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Potential toxicity assessment of novel selected pesticides against sand termite, *Psammotermes hypostoma* Desneux workers (Isoptera: Rhinotermitidae) under field conditions in Egypt

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Abstract: The sand termite, *Psammotermes hypostoma* Desneux is a major pest in the New Valley Governorate, Egypt. Great efforts have been taken to control the pest. We evaluated the toxicity of four selected neonicotinoids [acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG)], and one organophosphate pesticide [chlorpyrifos (48% EC)] against *P. hypostoma* workers. The investigation was done under field conditions, using the palm fronds method. However, the reduction percentages in palm fronds was recorded as the height of the sandy clay formed by termite workers on palm frond surfaces, when the fronds had been treated with a different concentration of each pesticide. The results were recorded after 15, 30, 45, and 60 days. Chlorpyrifos (48% EC) was considered the most potent pesticide among all the pesticides tested, but acetamiprid (20% SP) was considered the most toxic among the neonicotinoid pesticides tested. Moreover, the formulation of thiamethoxam (18.6% SC) was more powerful than thaimethoxam (40% WG). Furthermore, the reduction percentages on palm fronds increased significantly with an increase of the exposure period (from 15 to 60 days), as a result of the surface foraging activity of the sand termite workers. These results are given as unique and encouraging trends in controlling sand termites in Egypt.

Key words: *Psammotermes hypostoma* Desneux, neonicotinoids, toxicity, subterranean termite

Introduction

The subterranean termite is the most common termite found in the New Valley Governorate, Egypt. The subterranean termite is the most economically important wood destroying insect not only in Egypt but also in United States. They are classified as serious pests which cost millions of dollars in annual control (Su and Scheffrahn 1990; Su 1994; Su and Scheffrahn 1998; Ahmed *et al*. 2014).

Presently, there are eight species of subterranean termites in Egypt; four of them belong to the genus *Psammotermes*. The common abundance of subterranean termites is in arid and semiarid areas of upper Egypt. However, these termites prefer to infest the places that have a high moisture content and warm temperature (Hafez 1980; Moharram *et al*. 1992). The sand termite, *Psammotermes hypostoma* Desneux, can do great damage to unprotected buildings, other wooden structures, and almost anything containing cellulose. Therefore, attention must be paid to the effects of the various control methods (Abdel-Galil and Kelany 1988; Ahmed *et al*. 2014).

The use of chemical compounds (e.g., pesticides) is of great importance for termite control. However, the extensive reliance on conventional pesticides in pest control led to the eventual rise of pest resistance that could make many of the pesticides ineffective. Great efforts must be done to formulate different strategies to avoid the development of the resistance issue. The use of the new pesticide groups, such as neonicotinoids, are considered the right path towards developing a new strategy to preclude the presence of the pest resistance (Rust and Saran 2008; Smith *et al*. 2008; Ahmed and Matsumura 2012; Ahmed *et al*. 2014; Ahmed and Saba 2014). For instance, chlorpyrifos, an example of an organophosphate pesticide, is more toxic than neonicotinoid pesticides. Yet, chlorpyrifos is not preferred for termite control because neonicotinoid pesticides and their mammalian toxicities are generally low and show low acute toxicities to birds and fish compared to other common pesticide groups. Chlorpyrifos, though, displayed significant toxicities to bees (Parman and Vargo 2010; Neoh *et al*. 2014; Rondeau *et al*. 2014).

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In this study, we evaluated the potential toxicity of four selected neonicotinoids [acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG)], and one organophosphate pesticide [chlorpyrifos (48% EC)] against *P. hypostoma* workers under field conditions using the palm fronds method.

Materials and Methods

The sand termite, *P. hypostoma* was investigated. The formulations of imidacloprid (20% SL), acetamiprid (20% SP), thaimethoxam (40% WG, and 18.6% SC), and chlorpyrifos (48% EC) were used in the field experiments. These pesticides were obtained from the Central Agricultural Pesticides Laboratory (CAPL) in Dokki, Giza, Egypt as gifts (Table 1).

