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Reham, E. Ali, S. Sh. Hafez and Mona, K. El-Hadek

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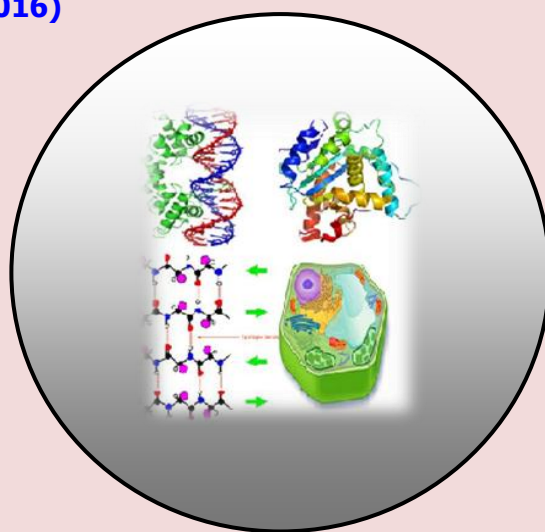
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Dr. Reham E. Ali

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Resistance Coefficient and Biochemical Effects of four Pyrethroid Insecticides on Pink Bollworm (*Pectinophora Gossypiella*) (Saunders) in 2016 Cotton Season

Reham, E. Ali, S. Sh. Hafez and Mona, K. El-Hadek

Central Agricultural of Pesticides Laboratory, Agricultural Research Center,
Dokki- Giza Egypt**ABSTRACT**

The toxicity of four pyrethroid insecticides against field strains of Pectinophora gossypiella collected from four Governorates (Fayoum, Menofia, Dakahlia and Sharkia) was investigated. The results indicated that resistance coefficient (RC) varied from one governorate to another. Cypermethrin showed low resistance in Dakahlia, Menofia and Sharkia but in Fayoum had medium resistance was obtained. Cypermethrin El-nasr showed low resistance in Dakahlia, Fayoum and Sharkia but in Menofia medium resistance was recorded. In Fayoum, Menofia and Dakahlia, resistance to sumi-alfa was medium. But in Dakahlia had high resistance on the other hand Cypermethrin was high resistance in Dakahlia, Menofia and Sharkia but in Fayoum was low resistance. Acetylcholinesterase (AChE), transaminases (GOT and GPT) activity and total protein content in four field strain and one laboratory strain were determined. Data showed that field strain exhibited higher levels of AChE activity than the laboratory strain. High significant reduction in GOT activity in all strains was found as compared with laboratory. The same trend was showed in case of GPT. Total protein was decreased in field strain compared with the laboratory strain.

Keywords: Toxicity Pyrethroids Insecticides, Biochemical Enzymes, GPT (ALT), GPT (AST), AChE and Total Protein.

INTRODUCTION

The pink bollworm *Pectinophora gossypiella* is considered to be one of the most destructive cotton pests. It causes serious damage in cotton bolls resulting in high reduction in quantity and quality of cotton lint yield. The use of insecticides is still having an important role in controlling such insect-pest. Foliar applied insecticides are necessary when the economic threshold is exceeded. Therefore, insecticides are currently the principle method for controlling the bollworms in Egypt and will likely continue to be used until more efficiently based management systems could be developed. Pyrethroids have been commonly used for managing bollworm for many years. However due to their lower toxicity and higher efficacy, recurrent application had led to occurrence of insecticide resistance in various governorates in Egypt (Abo Sholoua *et al.*, 1998). This potential resistance could threaten continued cotton production in Egypt. Thus, it is essential to monitor the effectiveness of four different common pyrethroids and their sustainability to manage such

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economically important insect pest. Early detection of pyrethroids resistance is a key to devising new resistance management strategies to restore the efficacy of pyrethroid-based programmes (Khidr *et al.*, 2002). The results showed that the level of resistance to the tested insecticides was higher in Fayoum governorate than in Dakahlia governorate. The activity of alkaline phosphatase, acetylcholinesterase and total protein were also determined in both field strains. The objectives of the present study were to evaluate the resistance levels in pink bollworm collected from four governorates in Egypt; Dakahlia, Fayoum, Sharkia and Menofia against four pyrethroid insecticides. Biochemical parameters related to reduce sensitivity of *P. gossypiella* against tested insecticides were also determined. These biochemical factors are namely: glutamate pyruvate transaminase (GPT), glutamic oxaloacetic transaminase (GOT), acetylcholinesterase (AChE) and total protein.

