

## Ecotoxicology

# Effects of Imidacloprid Applied Alone or in Combination With Organosilicone Surfactants on Biological Traits and Predatory Feeding of *Chrysoperla nipponensis* (Neuroptera: Chrysopidae)

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## Abstract

Organosilicone adjuvants are widely used to increase insecticide application on targeted surfaces. In this work, our aim was to investigate side effects of imidacloprid treatment, either applied alone or in combination with organosilicone compounds, against *Chrysoperla nipponensis*, an important predator of aphids. Four types of organosilicones were mixed with imidacloprid at different concentrations. The toxicity of the mixture to *C. nipponensis* was measured under laboratory conditions. The LC<sub>50</sub> and LC<sub>30</sub> of imidacloprid applied alone and in combination with 0.05% organosilicone were determined. Imidacloprid (LC<sub>30</sub>) applied alone or in combination with 0.05% organosilicone was used to treat second instar larvae of *C. nipponensis*; thereafter, its effects on the growth, development, longevity, reproduction, and predatory ability of *C. nipponensis* were evaluated. The results demonstrated that the organosilicone Silwet L-77 reduced the LC<sub>50</sub> and LC<sub>30</sub> of imidacloprid to 6.09 (95% CI: 2.31–9.42) and 10.95 mg/L (95% CI: 8.16–13.63), respectively, and enhanced imidacloprid toxicity to *C. nipponensis*, as reflected by the resulting extension of the growth and developmental period, reduction in female longevity, and inhibition of reproduction. When applied alone or in combination with an organosilicone, imidacloprid reduced the consumption of *Corcyra cephalonica* eggs by *C. nipponensis*. The functional response of *C. nipponensis* treated with imidacloprid alone or in combination with organosilicone was type II. Concomitantly, the attack rate was reduced and the handling time of prey increased.

**Key words:** imidacloprid, organosilicone surfactant, *Chrysoperla nipponensis*, synergistic effect

Imidacloprid is a systemic nitromethylene-analogue insecticide for crop protection, and is the most frequently used insecticide in apple orchards to control aphids every year (Hu and Prokopy 1998, Lowery et al. 2006). Due to the waxy nature of apple leaves, an appropriate adjuvant is often needed to help the spray solution to adhere to leaves. Compared to conventional surfactants, organosilicone adjuvants display low-equilibrium surface tension and surface energy, which is important for effective leaf wetting and spray-drop retention by leaf surfaces (Knoche 1994). Therefore, organosilicone surfactants are commonly supplemented with insecticides and herbicides to improve their physicochemical characteristics (Stevens 1993). This enhancement could greatly increase the efficacy of toxic active ingredients (Li et al. 2019). Furthermore, Silwet-type organosilicone

surfactants, i.e., polyalkyleneoxide-modified polydimethylsiloxane copolymers, have been widely applied to increase the efficacy of foliar applied agrochemicals, such as thiamethoxam and  $\lambda$ -cyhalothrin in soybean crop (Muraro et al. 2020) and imidacloprid in citrus orchards (Srinivasan et al. 2008). Imidacloprid is reportedly more toxic to *Phenacoccus solenopsis* (Hemiptera: Pseudococcidae) when combined with an organosilicone surfactant (Huang et al. 2014). Additionally, as an adjuvant, organosilicones also show insecticidal activity against several pests and nontarget species (Imai et al. 1995, Tipping et al. 2003).

The green lacewing, *Chrysoperla nipponensis* (Okamoto) (Neuroptera: Chrysopidae) is one of the most important, native, natural predators that are widely distributed in China, Japan, and

Korea (Brooks 1994). Larvae of this species prey on many species of aphids, thrips, leafhoppers, psyllids, caterpillars, cephalonica eggs, and mites that are agricultural and forest pests (Montoya-Alvarez et al. 2010, Memon et al. 2015). Given its high hunting and predatory ability, it is considered as a potential biological control agent in various field crops (Duelli 2001, Montoya-Alvarez et al. 2010, Memon et al. 2015). For example, in Korea, *C. nipponensis* was used to control *Pseudaulacaspis cockerelli* (Cooley) (Homoptera: Diaspididae) (Ham et al. 2013). Similarly, in China, *C. nipponensis* has been successfully mass-reared for application in biological control programs (Ji et al. 2011). Recent evidence supports observations that *C. nipponensis* larvae are highly resistant to the effects of pesticides in comparison with other predatory insects, which is advantageous for integrated pest management programs (Mochizuki and Mitsunaga 2004, Ham et al. 2019). Although imidacloprid toxicity to *C. nipponensis* has been reported (Ham et al. 2019), the synergistic effects of organosilicones on this reported toxicity are not yet fully understood.

