



Article Effects of Certain Pesticides on the Predatory Mite Typhlodromus ndibu Pritchard and Baker (Acari: Phytoseiidae)

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Abstract: *Typhlodromus ndibu* Pritchard and Baker (Acari: Phytoseiidae), collected from citrus orchards in the southern region of Vietnam, has been identified as a natural enemy of the citrus red mite *Panonychus citri* (McGregor) (Acari: Tetranychidae). It holds potential as a laboratory-reared predator for biological control purposes. However, the research on *T. ndibu* remains limited. This study focuses on investigating the effects of fenpyroximate, alpha-cypermethrin, pymetrozin, buprofezin, matrine, and azadirachtin on this predatory mite in laboratory conditions. Fenpyroximate was the most toxic substance against *T. ndibu*, affecting its fertility, egg-hatching rate, and population establishment ability. The mortality rate among adult female *T. ndibu* is (73.97 \pm 2.43%), and (89.09 \pm 0.00%) in the immature stages after 72 h of exposure to fenpyroximate. Matrine and azadirachtin—active ingredients of plant origin—were classified as slightly harmful to *T. ndibu* with mortality rates among adult females and the immature stages being less than 50%. The implications of the obtained results regarding the integration of biological and chemical control methods may facilitate the more effective development of integrated pest management (IPM) programs.

Keywords: azadirachtin; biological control; biopesticide; natural enemies; toxicology



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1. Introduction

Tetranychid mites are harmful phytophagous species found in greenhouses, fruit orchards, vegetable gardens, and ornamental plants, especially in warm and sunny weather conditions [1]. In citrus orchards, the citrus red mite (*Panonychus citri* (McGregor)) has been widely reported as a significant pest, causing damage to plants [2–4]. Chemical control is used as a common strategy worldwide for the management of mite infestations [5]; however, the long-term and frequent use of pesticides has led to resistance in the pests and negative effects on natural predators [6].

In order to combat this issue, alternative management strategies have been proposed, such as using predatory mites in combination with insecticides, rather than relying solely on chemical treatments in agricultural production areas [7]. Such an integrated approach aims to achieve effective pest control while minimizing the negative impacts on the environment and non-target organisms. Major pest species have been shown to be developing resistance to various pesticides [8,9], and the use of specialized and highly toxic pesticides often exacerbates the problem of pest resistance. New approaches in pest management, such as biological control using parasites, beneficial insects, or entomopathogenic fungi, are aimed at reducing/replacing the application of chemical insecticides [10,11]. Completely eliminating pesticide use immediately is often not feasible, as relying solely on biological control measures may not maintain pests below an economically damaging threshold [8,11]. Creating favorable conditions for the development of natural predators, reducing pesticide use, and minimizing the development of pest resistance are the primary objectives of IPM programs [12,13].

In recent years, studies have been conducted using biopesticides to mitigate adverse impacts on natural enemies and the environment [14]. Biopesticides are substances with

biological activity and effectiveness in controlling agricultural pests [15], particularly botanical pesticides that have gained widespread popularity [15,16] and have been successfully employed in pest management [14,16,17]. The neem tree (*Azadirachta indica*) is an evergreen tropical plant belonging to the Meliaceae family. It originates from East India and Myanmar and can be found throughout Southeast Asia and West Africa, with some recent plantings in the Caribbean and Central America, including Mexico [18]. The active ingredient in neem is azadirachtin, which acts as a repellent and destroyer of various species of many caterpillars, thrips, mites, and whiteflies [19]. Farmers are increasingly adopting neem as a natural insecticide to control pests and manage integrated pest issues [19].

Determining the impact of pesticides used in plant protection on beneficial organisms is crucial for the development of integrated pest management (IPM) strategies. Within an IPM program, it is essential to select plant protection products with the least impact on beneficial organisms. Therefore, studies investigating the effects of pesticides on beneficial organisms are of significant importance in the development of IPM programs and help to identify compounds that may be effectively used in IPM programs [20].

