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Efficacy of certain insecticides against black cutworm *Agrotis ipsilon* (Lepidoptera: Noctuidae) in cotton field

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Abstract

The insecticidal activity and biological effects of four compounds (Dolf-X, Indoprim, Kofrothrin, Terraguard) against the fourth instar larvae of the black cutworm, *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae), were examined by feeding and dipping techniques. In addition, its efficacy on cotton crops in open fields in two separate seasons (2023 and 2024). The results showed that all compounds exhibited good activity against larvae. Dolf-X has the lowest LC_{50} value (highest potency) for both feeding and dipping methods. In addition, dipping generally displayed higher LC_{50} values than feeding. The biological effects of all treatments prolonged larval duration, decreased pupation percentage, increased pupal malformation, and extended pupal duration compared to the control, where Terraguard showed the strongest effects on larval duration and pupation malformation (22.64 days and 59%, respectively) with the most significant impact on pupal weight and adult emergence (both normal and malformed). In the field evaluation, the data reported that all tested insecticides significantly reduced black cutworm larvae populations in cotton fields across two seasons ($p < 0.05$). Dolf-X and Terraguard consistently achieved the highest larval reduction percentages (90.44% and 98.83%, respectively), suggesting their potential as preferred choices for black cutworm control in these specific conditions. All four tested insecticides effectively controlled black cutworm larvae, but Dolf-X and Terraguard were the most potent.

Introduction

The black cutworm *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae), is a polyphagous pest of immense economic importance, attacks the seedlings various agricultural crops worldwide (Cranshaw, 2010). Its broad host range encompasses vegetables, fruits, legumes, cotton, and maize, impacting food security and causing significant economic losses to farmers (Ahmad *et al.*, 2023 and Talekar and Shelton, 2011).

In Egypt, *A. ipsilon* poses a particularly dire threat due to its favorable climatic conditions and extensive agricultural practices (Mohamed and Khattab, 2007, and El-Serafi *et al.*, 2017). The warmer temperatures and mild winters provide optimal conditions for year-round development and multiple generations of the pest (Archer *et al.*, 1980). The black cutworm, *A. ipsilon*, inflicts a significant economic burden on Egyptian agriculture due to its polyphagous nature and voracious appetite. Its broad host range encompasses vital crops like cotton, vegetables, and legumes, directly impacting food security and farmer livelihoods (El-Serafi *et al.,* 2017, and Amitava *et al.,* 2011). In cotton fields, *A. ipsilon* infestation can lead to yield loss exceeding 50%, translating to substantial economic losses for farmers (El-Serafi *et al.,* 2017). Control of *A. ipsilon* remains a formidable challenge due to its adaptability, polyphagy, and potential for rapid resistance development (Srinivasan *et al.,* 2015). Methods like chemical insecticides offer basic control tools, especially with high- density levels (Matthews *et al.,* 2014, and Elbert *et al*., 2008).

Therefore, the current study aims to compare the insecticidal activity of the four tested compounds to control *A. ipsilon* on cotton crop in Assiut Governorate.

Materials and methods

1. Tested insecticides:

Four compounds belonging to four different chemical groups were evaluated (Table 1). ` All tested insecticides were approved by the Agricultural Pesticides Committee (2023) of the Ministry of Agriculture, Egypt.

2. Laboratory experiment:

2.1. Rearing of *Agrotis ipsilon* larvae:

Fourth-instar larvae of *A. ipsilon* were individually housed in chambers formed by plastic grids, resembling 'ice cubes,' as described by Abdel-Salam (1980). The insects were reared under controlled conditions of 25°C and 60-70% relative humidity in the laboratory; they were fed fresh castor leaves until pupation. Once emerged, adult moths were paired for mating in large glass jars. In these jars, a 10% sugar solution-soaked cotton pad was placed as a food source for the moths and muslin cloths as an oviposition site for egg laying. Fourth-instar larvae were again employed for the subsequent bioassay test.

