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The effectiveness of some selected insecticides against cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) and their non-targeted predators

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Abstract
The invasive polyphagous cotton mealybug, *Phenacoccus solenopsis* Tinsley has emerged as a serious sucking pest of cotton. The laboratory and field bioassays of seven selected insecticides belonging to five toxicant groups were evaluated against *P. solenopsis* and their most abundant natural predators, *Chrysoperla carnea* (Steph.) and *Hyperaspis vinciguerrae* Capra. Based on the toxicity index the most toxic group of insecticides under laboratory conditions was esfenvalerate, acetamiprid and dimethoate after 24 h and 72 h of treatment followed by the oxadiazines metaflumizone, indoxacarb and finally the anti-moulting IGRs, diflubenzuron and chlorfluazuron against the 3rd instar nymphs of *P. solenopsis* using spraying method technique. Of the selected insecticides, dimethoate was significantly superior over the rest of treatments with a highest average reduction percentage in cotton mealybug population (98.26%) under field conditions followed by esfenvalerate (96.72%), indoxacarb (89.66%), metaflumizone (89.43%), acetamiprid (86.28%), diflubenzuron (81.51%), and the least one chlorfluazuron (76.82%). Field experiments recorded that the anti-moulting IGRs were the safer toxicants towards *C. carnea* with average reduction diflubenzuron (45.99%) and chlorfluazuron (25.68%) and towards *H. vinciguerra*, were diflubenzuron (49.39%) and chlorfluazuron (41.52%).

Keywords: Cotton mealybug, *Phenacoccus solenopsis*, insecticides, predators

Introduction
Cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) has been recorded to be a destructive and invasive sucking pest of cotton and wide range of host plants, it possessed a vast range of biological adaptations, morphological properties and ecological adaptability [1-4].

The mealybugs feed by sucking the plant sap from leaves and stems causing symptoms like retarded growth and drying of the host plant. Mealybugs are soft bodies cottony in appearance and cryptic in nature. They infested all plant parts, especially the early stages of plants. Like the aphids, mealybugs also produce the honeydew substance and contribute in the development of black sooty mould over plant surfaces and reduces the quality of the lint [3, 5, 6].

Management of cotton mealybug using intensively chemical insecticides is difficult as a result of the presence of white mealy wax covering their soft bodies. Also, the cotton mealybug's habit of sucking sap is a protective feeding strategy against chemical pesticides contact in addition to the high reproductive potential and the wide host range. Bio-pesticides, organophosphates and insect growth regulators (IGRs) have recently been recommended for controlling cotton mealybug [3].

Development of economically integrated control methods in suppressing *P. solenopsis* is urgently needed also, effective insecticide resistance management (IRM) strategies should include the rotation of insecticides with several mode of action to control *P. solenopsis*. Use of safer chemical insecticides are recommendable for integrated pest management programs aiming at the conservation of the important natural enemies in agro-ecosystems [5, 7].

A successful management strategy should directed to keep *P. solenopsis* under control without adversely impacting natural enemy populations.

The two main natural enemies recorded for *P. solenopsis* were Coccinellids and Chysopids on cotton [3, 5, 8].

The current study assessed the effectiveness of seven insecticides belonging to five different classes to control the cotton mealybug under both lab and field conditions. In addition, studying the effect of these toxicants against the natural enemies associated with the pest under investigation.

Materials and Methods

Insecticides

Commercial formulations of Ultraprid (Acetamiprid 20% SP, Shanghai hanfu biotechnology co. ltd, China), Somi-gold KZ (Esfenvalerate 20% EC, Sumitomo Chemical Ltd, Japan), Camvaal (Indoxacarb 15% EC, Jiangsu Flag Chemical Industry Co., Ltd. China), Topron (Chlorfluazuron 5% EC, Agrochem Co., Alexandria, Egypt), Metazone (Metaflumizone 24% SC, Jingbo Agrochemicals Technology Co, Ltd. , China), Dimilin (Diflubenzuron 48% SC, Arysta Life Science Benelux SPRL, Belgium) and Dancothoate (Dimethoate 40% EC, Jiangsu Tenglong Biological & Medicinal Co., Ltd., China) were assessed for their insecticidal properties to *P. solenopsis* under laboratory and field conditions and also against the associated predators under field consideration only.