In this study, we investigated the efficacy of organosilicone compounds on *C. nipponensis*, namely, Silwet 618, STIK2, ECO, and L-77, when combined with imidacloprid. Additionally, we studied the sublethal effects of imidacloprid, either alone or combined with organosilicones, on growth, development, longevity, reproduction, and functional response of *C. nipponensis*. These results provide a theoretical basis for scientifically-based application of organosilicones for the protection of *C. nipponensis*, whereby such application may contribute to an adequate and effective coordination between insect-pest chemicals and biological control measures.

## Materials and Methods

### Experimental Organisms

The green lacewing, *C. nipponensis*, was obtained from apple orchards in Wanrong County, Shanxi Province, China (35°18'N, 110°41'E) in August 2012. Adult *C. nipponensis* (10 individuals) were maintained in a breeding jar (height, 15 cm; diameter, 6 cm) containing brewer's yeast and sucrose at a ratio of 5:4 (weight) (Zhou et al. 1981). A flat layer of gauze was placed in the bottom of the jar for egg mass collection. To limit visual disturbances and to allow the insects to remain undisturbed, the breeding jar was placed in an environmental growth chamber at  $22 \pm 3^\circ\text{C}$ ,  $70 \pm 5\%$  RH, and a photoperiod of 16:8 (L:D) h. The gauze with egg masses was transferred to Petri dishes (1 cm in height, 9 cm in diameter). Newly hatched larvae were individually maintained in Petri dishes to avoid cannibalism and reared on eggs of rice moth *Corcyra cephalonica* (Stainton) (Lepidoptera: Pyralidae).

### Chemical Tests

The insecticide tested in this study was technical grade imidacloprid (95.8% purity) (Hebei Veyong Bio-Chemical Co., Ltd., Hebei, China), which is the most commonly used chemical to control aphids in apple orchards in China (Yang et al. 2013). Four different commercial organosilicone surfactants (Silwet 618, STIK2, ECO, and L-77) were purchased from Momentive Performance Materials Inc. (CT).

### Synergistic Action Bioassay

The toxicity of tested chemicals against *C. nipponensis* was investigated using an established method with the second instar larvae (Desneux et al. 2006). Since the tested organosilicones had no lethal or physical biocativities against *C. nipponensis* larvae according to

the preliminary experiments, the tests were therefore performed by assaying the lethal effect of varying concentrations of imidacloprid alone (IMI group) or in combination with organosilicones (Silwet 618, Silwet L-77, STIK2, or ECO) at 0.05%, which is the recommended concentration for field application. The insecticide solutions were stirred to obtain a uniform mixture before conducting the larvae bioassays. Individual larvae were anesthetized with  $\text{CO}_2$  for 10 s, and 1  $\mu\text{l}$  of the mixture was then smeared on the thoracic dorsum of the second instar larvae using a micro-applicator (Burkard Scientific, Uxbridge, UK), as previously described (Tan et al. 2012). A control group, which received only distilled water, was included in each series of tests. The mortality rate was assessed after 24 h of exposure. Larvae were considered dead when they were unresponsive to touching with a fine brush and could not move properly. Three replicates were performed for all assays, with  $\geq 30$  larvae tested per dose. Dose-mortality regression lines were determined according to the effective concentrations that caused 10–90% mortality of the insects in the test. The  $\text{LC}_{50}$  and  $\text{LC}_{30}$  values were estimated from linear regression.

### Growth, Longevity, and Reproduction of *C. nipponensis*

Newly molted second larval instar individuals of *C. nipponensis* were exposed to  $\text{LC}_{30}$  of imidacloprid or to a mixture of  $\text{LC}_{30}$  of imidacloprid and 0.05% organosilicone. After 24 h of exposure, all surviving larvae were transferred to a new Petri dish and supplied with sufficient *C. cephalonica* eggs for subsequent observations previously described methods (Tan et al. 2012). All Petri dishes were checked twice daily to record developmental time. The duration of the third larval stage of at least 20 larvae, pupal weight, pupal duration, and percentage of adults that emerged successfully from pupae were recorded for each treatment. To further assess female longevity, pre-oviposition period, and oviposition, at least 10 adult couples that emerged from the same treatment were kept separately in different oviposition plastic containers and were watched daily until death. Fecundity was evaluated as the total number of eggs laid per female.