2.2. Bioassay test:

2.2.1. Feeding method

Castor leaves were dipped for 20 seconds in the solutions of the four tested compounds at different concentrations, and the leaves were left to dry under laboratory conditions for about one hour before larval feeding (Abdel-Salam, 1980). All concentrations were prepared on the active ingredient basis (ppm) as shown in Table 1. Where was tested at Dolf-X was tested at 0.25, 0.5, 0.75, 0.90, and 1.0 ppm; Indoprim was tested at 0.39, 0.79, 1.0, 1.25, and 1.5 ppm, Kofrothrin was tested at 0.125, 0.25, 0.5, 0.75, and 1.0 ppm; and Terraguard was tested at 0.05, 0.10, 0.20, 0.25, and 0.40 ppm. The castor leaves that dipped in distilled water only used as a control. About fifty larvae in three replicates of the fourth instar fed on the treated and control leaves were placed individually within the rearing chambers. After 24 h, other untreated ones replaced the treated leaves, and the larvae were continued to be fed until the pupation (Wang *et al.,* 2013).

2.2.2. Dipping method:

The fourth-instar larvae were carefully dipped with a dip net into the different dilutions of the four tested compounds for five seconds for completion. The treated larvae were then poured through fine muslin suspended over an empty beaker and allowed to dry for ten minutes. Afterward, the treated larvae were transferred one by one into rearing chambers where they fed on untreated castor leaves until pupation (Shinde *et al.,* 2011). On the other side, the control larvae were dipped in distilled water for five seconds and then received untreated castor leaves until they pupated. For each treatment and control, fifty larvae in three replicates were used. Biological parameters were determined daily, including the length of the larval and pupal stages, the percentage of pupation adult emergence, the pupal weight, and the LC_{50} values of the tested compounds that were calculated. In addition, the detected malformations were also noted.

Table (1): Tested insecticides.

Common name	Active ingredient	Chemical group	Mode of action	
			Primary Effects	**Secondary Effects**
Dolf-X	Lambda-cyhalothrin 5% EC	Pyrethroids	Disrupts sodium channel function in nerve cells, leading to paralysis and death.	Excitatory tremors, rapid knockdown, increased sensitivity in mammals.
Indoprim	Indoxacarb30% WG	Oxadiazine	Activates glutamate receptors in nerve cells, causing overstimulation and paralysis.	Slow kill, feeding cessation, molting disruption, reduced egg-laying.
Kofrothrin	Deltamethrin 25% EC	Pyrethroids	Binds to sodium channels in nerve cells, disrupting nerve signal transmission and causing paralysis.	Rapid knockdown, tremors, excitatory behavior, potential skin irritation.
Terraguard	Chlorpyrifos 48% EC	Organophosphate	Inhibits acetylcholinesterase enzyme, leading to acetylcholine buildup and overstimulation of muscles, causing paralysis and death.	Sweating, dizziness, nausea, muscle twitching, respiratory problems (toxic to humans).

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3. Field experiment:

The experiment was conducted in two separate seasons (2023 and 2024) during the spring season at Assiut governorate, Al-Fath region, within a history of high population of cutworm *A. ipsilon* throughout May in the cotton field. Tested compounds were evaluated using the recommended rate, where Dolf-X in a rate of 750 cm/feddan (1 feddan = 0.42 hectare), Indoprim in a rate of 25 g/100 L water, Kofrothrin in a rate of 350 cm/feddan, and Terraguard in a rate of 1.25 L/feddan.

The cutworm larvae that were used for open plot experiment depended on the natural infection population. The experimental area was 0.5 feddan cultivated with cotton seedling cultivar Giza 86. Three plots (the Area of each is 175m^2) were treated with baits of each tested component at recommended rate, and one untreated served as a control. Cutworm infection usually begins when cotton plants contain 2-3 leaves, which is the period in which the evaluation was conducted. The evaluation was based on the number of damaged plants with cuts on the root or shoot systems and marked before and after treatments.

After 72 hrs., the randomly selected plants in each treatment were dug up to depth of 5 to 10 cm, and the soil was inspected for the presence of live and dead cutworm larvae. The percentage of reduction in the number of larvae was calculated according to Henderson and Tilton (1955).