MATERIALS AND METHODS

1- Field strains

The field populations of *P. gossypiella* were collected from cotton fields in four Governorates; Dakahlia, Fayoum, Sharkia and Menofia in 2016 cotton season. The full grown larvae were released from infested bolls and kept under laboratory conditions till pupation. The emerged adults were used only in bioassay tests.

2- Tested insecticides

Table 1. The common name, trade name and application rate of the tested pyrethroid insecticides.

Trade name	common name	Rate ml/ fedddan
Kfrothren 2.5 % EC	Deltamethrin	350
Kothren Elnasr 2.5 % EC	Deltamethrin	750
Cumi-alfa 5% EC	Es- fenvalerate	400
Cyperico 20 % EC	cypermethrin	300

3-Bioassay

P. gossypiella adults were tested with the tested insecticides using residual thin film method for 20 sec. six aqueous series concentrations of each tested insecticides were used to evaluate their toxicity against adults of *P. gossypiella*. Ten adults (1-2 days old) were added to each treated glass vial. Each treatment was replicated five times in addition to the control in which insects were treated with distilled water. Mortality percentages were estimated after 24 hrs. and corrected according to the Abbott's formula (Abbott, 1952). The LC_{50} values were calculated using probit analysis statistical method of Finney, 1971. The resistance coefficient (RC) values were calculated according to Joann *et al.* 2013 as follows:

$RC = LC_{95} / \text{recommended field dose}$.

The following criteria for resistance assessment were assumed

$RC \leq 1$ the lack of resistance

$RC = 1.2 - 2$ low resistance

$RC = 2.1 - 5$ medium resistance

$RC = 5.1 - 10$ high resistance

$RC > 10$ very high resistance

4-Biochemical Assay

Parent of both the laboratory and field adults of *P. gossypiella* were used for the biochemical assays as 100 mg of the moths were homogenized in tris buffer pH 7.8. The cold crude extracts were centrifuged at 10000 rpm for 30 min at 4°C. The supernatant was taken carefully into new tubes and preserved at -20°C until the biochemical determination. Total protein was determined colorimetrically by using Buriet reaction (Gornal *et al.*, 1949). Activity of acetylcholinesterase (AChE) was determined according to Ellman *et al.*, (1961). Glutamic Oxaloacetic Transaminase (GOT) and Glutamic Pyruvic Transaminase (GPT) activities were determined colorimetrically according to Reitman and Frankel (1957).

Statistical Analysis

Statistical analysis was carried out according to Fisher (1970) LSD (Least squares difference) test was used to compare the significant differences between means of treatment (Waller and Duncan, 1969)

RESULTS AND DISCUSSION

Resistance coefficient (RC) of the tested insecticides against four field strains of the *P. gossypiella* collected from Dakahlkia, Fayoum, Menofia and Sharkia Governorates in 2016 cotton season is shown in Tables 2,3,4 and 5. The results indicated that the resistance coefficient (RC) varied among the Governorates. Field strains exhibited low levels of resistance to katrothrin as the RC value was 1.97, 1.81 and 1.11 in Dakhliya, Menofia and Sharkia, respectively. Whereas, in Fayoum governorate, medium resistance with RC (2.02) was observed. On the other hand, Kothren-Elnasr had low resistance in Dakahlkia, Fayoum Sharkia and Menofia as RC values were 0.80, 0.72, 0.66 and 1.34, respectively. In case of sumi-alfa medium resistance was recorded in Fayoum Menofia and Sharkia Governorates as RC values were 2.70, 2.70 and 2.7, respectively. But the highest RC was in Dakahlkia (RC= 5.38.) Finally, very high resistance to cyperco was obtained in Dakahlkia, Menofia and Sharkia; RC values were 12.93, 13.9 and 7.28, respectively. In Fayoum governorate low resistance (RC= 1.15) was showed. In conclusion, katrothrin, kothren Elnasr and sum-alfa were effective in controlling *P. gossypiella* as they recorded low and medium levels of resistance, but cyperco showed high level of resistance. Therefore, in order to delay or mitigate the occurrence of resistance in pink boll worm, pyrethroid insecticides could be alternated with other insect pest controlling means into an integrated insect management program. Similar results were also obtained by Mostafa *et.al.* (2014) who found that in field strains of cotton leaf worm collected from Gharbya, Kafr El-Sheikh and Behera governorates, RC varied from one year to another and from Governorates to another.