### Assessment of Feeding Behavior of *C. nipponensis* on *C. cephalonica*

Second instar larvae of *C. nipponensis* were treated with imidacloprid at the  $\text{LC}_{30}$  concentration, either alone or combined with 0.05% organosilicone, to evaluate potential changes in the feeding behavior of *C. nipponensis* upon treatment exposure. Then, all surviving larvae were collected 24 h after initial exposure, followed by starvation for 12 h, and then were placed individually in Petri dishes containing *C. cephalonica* eggs at densities of 40, 80, 120, 160, 200, and 240 eggs per dish. After 24 h of predation, *C. nipponensis* larvae were removed, and the number of consumed prey ( $N_d$ ) was counted. Each experiment was performed three times.

### Statistical Analysis

A linear regression model for mortality curves was generated using logistic transformation of concentrations and probit transformation of mortalities and these models were considered valid when there was absence of significant deviation ( $P > 0.05$ ) (Finney 1971).  $\text{LC}_{50}$  and  $\text{LC}_{30}$  values were estimated using the resulting regression lines.  $\text{SR}_{50}$  was calculated by dividing the  $\text{LC}_{50}$  value of imidacloprid by that of the imidacloprid-organosilicone (Silwet 618, Silwet L-77, STIK2, or ECO) combination treatment. A synergistic effect was considered significant at  $P < 0.05$ . A one-way ANOVA followed by

Fisher's LSD test was used to compare differences in growth and reproduction (SAS Institute 1990). When required, data were transformed to satisfy assumptions of normality and homoscedasticity. All figures were generated using GraphPad Prism software (GraphPad, San Diego, CA).

The type of functional response (Type II or III) was determined using a cubic logistic regression analysis between the proportion of prey consumed ( $N_a$ ) in relation to the prey offered ( $N_0$ ) equation 1 (Juliano 2001).

$$\frac{N_a}{N_0} = \frac{\exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)}{1 + \exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)} \quad (1)$$

where  $N_a$  is the number of prey consumed,  $N_0$  is the number of prey offered, and  $P_0$ ,  $P_1$ ,  $P_2$ , and  $P_3$  are the intercepts of the linear, quadratic, and cubic parameters, respectively, which were estimated using the maximum likelihood method. A negative  $P_1$  indicated a type-II functional response, whereas a type-III functional response was otherwise presumed. Our data fit the type-II functional response; the attack rate ( $a$ ) and handling time per prey item ( $T_h$ ) were estimated using the random predator equation (2) (Rogers 1972).

$$N_a = N_0 \{1 - \exp[a(T_h N_a - T)]\} \quad (2)$$

where  $a$  is the instantaneous attack rate,  $T_h$  is the handling time per prey item, and  $T$  is the total time available for the predator (in this study,  $T = 24$  h). Parameters were estimated using the PROC NLIN procedure of SAS and compared based on 95% confidence limits (Burnham 1989).

## Results

### Toxicity of Imidacloprid Alone or in Combination With Organosilicone to *C. nipponensis*

The synergistic toxicity of organosilicone-imidacloprid combination treatments to second instar larvae of *C. nipponensis* is shown in Table 1. Imidacloprid by itself was the least potent treatment tested ( $LC_{50} = 18.59$  mg/L after 24 h of exposure), followed by imidacloprid in combination with Silwet 618 ( $LC_{50} = 8.13$  mg/L), STIK2 ( $LC_{50} = 6.74$  mg/L), ECO ( $LC_{50} = 6.40$  mg/L), and Silwet L-77 ( $LC_{50} = 6.09$  mg/L) after 24 h of exposure. These findings indicate that organosilicones significantly synergized imidacloprid ( $P < 0.001$ ). The lowest synergistic effect after 24 h of exposure was observed for Silwet 618, whereas the highest was observed for Silwet L-77. The  $LC_{50}$  value for imidacloprid was 10.95 mg/L (95% CI: 8.16–13.63 mg/L); therefore, this was the concentration of imidacloprid used thereafter.