Phytoseiid mites—specifically, the small predatory mites of the Phytoseidae family—are important biological control agents for various pest insects in greenhouses, flower farms, fruit orchards, and citrus groves [3,21–31]. The Phytoseidae family, with around 2557 documented species, stands among the most researched and utilized clusters within the Acari class. [32,33].

Typhlodromus ndibu Pritchard and Baker was first described in the literature by Pritchard and Baker (1962) [34] from Congo and Rwanda. It has been recorded in Nigeria [35], Indonesia [36], sub-Saharan Africa [37], Kenya [38], and La Réunion [39]; however, biological studies of this species still largely remain scarce [39].

We collected *T. ndibu* from citrus cultivation areas in the southern part of Vietnam and found that they have good potential for controlling *P. citri* under laboratory conditions. They were collected, reared in a laboratory, and yielded promising results. Therefore, further research was conducted with the aim of using this predatory mite species within an integrated pest management (IPM) model for citrus tree fields in Vietnam. To successfully utilize *T. ndibu* in the IPM model, we conducted studies to assess the impact of insecticides that are commonly used in citrus groves on this predatory mite.

2. Materials and Methods

2.1. Predatory Mite

The population of *T. ndibu* was collected from citrus orchards in the southern region of Vietnam in March 2021. *P. citri*, the citrus red mite, was gathered from citrus trees and cultivated on kidney bean plants (*Phaseolus vulgaris*) in greenhouses at the Institute of Tropical Biology.

The predatory mites were cultured on bean leaf situated atop water-saturated cotton wool within plastic Petri dishes (90 × 15 mm) inside a plastic box (26 cm × 16 cm × 7 cm). A water-saturated sponge was placed in the container to maintain humidity. Water was added for the purpose of preserving the leaf's freshness and preventing the mites' escape. At intervals of 2 days, bean leaves hosting *P. citri* were introduced into the rearing facility. This cultivation was commenced three months before the onset of the experiments. The maintenance of the rearing unit took place within a growth chamber at 25 ± 1 °C and 75 ± 5% relative humidity, with a photoperiod of 16 h of light and 8 h of dark.

2.2. Pesticide

Six types of pesticides were used, including two biopesticides. These pesticides are known to be effective against the target pests in citrus orchards in the southern region of Vietnam (Table 1). Each pesticide was diluted to the recommended dose with 500 mL of distilled water. The control treatments were prepared using distilled water only.

Common Names	Active Ingredients	Concentration Rates/16 L *	Manufacturing Company
Ortus 5EC	Fenpyroximate 5%	19–27 mL	Central plant protection joint stock company 1 Le trong Tan, Ha Noi, Vietnam
Fastac 5EC	Alpha-cypermethrin 5%	10–20 mL	UPL Vietnam Co., Ltd. Ho chi Minh City, Vietnam.
Chess 50WG	Pymetrozin 500 g/kg	15 g	Syngenta, VN Ho Chi Minh City, Vietnam.
Applaud 10WP	Buprofezin 100 g/kg	40 g	Nihon Nohyaku C., Ltd. Tokyo, Japan
Kobisuper 1SL	Matrine 10g/L	19 mL	Nam Bac Co., Ltd. Ho Chi Minh City, Vietnam.
NeemNim	Azadirachtin 0.3%	16 mL	Ngan Anh Co., Ltd. Ho Chi Minh City, Vienam

Table 1. Common names, active ingredients, concentration rates, and manufacturing company of 6 pesticides used for the toxicity test on the predatory mite, *Typhlodromus ndibu*.

* The recommended dosage from the supplier is mixed with 16l of water.

2.3. Toxicity Assessment of Pesticides on Adult Females and Immature Stages of T. ndibu

We conducted toxicity tests of the pesticides on *T. ndibu* at the immature and adult female stages.