The field evaluation was carried out in Om El-Naseem (the New Valley Governorate, Egypt). Each concentration of the insecticides used was prepared by dissolving the pesticide in distilled water. The concentrations used in this study were: 0.1, 1, 10, 100, and 1,000 mg · l^{-1} based on laboratory evaluations that were conducted before (Ahmed *et al*. 2014). Palm fronds (10 cm) were dipped in each concentration for 10 sec and were left to dry for 20 min. Then, these dried-off fronds were placed inside heavily infested holes. Two holes, almost homogenate holes, were used as two replications for each concentration of each pesticide. As the control, palm fronds were dipped in distilled water only. The percentage of reduction in the termite population when using treated palm fronds was estimated based on the equation below. The experiment was conducted during the period starting from December 5th, 2013 to February 5th, 2014.

Reduction % = 1 [the height (cm) of the palm frond used in the treatment/the height (cm) of the palm frond used in the control] × 100.

Statistical analysis

The statistical analysis of the reduction in height (cm) of selected neonicotinoid pesticides and chlorpyrifos against the sand termite, *P. hypostoma*, by using the surface foraging activity of termite workers on palm fronds, at a concentration of 100 mg · l^{-1}, was done by using Multifactor Regression Analysis by means of the Advance Statistical Analysis Package (ASAP).

Results and Discussions

The reduction in palm fronds (recorded as the height to the coverage of the sandy clay, formed by termite workers on the surface of the palm fronds) after 15 days, was shown in table 2. At a concentration of 0.1 mg · l^{-1}, the reduction percentages using chlorpyrifos (48% EC), acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG) were: 45.00, 41.67, 18.42, 20.00, and 01.96%, respectively. In the case of the higher concentrations, (100 mg · l^{-1}), the reduction percentages were: 100, 100, 86.84, 80.00, and 60.78%, and were: 100, 100, 100, 100, and 80.39% at 1,000 mg · l^{-1}, respectively. After 30 days (Table 3), when a concentration of 0.1 mg · l^{-1} was used, the reduction percentages for chlorpyrifos (48% EC), acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG) were: 58.67, 51.43, 45.78, 35.00, and 14.28%, respectively. While, at high concentrations, 100 and 1,000 mg · l^{-1}, the reduction percentages were: 89.33, 85.71, 78.31, 75.00, and 57.14% for 100 mg · l^{-1}, and 100, 100, 87.95, 87.50, and 71.43% for 1,000 mg · l^{-1}, respectively. After 45 days (Table 4), at the lower concentration of 0.1 mg · l^{-1}, the percentages of reduction for chlorpyrifos (48% EC), acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG)

Table 1. Pesticides used in the field experiments

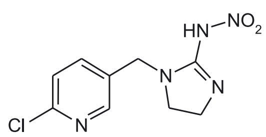
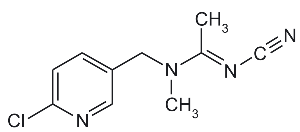
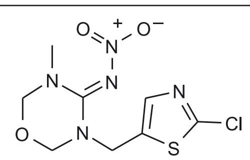
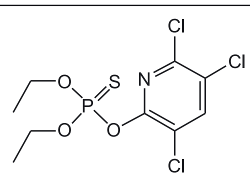
Common name	Group	Chemical name	Chemical structure
Imidacloprid	neonicotinoids	1-(6-chloro-3-pyridylmethyl)-*N*-nitroimidazolidin-2-ylideneamine	
Acetamiprid	neonicotinoids	*N*-[(6-chloro-3-pyridyl)methyl]-*N′*-cyano-*N*-methyl-acetamidine	
Thiamethoxam	neonicotinoids	3-[(2-chloro-1,3-thiazol-5-yl)methyl]-5-methyl-*N*-nitro-1,3,5-oxadiazinan-4-imine	
Chlorpyrifos	organophosphates	*O,O*-Diethyl*O*-3,5,6-trichloropyridin-2-yl phosphorothioate	

