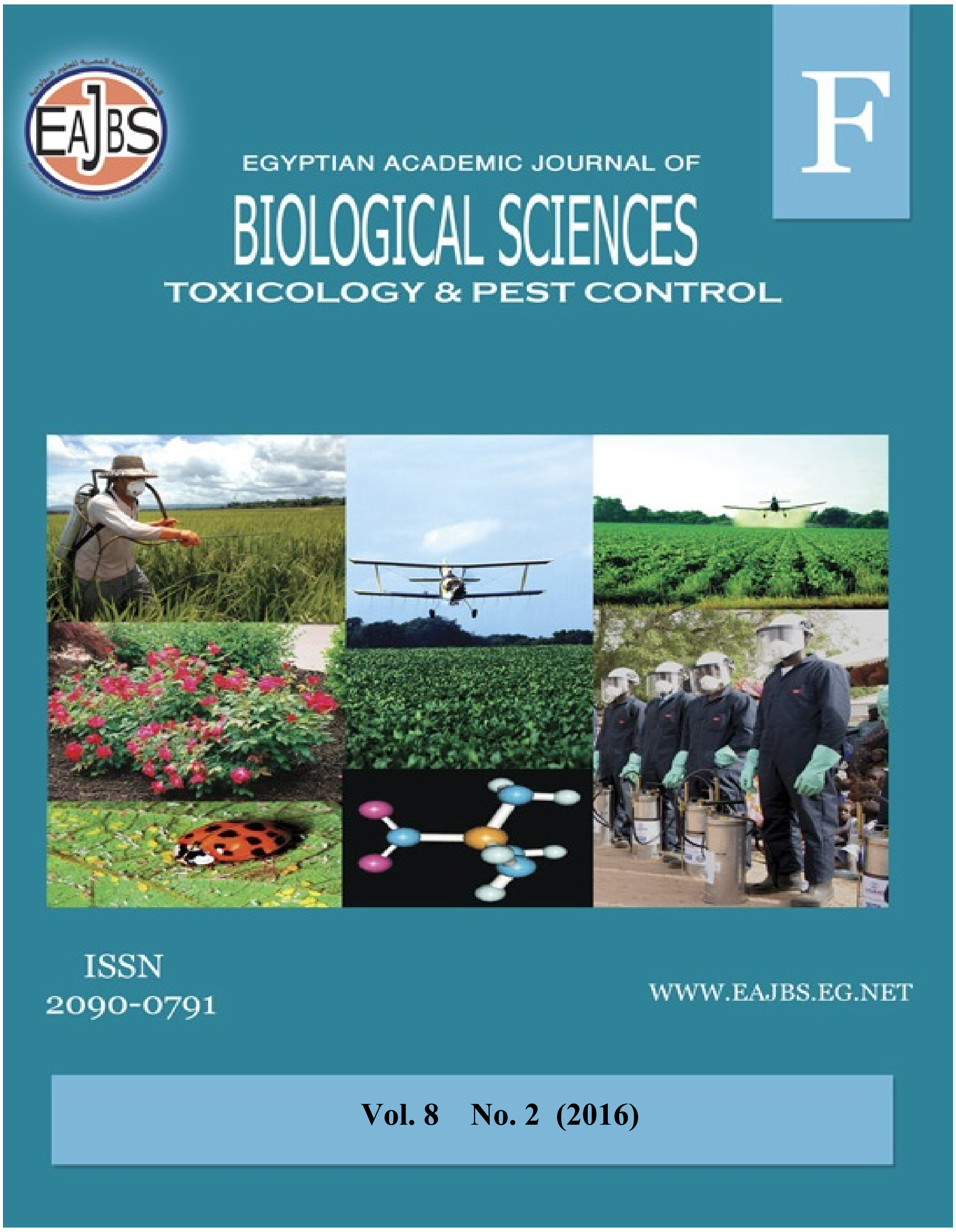

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Monitoring *Aphis gossypii* Glover Resistance to Certain Insecticides in Cotton Fields and Activity of Some Detoxification Enzymes