4. Statistical analysis

After 24 hrs. of larval treatment of the fourth instar using either a leaf dip or a larval dip, the total mortality percent of the four investigated compounds was recorded. Data was adjusted using the Abbott formula (Abbott, 1925). LC_{50} values for the four tested compounds were calculated. At the LC_{50} values, the various biological parameters were determined. The acquired data was subjected to statistical analysis using the SPSS statistical program to calculate the F-, P-, and LSD 0.05 levels. In the field experiment, all data were subjected to a one-way analysis of variance (ANOVA), according to the Duncan multiple range test (DMRT), to determine the significant differences at 0.05 (Duncan, 1955).

Results and discussion

Data in Tables 2 and 3 showed the insecticidal activity and biological effects of four tested compounds: Dolf-X (Lambda-cyhalothrin 5% EC), Indoprim (Indoxacarb 30% WG), Kofrothrin (Deltamethrin 25% EC) and Terraguard (Chlorpyrifos 48% EC) against the 4th instar larvae of the black cutworm, *A. ipsilon.*

1. Insecticidal activity (LC_{50} Values):

1.1. Feeding method:

All tested insecticides exhibited significant insecticidal activity against black cutworm larvae via the feeding method, consistent with previous studies (Shakur *et al.*, 2007; Abd-El-Aziz *et al.*, 2019, and Ismail and Said, 2021. Dolf-X displayed the highest potency with the lowest LC~50~ value (27.2 ppm), followed by Indoprim (33.6 ppm), Kofrothrin (38.8 ppm), and Terraguard (80.4 ppm). This suggests that Dolf-X requires the least concentration to achieve 50% larval mortality, making it potentially more efficient for controlling black cutworm larvae through ingestion (Srinivasan *et al.*, 2015). These findings align with those reported by Abbas *et al.*, (2023), who also found Dolf-X to be highly effective against lepidopteran pests.

Table (2): Insecticidal activity of tested components as LC_{50} values against the 4th instar larvae of black cutworm *Agrotis ipsilon*

Insecticides	Method of treatment							
	Feeding				Dipping			
	LC_{50} values (ppm)	Slope	95% confidence limit		LC_{50} values (ppm)	Slope	95% confidence Limit	
			Lower	Upper			Lower	Upper
Dolf-X	27.2	3.99	17.28	28.96	80	5.17	36.96	208
Indoprim	33.6	5.27	10.50	87.52	500	4.19	187.56	1600
Kofrothrin	38.8	6.87	36.88	75.74	600	3.61	1570.33	245.42
Terraguard	80.4	3.38	81.65	99.63	800	3.89	272.70	340.64

1.2. Dipping method:

Similar to the feeding method, all insecticides exhibited insecticidal activity when applied via dipping (Chandrasekaran *et al.*, 2020). However, the LC~50~ values were generally higher compared to the feeding method. Dolf-X again presented the lowest LC~50~ value (80 ppm), followed by Indoprim (500 ppm), Kofrothrin (600 ppm), and Terraguard (800 ppm). This indicates that the dipping method was less effective compared to feeding for these specific insecticides (Talekar and Shelton, 2011). The higher LC~50~ values in the dipping method suggest that direct ingestion of treated leaves may be a more efficient route of exposure for these compounds.

2. Biological Effects

2.1. Larval duration:

All tested insecticides extended the larval duration compared to the control, suggesting a disruption in the normal development process (Ahmad *et al.*, 2019). The effect was more pronounced with dipping compared to feeding for most insecticides. Terraguard exhibited the highest increase in larval duration for both methods, which could be due to reduced feeding ability or sublethal effects of the insecticides on larval development (Talekar and Shelton, 2011). This delay in larval development may reduce the overall fitness of the larvae, potentially impacting their survival and reproductive success.