Table 2. Resistance coefficient of four pyrethroid insecticides against *P. gossypiella* in Dakahlkia Governorate in 2016 cotton season.

No	Insecticides	LC ₅₀	Slope	LC ₉₅	RC
1	katrothrin	88.3	1.83	692.5	1.97
2	Kothren Elnasr	71.50	1.78	600.35	0.80
3	Sum-alfa	154.95	1.43	2154.47	5.38
4	cyperco	494.9	1.83	3879.9	12.93

Table 3. Resistance coefficient of four pyrethroid insecticides against *P. gossypiella* in Fayoum Governorate in 2016 cotton season.

No	Insecticides	LC ₅₀	Slope	LC ₉₅	RC
1	katrothrin	80.21	1.46	1059.24	3.02
2	Kothren Elnasr	70.51	1.85	542.22	0.72
3	Sum-alfa	159.2	1.97	1080.85	2.70
4	cyperco	2.76	0.78	345.7	1.15

Table 4. Toxicity and resistance coefficient of four pyrethroid insecticides against *P. gossypiella* in Menofia Governorate in 2016 cotton season.

No	Insecticides	LC ₅₀	Slope	LC ₉₅	RC
1	katrothrin	78.3	1.80	635.01	1.8
2	Kothren Elnaser	89.28	1.56	1005.80	1.34
3	Sumi-alfa	159.20	1.97	1080.85	2.70
4	cyperco	360.01	1.54	4173.42	13.9

Table 5. Resistance coefficient of four pyrethroid insecticides against *P. gossypiella* in Sharkia Governorate in 2016 cotton season.

No	Insecticides	LC ₅₀	Slope	LC ₉₅	RC
1	katrothrin	63.50	2.08	390.46	1.11
2	Kothren elnaser	46.21	1.58	501.156	0.66
3	Sumi-alfa	141.02	1.58	1084.58	2.71
4	cyperco	355	2.08	2186.90	7.28

Biochemical studies**1. AchE activity**

Biochemical assays in pink bollworm adults indicated that field strains exhibited higher levels of AchE activity than in the laboratory strain, representing 2.9, 3.2 and 3.1 $\mu\text{mol/L/min}$ for Dakahlia, Menofia and Fayoum, respectively as compared with laboratory strain (2.6715 $\mu\text{mol/L/min}$). On the other hand, moths from Sharkia governorate had a decreased activity in AchE reached 1.7 %; while moth had the highest AchE activity (21.08 %) as compared with laboratory strain. These results agree with those obtained by Sabry *et al.*, (2013) who found that AchE activity increased after exposure of pink bollworm to spinosad, colony and buprofezin, pyriproxyfen, lambda-cyhalothrin, thiamethoxam, chlorpyrifos and carbaryl. Nour El-Hoda *et al.*, (2012) illustrated that field strains of *P. gossypiella* expressed higher levels of AchE activity than laboratory strain. With regards to transaminase enzymes GOT and GPT, data in Table (6) show that there was a high significant reduction in GOT activity in all field strains moths as compared with laboratory strain. The levels of reduction in GOT activity reached to 28.33, 34.00, 30.00 and 25.00 ($\mu\text{mol/L/min}$) for Dakahlia, Menofia, Sharkia and Fayoum Governorates, respectively as compared with laboratory strain (36.00 $\mu\text{mol/L/min}$). Data also illustrated that Fayoum strain had the highest reduction in GOT activity (30.56 %), while Menofia recorded the lowest reduction in activity (5.56 %). As for GPT activity, data showed the same trend. GPT activities were 62.00, 53.33, 60.00 and 51.33 units /ml for field strain from Dakahlia, Menofia, Sharkia and Fayoum, respectively when compared with laboratory strain (85.5 units/ml). Data revealed that moths collected from Fayoum governorate had the most decreased GPT activity (40.07%), while moths from Dakahlia had the lowest reduction in enzyme activity (27.6 %). These results agree with those obtained by Kandil *et al.* (2013), who found that there was a reduction in GOT and GPT activities in pink bollworm adults after treatments with insect growth regulators (IGRs), hexaflumuron and chlorfluazuron. Also Salem (2015) reported that, treatments of *P. gossypiella* with pesticides caused a reduction in transaminase enzymes activity.