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ABSTRACT

Aphids resistance to insecticides is an increasing problem because insecticides are an integrated part of high–yielding productive of cotton. Therefore, aphid population, *Aphis gossypii* Glover collected from cotton fields at the Sakha Agric. Res. Station Farm, Kafr El-Sheikh, Egypt, in 2014 and 2015 seasons, was screened for resistance to five insecticides belonged to different groups in addition to activity of some detoxification enzymes. Generally, aphid population was less susceptible to all the tested insecticides in season of 2015 than 2014. Results of LC_{50} s indicated that carbosufan was the most toxic insecticide against laboratory strain of *A. gossypii.* Malathion in season of 2014 and carbosulfan, imidacloprid and chlorpyrifos in season of 2015 gave the highest toxic effect against the field population, while lambda-cyhalothrin exhibited the least toxic effect against both laboratory strain and the field population. Field population showed high resistance to carbosulfan in the two seasons, while resistance to malathion was moderate in the second season only. The resistance to lambda-cyhalothrin, chlorpyrifos and imidacloprid was low in the two study seasons. Based on the susceptibility factor, chlorpyrifos only could be effective against aphids at recommended field rate in the two study seasons, while malathion was effective in season of 2014 only. Activity of AChE and α-esterase was higher in the field population than the laboratory strain in the two seasons. The field aphids exhibited high concentration of total protein compared to the laboratory strain, while the reverse was found in case of lipids.

However, the obtained results could be useful in developing an integrated insect management programs in cotton fields.

INTRODUCTION

Cotton aphid, *Aphis gossypii* Glov. (Homoptera: Aphididae) is one of the most important insect pests on cotton. High aphid population may stunt and retard cotton seedling growth and development because of direct feeding. Late-season populations can cause a decrease in fiber quality because of stickiness and development of sooty mold associated with honeydew dropped on open cotton bolls (Blackman and Eastop, 1984 and Forlow and Henneberry, 2001). Chemical control of cotton aphid is primarily depended on the application of broad-spectrum insecticides such as organophosphates, carbamates and pyrethroids (Li *et al*., 2001).

Due to widespread intensive application of the insecticides, cotton aphids have developed different levels of resistance to these insecticides (Takada and Murakami, 1988; O'Brien *et al.,* 1992; Nauen and Elbert, 2003; Andrew *et al*., 2006; Ahmed and Arif, 2008; Cao *et al*., 2008 and Tabacian *et al*., 2011).

However, mechanisms in which insects develop resistance to insecticides include decrease in insecticide penetration through cuticle which reduce target site sensitivity and enhance metabolism (Oppenoorth, 1985). Also, detoxification enzymes; cytochrome P450 monooxygenases, glutathione-S-transferases and esterases are of major factors in resistance to insecticides in several aphid species (Devonshire, 1989 and Abdel-Aal *et al*., 1990).

Acetylcholinesterase (AChE) is a key enzyme involved in the termination of nerve transmission at cholinergic synapses by hydrolyzing acetylcholine released from the presynaptic terminal, making it an effective target for organophosphate and carbamate insecticides. Several researchers demonstrated that alteration of AChE to an insensitive form in cotton aphid indicated as an important mechanism for resistance toward organophosphate and carbamate insecticides (Devonshire, 1989; Moores *et al.,* 1996; Benting and Nauen, 2004; Andrews *et al*., 2006 and Shang *et al*., 2012) . In contrast, an increase in its activity was reported for the resistant strains in some cases (Zhu and Gao, 1999 and Gao and Zhu, 2002).

Therefore, the continuous monitoring insect resistance to the different insecticides should be conducted on a regional scale to identify the efficiency of these insecticides, as the information about possible resistance to several insecticide classes may be useful in developing an integrated pest management program for the cotton aphid.

The present work was carried out to monitor resistance of *A. gossypii* collected from cotton fields to five insecticides belonging to different groups at Kafr El-Sheikh Governorate during season of 2014 and 2015. The study also involved determination of some detoxification enzymes activity.

MATERIALS AND METHODS

Aphid culture:-

Colonies of *Aphis gossypii* Glov. were orginated from unsprayed cotton fields at Sakha Agric. Res. Station Farm. The strain was reared on cotton seedlings (Giza 86) grown in plastic pots under laboratory conditions of 25±2 C^0; 65±5% relative humidity and a photoperiod of 14 light: 10 dark as described by Norman and Suttan (1967). These colonies were kept on the rearing seedlings for one year without any exposure to insecticides. Every week, the seedlings were replaced with new ones in order to keep aphid colonies alive. This laboratory strain was used as a reference strain.