### Effects of Imidacloprid Alone or in Combination With Organosilicone on the Development of *C. nipponensis*

Notably, imidacloprid alone extended development duration of the third larval instar ( $P < 0.05$ ) (Fig. 1A). Furthermore, organosilicones Silwet 618, ECO, and L-77 significantly inhibited this effect ( $P < 0.05$ ) (Fig. 1A). Compared to control pupae, pupal developmental duration was remarkably prolonged after exposure to imidacloprid alone ( $P < 0.05$ ) (Fig. 1B). In turn, an increased duration of pupal development was also observed when *C. nipponensis* was exposed to a combination of imidacloprid and Silwet 618, ECO, or Silwet L-77 during the second larval instar stage, compared to the use of imidacloprid alone ( $P < 0.05$ ) (Fig. 1B). Similarly, treatment with imidacloprid alone and in combination with Silwet 618, ECO, or Silwet L-77 resulted in a significant reduction in pupal weight; particularly, there was a significant difference between treatment with imidacloprid alone and treatment with a combination of imidacloprid with Silwet 618, STIK2, ECO, or Silwet L-77 ( $P < 0.05$ ) (Fig. 1C). The emergence rate of *C. nipponensis* pupae is shown in Fig. 1D. Imidacloprid did not affect the emergence rate of *C. nipponensis*, compared to the control group ( $P > 0.05$ ). In contrast, the imidacloprid–Silwet L-77 combination treatment reduced the emergence rate of *C. nipponensis* ( $P < 0.05$ ), whereas the emergence rate was not affected by using Silwet 618, STIK2, or ECO ( $P > 0.05$ ) (Fig. 1D).

### Effects of Imidacloprid Alone or in Combination with Organosilicone on Adult Longevity and Reproduction of *C. nipponensis* Adults

The female lifespan is shown in Fig. 2. The longevity of *C. nipponensis* female adults decreased significantly after exposure to imidacloprid at  $LC_{50}$  during the second larval instar stage ( $P < 0.05$ ). Furthermore, a significant difference in female longevity was observed between treatment with imidacloprid alone or in combination with Silwet L-77.

Additionally, the adult pre-oviposition period of *C. nipponensis* significantly increased after exposure to imidacloprid combined with Silwet L-77 during the second larval instar stage ( $P < 0.05$ ), compared with other treatments (Fig. 3A). A significantly reduced oviposition period was observed after treatment with imidacloprid alone or in combination with organosilicones Silwet 618, Silwet L-77, STIK2, or ECO, compared to the control group ( $P < 0.05$ , Fig. 3A). Additionally, a significant decrease in oviposition period was recorded for *C. nipponensis* when treated with the imidacloprid–Silwet L-77 combination treatment, compared with imidacloprid treatment ( $P < 0.05$ , Fig. 3A). And the total number of eggs laid per

**Table 1.** Summary of data on the toxicity ( $LC_{50}$ ) of imidacloprid in combination with organosilicone to second instar of *C. nipponensis*

Treatment	$LC_{50}^b$ (95% CI)	Regression	$R^2$	$SR_{50}^c$
IMI	18.59 (15.12–22.24)	$2.1029 + 2.2826x$	0.9692	1.00
IMI + 618 <sup>a</sup>	8.13 (3.80–11.60)	$3.4734 + 1.6778x$	0.9592	2.29 <sup>d</sup>
IMI + STIK2 <sup>a</sup>	6.74 (3.76–9.41)	$3.2482 + 2.1135x$	0.9766	2.76 <sup>d</sup>
IMI + ECO <sup>a</sup>	6.40 (2.70–9.87)	$3.8090 + 1.4770x$	0.9814	2.90 <sup>d</sup>
IMI + L-77 <sup>a</sup>	6.09 (2.31–9.42)	$3.8376 + 1.4811x$	0.9501	3.05 <sup>d</sup>