2.3.1. Toxicity Assessment of Pesticides on Adult Female T. ndibu

To evaluate the toxicity of the pesticides, we used the leaf-dip method described by Croft and Nelson (1972) [40], Overmeer (1985) [41], and Zhang and Sanderson (1990) [1], with some modifications. Clean grapefruit leaves, grown in a screened greenhouse at the Institute of Tropical Biology, were trimmed to a size of 3 cm \times 4 cm. These leaves were immersed in the diluted pesticide solutions (according to the rates recommended by the manufacturers) for 10 s, while gently stirring the solution with a glass rod to ensure complete absorption. Subsequently, the pesticide-treated leaves were briefly placed on a silk paper to remove excess solution and then left to air-dry naturally for one hour at room temperature (27–30 °C). After drying, the treated leaves were placed on a moist cotton pad in a Petri dish (90 \times 15 mm) within a plastic box (26 cm \times 16 cm \times 7 cm). Water was introduced to the plastic container to create a barrier that prevented the predatory mites from escaping from the treated environment (considered as a rearing unit).

First, we transferred 30 *P. citri* protonymphs to serve as prey for the predatory mites in each rearing unit containing leaves treated with pesticides. Then, we introduced one adult female *T. ndibu* to each rearing unit. The predators and prey were kept on the pesticide-treated leaf discs under fluorescent light with a 16:8 h light/dark photoperiod at a constant temperature of 25 ± 1 °C and 75 ± 5 % relative humidity.

Experimental setup: The experiment consisted of seven treatments (six types of pesticides and a control with distilled water). Each treatment had 10 rearing units per replicate, and the entire experiment was replicated three times.

Mortality rates were recorded at 24 h, 48 h, and 72 h after treatment. Predatory mites that did not respond when gently probed with a soft brush were considered to be dead. Any predatory mites that died due to drowning or escape were excluded from the data.

The efficacy of the pesticide was calculated using the Henderson–Tilton Formula [42]:

$$H(\%) = \left(1 - \frac{\text{Ta} \times \text{Cb}}{\text{Ca} \times \text{Tb}}\right) \times 100 \tag{1}$$

where Tb is the number of live individuals in the treatment group before surface exposure, Ta is the number of live individuals in the treatment group after surface exposure, Cb is the number of live individuals in the control group before surface exposure, and Ca is the number of live individuals in the control group after surface exposure.

Based on the toxicity evaluation, the toxicity of the pesticide to the predatory mites was categorized into four levels, according to the IOBC/WPRS four-tier scale (Hassan1994) [43], as follows:

Level 1: Non-toxic (mortality rate of predatory mites < 30%); Level 2: Slightly toxic (mortality rate of predatory mites from 30% to 79%); Level 3: Moderately toxic (mortality rate of predatory mites from 80% to 99%); Level 4: Highly toxic (mortality rate of predatory mites > 99%).

2.3.2. Toxicity Assessment of Pesticides on Immature Stages

We conducted similar experiments as described in the Toxicity Assessment of Pesticides on Adult Female *T. ndibu* section but, instead of using adult female predatory mites, we used mites in the immature stages.

2.4. Assessment of the Effects of Pesticides on the Biological Traits of Adult Female Predatory Mites

We prepared clean grapefruit leaves immersed in pesticide solutions and placed them in the rearing units (as described above). We then introduced adult female predatory mites onto the grapefruit leaves that had been treated with the pesticide solution for 24 h. After 24 h, we removed the surviving adult female predatory mites and individually placed them on clean grapefruit leaves, which were placed on a moist cotton pad in a Petri dish (90 × 15 mm) within a plastic box (26 cm × 16 cm × 7 cm) surrounded by water. We paired the female predatory mites that had been exposed to the pesticide with male predatory mites that had not been exposed to the pesticide for mating. The experiment was set up in a completely randomized design with seven treatments (six pesticide treatments and one control treatment with distilled water). Each treatment included 10 rearing units per replicate, and the entire experiment was replicated three times.

The parameters monitored during the experiment included the number of days of the pre-oviposition, oviposition, and post-oviposition periods, as well as the total number of eggs laid per female predatory mite, female adult periods, hatching rate, and sex ratio.

2.5. Statistical Analysis

The processing of the data was carried out using the SigmaPlot 11.0 software. The Tukey test was employed to compare different factors, and the results are presented as mean values. In all tests, *p* values less than or equal to 0.05 were considered significant.