In this study, the cotton leaves infested with cotton aphid, *A. gossypii* were collected from cotton fields at Sakha Agric. Res. Station Farm, Kafr El-Sheikh, Egypt, in August 2014 and 2015 and transferred to the laboratory to carry out the toxicity tests.

The tested insecticides:-

In this study, the commercial formulations of the tested insecticides were used.

Organophosphates

Chlorpyrifos (Dursban 48% EC) was obtained from Dow Agro Sciences.

Malathion (Malatox 57% EC) was obtained from Chemenova denemark Co.

Carbamates

Carbosufan (Marshal 20% EC) was obtained from FMC Corporation- USA.
Synthetic pyrethroids
Lambda-cyhalothrin (Kaput 5% EC) was obtained from AKROB.V. Holand.
Neonicotenoid Imidacloprid (Confidor 35% SC) was obtained from Bayer Co.

Table 1: The insecticides used in the periodical spray program applied sequently in cotton season of 2014 and 2015 at Kafr El-Sheikh Governorate.

No. of spray	2014 season	Rate/ feddan	2015 season	Rate/ feddan
1	Chlorpyrifos (Dursban 48% EC)	1 Liter	Chlorpyrifos (Dursban48%EC)	1 Liter
2	Lambda-cyhalothrin (Kaput 5% EC)	750 cm^3	Methomyl (Neomyl 90%SP)	300 gm
3	Profenfos (Selyan 72% EC)	750 cm^3	Alpha-cypermethrin (Cypermethrin 10%EC)	250 cm^3

Toxicity tests:

The leaf-dip technique described by Moores *et al*. (1996) was used to compare susceptibility of the laboratory and field colonies of *Aphis gossypii* Glov. to certain insecticides during 2014 and 2015 cotton seasons. Desired concentrations of each insecticide were prepared by diluting the commercial formulations with distilled water. Cotton seedlings leaves were dipped in the insecticidal solution for 10 seconds. The excess of insecticide was allowed to drain off and the leaves were dried on paper towel in shade. For each strain of *A. gossypii,* ten apterous adults of the same age and size, prestarved for 4 hours were placed on the treated leaves in petri-dishes (9cm) with the help of a soft camel hairbrush. Also, control treatment was done using cotton leaves dipped in water only. Three replicates were used for each concentration of the tested insecticides and the control. All Petri dishes were maintained under laboratory conditions (25±2^{0}C; 65±5% RH and a photoperiod of 14 light:10 dark). Mortality counts were recorded after 24 hours of aphid release on the treated leaves and were adjusted for mortality in the control by Abbott's correction (Abbott, 1925) wherever was necessary. Aphids that failed to move when touched with camel hairbrush were considered as dead. The corrected mortalities were plotted on a log-dosage probit paper and the regression lines, values of LC_{50}, LC_{90} for the tested insecticides were calculated according to Finney (1971). The resistance ratio was determined by dividing LC_{50} of the field strain over that of the laboratory strain according to Georghiou (1972).

Biochemical assay:-

Activity of acetylcholinestrase (AChE), non speciefic estrases (α-esterase), total protein and lipids were assayed in the field strain of cotton aphid collected from the cotton fields during season of 2014 and 2015 as well as in the laboratory strain. The insects were prepared as described by Amin *et al.* (1998) and homogenized with cold phosphate buffer PH 7.2 (50 mg/ 1 ml) using a glass homogenizer. The cold crude extracts were centrifuged at 8000 rpm for 15 minutes at 2 C^o in a refrigerated centrifuge, and then passed through glass wool to remove the last of insoluble cell debris. The supernatant was kept in deep freezer at -20 C^o until determination the total protein was determined according to the method of (Bradford, 1976), total lipids (Knight *et al*., 1972), AChE activity (Simpson *et al*., 1964), and non specific esterases (Van Asperen, 1962).

RESULTS AND DISCUSSION

The present study was carried out to monitor susceptibility of aphid colonies, *Aphis gossypii* Glov. collected from cotton fields to five insecticides belonging to different groups at Kafr El-Sheikh Governorate during season of 2014 and 2015. The study also involved determination of some detoxification enzymes activity.