Insecticidal test against *P. solenopsis* under laboratory conditions

P. solenopsis was transferred from untreated infested cotton field plants by the authors in Aga district, Dakahalia governorate, Egypt during summer 2022 and identified at Scale Insect Department, Plant Protection Research Institute, Agric. Res. Center, Giza, Egypt as *P. solenopsis.* Cotton mealybug was brought and reared according to Abd El-Mageed *et al*. (2018) [3]. The laboratory experiments were performed on the newly moulted 3rd instar nymphs [2, 3, 9].

Ten reared laboratory 3rd instar nymphs of *P. solenopsis* were released to a cotton leaf, placed in a culture Petri dish and ready for the insecticidal applications. In addition to control, seven treatments were replicated thrice and by using spray method technique, five diluted concentrations of each commercial insecticide were evaluated [2, 10]. By using Abotts formula the mortality was recorded and corrected after 24 and 72 hours post-treatment [11] and LC_{50}, LC_{90} and slope values were analyzed in accordance with Finney (1971) [12]. Toxicity index of the seven insecticides was computed by using Sun's equation [13].

Field bioassay of the selected insecticides against both *P. solenopsis* and its associated natural enemies

Field study was laid out to assess the effectiveness of seven toxicants during summer 2022 against *P. solenopsis* on cotton plants Giza 86 at the field of Aga, Dakahalia, Egypt. Field experiments was carried out to eight treatments (seven insecticides plus a control) in a randomized complete block design. Three replications of each treatment (each 42 m^2) were performed per plot.

Twenty cotton plants were chosen randomly and tagged from each replicate for observation to count the *P. solenopsis* population. Field application was performed by a knapsack sprayer 200 l water/feddan. In accordance with the procedure outlined by (Abel El-Mageed *et al*., 2018) [3], mealybugs were counted.

Observation a sufficient number of mealybug population imposed the field application in the experimental blocks. Observations regarding the *P. solenopsis* population were made a day before spray as well as 1, 3, 7, 14 and 21 days post spraying. Data concerning mealybug population reduction in the all treatments was performed based on Henderson and Tilton (1955) [14].

The impact of the selected toxicants on the related predators, *H. vinciguerra* and *C. carnea*, which were identified at the Scale Insect Department of the Plant Protection Research Institute, Agriculture Research Center, Giza, Egypt, was also observed day before spraying as well as 3, 7, 14, and 21 afterwards days.

Statistical analysis.

One way analysis of variance (ANOVA) was subjected to the obtained data also, Duncan's Multiple Range Test at $p< 0.05$ separated the means of the gathered data [15].

Results and discussion

The toxicity effect of seven insecticides belonging to five different chemical groups was evaluated against the 3rd instar nymphs of *P. solenopsis* under laboratory condition using spraying method technique (Table 1). Of the tested insecticides, esfenvalerate (pyrethroids) recorded the highest degree of effectiveness after one day of application on the basis of toxicity index followed by acetamiprid (neonicotinoids), dimethoate (organophosphates), metaflumizone (Semicarbazone oxadiazoles), indoxacarb (oxadiazines) and the benzoylurea IGRs chlorfluazuron and diflubenzuron (chitin synthesis inhibitors). LC_{50} values were 4.24, 6.59, 15.04, 45.54, 53.44, 457.60 and 629.91 ppm, respectively. After three days of treatment little variation in the order of effectiveness was only recorded, esfenvalerate was consistently the most potent followed by acetamiprid, dimethoate, indoxacarb, metaflumizone, diflubenzuron and chlorfluazuron. LC_{50} values were 1.97, 2.13, 6.85, 7.72, 21.48, 271.43 and 306.58 ppm, respectively.

The study revealed drastic variations between the five tested chemical groups in their degree of toxicity against *P. solenopsis*. Our results revealed that esfenvalerate, acetamiprid and dimethoate found to be the most toxic group in laboratory. According to Saeed *et al*., (2007) [16], the 3rd instar nymphs of *P. gossypiphilous* was susceptible to esfenvalerate using leaf dip method and the LC_{50} values were (27.7 and 24.2 ppm) after 24 h and 48 h in laboratory. Padaliya *et al*., (2022) [17] showed the effectiveness of acetamiprid 20% SP at 0.004% which recorded 80.77% mortality of *P. solenopsis* after 72h of exposure. Saminathan and Jayaraj, (2001) [18] evaluated dimethoate efficacy using leaf dip method, the mortality percentages were 63.33% at 48 h, and 66.67% at 72 h against *Ferrisia virgata*, the mealybug on cotton. While, indoxacarb and metaflumizone were the modertly active toxicants, indoxacarb showed LC_{50} value 21.04 ppm against the 2nd instar nymphs of *P. solenopsis* after 72 h of treatment [19]. Metaflumizone belonging to semicarbazone oxadiazine insecticide group which are voltage-dependent sodium channel blockers on nerve axons [20]. Mohamed and Bakry (2019) [21] recommended the anti moulting compounds like diflubenzuron for controlling mealybug infested guava trees *Icerya seychellarum* and *Ferrisia virgata*.