For percentage values within the range of 0–100%, the conversion to $\operatorname{Arcsin}_{\sqrt{x}}$ was accomplished using the formula in Excel as follows: =ASIN(SQRT(A1/100)) × 180/PI() (A1: represents the percentage value to be converted), where 0% was replaced by (1/4n)%, and 100% was replaced by (100 - 1/4n)% (*n* is the number of samples within experiment) [44].

3. Results

3.1. Evaluation of Pesticide Efficacy on Predatory Mites

3.1.1. Evaluation of Pesticide Efficacy on Adult Female Typhlodromus ndibu

The data in Table 2 indicate statistically significant different mortality rates for adult female *T. ndibu* among the tested pesticide treatments.

Ortus 5EC (fenpyroximate 5%) and Fastac 5EC (alpha-cypermethrin 5%) presented mortality rates ranging from 61.46% to 73.97% in adult female *T. ndibu*. Chess 50WG (pymetrozin 500 g/kg) resulted in mortality rates fluctuating between 58% and 67% in adult female *T. ndibu*. Applaud 10WP (buprofezin 100 g/kg) led to mortality rates varying from 58% to 65% in adult female *T. ndibu*. On the other hand, Kobisuper 1SL (matrine 10 g/L) led to mortality rates varying from 28% to 37% in adult female *T. ndibu* and NeemNim (azadirachtin 0.3%) exhibited mortality rates below 30% (ranging from 21% to 29%) in adult female *T. ndibu*.

Regarding the toxicity of the tested pesticides, the data in Table 2 indicate that five of the pesticides—namely, Ortus 5EC, Fastac 5EC, Chess 50 WG, Applaud 10 WP, and Kobisuper 1 SL—were all slightly harmful to adult female *T. ndibu* (all at toxicity level 2).

	%				
Pesticide –	24 h *	48 h *	72 h *	IUDC Category -	
Ortus 5EC (Fenpyroximate 5%)	61.46 ± 1.09 a	67.06 ± 1.58 a	73.97 ± 2.43 a	2	
Fastac 5EC (Alpha cypermethrin 5%)	$58.75\pm0.82~\mathrm{ab}$	$62.64\pm1.17~\mathrm{ab}$	$67.86 \pm 1.54~\mathrm{ab}$	2	
Chess 50WG (Pymetrozin 500 g/kg)	$58.79\pm0.88~\mathrm{abc}$	$59.86\pm0.96\mathrm{b}$	$65.32\pm1.14~\mathrm{b}$	2	
Applaud 10WP (Buprofezin $100 \text{ g/kg})$	$54.62\pm1.29~\mathrm{b}$	$58.19\pm1.31~\mathrm{b}$	$60.57\pm1.19\mathrm{b}$	2	
Kobisuper 1SL (Matrine 10 g/L)	$28.81\pm1.74~\mathrm{c}$	$34.87 \pm 1.76 \text{ c}$	$37.85 \pm 2.14 \text{ c}$	2	
NeemNim (Azadirachtin 0.3%)	$21.61\pm2.42~d$	$27.37\pm2.37~d$	$29.04\pm2.29~d$	1	
Control	0.91 e	0.91 e	0.91 e	1	
df	5	5	5		
F	133.29	105.43	93.61		

Table 2. Mortality rates of adult female *Typhlodromus ndibu*.

¹ Means within a column followed by the same letter are not significantly different (p > 0.05). According to a Tukey test (24 h, 48 h, 72 h), F, df, and p values refer to one-way ANOVAs. * 24 h, 48 h, and 72 h after predatory mite treated with the pesticide solution. ² 1: Non-toxic (mortality rate of predatory mites <30%); 2: slightly toxic (mortality rate of predatory mites from 30% to 79%); 3: moderately toxic (mortality rate of predatory mites from 80% to 99%); 4: highly toxic (mortality rate of predatory mites >99%).