- Susceptibility of *A. gossypii* to certain insecticides:-

The results in Table (2) cleared that carbosufan was the most toxic insecticide against the laboratory strain with LC_{50} value of 1.064 ppm while; lambada-cyhalothrin was the least toxic one with LC_{50} value of 367.348 ppm. Meanwhile, malathion, imidacloprid and chlorpyrifos gave a moderate toxic effect without significant differences between them because of the overlap between individual values of confidence limits.

Table 2: Toxicity of certain insecticides against laboratory strain and aphid colonies, *Aphis gossypii* Glov. collected from cotton fields during 2014 and 2015 season under laboratory conditions.

Insecicides	Laboratory strain			Field strain of 2014				Field strain of 2015			
	LC50 (ppm)	C.L. of LC50*	Slope ± SE	LC50 (ppm)	C.L. of LC50*	Slope ± SE	R.R.**	LC50 (ppm)	C.L. of LC50*	Slope ± SE	R.R.**
Chlorpyrifos	26.573	20.799-34.836	1.534± 0.197	50.489	38.225-67.052	1.346± 0.183	1.90	102.689	84.116-124.280	1.936± 0.268	3.86
Malathion	13.209	7.634-22.100	1.312± 0.212	33.859	29.101-37.845	2.473± 0.299	2.56	376.327	320.909-440.771	2.374± 0.352	28.49
Carbosufan	1.064	0.715-1.608	0.962± 0.138	62.759	44.296-86.550	1.223± 0.218	59.00	81.963	62.041-108.640	1.344± 0.185	77.06
Lambda-cyhalothrin	367.348	289.503-417.09	1.647± 0.201	397.261	347.546-449.327	3.538± 0.446	1.08	725.409	598.254-892.533	2.056± 0.326	1.97
Imidacloprid	15.611	12.512-19.834	1.841± 0.243	51.022	41.223-63.422	1.745± 0.253	3.27	84.883	68.013-105.427	1.717± 0.229	5.44

C.L. of LC50*= confidence limits R.R.**= resistance ratio

As for field-collected aphids, the population differed in their susceptibility to the tested insecticides from one season to another and within the season (Table 2). In general, field aphid population was less susceptible to all the tested insecticides in 2015 season than 2014 one. This may be due to indirect effect of insecticides used for controlling the different cotton pests as shown in Table (1).

With regard to season of 2014, malathion was the most toxic insecticide with LC_{50} value of 33.859 ppm while; lambda-cyhalothrin was the least toxic one (LC_{50} of 397.261 ppm). The rest insecticides exhibited a moderate effect without significant differences, as there was overlap between values of confidence limits. As for season of 2015, carbosulfan, imidacloprid and chlorpyrifos gave the highest toxic effect with LC_{50} values of 81.963, 84.883 and 102.689 ppm, respectively, without significant differences because of overlap between their confidence limits, while lambda-cyhalothrin was the least toxic one with LC_{50} of 725.409 ppm. This result agreed with the finding of Hugh *et al.* (2003) and Tabacian *et al.* (2011) who reported that *A. gossypii* exhibited high susceptibility to lmidacloprid.

The slope values of toxicity lines were taken as an indication of the degree of homogeneity of the aphid population to the tested insecticides. The results indicated that the laboratory strain to a great extent did not show homogeneity in its response to the tested insecticides, as the slope values ranged from 0.9620±.138 to 1.84±10.243.

As for the aphids collected from cotton fields, the population reflected different degrees of homogeneity in response to the tested insecticides. It was obvious that the slopes of malathion and lambda-cyhalothrin regression lines in the two seasons were steeper than those of other insecticides indicating less variation in the susceptibility of aphids to the two insecticides. On the other side, there was no homogeneity in response of the population to carbosulfan that had the least slope value in the two seasons.

These results agreed with the finding of O' Brien *et al*. (1992) who indicated higher homogeneity of field colony aphids, *A. gossypii* in response to chlorpyrifos compared with susceptible aphids, as the slope value of the line for field colony aphids was significantly greater than that of susceptible strain.

However, the slope value is considered as a reaction indicator between the chemical compound and the target organism. In other words, the highest slope value means more homogeneity in the response of the organism towards the pesticide and in the same time, the pesticide is acting as a selection factor producing an organism strain as genetically pure as possible, while the low slope value indicates heterogeneity of organism population response to the pesticide. Also, one of the first signs in the development of a resistant strain is the decrease in the slope value (Hoskins and Gordon, 1956).