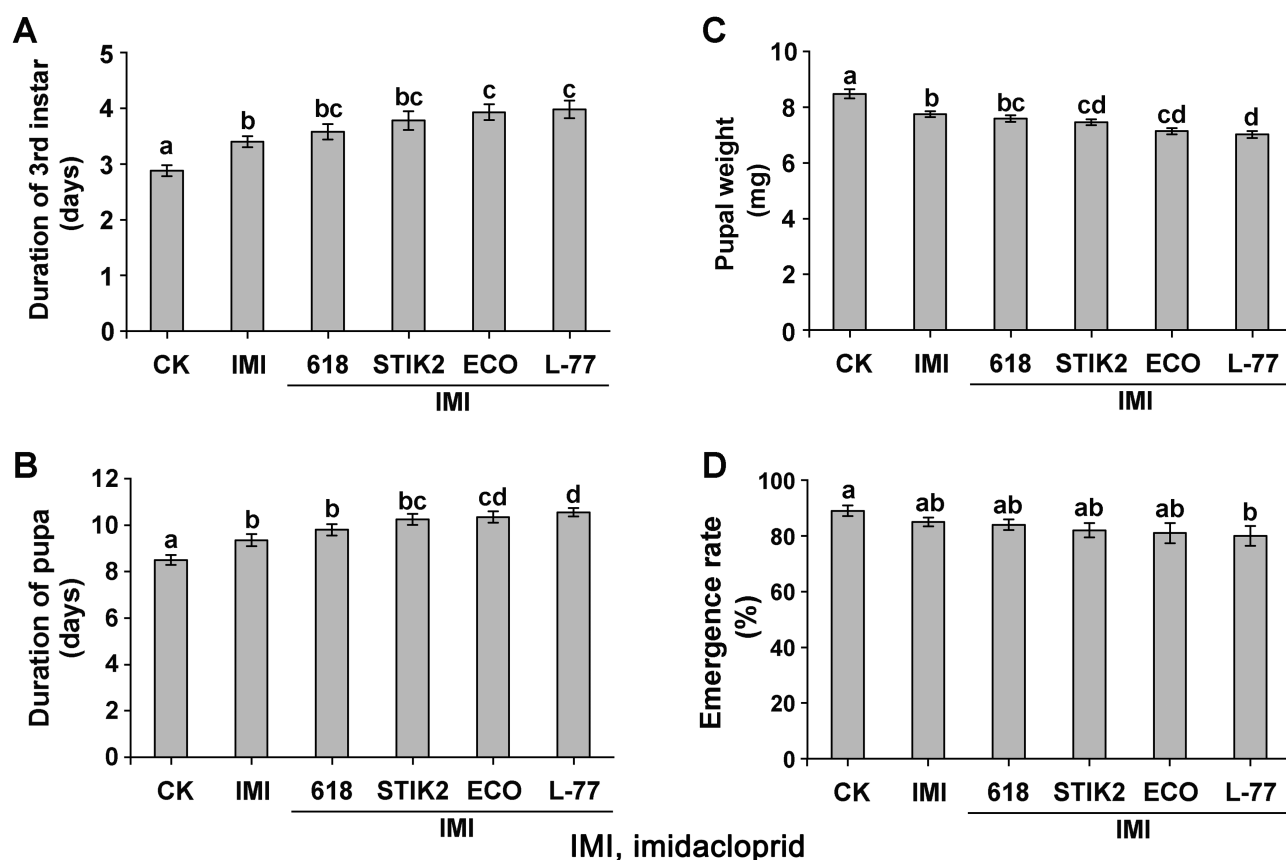
<sup>a</sup>Concentration of organosilicone (Silwet 618, STIK2, ECO, and L-77) was 0.05%.

<sup>b</sup>Concentration is expressed in mg/L, and the response was examined after 24 h of the initial exposure.

<sup>c</sup> $SR_{50}$  is calculated by dividing the  $LC_{50}$  of IMI by the  $LC_{50}$  of IMI + organosilicone, and significantly different from IMI group without organosilicone ( $SR_{50} = 1$ ) at  $P$ -value less than 0.05.

<sup>d</sup> $SR_{50}$  is significantly different from IMI without organosilicone ( $SR = 1$ ) at  $P < 0.05$ .

IMI, imidacloprid; CI, 95% confidence interval;  $SR_{50}$ , Synergistic ratio based on  $LC_{50}$ .



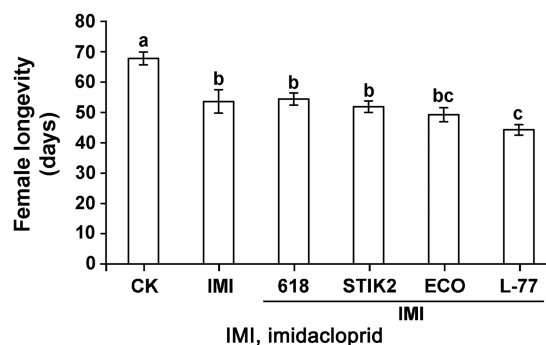
**Fig. 1.** Effects of imidacloprid alone or in combination with organosilicone surfactants on the development of *C. nipponensis*. (A) Mean duration of the third larval instar, (B) mean duration of the *C. nipponensis* pupal stage, (C) mean weight of *C. nipponensis* pupa, and (D) mean emergence proportion of *C. nipponensis*. *C. nipponensis* treated with LC<sub>50</sub> of imidacloprid alone or in combination with 0.05% organosilicone. Bars with different letters indicate significant differences according to one-way ANOVA, followed by Fisher's LSD test ( $P < 0.05$ ).  $n = 20$ .

female significantly decreased after treatment with the combination of imidacloprid and Silwet L-77 ( $P < 0.05$ , Fig. 3B).