3.1.2. Evaluation of Pesticide Efficacy on Immature Predatory Mites

The data in Table 3 indicate statistically significant differences in the mortality rates of immature *T. ndibu* among the tested pesticide treatments. Ortus 5EC (fenpyroximate 5%) caused high mortality rates, ranging from 73% to 89%, in the immature *T. ndibu*. Notably, after just 2 days of treatment, the immature predatory mites had been completely killed with Ortus 5EC (fenpyroximate 5%). Fastac 5EC (alpha-cypermethrin 5%) caused mortality rates of 70% to 83%, and Chess 50WG (pymetrozin 500 g/kg) caused mortality rates of 58% to 71% in immature *T. ndibu*. Applaud 10WP (buprofezin 100 g/kg) led to mortality rates ranging from 55% to 67% in immature *T. ndibu*.

Table 3. Mortality rates of immature stages of *Typhlodromus ndibu*.

D	%				
Pesticide –	24 h *	48 h *	72 h *	IOBC Category -	
Ortus 5EC (Fenpyroximate 5%)	73.44 ± 0.97 a	89.09 ± 0.00 a	89.09 ± 0.00 a	3	
Fastac 5EC (Alpha cypermethrin 5%)	$70.83 \pm 2.03~\mathrm{ab}$	$74.83 \pm 2.75 \mathrm{~ab}$	$83.57\pm2.33~\mathrm{ab}$	3	
Chess 50WG (Pymetrozin 500 g/kg)	$58.79 \pm 1.10~\mathrm{abc}$	$63.87\pm1.26~\mathrm{abc}$	$71.91 \pm 1.29~\mathrm{abc}$	2	
Applaud 10WP (Buprofezin 100 g/kg)	55.43 ± 1.13 bcd	$62.67\pm1.23\mathrm{bc}$	67.98 ± 1.19 bcd	2	
Kobisuper 1SL (Matrine 10 g/L)	40.23 ± 3.33 cd	$45.48 \pm 3.32 \text{ cd}$	49.96 ± 3.26 cd	2	
NeemNim (Azadirachtin 0.3%)	$26.71\pm1.67~d$	$30.73\pm1.96~d$	$33.14\pm1.93~d$	2	
Control	0.91 e	0.91 e	0.91 e	1	
df	5	5	5		
Н	53.10	53.59	54.25		

¹ Means within a column followed by the same letter are not significantly different (p > 0.05), according to a Rank test; H, df, and p values refer to one-way ANOVAs. * 24 h, 48 h, and 72 h after predatory mite treated with the pesticide solution. ² 1: Non-toxic (mortality rate of predatory mites <30%); 2: slightly toxic (mortality rate of predatory mites from 30% to 79%); 3: moderately toxic (mortality rate of predatory mites from 80% to 99%); 4: highly toxic (mortality rate of predatory mites >99%).

Kobisuper 1SL caused mortality rates ranging from 40% to 49% in immature stages of *T. ndibu* after 24 to 72 h of exposure, indicating level 2 toxicity for the immature predatory mites. The biopesticide NeemNim was found to be slightly harmful to immature *T. ndibu*, causing mortality rates ranging from 26% to 33%. Overall, the results demonstrate that the different pesticide treatments had varying effects on the mortality rates of immature *T. ndibu*, with some pesticides showing high toxicity while others were less toxic to the immature predatory mites.

Regarding the toxicity of the tested pesticides in immature *T. ndibu*, the results in Table 3 demonstrate that Ortus 5EC (Fenpyroximate 5%) is harmful to immature *T. ndibu*, classified as toxicity level 3.

Analyzing both Tables 2 and 3, we can observe that Applaud 10WP and Chess 50WG had relatively similar toxicity in both adult and immature *T. ndibu*. These pesticides presented toxicity level 2 (mortality rates from 60% to 65% after 72 h) in adult *T. ndibu* (Table 2) and similar toxicity in immature *T. ndibu*, with mortality rates ranging from 67% to 71% after 72 h (Table 3). The two biopesticides—Kobisuper 1SL and NeemNim—presented low toxicity at both stages, based on the classes of toxicity adopted by the IOBC, NeemNim is rated as harmless (class 1: both percentages were <30%) to female adult *T. ndibu*.

