggs for 24 h, and removed. The number of eggs was then adjusted to 10 per disc on each of 5 leaf discs for each insecticide tested. The leaf discs with predator eggs were sprayed with an aqueous solution of each insecticide or distilled water (control) and then allowed to air-dry for 1 h. Observations on the egg hatch were made daily. To assess the direct larvicidal toxicity of insecticides, adult female predators were transferred to each of five leaf discs and allowed to oviposit for 24 h. The *T. urticae* eggs served as the food source for *N. californicus*. After 24 h, adult female predators were removed and *N. californicus* eggs were allowed to hatch. At this time, any unhatched *N. californicus* eggs were removed, and the number of larvae was then adjusted to 10 per disc on each of 5 leaf discs. Each treatment was replicated five times. The leaf discs with *N. californicus* larvae were sprayed as described previously. Mortality was evaluated after 24 h.

To evaluate the effects of insecticidal residues on immature predators, the leaf discs were sprayed with an aqueous solution of each insecticide or distilled water (control) and then allowed to air-dry for 1 h before being placed in the Petri dishes. Fifty eggs of *N. californicus* (0–24 h old, 10 eggs per leaf disc) were transferred to the leaf discs that had been treated with each insecticide or distilled water. A surplus of all stages of *T. urticae* was placed on each disc when the predator eggs began to hatch. Immature survival to adulthood was observed daily and was assessed by counting the number of subsequent stages. Observations were discontinued when all predators reached adulthood.

Statistical analyses. Data were analyzed using analysis of variance (ANOVA) and Tukey's range test (SAS Institute 2002). Data in the form of percentages were transformed to arcsine values for ANOVA before analysis and were reconverted for reporting.

Results and Discussion

Topical toxicity of the insecticides tested on the survival of *N. californicus* adult females at different time intervals after application is shown in Table 1. Generally, the survival rates of *N. californicus* adult females in all treatments decreased over time after exposure. After 168 h, 86–92% of *N. californicus* adult females survived in treatments with the insecticides; these survival rates were not statistically different

Table 1. Survival of adult females of *Neoseiulus californicus* on bean leaf discs treated directly with different insecticides.

Insecticides Treated	Treatment Rate	% Survival (Mean \pm SEM) After*			
		24 h	72 h	120 h	168 h
Chlorantraniliprole	0.5 g/L	100.0 \pm 0 a	98.0 \pm 2.0 a	94.0 \pm 4.0 a	92.0 \pm 3.7 a
Flonicamid	0.5 g/L	100.0 \pm 0 a	98.0 \pm 2.0 a	94.0 \pm 2.5 a	90.0 \pm 3.2 a
Flubendia-mide	0.5 ml/L	100.0 \pm 0 a	96.0 \pm 2.5 a	92.0 \pm 3.7 a	92.0 \pm 3.7 a
Indoxacarb	1 ml/L	100.0 \pm 0 a	100.0 \pm 0 a	96.0 \pm 2.5 a	92.0 \pm 3.7 a
Metaflumi-zone	0.67 ml/L	100.0 \pm 0 a	96.0 \pm 2.5 a	92.0 \pm 4.9 a	90.0 \pm 4.5 a
Pymetrozine	0.33 g/L	100.0 \pm 0 a	98.0 \pm 2.0 a	96.0 \pm 2.5 a	92.0 \pm 2.0 a
Spinetoram	0.5 g/L	96.0 \pm 4.0 a	94.0 \pm 4.0 a	92.0 \pm 3.7 a	86.0 \pm 4.0 a
Thiacloprid	0.5 ml/L	98.0 \pm 2.0 a	94.0 \pm 4.0 a	90.0 \pm 4.5 a	86.0 \pm 4.0 a
Control	—	100.0 \pm 0 a	100.0 \pm 0 a	98.0 \pm 2.0 a	94.0 \pm 2.5 a

* Means in the same column followed by the same letter are not significantly different ($P = 0.05$, Tukey test). Mortality was transformed to arcsine value before analysis of variance. Means of untransformed data are reported.

Table 2. Reproduction of *Neoseiulus californicus* adult females on bean leaf discs treated with different insecticides and percentages of eclosion.

Insecticides Treated	Number of Eggs per Leaf Disc (Mean ± SEM)*	% Eclosion (Mean ± SEM)*
Chlorantraniliprole	94.4 ± 1.1 a	100.0 ± 0 a
Flonicamid	92.2 ± 1.7 ab	100.0 ± 0 a
Flubendiamide	92.6 ± 1.7 a	100.0 ± 0 a
Indoxacarb	95.0 ± 0.6 a	100.0 ± 0 a
Metaflumizone	83.4 ± 0.8 cd	100.0 ± 0 a
Pymetrozine	86.2 ± 1.8 bc	99.8 ± 0.2 a
Spinetoram	80.6 ± 1.5 cd	100.0 ± 0 a
Thiacloprid	79.8 ± 1.4 d	99.7 ± 0.3 a
Control	97.0 ± 0.9 a	100.0 ± 0 a