#### Effects of Imidacloprid Alone or in Combination With Organosilicone on the Functional Response of *C. nipponensis* Larvae

The functional response of the second instar larvae of *C. nipponensis* to *C. cephalonica* eggs was determined. The results showed that the number of prey items consumed by *C. nipponensis* larvae decreased significantly after treatment with imidacloprid alone, or in combination with organosilicones Silwet 618, STIK2, ECO, or L-77 at all tested densities ( $P < 0.05$ ) (Table 2). The logistic regression for all prey consumed by second instar larvae of *C. nipponensis* showed a significant linear ( $P_1 < 0$ ) and a quadratic ( $P_2 > 0$ ) trend, indicating that the functional response of *C. nipponensis* treated with imidacloprid alone or in combination with organosilicones was a type-II functional response (Table 3).

Furthermore, the attack rate and prey handling time were estimated using the random-predator equation 2 to determine the predatory ability of the second instar larvae of *C. nipponensis* on *C. cephalonica* eggs. As shown in Table 4, imidacloprid reduced the attack rate significantly, especially when combined with organosilicones Silwet 618, STIK2, ECO, or L-77. Furthermore, *C. nipponensis* exposed to the imidacloprid–Silwet L-77 combination treatment showed the lowest attack rate. In contrast, there was no significant difference in prey handling time between the imidacloprid-treated and the control groups ( $P > 0.05$ ). However,

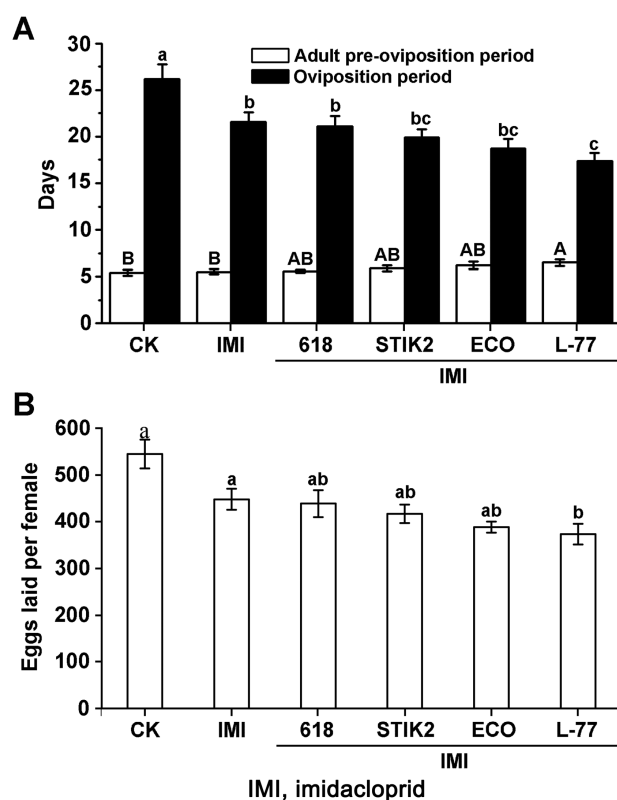


**Fig. 2.** Effects of imidacloprid alone or in combination with organosilicone surfactants on the longevity of *C. nipponensis* female adults. Bars with different letters indicate significant differences according to one-way ANOVA, followed by Fisher's LSD test ( $P < 0.05$ ).  $n = 10$ .

imidacloprid combined with an organosilicone (Silwet 618, STIK2, ECO, or L-77) significantly increased the *C. nipponensis* prey handling time ( $P < 0.05$ , Table 4).

#### Discussion

Green lacewing, *C. nipponensis*, larvae prey upon various insect pests including, caterpillars, mealy bugs, whiteflies, thrips, and aphids; thus, it is a valuable potential biocontrol agent with high efficacy in agricultural ecosystems and in nature. As a member of the



**Fig. 3.** Effects of imidacloprid alone or in combination with organosilicone surfactants on fecundity and fertility of *C. nipponensis* female adults. (A) Mean adult pre-oviposition period and oviposition period of *C. nipponensis* females, and (B) mean fecundity of *C. nipponensis* adults. Bars with different letters indicate significant differences according to one-way ANOVA, followed by Fisher's LSD test ( $P < 0.05$ ).  $n = 10$ .

neonicotinyl class of insecticides, imidacloprid has been shown to be effective against aphids (Mullins 1993). Organosilicones have been shown to improve the toxicity of imidacloprid to *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) (Srinivasan et al. 2008). However, whether synergistic effects occur in *C. nipponensis* remains unclear. In this study, we exposed a laboratory population of *C. nipponensis* to imidacloprid, either alone or in combination with several different organosilicone compounds. Our results showed that the organosilicones tested herein increased the toxicity of imidacloprid to *C. nipponensis* larvae and adversely affected their growth, development, reproduction, and functional response.