3.2. Evaluation of The Impact of Plant Protection Agents on the Biological Characteristics of Adult Predatory Mites

Based on Table 4, it can be observed that the number of eggs laid per day by female *T. ndibu* presented no statistically significant differences among the Ortus 5EC, Fastac 5EC, and Chess 50WG, but there were significant differences compared to the control treatment. Meanwhile, the daily egg-laying rate did not present statistically significant differences between the two biopesticide treatments and the control treatment.

Table 4. Mean (\pm SE) of pre-oviposition, oviposition, and post-oviposition periods (days), fecundity (eggs per female), oviposition rate (eggs/female/day), and female adult periods (days) of *T. ndibu* after contact with residues of pesticides.

	Pre- Oviposition Period (Days) *	Oviposition Period (Days) *	Post- Oviposition Period (Days) *	Female Adult Periods (Days) *	Oviposition Rate (Eggs/ Female/Day) *	Total Number of Eggs (Eggs/Female) *	Hatching Egg Proportion of the Progeny (%) *	Female Proportion of the Progeny (%) *
NT1	$4.53\pm0.18~\mathrm{a}$	$9.03\pm0.35b$	$3.00\pm0.15~d$	$16.57 \pm 0.35 \text{ e}$	$1.09\pm0.02~\mathrm{a}$	$10.03\pm0.49~\mathrm{b}$	$53.27 \pm 1.704 \text{ b}$	$46.42\pm0.53~\mathrm{c}$
NT2	3.70 ± 0.23 b	$9.89\pm0.45\mathrm{b}$	$3.13\pm0.13~\mathrm{cd}$	$16.73 \pm 0.41 \text{ e}$	$1.18\pm0.03~\mathrm{a}$	$11.90\pm0.79\mathrm{b}$	$59.04\pm0.68\mathrm{b}$	$51.34\pm0.50~{ m bc}$
NT3	$2.67\pm0.18~{\rm c}$	$10.2\pm0.31\mathrm{b}$	$4.37\pm0.24~\mathrm{b}$	17.23 ± 0.27 de	$1.19\pm0.04~\mathrm{a}$	$12.30\pm0.45\mathrm{b}$	$64.89\pm0.63\mathrm{b}$	55.01 ± 0.63 bc
NT4	$2.60\pm0.11~{ m c}$	$12.03\pm0.23~\mathrm{ab}$	$4.19\pm0.21~\mathrm{b}$	$18.83\pm0.39~\mathrm{cd}$	$1.19\pm0.10~\mathrm{ab}$	$14.37\pm0.47~\mathrm{ab}$	$70.33\pm0.65~\mathrm{ab}$	$58.52\pm1.15~\mathrm{ab}$
NT5	$2.20\pm0.17~{ m c}$	$13.70 \pm 0,28$ a	$4.57\pm0.29~\mathrm{b}$	$20.47\pm0.35\mathrm{bc}$	1.31 ± 0.03 a	$18.03\pm0.41~\mathrm{a}$	$74.93 \pm 1.01 \text{ a}$	63.43 ± 1.06 a
NT6	$1.97\pm0.19~{ m c}$	14.43 ± 0.41 a	6.30 ± 0.29 a	22.67 ± 0.58 a	$1.32\pm0.07~\mathrm{a}$	19.30 ± 1.10 a	78.35 ± 0.85 a	64.68 ± 0.99 a
NT7	$2.10\pm0.14~\mathrm{c}$	15.13 ± 0.43 a	$6.00\pm0.32~\mathrm{a}$	$23.23\pm0.27~\mathrm{a}$	$1.35\pm0.05~\mathrm{a}$	$20.63\pm0.94~\mathrm{a}$	81.47 ± 0.71 a	63.78 ± 1.78 a
F	30.38	57.01	27.61	51.68	22.81	54.53	64.52	55.82
df	6	6	6	6	6	6	6	6
р	< 0.001	< 0.001	< 0.001	< 0.001	0.614	0.248	0.044	0.541