2.2. Pupation:

Both pupation percentage and pupal malformation were affected by the insecticides (Figure 1). Pupation percentage generally decreased compared to the control, with Terraguard showing the lowest pupation rate for both methods, consistent with findings by Chandrasekaran *et al.* (2020). This suggests that the insecticides interfere with the pupation process, potentially reducing the population (Srinivasan *et al.*, 2015). Pupal malformation increased in all treatments compared to the control, indicating developmental abnormalities caused by the insecticides (Abbas *et al.*, 2023). These malformations could lead to reduced fitness and survival of emerged adults, further contributing to population decline.

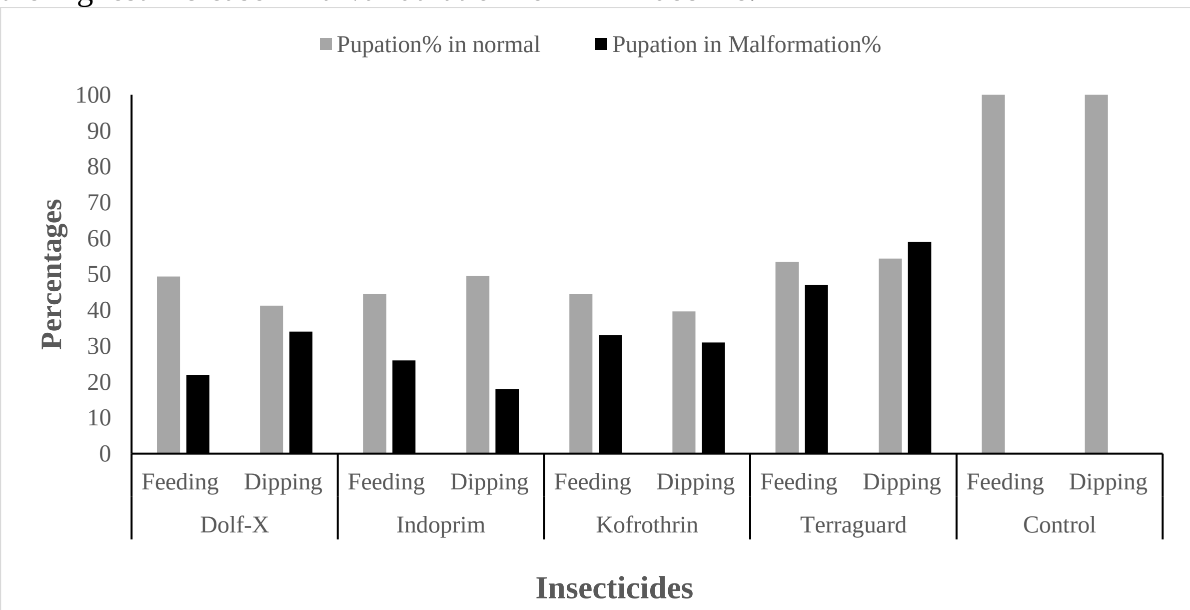

Figure (1): Effect of tested insecticides at their LC_{50} values using two application methods on pupation of 4th instar larvae of *Agrotis ipsilon.*

2.3. Pupal duration:

Similar to larval duration, pupal duration also increased in all treatments compared to the control (Talekar and Shelton, 2011). This further suggests delays in insect development due to the insecticidal effects (Chandrasekaran *et al.*, 2020). The extended pupal duration may result in a longer developmental cycle, potentially reducing the number of generations per season and thus limiting population growth.

2.4. Pupal weight:

Pupal weight generally decreased in all treatments compared to the control (Ahmad *et al.,* 2019). This could be due to reduced feeding during the larval stage and energy reserves as a result of exposure to insecticides (Abbas *et al.*, 2023). Lower pupal weight may indicate reduced energy reserves, which could negatively impact adult emergence and reproductive success.

2.5. Adult emergence:

Both normal and malformed adult emergence decreased in all treatments compared to the control (Talekar and Shelton, 2011). Terraguard displayed the lowest adult emergence rate for both methods, indicating significant disruption in metamorphosis and adult development (Chandrasekaran *et al.*, 2020). This could ultimately lead to population decline due to reduced reproductive potential (Srinivasan *et al.*, 2015). The reduction in adult emergence, particularly of normal adults, suggests that the insecticides not only kill larvae but also impair the development of surviving individuals, further contributing to pest control.