Table 6. Biochemical study in laboratory strain and four field strains of *P.gossypiella* in 2016 cotton season.

Insect population	Acetylcholinesterase ($\mu\text{mole/l. per min}$)		GOT (Units/ml)		GPT (Units/ml)		Total Protein (g/dl)	
	Activity Mean $\pm$ S.E.	Change (%)	Activity Mean $\pm$ S.E.	Change (%)	Activity Mean $\pm$ S.E.	Change (%)	Activity Mean $\pm$ S.E.	Change (%)
Dakahlia	2.9288 $\pm$ 0.2550	(+) 9.6313	28.33 $\pm$ 2.0276**	(-) 21.30	62.00 $\pm$ 1.1547***	(-) 27.62	12.6466 $\pm$ 0.1438	(-) 4.3128
Menofia	3.2348 $\pm$ 0.0560	(+) 21.0860	34.00 $\pm$ 2.5166	(-) 5.560	53.33 $\pm$ 2.4037***	(-) 37.74	13.1767 $\pm$ 0.2161	(-) 0.3026
Sharkia	2.6240 $\pm$ 0.0204	(-) 1.7780	30.00 $\pm$ 0.8819**	(-) 16.67	60.00 $\pm$ 1.1547***	(-) 29.95	12.5866 $\pm$ 0.1084	(-) 4.7667
Fayoum	3.1030 $\pm$ 0.0589	(+) 16.1520	25.00 $\pm$ 1.7321**	(-) 30.56	51.33 $\pm$ 1.7638***	(-) 40.07	12.7033 $\pm$ 0.1146	(-) 3.8838
Laboratory	2.6715 $\pm$ 0.1274		36.00 $\pm$ 1.1547		85.66 $\pm$ 1.4529		13.2166 $\pm$ 0.1919	

S.E. = Standard Error

(+) increase

(-) decrease

Change % = mean activity of field strain - mean activity of laboratory strain / mean activity of laboratory strain $\times$ 100.**2. Total protein**

The level of total protein in both laboratory and field moth strains was determined as shown in Table (6). The results showed that the total protein decreased in field strain compared with the laboratory one.

Values recorded were 12.64, 13.17, 12.58 and 12.70 g/dl for Dakahlia, Menofia, Sharkia and Fayoum, respectively as compared with in laboratory strain (13.2166 g/dl). Data also illustrated that, Sharkia governorate had the most decreased protein content (4.76 %), whereas, Menofia governorate had the lowest decreased protein content (0.31 %). Similar results were also obtained by Kandil *et al.*, (2013), who found that treatment of *P. gossypiella* adults with insecticides, decreased total protein content. The relationships between protein synthesis and transaminase levels were affected by the hormonal control of protein synthesis and neurosecretory hormones which are involved in the regulation of transaminase levels on the other hand (Megahed *et al.*, 2013) Amira M. Rashad *et al.*, (2015) reported that there was a reduction in total protein in *P. gossypiella* after treatment with insecticides and also they concluded that this reduction may be due to inhibition of DNA and RNA synthesis. However, the decrease of total protein may reflect the decrease in the enzymatic activities of various enzymes.

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Corresponding author: K. El-Hadek, Central Agricultural of Pesticides Laboratory, Agricultural Research Center, Dokki- Giza Egypt

Email: monahazek@yahoo.com