* Means within a column followed by the same letter are not significantly different (p > 0.05). According to a Tukey test (preoviposition period, oviposition period, post-oviposition period, oviposition rate, Female adult periods (days)) and according to a Rank test (total number of eggs, hatching egg proportion of the progeny, female proportion of the progeny), F-, df- and p values refer to one-way ANOVAs. NT1: Ortus 5EC (Fenpyroximate 5%), NT2: Fastac 5EC (Alpha cypermethrin 5%), NT3: Chess 50WG (Pymetrozin 500 g/kg), NT4: Applaud 10WP (Buprofezin 100 g/kg), NT5: Kobisuper 1SL (Matrine 10 g/L), NT6: NeemNim (Azadirachtin 0.3%), NT7: control (distilled water).

The oviposition period varied with the different pesticide treatments, resulting in different total egg counts per female in each treatment. The shortest time taken to lay eggs was observed in the adult female *T. ndibu* affected by the Ortus 5EC (fenpyroximate 5%) pesticide treatment (9.03 ± 0.35 days). The time taken to lay eggs showed statistically significant differences between the pesticide treatments (Ortus 5EC, Fastac 5EC, and Chess 50WG) and biopesticide treatments (Kobisuper 1SL and NeemNim). However, there was no statistically significant difference in the time taken to lay eggs between the NeemNim pesticide treatment and the control treatment.

The total number of eggs per female when affected by the Ortus 5EC (fenpyroximate 5%) pesticide was the lowest (10.03 \pm 0.49 eggs), and reached half of the number when compared to females in the control treatment.

Both the hatching rate and the female proportion in the pesticide-treated groups (Ortus 5EC, Fastac 5EC, and Chess 50WG) showed statistically significant differences compared to the control treatment. The hatching rate was low in the three pesticide-treated groups affected by Ortus 5EC (Fenpyroximate 5%), Fastac 5EC (alpha-cypermethrin 5%), and Chess 50WG (pymetrozin 500 g/kg), all below 65%.

Thus, Ortus 5EC (fenpyroximate 5%), Fastac 5EC (alpha-cypermethrin 5%), and Chess 50WG (pymetrozin 500 g/kg) were all found to have a negative impact on the reproductive capability of *T. ndibu*. This will lead to an effect on establishment of the *T. ndibu* population,

with the strongest impact being that of Ortus 5EC (fenpyroximate 5%), followed by Fastac 5EC (alpha-cypermethrin 5%) and Chess 50WG (pymetrozin 500 g/kg), respectively.

4. Discussion

Neem is rich in a variety of bioactive compounds. The most essential active ingredient that comprises a significant portion of neem is azadirachtin [45]. Azadirachtin within neem is considered the most biologically significant compound, as it accounts for over 90% of neem's pest control effects [19]. The components of neem impact the insect's endocrine system rather than its nervous or digestive system, unlike chemical insecticides, thereby hindering the development of resistance in successive generations [19]. The molecular structure of azadirachtin, a limonoid compound, is a key chemical constituent of neem [46]. The active ingredient azadirachtin repels, disrupts feeding, weakens development, and hampers the reproduction of insects instead of rapidly killing them [19]. The azadirachtin active compound in neem has demonstrated insect growth regulating (IGR) activities, particularly during the larval stages of insects [19].

Botanical pesticides, such as neem oil, have been shown to be highly effective against pest mites while exhibiting low toxicity to beneficial predators [47]. In our study, we demonstrated that the active ingredient azadirachtin extracted from neem trees is slightly toxic to both the adult and immature stages of *T. ndibu*, and it did not adversely affect the reproductive capability of the F0 generation, including the hatching rate and sex ratio of the F1 generation. Similarly, Venzon et al. (2005) did not find any negative effects of neem on the species *Iphiseiodes zuluagai* Denmark and Muma (Acari: Phytoseiidae) [48]. Silva et al. (2013) also reported that azadirachtin is not toxic to *Proprioseiopsis neotropicus* (Acari: Phytoseiidae) [49].