Management of cotton mealybug *P. solenopsis* using some selected insecticides under field conditions

The effect of some selected insecticides on the cotton mealybug population in the field experiment was conducted and presented in tables 2. Observations on mealybug population were made on a day before, 1, 3, 7, 14 and 21 days after initial application.

The pretreatment population of *P. solenopsis* ranged from 271.67 to 404.67 per 10 inches apical shoot. On the day before the treatment, there was no statistically significant difference in the population of mealybugs between the plots. (Table 2).

One day after spray esfenvalerate was significantly superior over the rest of treatments with a reduction percentage (97.45%) followed by dimethoate (95.80%), indoxacarb (95.27%), metaflumizone (80.52%), chlorfluazuron (77.65%), acetamiprid (77.09%) and diflubenzuron (75.02%). The superiority of dimethoate was significantly recorded after three days of treatment with a population reduction (99.51%) and up to twenty one days to reach (99.63%). The highest average reduction percentage was recorded for dimethoate (98.26%) followed by esfenvalerate (96.72%), indoxacarb (89.66%), metaflumizone (89.43%), acetamiprid (86.28%), diflubenzuron (81.51%), and chlorfluazuron (76.82%).

Our findings were in accordance with Ghanim and Elgohary (2015) [22], who demonestrsted that imidacloprid and dimethoate were effective toxicants under field conditions against the citrus mealybug. Saeed *et al*., (2007) [16] recorded that methomyl was the most potent for controlling the mealybug *Phenacoccus gossypifolia* followed by esfenvalerate and thiodicarb after 24 h of treatment. Neonicotinoid insecticides acetamiprid, imidacloprid and thiamethoxam are highly toxic insecticides against *P. solenopsis* [6]. Indoxacarb's field activity against some Homoptera and Hemiptera, while potent, is less than that observed in Lepidoptera, in part due to lower inherent sensitivity, slower bioactivation, and also due to physical characteristics that are less suitable for sucking insect orally [23]. Application of the non-systemic insect growth regulator diflubenzuron as a foliar spray reduced the striped mealybugs *Ferrisia virgata* (Cockerel) by about half with no phytotoxicity [24]. Chlorfluazuron is one of potent benzoylphenyl ureas (BPUs) that have been developed to be significantly more effective than diflubenzuron in reducing insect pests of cotton and vegetable crops [25].

The conservation of natural enemies was one of the key elements that made it possible to launch the IPM program. The inclusion of selective insecticides targeted *P. solenopsis* and allowed the preservation of arthropod predators to give the chance for their biological control called integrated control [26, 27]. After 3, 7, 14 and 21 days post-treatment, the toxicological effects of the selected insecticides on the associated predators, ladybird beetles *H. vinciguerra* and green lacewings *C. carnea*, were assessed in the field (Table 3).

Field bioassays with dimethoate revealed slightly harmful effects towards *C. carnea* with average reduction (64.82%) followed by esfenvalerate (57.35%), indoxacarb (49.74%), metaflumizone (47.27%), acetamiprid (46.12%), diflubenzuron (45.99%) and finally chlorfluazuron (25.68%). While against the second associated predacious *H. vinciguerra*, dimethoate recorded the highest average reduction (74.64%) followed by esfenvalerate (68.52%), acetamiprid (61.11%), metaflumizone (58.46%), indoxacarb (50.05%), diflubenzuron (49.39%) and the least toxic chlorfluazuron (41.52%).

Results of the present study on effect of insecticides against the natural enzymes associated with cotton mealybugs were on par with those reported by Abel El-Mageed *et al*., (2018) [3] who recorded the tolerance of the most common predator in agroecosystem *C. carnea* against insecticides compared with ladybird beetles *H. vinciguerra*.