Organosilicones have been reported to show efficient insecticidal activity (Liu and Stansly 2000). Furthermore, organosilicones were confirmed to exhibit insecticidal potency against *C. nipponensis* (Saxena and Saxena 1989). Our data showed that a combination of 0.05% Silwet L-77 with imidacloprid elicited the highest toxicity to the second larval instar of *C. nipponensis* ( $LC_{50}$  of imidacloprid: 6.09 mg/L, 95% CI: 2.31–9.42), suggesting that the organosilicone increased imidacloprid-toxicity to *C. nipponensis*. This efficient insecticidal activity of organosilicones may be based on their excellent wettability on the leaf surface where insect pests are located (Liu and Stansly 2000). Additionally, as surfactants, organosilicones can spread over the aphid body surface and even the respiratory system because of their low surface tension (Imai et al. 1995). Furthermore, organosilicones induce imidacloprid deposition on the interfaces of the targets, which promotes insecticide translocation into the inner parts of targets, thereby increasing application efficiency (Cao et al. 2014).

**Table 2.** Functional response of second instar of *C. nipponensis* exposed to imidacloprid alone or in combination with organosilicone to different densities of *C. cephalonica* eggs

Treatment	Densities of <i>C. cephalonica</i> eggs				
	40	80	120	160	200
CK	30.57 ± 1.04a	56.03 ± 1.40a	73.40 ± 1.16a	91.87 ± 2.73a	108.63 ± 3.09a
IMI <sup>a</sup>	25.20 ± 0.20b	46.13 ± 0.26b	60.93 ± 0.42b	76.97 ± 0.94b	93.43 ± 1.89b
IMI + 618 <sup>b</sup>	23.40 ± 0.35c	41.23 ± 0.74c	53.33 ± 2.62c	67.77 ± 0.84c	81.23 ± 1.23c
IMI + STIK2 <sup>b</sup>	21.17 ± 0.65d	37.81 ± 0.56d	48.27 ± 2.15cd	65.67 ± 0.35c	75.83 ± 1.44c
IMI + ECO <sup>b</sup>	18.40 ± 0.17e	33.27 ± 0.15e	46.10 ± 1.24d	56.53 ± 1.25d	66.23 ± 3.08d
IMI + L-77 <sup>b</sup>	15.10 ± 0.12f	28.33 ± 1.37f	37.57 ± 1.93e	46.50 ± 0.71e	60.63 ± 2.01d
					105.07 ± 2.65a
					89.90 ± 0.06b
					79.50 ± 1.95c
					72.57 ± 0.91d
					64.13 ± 2.26e
					55.09 ± 1.04f

The data were presented as the mean ± standard deviation, and different lowercase letters indicate the difference between the groups at  $P < 0.05$  by one-way ANOVA, followed by Fisher's LSD test. IMI, imidacloprid.

<sup>a</sup>Concentration of imidacloprid was  $LC_{50}$ ; the concentration is expressed in mg/L, and the response was examined after the treatment described above.

<sup>b</sup>Concentration of organosilicone (Silwet 618, STIK-2, ECO, and L-77) was 0.05%.



**Table 3.** Results of logistic regression analysis of the proportion of *C. cephalonica* eggs eaten by second instar of *C. nipponensis* treated with imidacloprid alone or in combination with organosilicone

Treatment	Parameters	Estimate	SE	$\chi^2$	P-values
CK	Constant ( $P_0$ )	1.9361	0.1851	0.3006	0.0090
	Linear ( $P_1$ )	-0.0236	0.0053	0.3023	0.0465
	Quadratic ( $P_2$ )	0.0001	0.0000	0.4250	0.0844
	Cubic ( $P_3$ )	0.0000	0.0000	0.1682	0.0253
IMI <sup>a</sup>	Constant ( $P_0$ )	1.0313	0.1563	0.5171	0.0028
	Linear ( $P_1$ )	-0.0154	0.0045	0.4936	0.6432
	Quadratic ( $P_2$ )	0.0001	0.0000	0.4629	0.1315
	Cubic ( $P_3$ )	0.0000	0.0000	0.4094	0.1298
IMI + 618 <sup>b</sup>	Constant ( $P_0$ )	-0.6849	0.1812	0.0324	0.0634
	Linear ( $P_1$ )	-0.0134	0.0052	0.1635	0.1217
	Quadratic ( $P_2$ )	0.0001	0.0000	0.4560	0.1962
	Cubic ( $P_3$ )	0.0000	0.0000	0.4232	0.1865
IMI + STIK2 <sup>b</sup>	Constant ( $P_0$ )	0.6290	0.1442	0.0450	0.0488
	Linear ( $P_1$ )	-0.0153	0.0041	0.0757	0.0648
	Quadratic ( $P_2$ )	0.0001	0.0000	0.1203	0.1044
	Cubic ( $P_3$ )	0.0000	0.0000	0.1658	0.0134
IMI + ECO <sup>b</sup>	Constant ( $P_0$ )	-0.2467	0.1401	0.0239	0.2202
	Linear ( $P_1$ )	-0.0129	0.0040	0.0145	0.0840
	Quadratic ( $P_2$ )	0.0001	0.0000	0.0021	0.1321
	Cubic ( $P_3$ )	0.0000	0.0000	0.0022	0.1253
IMI + L-77 <sup>b</sup>	Constant ( $P_0$ )	-0.1485	0.1189	0.0416	0.3378
	Linear ( $P_1$ )	-0.0099	0.0034	0.0842	0.0993
	Quadratic ( $P_2$ )	0.0001	0.0000	0.1726	0.1603
	Cubic ( $P_3$ )	0.0000	0.0000	0.4260	0.1435