Table 2. The reduction (%) of selected neonicotinoid pesticides as compared with chlorpyrifos against the sand termite, *Psammotermes hypostoma*, by using the surface foraging activity on palm fronds method, after 15 days of palm frond treatment

Concentration [mg · l⁻¹]	Chlorpyrifos (48% EC)		Acetamiprid (20% SP)		Imidacloprid (20% SL)		Thiamethoxam (18.6% SC)		Thiamethoxam (40% WG)	
	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]
The control	4.0	00.00	4.8	00.00	3.8	00.00	5.0	00.00	5.1	00.00
0.1	2.2	45.00	2.8	41.67	3.1	18.42	4.0	20.00	5.0	01.96
1	1.3	67.50	1.7	64.58	2.1	44.74	2.9	42.00	4.2	17.96
10	0.5	87.50	1.0	79.17	1.2	68.42	2.0	60.00	3.0	41.18
100	0.0	100	0.0	100	0.5	86.84	1.0	80.00	2.0	60.78
1,000	0.0	100	0.0	100	0.0	100	0.0	100	1.0	80.39

*the reduction percentages were recorded as the height (cm) of the sandy clay galleries of termites formed by the workers on the surface of the palm fronds

Table 3. The reduction (%) of selected neonicotinoid pesticides as compared with chlorpyrifos against the sand termite, *Psammotermes hypostoma*, by using the surface foraging activity of termite workers on palm fronds after 30 days of treatment

Concentration [mg · l⁻¹]	Chlorpyrifos (48% EC)		Acetamiprid (20% SP)		Imidacloprid (20% SL)		Thiamethoxam (18.6% SC)		Thiamethoxam (40% WG)	
	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]
The control	7.5	00.00	7.0	00.00	8.3	00.00	8.0	00.00	7.0	00.00
0.1	3.1	58.67	3.4	51.42	4.5	45.78	5.2	35.00	6.0	14.28
1	2.2	70.67	2.8	56.62	3.6	56.62	4.1	48.75	5.2	25.71
10	1.6	78.67	2.1	67.47	2.7	67.47	3.0	62.50	4.0	42.86
100	0.8	89.33	1.0	78.31	1.8	78.31	2.0	75.00	3.0	57.14
1,000	0.0	100	0.0	87.95	1.0	87.95	1.0	87.50	2.0	71.42

*the reduction percentages were recorded as the height (cm) of the sandy clay galleries of termites formed by the workers on the surface of the palm fronds

Table 4. The reduction (%) of selected neonicotinoid pesticides as compared with chlorpyrifos against the sand termite, *Psammotermes hypostoma*, by using the surface foraging activity of termite workers on palm fronds after 45 days of treatment

Concentration [mg · l⁻¹]	Chlorpyrifos (48% EC)		Acetamiprid (20% SP)		Imidacloprid (20% SL)		Thiamethoxam (18.6% SC)		Thiamethoxam (40% WG)	
	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]
The control	00.00	8.0	8.0	00.00	9.5	00.00	9.5	00.00	9.5	00.00
0.1	47.50	4.2	5.0	37.50	5.4	43.16	7.0	26.31	8.0	15.79
1	60.00	3.2	3.7	53.75	4.5	52.63	5.6	41.05	6.6	30.53
10	71.25	2.3	3.0	62.50	3.6	62.10	4.4	53.68	5.5	42.10
100	81.25	1.5	2.0	75.00	2.7	71.58	3.3	65.26	4.2	55.79
1,000	100	0.0	1.0	87.50	1.5	84.21	2.0	78.95	3.0	68.42

*the reduction percentages were recorded as the height (cm) of the sandy clay galleries of termites formed by the workers on the surface of the palm fronds

were: 47.50, 37.50, 43.16, 26.31, and 15.79%, respectively. In contrast, at high concentrations, the reduction percentages were: 81.25, 75, 71.58, 65.26, and 55.79% at 100 mg · l^{-1}, and 100, 87.5, 84.21, 78.95, and 68.42% at 1,000 mg · l^{-1}. The same trend of the results was also observed after a 60-day exposure period (Table 5).