Our data are closely parallel to those presented by Bayoun *et al*., (1995) [28], dimethoate and esfenvalerate were also toxic to natural enemies. The IGRs insecticides were found to be safer against both *C. carnea* and *H. vinciguerra* [3]. Predator densities were reduced in the acetamiprid plots compared with the IGR plots, the activity including predatory beetles, green lacewing, predaceous and omnivorous bugs and predatory flies [29]. Several studies reported the higher mortality of metaflumizone than indoxacarb on the Hemiptera predators and this may be due to the different chemical structure [30].

Table 1: Toxicity of some selected insecticides against the 3rd instar nymphs of *P. solenopsis* under laboratory conditions

Tested Compounds	After 24h of treatment					After 72h of treatment				
	LC_{50} (ppm) and confidence limits at 95%	LC_{90} (ppm) and confidence limits at 95%	Slope ± SE	X^2	Toxicity index*	LC_{50} (ppm) and confidence limits at 95%	LC_{90} (ppm) and confidence limits at 95%	Slope ± SE	X^2	Toxicity index*
Esfenvalerate	4.24 (0.75 8.16)	60.14 (34.47 229.94)	1.112±.0.29	0.76	100.0	1.97 (0.12 4.28)	18.70 (11.84 46.91)	1.313 ±0.40	0.72	100.0
Acetamiprid	6.59 (2.76 10.71)	126.61 (51.50 1627.55)	0.999 ±0.26	0.33	64.28	2.13 (0.49 3.84)	20.63 (12.80 57.57	1.299±0.32	0.97	92.76
Dimethoate	15.04 (12.20 18.56)	41.99 (31.73 64.37)	2.875±0.38	0.58	28.16	6.85 (4.14 9.53)	39.63 (23.59 135.26)	1.681±0.38	0.63	28.81
Indoxacarb	53.44 (27.19 413.67)	2019.5863 (308.40 2891655.76)	0.813 ±0.25	0.25	7.93	7.72 (2.42 14.36)	510.32 (113.45 209200.06)	0.704 ±0.22	0.72	25.57
Metaflumizone	45.54 (16.44 86.71)	1996.06 (479.68 741971.83)	0.781 ±0.25	2.85	9.30	21.48 (5.13 38.12)	536.02 (213.58 10384.45)	0.917 ±0.26	0.49	9.19
Chlorfluazuron	457.60 (236.32 1361.66)	6598.42 (1976.73 75673.36)	1.106 ±0.20	0.22	0.93	306.58 (114.05 2464.01)	53570.81 (4986.21 21360286.34)	0.572 ±0.13	0.05	0.64
Diflubenzuron	629.91 (253.14 4149.10)	22412.01 (3596.12 1677145.90)	0.826±0.18	0.40	0.67	271.43 (146.44 824.11)	4920.81 (1369.73 70584.88)	1.019±0.19	0.35	0.73

*Toxicity index = LC_{50} of the most effective compound/ LC_{50} of the tested compound × 100

Table 2: Effectiveness of some selected insecticides against *P. solenopsis* population under field conditions

Insecticide	Field recommended rate*	Pre-spray	Mean number per plant and percent reduction of *P. solenopsis* Days after insecticide treatment: 1		3		7		14		21		Overall Mean	
			Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.
Dimethoate	150 cm^3/100L	280.00^{a}	16.33^{c}	95.80	2.00^{e}	99.51	2.00^{d}	99.06	6.33^{e}	97.29	1.00^{d}	99.63	5.53	98.26
Esfenvalerate	150 cm^3/fed	404.67^{a}	14.33^{c}	97.45	25.00^{de}	95.74	4.00^{d}	98.70	15.00^{e}	95.56	15.67^{cd}	96.14	14.80	96.72
Indoxacarb	100 cm^3/fed	339.33^{a}	22.33^{c}	95.27	41.33^{cd}	91.59	14.33^{cd}	94.45	31.67^{d}	88.81	74.33^{b}	78.17	36.80	89.66
Metaflumizone	300 cm^3/fed	380.33^{a}	103.00^{b}	80.52	74.00^{c}	86.57	9.33^{d}	96.78	31.67^{d}	90.02	25.67^{c}	93.28	48.73	89.43
Acetamiprid	25 g/100L	283.67^{a}	90.33^{b}	77.09	116.67^{b}	71.62	17.67^{cd}	91.81	8.00^{e}	96.62	16.33^{cd}	94.26	49.80	86.28
Chlorfluazuron	400 cm^3/ fed	285.33^{a}	88.67^{b}	77.65	59.67^{cd}	85.57	52.33^{b}	75.89	78.00^{b}	67.24	63.67^{b}	77.77	68.47	76.82
Diflubenzuron	125 cm^3/fed	294.67^{a}	102.33^{b}	75.02	62.33^{c}	85.40	42.00^{bc}	81.26	55.33^{c}	77.50	34.33^{c}	88.39	59.26	81.51
Control		271.67^{a}	377.67^{a}		393.67^{a}		206.67^{a}		226.67^{a}		272.66^{a}		295.47	
LSD $_{0.05}$		169.77	56.37		37.06		31.02		15.55		21.42			