IMI, imidacloprid.

<sup>a</sup>Concentration of imidacloprid was LC<sub>30</sub>; the concentration is expressed in mg/L, and the response was examined after 24 h of exposure.

<sup>b</sup>Concentration of organosilicone (Silwet 618, STIK2, ECO, and L-77) was 0.05%.

**Table 4.** Attack rate (a) and handling time ( $T_h$ ) of *C. nipponensis* treated by LC<sub>30</sub> of imidacloprid alone or in combination with organosilicone

Treatment	a (h <sup>-1</sup> )	$T_h$ (h)
CK	0.8751 ± 0.0333a	0.0040 ± 0.0002a
IMI <sup>a</sup>	0.7113 ± 0.0086b	0.0044 ± 0.0017a
IMI + 618 <sup>b</sup>	0.6769 ± 0.0184b	0.0058 ± 0.0047b
IMI + STIK2 <sup>b</sup>	0.6043 ± 0.0243c	0.0059 ± 0.0005b
IMI + ECO <sup>b</sup>	0.5219 ± 0.0127d	0.0062 ± 0.0006b
IMI + L-77 <sup>b</sup>	0.4207 ± 0.0081e	0.0065 ± 0.0004b

The data were presented as the mean ± standard deviation, different lowercase letters indicate the difference between the groups at  $P < 0.05$  by one-way ANOVA, followed by Fisher's LSD test. IMI, imidacloprid.

<sup>a</sup>Concentration of imidacloprid was LC<sub>30</sub>; the concentration is expressed in mg/L, and the response was examined after 24 h of exposure.

<sup>b</sup>Concentration of organosilicone (Silwet 618, STIK2, ECO, and L-77) was 0.05%.

Mechanically, the addition of an organosilicone enhances the proportion of drops susceptible to spray drift, decreases the median droplet diameter, and reduces the surface tension (Costa et al. 2017). Additionally, organosilicone surfactants are known to increase the contact toxicity of pesticides by decreasing the equilibrium surface tension, thereby improving insecticide retention (Knoche 1994, Gaskin et al. 2000). Specifically, organosilicone Silwet L-77 reportedly exhibits aphicidal effects (Imai et al. 1995). However, the functional mechanism remains largely unknown. Additional studies are recommended to elucidate the synergistic mechanisms. We speculate that organosilicones increase the permeability of insecticides, thereby affecting insect metabolic processes.

Imidacloprid alone prolongs the duration of third larval instar and pupae, and decreases the pupal weight of *C. nipponensis* (Liu et al. 2016). Here, imidacloprid in combination with organosilicone

compounds significantly increased the developmental duration of the third instar larvae and pupae of *C. nipponensis*. Thus, a significant decrease in pupal weight and emergence rate was observed after exposure of the second larval instar stage to an imidacloprid–organosilicone combination treatment. Furthermore, organosilicones intensified the adverse effects of imidacloprid on the longevity and fertility of *C. nipponensis* female adults. The functional response was investigated using second instar larvae of *C. nipponensis*. Our results indicate that organosilicone compounds and imidacloprid synergistically inhibited the predatory behavior of *C. nipponensis* on *C. cephalonica* eggs. Therefore, the application of imidacloprid, especially together with silicone additives, should be avoided during the peak period of lacewing population growth. Here, we primarily studied the synergistic efficacy of imidacloprid and organosilicone compounds on the second larval instar of *C. nipponensis*. Given that insects at various life stages showed different sensitivities to

imidacloprid (Salehi et al. 2013), further studies are needed to determine whether the synergistic effects observed herein would occur at other stages of *C. nipponensis* development as well.

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