However, it is worth noting that the active ingredient azadirachtin from neem oil may take some time—ranging from one week to three months—to transform under field conditions [50–52]. The considered exposure time was limited to only 72 h, and the exposure method involved dipping the leaves in the pesticide solution, rather than other methods such as direct spraying on small predatory mites or feeding them prey that had been in direct contact with the active ingredient. This approach may lead to advantages in laboratory testing, rather than accurately reflecting the mortality rate in the field [49].

The active ingredient alpha-cypermethrin was found to be toxic to the predatory mite *T. ndibu* and negatively affected its reproductive capacity, as well as having adverse effects on the F1 generation of *T. ndibu*. In laboratory conditions, alpha-cypermethrin has been shown to cause varying levels of mortality (from moderate to high) in common predatory mites such as *Walzia australica* Womersley (Anystidae), *Bdellodes affinis* Atyeo (Bdellidae), *Balaustium murorum* (Hermann) (Erythraeidae), and *Parasitus fimetorum* (Berlese), which are common predators of *Halotydeus destructor* (Tucker) [53]. Alpha-cypermethrin has also been observed to cause mild to moderate toxicity in *Neoseiulus idaeus* Denmark and Muma [54].

The active ingredient fenpyroximate exhibited toxicity against *T. ndibu*, being quite toxic to adult individuals and toxic to the immature stage of *T. ndibu*, while also negatively impacting its reproductive capacity. Other studies have also reported certain effects of this active ingredient on various predatory mite species. Fenpyroximate is not toxic to adult females of *Amblyseius longispinosus*, but it resulted in high mortality rates in the juvenile stage [55]. Mochizuki (2003) and Amano et al. (2004) have reported that fenpyroximate has adverse effects on *Neoseiulus womersleyi* Schicha [56,57], while Kim and Paik (1996) found that fenpyroximate does not harm the eggs and pre-adult stage of *N. womersleyi* [58]. Fenpyroximate prolonged the duration of the egg and nymph stages, reducing the longevity of *Ambylseius swirskii* when used at a concentration of 50 mg/L [59]. The LC₅₀ concentration of fenpyroximate significantly affects the reproductive capacity and lifespan of female *Phytoseius plumifer*, leading to a decrease in population growth [11].

Buprofezin does not cause toxicity to *A. longispinosus* [55] and has a moderately toxic effect in *A. eharai* [60]. However, in this study, it exhibited toxicity in the immature and slightly toxicity in adult female stages of *T. ndibu*.

Matrine is a highly effective alkaloid extracted from the plant *Sophora flavescens* Ait [61]. In our study, we observed that this active ingredient is of low toxicity to adult female *T. ndibu* and immature *T. ndibu*. Similarly, in the study conducted by Fang et al. (2017), matrine was found to only be slightly toxic to *Neoseiulus cucumeris* Oudemans [62].

Biological control through the recruitment and conservation of a diverse set of natural enemies of pests offers the greatest potential for pest management, and combining the use of pesticides with low or no toxicity to the existing population of natural enemies is considered the most optimal approach [62].

5. Conclusions

Three insecticides—Ortus 5EC (fenpyroximate 5%), Fastac 5EC (alpha-cypermethrin 5%), and Chess 50WG (pymetrozin 500 g/kg)—exhibited toxicity against the predatory mite *T. ndibu* at both the immature and female adult stages, thereby inhibiting the establishment of *T. ndibu* populations.

The biopesticide Kobisuper 1SL (matrine) exhibited low toxicity in both adult female *T. ndibu* and immature *T. ndibu*. On the other hand, the biopesticide NeemNim, derived from the neem plant, was not toxic to both the adult and pre-adult stages of *T. ndibu*. Furthermore, it did not affect the reproductive capability of *T. ndibu* in terms of the egghatching rate and the sex ratio in the F1 generation. Therefore, the use of biopesticides might be considered as a key strategy for the control of pests while not harming the existing population of predatory mites in citrus orchards in Southern Vietnam.

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