The statistical analysis of the reduction (cm) at 100 mg · l^{-1} for all tested pesticides was shown in table 6. An interesting finding was that the most significant toxic pesticide was chlorpyrifos (48% EC) followed by acetamiprid (20% SP), imidacloprid (20% SL), thiamethoxam (18.6% SC), and thiamethoxam (40% WG).

Table 5. The reduction (%) of selected neonicotinoid pesticides as compared with chlorpyrifos against the sand termite, *Psammotermes hypostoma*, by using the surface foraging activity of termite workers on palm fronds after 60 days of treatment

Concentration [mg · l⁻¹]	Chlorpyrifos (48% EC)		Acetamiprid (20% SP)		Imidacloprid (20% SL)		Thiamethoxam (18.6% SC)		Thiamethoxam (40% WG)	
	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]	height [cm]	red.* [%]
The control	10.0	00.00	10.0	00.00	10.0	00.00	10.0	00.00	10.0	00.00
0.1	05.4	46.00	06.0	40.00	07.0	30.00	08.0	20.00	10.0	00.00
1	04.5	55.00	05.0	50.00	05.8	42.00	07.1	29.00	08.0	20.00
10	03.3	67.00	04.0	60.00	04.7	53.00	06.0	40.00	06.7	33.00
100	02.4	76.00	03.0	70.00	03.6	64.00	04.8	52.00	05.2	48.00
1,000	00.0	100	02.2	78.00	02.3	77.00	03.2	68.00	04.0	60.00

*the reduction percentages were recorded as the height (cm) of the sandy clay galleries of termites formed by the workers on the surface of the palm fronds

Table 6. The statistical analysis of the reduction in height (cm) of selected neonicotinoid pesticides as compared with chlorpyrifos against the sand termite, *Psammotermes hypostoma*, by using the surface foraging activity of termite workers on palm fronds. The pesticide had a concentration of 100 mg · l⁻¹

Treatment	15-day	30-day	45-day	60-day
The control	4.80 A	7.00 A	9.50 A	10.00 A
Thaimethoxam (40% WG)	1.96 B	3.00 B	4.20 B	05.20 B
Thaimethoxam (18.6 % SC)	1.00 C	2.00 CD	3.30 C	04.80 C
Imidacloprid (20% SL)	0.50 D	1.80 D	2.66 D	04.60 D
Acetamiprid (20% SP)	0.00 E	1.00 EF	2.00 E	03.00 E
Chlorpyrifos (48% EC)	0.00 E	0.80 F	1.20 F	02.26 F

The same letter within the same column are not significantly different at a 0.05% level of probability