*The used concentrations were determined based on the recommendations of Egyptian Ministry of Agriculture. The figures superscripted with same alphabets in the same columns do not significantly differ from each other as per Duncan's multiple range test

Table 3: Toxicity of applied insecticides to adults of *C. carnea* and *H. vinciguerra*.

Insecticide	Pre-spray	Mean population per plant and percent reduction of associated predators Days after insecticide treatment: 3		7		14		21		Overall Mean	
		Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.	Mean No.	% Reduc.
Chrysoperla carnea											
Dimethoate	7.67^{a}	3.00^{c}	67.98	6.33^{b}	52.06	4.00^{bcd}	62.44	2.67^{d}	76.79	4.00^{cd}	64.82
Esfenvalerate	5.33^{abc}	2.33^{c}	64.22	4.00^{cd}	56.41	3.67^{cd}	50.40	3.33^{cd}	58.35	3.33^{d}	57.35
Acetamiprid	6.67^{ab}	4.67^{b}	42.69	7.00^{b}	39.04	5.00^{bc}	46.01	4.33^{bc}	56.72	5.25^{bc}	46.12
Indoxacarb	7.33^{a}	5.00^{b}	44.16	7.50^{b}	40.57	5.33^{b}	47.62	3.67^{cd}	66.62	5.38^{bc}	49.74
Metaflumizone	7.67^{a}	7.00^{a}	25.29	5.67^{bc}	57.06	5.00^{bc}	53.05	5.33^{b}	53.67	5.75^{b}	47.27
Diflubenzuron	4.33^{bc}	3.67^{bc}	30.62	3.00^{d}	59.76	3.00^{d}	50.10	3.67^{cd}	43.49	3.34^{d}	45.99
Chlorfluazuron	3.67^{c}	3.67^{bc}	18.14	5.33^{bc}	15.64	2.67^{d}	47.60	4.33^{bc}	21.34	4.00^{cd}	25.68
Control	6.00^{abc}	7.33^{a}		10.33^{a}		8.33^{a}		9.00^{a}		8.75^{a}	
LSD $_{0.05}$	2.69	1.58		2.65		1.66		1.12		1.71	
Hyperaspis vinciguerrae											
Dimethoate	5.33^{bc}	1.33^{e}	78.45	1.67^{d}	74.14	2.67^{d}	73.58	3.33^{b}	72.40	2.25^{d}	74.64
Esfenvalerate	5.00^{c}	1.67^{e}	71.16	2.00^{d}	66.99	3.00^{cd}	68.35	3.67^{b}	67.58	2.59^{cd}	68.52
Acetamiprid	5.67^{bc}	3.00^{d}	54.31	3.00^{cd}	56.33	4.33^{bcd}	59.72	3.33^{b}	74.06	3.42^{bcd}	61.11
Indoxacarb	6.00^{bc}	5.33^{b}	23.29	4.00^{bc}	44.98	4.67^{bc}	58.94	3.67^{b}	72.98	4.50^{bc}	50.05
Metaflumizone	7.33^{a}	4.67^{bc}	44.98	4.00^{bc}	54.96	5.00^{c}	64.02	5.00^{b}	69.87	4.67^{b}	58.46
Chlorfluazuron	5.00^{c}	3.67^{cd}	36.61	4.67^{b}	22.92	4.67^{bc}	50.73	5.00^{b}	55.83	4.50^{bc}	41.52
Diflubenzuron	5.67^{bc}	4.00^{cd}	39.08	4.00^{bc}	41.78	5.33^{b}	50.41	4.33^{b}	66.27	4.42^{bc}	49.39
Control	6.33^{ab}	7.33^{a}		7.67^{a}		12.00^{a}		14.33^{a}		10.33^{a}	
LSD $_{0.05}$	1.17	1.22		1.37		2.00		2.87		2.00	

The figures superscripted with same alphabets in the same columns do not significantly differ from each other as per Duncan's multiple range test

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