For calculating resistance ratio, the laboratory strain was used as a susceptible strain and the term" susceptible strain" was used as a relative term, consequently, the resistance ratio might be greater if a more susceptible population had been used. The results in Table (2) indicated that the field-collected aphids showed low resistance to lambda-cyhalothrin, chlorpyrifos and imidacloprid in the two study seasons, as the resistance ratio ranged from 1.08 to 3.27 fold in season of 2014 and from 1.97 to 5.44 fold in season of 2015. Moreover, the aphid resistance to malathion increased sharply from low level (2.56 fold) in season of 2014 to reach a moderate resistance (28.49 fold) in season of 2015. Also, it was found that aphid population exhibited high level of resistance to carbosufan by 59.00 and 77.06 fold in the two seasons, respectively. This result might due to the annually wide use of carbosulfan and to some extent malathion either against cotton insects or against other arthropods at Kafr El-Sheikh Governorate. Thus, it can be concluded that carbosulfan had more affinity to mechanism (s) of resistance than malathion, since aphids had acquired resistance to carbosulfan by about 23.05 and 2.70 fold more than that of malathion in season of 2014 and 2015, respectively.

The obtained results agreed with those of Ahmed and Arif (2008) who reported that the Pakistani field population of *A. gossypii* exhibited a very low resistance to organophosphates; monocrotophos, profenofos, chlorpyrifos and pirimiphos-methyl and a low to moderate resistance to carbamate compound; thiodicarb, while, they found no resistance to carbamate aphicides; carbosulfan and furathiocarb. Nauen and Elbert (2003) found that *A. gossypii* had no resistance to imidacloprid, but it had a strong resistance to pirimicarb and oxydemeton-methyl and to a lesser extent to cyfluthrin. O'Brien *et al.* (1992) detected resistance to the systemic carbamate; aldicarb in cotton aphid from Mississippi.

Opposite results were reported by Wang *et al.* (2007) when determined resistance of *A. gossypii* collected from four leading cotton producing regions and one non-cotton producing region in Shandong, China, in 1985,1999 and 2004 to fenvalerate, omethoate, imidacloprid, acetamiprid, carbosufan and endosulfan on cotton. They reported that *A. gossypii* became highly resistant to fenvalerate from different regions as compared with the susceptible population. The insects also, exhibited strong resistance to imidacloprid. In contrast, the resistance to carbosulfan did not significantly increase from 1999 to 2004 in all regions. However, the contradiction in results may be due to the differences in susceptibility of aphids in the different geographical regions.

From the mentioned results, it can be concluded that the low slope value of carbosulfan was accompanied by high resistance ratio. The foregoing results are supported by those of Hoskins and Gordon (1956) who reported that the first sign in developing resistance was the change in the slope values. On the other side, the slope values of the other insecticides did not coincide with the level of resistance ratio.

To predict the ability of the tested insecticides at their recommended field rates (diluted in 300 L water/feddan) to control *A. gossypii* population in cotton fields, susceptibility factor was calculated as described by Nazer *et al.*, (1983) and the insecticides expected to be effective under field conditions are those having values less or equal to 0.5 only. The results in Table (3) generally showed variation in susceptibility factor depending on the tested insecticides. Chlorpyifos could be effective to control aphids at recommended field rate in the two seasons, as the susceptibility factor value was less than 0.5. Thus, this treatment might delay the development of aphid resistance to this insecticide, while malathion was effective against aphids in season of 2014 only. In contrast, the other insecticides should not be used to control aphids in cotton fields during the two study seasons to avoid control failure, since susceptibility factor values were more than 0.5.

However, the absolute comparison should be taken with more care because of the complete differences between both of laboratory and field conditions. Also, the susceptibility factor can be modulated by reducing the total spray volumes. Therefore, spraying equipments of reduced volumes is preferred.