Table (3): Biological activities of tested insecticides at their LC_{50} values using two application methods against the 4^{th} instar larvae of black cutworm *Agrotis ipsilon*

Insecticides	Method	Larval duration (days) ± SD	Pupation (%)		Pupal duration (days) ± SD	Pupal weight (mg) ±S.D	Adult Emergence (%) +SD	
			Normal Mean +SD	Malformation%			Normal	Malformation%
Dolf-X	Feeding	$16.632±1.06^{d}$	$49.40±1.62^{bc}$	22	$12.97±1.09^{cd}$	$445.52±1.41^{c}$	$83.23±3.59^{b}$	14
	Dipping	$19.71±2.89^{c}$	$41.22±3.51^{bc}$	34	$15.8±1.24^{a}$	$367.29±3.42^{e}$	$85.41±4.24^{b}$	23
Indoprim	Feeding	$16.335±2.90^{d}$	$44.55±2.69^{bc}$	26	$12.28±0.05^{d}$	$402.93±1.96^{e}$	$81.82±8.20^{b}$	19
	Dipping	$21.21±3.36^{b}$	$49.51±5.02^{bc}$	18	$14.26±2.94^{b}$	$399.96±3.51^{e}$	$41.93±2.06^{d}$	20
Kofrothrin	Feeding	$21.08±3.52^{b}$	$44.45±0.88^{bc}$	33	$12.74±0.96^{d}$	$413.82±2.69^{d}$	$66.51±4.94^{c}$	16
	Dipping	$20.39±3.0^{8}$	$39.60±1.42^{c}$	31	$13.35±1.59^{c}$	$306.90±2.62^{f}$	$89.42±7.37^{b}$	22
Terraguard	Feeding	$21.97±1.20^{b}$	$53.45±4.70^{b}$	47	$15.05±2.23^{ab}$	$431.16±3.02^{c}$	$34.37±8.21^{e}$	32
	Dipping	$22.64±1.62^{a}$	$54.37±1.41^{b}$	59	$15.6±3.36^{a}$	$438.27±4.70^{c}$	$31.43±3.10^{e}$	36
Control	Feeding	$11.60±4.89^{e}$	$100±1.56^{a}$	0	$11.10±0.12^{e}$	$518.00±3.80^{a}$	$100.00±0.13^{a}$	0
	Dipping	$12.46±3.55^{e}$	$100±1.80^{a}$	0	$12.80±2.44^{d}$	486±1.56b	$100.00±0.45^{a}$	0
F value	Feeding	38.2	5474	-	9.33	14,91	989	-
	Dipping	58.9	2568	-	65.71	25.11	16652	-
P value	Feeding	0.00017	0.00144	-	0.003	0.002	0.0268	-
	Dipping	0.000021	0.00566	-	0.0008	0.0141	0.0275	-
LSD (0.05)	Feeding	1.3	3.81	-	1.43	59.58	3.34	-
	Dipping	1,9	8.37	-	1.61	61.72	6.74	-

3. Efficacy of the tested insecticides on reduction percentages of black cutworm *Agrotis ipsilon* larvae 3 days after application during seasons 2023 and 2024 on cotton field in Assiut Governorate:

Data in Table (4) and Figure (2) demonstrated the significant effectiveness of all four tested insecticides (Dolf-X, Indoprim, Kofrothrin, Terraguard) in reducing black cutworm (*A. ipsilon*) larvae population in cotton fields across two seasons (2023 and 2024). This finding is crucial for cotton farmers facing black cutworm infestations and emphasizes the value of insecticide application as a control measure (Chandrasekaran *et al.*, 2020). Dolf-X and Terraguard consistently exhibited the highest larval reduction percentages across both seasons (90.44% - 98.83% in 2023 and 92.27% - 98.25% in 2024, respectively). This suggests their potential as preferred choices for black cutworm control in these specific cotton field conditions, possibly due to higher potency or compatibility with the environment (Oloumi-Sadeghi *et al.*, 1991 and Srinivasan *et al.*, 2015).