Certain studies conducted on the control of the subterranean termite showed promising results which were also in agreement with our present study. Rust and Saran (2008) evaluated the insecticidial and biological activity of the cyano-substituted neonicotinoid acetamiprid against the western subterranean termite, *Reticulitermes hesperus* Banks (Isoptera: Rhinotermitidae). They revealed that acetamiprid was very active against termites in topical applications; LD_{50} = 0.02 ng · termite⁻¹. Even though acetamiprid is extremely toxic in topical applications, deposits ≥ 50 ppm on sand were required to consistently provide > 90% kill of termites within 7 d after a 1-h exposure. Moreover, they demonstrated that termites were quickly affected by short exposures to sand treated with acetamiprid (1 ppm) and within 1 h, their locomotion was dramatically impaired. One important finding which they investigated is that acetamiprid was transferred from donors to recipients only when donors were held on deposits ≥ 50 ppm for 1 h. Deposits even as low as 1 ppm were repellent with termites failing to tunnel into the treated sand, and there was no significant mortality. Exposure to acetamiprid impaired the locomotion of termites as did other slow-acting neonicotinoids, such as imidacloprid. Acetamiprid was repellent at all concentrations tested, which is in strong agreement with our field results, acting like type I pyrethroid treatments in soil. In another study, Fei and Henderson (2005) found that the formosan subterranean termite exposed to imidacloprid had greater rates of mortality than those exposed to acetamiprid. Mao *et al*. (2011) evaluated the toxicity of seven pesticides including imidacloprid, using both topical application and substrate (sand) treatments to determine the LD_{50} and LC_{50} against two economically important subterranean termite species; the eastern subterranean termite, *Reticulitermes flavipes* (Kollar), and the Formosan subterranean termite, *Coptotermes formosanus* Shiraki. The LD_{50} rankings for *R. flavipes* from highest to lowest were: fipronil > bifenthrin > chlorantraniliprole > cyantraniliprole > > imidacloprid > chlorfenapyr > indoxacarb; however, the rankings for *C. formosanus* were: fipronil > imidacloprid > > chlorantraniliprole > cyanthraniliprole > bifenthrin > > chlorfenapyr > indoxacarb. In contrast with our study, Smith *et al*. (2008) tested three termiticides: thiamethoxam, acetamiprid, and a combination of acetamiprid + + bifenthrin, on field-collected *R. flavipes* termites. It was found that thiamethoxam treatments caused consistently greater termite mortality than acetamiprid treatments, which is in disagreement with our findings. Their data concluded that acetamiprid prevented soil penetration by termites more than thiamethoxam, although both were less repellent compared with bifenthrin alone, which causes little termite mortality at the tested doses. Moreover, Remmen and Su (2005a) conducted the time trends in the mortality method for the Formosan subterranean termite, *C. formosanus*, and the eastern subterranean termite, *R. flavipes*, that were determined for thiamethoxam and fipronil. Filter paper treated with 50 ppm thiamethoxam led to > 80% mortality in 2–4 days for *R. flavipes*, whereas 5 ppm thiamethoxam resulted in > 80% mortality in 2–3 days for *C. formosanus*. Filter paper treated with 1 ppm fipronil resulted in > 80% mortality in 5 days for *R. flavipes* and 9 days for *C. formosanus*, which illustrated that thiamethoxam is faster acting than fipronil. Therefore, they stress the fact that as concentration decreases for slow-acting termiticides, the time required for adverse effects to be fully expressed increases. Also, Remmen and Su (2005b) examined the termiticidal properties of thiamethoxam and fipronil against the Formosan subterranean termite, *C. formosanus*, and the eastern subterranean termite, *R. flavipes*. They found that concentrations ≥ 8 ppm thiamethoxam and ≥ 1 ppm fipronil provided an effective barrier against *C. formosanus* and *R. flavipes*. Sand was penetrated to some degree at all concentrations of thiamethoxam (0–800 ppm for *C. formosanus* and 0–1,000 ppm for *R. flavipes*) and fipronil (0–64 ppm for both *C. formosanus* and *R. flavipes*) tested, indicating that both termiticides are nonrepellent which is in disagreement with our results. Moreover, they revealed that thiamethoxam was to be more toxic against *C. formosanus* than *R. flavipes*, whereas fipronil showed similar toxicity for both species. Higher mortality prevented termites from penetrating the entire 5-cm segment of treated sand.

Conclusions

To sum up, the selected neonicotinoid pesticides that have been assessed in our study showed effectiveness against subterranean termites. Interestingly, imidacloprid, acetamiprid, and thiamethoxam (the newer types of neonicotinoids) are more persistent and show slower acting effects as compared with the older counterparts of conventional pesticides such as chlorpyrifos. The major advantage of the present findings is that the toxicological and repellent activity of the selected neonicotinoid pesticides were found to be promising tools in controlling termites with the advantage of having a low toxicity on environmental aspects. The study also provided an opportunity to directly compare toxicity, action speed, and bioavailability among this group of newer generation neonicotinoid pesticides against termites. Thus, the efforts to understand the biochemical and molecular basis of their mechanism of actions should be studied in the future for obtaining a better understanding of the mode of action of neonicotinoid pesticides against termites.

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