Table 3: Susceptibility factor of some insecticides against colonies of *Aphis gossypii* Glov. collected from cotton fields during 2014 and 2015 seasons at Kafr El-Sheikh Governorate

Insecticides	Recommended field conc. (ppm)	LC_{90} of field strain (ppm)		Susceptibility factor	
		Season of 2014	Season of 2015	Season of 2014	Season of 2015
Chlorpyrifos	1600	452.108	471.604	0.282	0.295
Malathion	570	111.681	970.045	0.196	1.702
Carbosufan	66.67	701.632	736.559	10.524	11.048
Lambda-cyhalothrin	66.67	975.595	1216.53	14.633	18.247
Imidacloprid	175.33	276.937	473.318	1.580	2.699

$$\text{Susceptibility factor} = \frac{LC_{90}\text{ of tested insecticide from LCP line}}{\text{Recommended field concentration in ppm}}$$

2- The specific activity of some enzymes in laboratory and field strain of *Aphis gossypii* Glov.:

acetylcholinesterase (AChE) activity:-

The data presented in Table (4) show acetylcholine esterase, α-esterase activity, total protein, total lipids in laboratory strain of *A. gossypii*, and field population collected from cotton fields during 2014 and 2015 at Kafr EL-Sheikh governorate. The results revealed significant increase in AChE activity in the field population of *A. gossypii* during the two study seasons compared to the laboratory one. Also, there was insignificant difference in AChE activity of the field strain in the two seasons, in spite of significant variations in its susceptibility degree to the tested insecticides. The activity of AChE in the field strain was 1.184 and 1.37 fold higher than that of the laboratory strain in season of 2014 and 2015, respectively.

It can be stated that there was a possible positive correlation between AChE activity in the field population and its high resistance toward carbosulfan and malathion, which were known as cholinesterase inhibitors.

Table 4: Acetylcholinesterase, α-esterase, total protein and lipids activity in laboratory and field population of *Aphis gossypii* Glov. collected from cotton fields during 2014 and 2015 seasons at Kafr El-Sheikh Governorate

Strain	Total protein (mean±SE) (mg/ml)	AChE activity (mean± SE) (MgAChBr/min/mg protein)	α-esterase (mean± SE) (mg α- naphthol/ min/ mg protein)	Total lipids (mean± SE) (O.D./mg/ml)
Laboratory strain	1.966±0.071 c	1.822±0.567 b	1.198±0.170 c	3.165±0.080 a
Field strain 2014	3.493±0.050 a	2.157±0.189 a	3.429±0.346 b	2.740±0.056 b
Field strain 2015	3.164±0.126 b	2.490±0.252 a	6.790±0.299 a	2.350±0.049 c

The obtained results agreed with those of Devonshire (1989) who revealed that *A. gossypii* had high tolerance to the carbamate compound; pirimicarb because of existence of a mutant form of acetylcholinesterase that is less sensitive to inhibition by this insecticide. Also, (Zhu and Gao, 1999 and Gao and Zhu, 2002) found an increase in the activity of AChE in organoposphate resistant strain of *Schizaphis graminum* (Rondani) and it was the result of over-expression of AChE.

Opposite results were obtained by Shang *et al*., (2012) who reported that the resistant strain of *A. gossypii* exhibited significantly lower specific AChE activity compared to the susceptible strain.

However, modification of AChE to an insensitive form can be related to the increased AChE activity and has been demonstrated as the most important mechanism providing resistance to the organoposphates and/ or carbamates in *Schizaphis graminum* (Rondani) (Zhu and Gao, 1999). Tsagkarakou *et al.* (2002) reported insignificant differences in AChE activity between resistant and susceptible populations in several cases.

b – α-esterase activity:-

Concerning the activity of α-esterase, aphid population showed significantly higher activity of α-esterase in season of 2015 than in season of 2014, while the laboratory strain exhibited the least α-esterase activity. The activity of α-esterase in field strain was 2.862 and 5.668 fold higher than that of the laboratory strain in season of 2014 and 2015, respectively. It can be stated that high α-esterase activity in the field strain of aphid in the second season was accompanied with its low susceptibility to the tested insecticides.

However, in several aphid species, esterses appear to be significant factors in resistance to insecticides, especially to organophosphates (Devonshire, 1989; Abdel-Aal *et al*., 1990 and Cao *et al*., 2008). Esterase enzymes play an important role in conferring or contributing to insecticide resistance in insect (Field and Devonshire, 1998). Also, Takada and Murakami (1988) using electrophoresis, detected esterase pattern of resistant *A. gossypii* to malathion and the resistance of *A. gossypii* to malathion was positively correlated with high esterase activity.