* Means in the same column followed by the same letter are not significantly different ($P=0.05$, Tukey test).

from the control. Based on the International Organization of Biological Control established categories (Hassan 1994), eight of the insecticides tested were in Category 1 (harmless, <30% mortality), indicating that these insecticides had little or no significant effects on the survival of *N. californicus* in these bioassays. Recently, chlorantraniliprole and flubendiamide have been reported to be nontoxic to immature and adult *Neoseiulus fallacis* (Garman) (Lefebvre et al. 2012). Colomer et al. (2011) documented that flonicamid had no impact on *Amblyseius swirskii* Athias-Henriot in the field. Pymetrozine has been reported to be innocuous to *N. californicus* females and eggs (Castagnoli et al. 2005). Bostanian and Akalach (2006) and Bostanian et al. (2010) documented that indoxacarb and thiacloprid were virtually innocuous to *N. fallacis* adults. Metaflumizone has also been reported to be harmless to *A. swirskii* adults (Gradish et al. 2011). Spinetoram was not toxic to *N. californicus* adult females and immatures in our study. In contrast, Lefebvre et al. (2012) reported that spinetoram was toxic to *N. fallacis* adults and larvae but not toxic to eggs of this predator. Beers and Schmidt (2014) also found that spinetoram caused high mortality of *Galendromus occidentalis* (Nesbitt) adults whereas larvae exhibited little mortality. Cuthbertson et al. (2012) and Lefebvre et al. (2012) referred to the response variability of phytoseiids to insecticide exposure and the need to evaluate insecticides on each species of acarine biocontrol agent.

Chlorantraniliprole, flonicamid, flubendiamide, and indoxacarb did not significantly affect the oviposition of *N. californicus* adult females (Table 2). Oviposition of the predators exposed to metaflumizone, pymetrozine, spinetoram, and thiacloprid was less than that of the control; however, treated females produced 82–89% as many eggs as did control females. These results suggest that the insecticides tested do not greatly influence the reproduction of surviving adult female predators. Moreover, high percentages of egg hatch (>98%) were recorded in both treated

Table 3. Effects of different insecticides on eggs and larvae of *Neoseiulus californicus*.

Insecticides Treated	% Hatchability (Mean \pm SEM)*	% Survival of Larvae (Mean \pm SEM)*
Chlorantraniliprole	100.0 \pm 0 a	92.0 \pm 2.0 a
Flonicamid	98.0 \pm 2.0 a	90.0 \pm 4.5 a
Flubendiamide	100.0 \pm 0 a	88.0 \pm 3.7 a
Indoxacarb	100.0 \pm 0 a	90.0 \pm 4.5 a
Metaflumizone	100.0 \pm 0 a	86.0 \pm 2.5 a
Pymetrozine	100.0 \pm 0 a	90.0 \pm 4.5 a
Spinetoram	100.0 \pm 0 a	84.0 \pm 2.5 a
Thiacloprid	98.0 \pm 2.0 a	88.0 \pm 3.7 a
Control	100.0 \pm 0 a	98.0 \pm 2.0 a

* Means in the same column followed by the same letter are not significantly different ($P = 0.05$, Tukey test).

and untreated females, and topical applications of the insecticides had low toxicity to *N. californicus* immatures with survival rates of 84–92% (Table 3). Placement of *N. californicus* immatures on treated leaf disc surfaces showed that the residues of the insecticides tested did not affect the survival of immatures (Table 4). The

Table 4. Effects of different insecticide residues on immature stages of *Neoseiulus californicus*.

Insecticides Treated	% Mortality (Mean \pm SEM) at*			% Survival to Adulthood*
	Egg Stage	Larval Stage	Nymphal Stage	
Chlorantraniliprole	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Flonicamid	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Flubendiamide	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Indoxacarb	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Metaflumizone	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Pymetrozine	0.0 \pm 0	0.0 \pm 0	6.0 \pm 4.0	94.0 \pm 4.0 a
Spinetoram	0.0 \pm 0	0.0 \pm 0	2.0 \pm 2.0	98.0 \pm 2.0 a
Thiacloprid	0.0 \pm 0	0.0 \pm 0	4.0 \pm 2.5	96.0 \pm 2.5 a
Control	0.0 \pm 0	0.0 \pm 0	0.0 \pm 0	100.0 \pm 0 a

* Means in the same column followed by the same letter are not significantly different ($P = 0.05$, Tukey test).

numbers of immature predators that reached adulthood in all insecticidal treatments were not significantly different from that of the control.

These laboratory studies indicate that the insecticides tested have an excellent margin of safety for *N. californicus* adult females and immatures and have excellent potential for use in an IPM program designed to utilize this predatory mite. Care should be exercised in translating laboratory tests into predictions of field performance (Lucas et al. 2004, Villanueva-Jimenez and Hoy 1998). Thus, field trials are needed to further evaluate the impact of these and other insecticides on *N. californicus* and the predatory mite's potential use in IPM programs.

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