Table (4): Reduction percentages in black cutworm, *Agrotis ipsilon* larvae, post 3 days after application with the four tested insecticides during seasons 2023 and 2024 in cotton fields at Assiut Governorate.

Insecticides	Total population of *Agrotis ipsilon* larvae*					
	Season 2023			Season 2024		
	Before	After 3 days	Reduction%	Before	After 3 days	Reduction%
Dolf-X	147	11	90.44[b]	212	18	92.27[b]
Indoprim	206	14	86.02[c]	157	22	86.59[c]
Kofrothrin	241	21	91.12[b]	281	33	88.92[c]
Terraguard	209	4	98.83[a]	187	5	98.25[a]
Control	186	164	-	251	244	-
P-value	0.00013			0.00172		
F-value	481.39***			22.73***		
LSD (0.05)	1.71			3.12		

- *The numbers represent the total surveyed population of larvae which damaging or cutting leaves during seasons 2023 and 2024.
- Mean with the same letter are not significantly different.
- * represent high significant.**

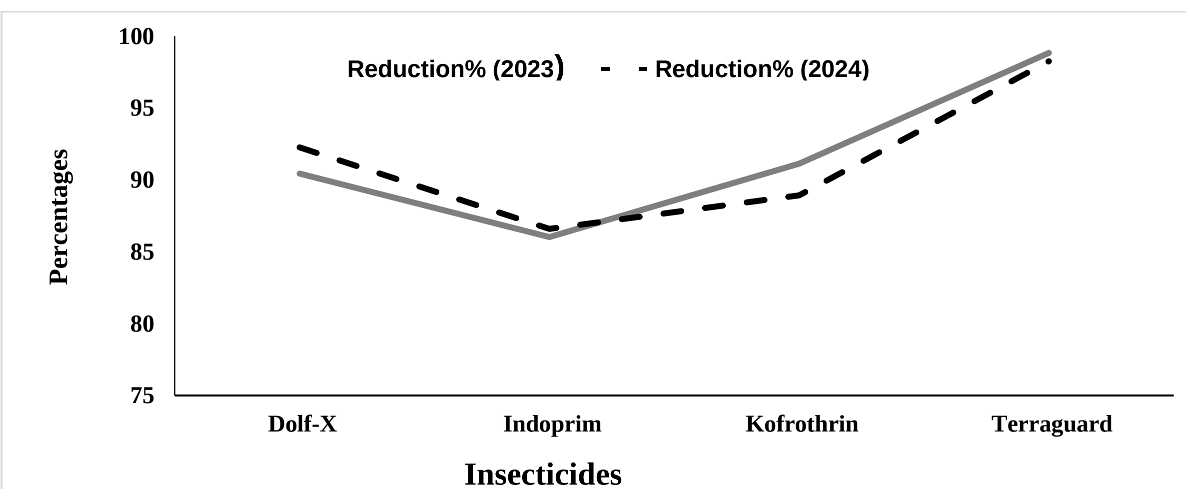

Figure (2): Reduction percentages in black cutworm, *Agrotis ipsilon* larvae, post 3 days after application with the four tested insecticides during seasons 2023 and 2024 in cottons field at Assiut Governorate.

3.1. Seasonal Variations:

While all insecticides remained effective across seasons, some variations were observed. Indoprim and Kofrothrin showed slightly higher reduction percentages in 2023 compared to 2024, while Dolf-X and Terraguard maintained consistent performance, in line with findings by Ismail (2021) and Abd-El-Aziz *et al.* (2019). This suggests potential climatic or environmental factors, such as temperature or rainfall, influencing the insecticides' effectiveness (Talekar and Shelton, 2011). On the other hand, the relatively stable larval populations in the control group emphasize the significant pest pressure exerted by black cutworms in these cotton fields (Talekar and Shelton, 2011). The minimal reduction observed further underscores the need for effective control measures like the tested insecticides to minimize crop damage and economic losses.

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