Regarding the concentration of total protein, the results in Table (4) showed that the field population in season 2014 exhibited the highest concentration of total protein followed by that in season of 2015, while the least concentration was found in the laboratory strain. However, the increase in the total protein of the field population may reflect the increase in the activity of various enzymes related to organophosphates, carbamates and pyrethroids because the proteins are among most important compounds of insects that bind with foreign compounds.

As for total lipids in the aphid population, the results indicated that lipid content significantly was higher in laboratory strain followed by aphid population in season of 2014, while, the least content of lipids was found in aphid population in season 2015. However, Gerami (2013) measured lipid and protein content in three strains of cotton aphid, *A. gossypii* (resistant to neonicotinoid, semi-sensitive and sensitive strains) in different exposure methods of spraying to neonicotinoids and showed that the total lipids in susceptible strain were decreasing more than resistant strain, whereas total proteins were increasing in resistant strain compared to sensitive strain.

Gerami and Heidari (2014) determined the total lipid and protein in three strains of cotton aphid, *A. gossypii* (resistant to neonicotinoid, semi-sensitive and sensitive strains). They found that total lipid in susceptible strain was increased in the counter of spraying and it was decreased in resistant and semi-sensitive strains, whereas total protein was decreased in all of the strains encountering with neonicotinoid stress. Chippindale *et al.* (2001) found that susceptible populations of cotton aphids accumulated high lipid and carbohydrate levels.

Finally, it can be concluded that the field population varied in their susceptibility to the tested insecticides from season to another. Malathion in season of 2014 and carbosulfan, imidacloprid and chlorpyrifos in season of 2015 gave the highest toxic effect against the field population, while lambda-cyhalothrin exhibited the least toxic effect against both laboratory strain and the field population. Also, the field population showed high resistance to carbosulfan in the two seasons, while the resistance to malathion was moderate in the second season only. Chlorpyrifos could be used effective to against aphids in the two study seasons and malathion in the first season. The activity of AChE and α-esterase was higher in the field population than laboratory strain in the two seasons.

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ARABIC SUMMERY

مراقبه مقاومة منّ القطن لبعض المبيدات في حقول القطن و نشاط بعض الإنزيمات الهادمة

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ان مشكلة مقاومة حشرات المن للمبيدات تزداد لأن المبيدات تمثل جزء هام فى الأنتاجية العالية للقطن ولذا يهدف البحث الى مراقبة حساسية من القطن فى حقول القطن بمزرعة محطة بحوث سخا بكفر الشيخ لخمسة مبيدات تتبع مجاميع كيماوية مختلفة خلال موسمى القطن ٢٠١٤، ٢٠١٥م وكذلك دراسة نشاط بعض الانزيمات الهادمة للمبيدات.
أشارت النتائج أن من القطن في موسم القطن ٢٠١٥ م كان اقل حساسية للمبيدات المختبرة عنه في موسم ٢٠١٤ ، وبناءاً علي التركيز القاتل ل٥٠% من الأفراد المعاملة كان كربوسلفان أكثر المبيدات فاعلية على السلالة المعملية ، وسجل ملاثيون أعلى سمية فى موسم القطن ٢٠١٥ ، وأظهر لمباداسيهالوثرين أقل تأثيرا على السلالة المعملية والحقلية فى الموسمين . وقد أظهرت الأفراد الحقلية درجة عالية من المقاومة فى حاله مبيد كربوسلفان خلال موسمى الدراسه ، بينما ملاثيون أعطى درجه متوسطه من المقاومة فى الموسم الثانى فقط ، وقد أوضحت الأفراد الحقلية درجة مقاومة منخفضة لمبيد لمباداسيهالوثرين وكلوربيروفوس واميداكلوبريد خلال موسمى الدراسة .
وبناءا على معامل الحساسية يمكن أن يكون كلوربيروفوس فعال ضد المن بالمعدلات الموصى بها خلال موسمى الدراسة ، بينما يكون ملاثيون فعال فى الموسم الأ ول فقط ،وكان نشاط انزيم استيل كولين استيريز وألفا استريز وكذلك كمية البروتين الكلية مرتفعة فى حالة السلالة الحقلية عنها فى السلالة المعملية ، بينما كانت كمية الليبيدات عكس ذلك .
وعموما فان النتائج المتحصل عليها تدل على أن متابعة درجة مقاومة المن للمبيدات لها أهمية كبيره فى تطوير برامج المكافحة المتكاملة للحشرات فى حقول القطن.