MONITORING OF RESISTANCE TO SEVERAL ORGANOPHOSPHORUS AND CARBAMATE INSECTICIDES IN *APHIS GOSSYPII* (GLOVER)

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ABSTRACT

The susceptibility of two field strains of the cotton aphid, *Aphis gossypii* Glover collected from Fayoum and Gharbia Governorates in Egypt representing Upper and Lower Egypt during 2005-2007 to eight organophosphorus and four carbamate insecticides was determined using a slide-dip technique.

The results indicated that both strains showed very low levels of resistance to chlorpyrifos methyl, chlorpyrifos ethyl and carbosulfan. In contract, resistance to profenofos, dimethoate, cyanofos, and thiodicarb was very high. Moderate to relative high resistance was observed for pirimiphos methyl, malathion, prothiophose, methomyl and pirimicarb. Generally, Gharbia strain exhibited higher resistance to profenofos, dimethoate and malathion than Fayoum strain, while the resistance level of prothiophose in Fayoum strain was higher than in Gharbia strain, but no significant difference in levels of resistance between the two strains was observed in the other insecticides.

The toxicity index of tested insecticides against the two field strains was also determined during 2005-2007. With regard to organophosphorus compounds, chlorpyrifos methyl was the most toxic action followed by chlorpyrifos ethyl and then pirimiphos methyl, while the other insecticides were least toxic. As for the carbamates, carbosulfan showed the most toxic effect followed by methomyl, while thiodicarb and pirimicarb were the least in both strains.

As a general conclusion, the OP_S chlorpyrifos methyl and chlorpyrifos ethyl and the carbamate carbosulfan which showed very low levels of resistance are preferred pesticides for controlling cotton aphid.

INTRODUCTION

The cotton aphid, *Aphis gossypii* Glover is one of the most serious pests attacking cotton at the early and end of its growing season in many cotton producing areas of the world. Resistance to organophosphorus (OP) and carbamate insecticides in *A. gossypii* field populations has been reported by several authors (Gubran *et. al*., 1992, Kerns and Gaylor, 1992, - Furk and Vedjhi. 1990, Melia and Blasco, 1990, Saito *et.al*., 1995, Wu and Liu, 1994, Li-Fei *et. al*., 2003, Jhansi and Subbaratnam, 2005). In Egypt, several OP and carbamate compounds had been used for control of this pest in cotton since 1970 until 2000 such as monocrotophos, methamidophos, fenitrothion, pirimiphos methyl, dimethoate, cyanofos, pirimicarb and aldicarb. The carbamate carbosulfan and the neonicotinoid had been used against this pest since 1997 until now.

No report on monitoring of resistance to OP and carbamate insecticides in the cotton aphid, *A. gossypii* had yet been done in Egypt. Annual evaluation of resistance monitoring data on field population is needed to provide an adequate data base that would allow more flexibility in choosing an appropriate insecticide for control of pests. Thus the present study was conducted as a survey of resistance to several OP and carbamate insecticides in the two field populations of the cotton aphid *Aphis gossypii* Glover collected form Fayoum and Gharbia Governorate during the cotton seasons 2005-2007 to determine resistance levels of tested pesticides for recommendation of pesticides showed least levels of resistance.

MATERIALS AND METHODS

Samples of cotton leaves infested with *Aphis gossypii* Glover were collected from field of Fayoum and Gharbia Governorates representing Upper and Lower Egypt during early cotton season of 2005-2007. Slide-dipping technique was used to evaluate the toxicity of the tested insecticides against the adult stage. Serial concentrations of each formulated insecticide were prepared by dilution in water. By means of fine brush, ten adults were affixed to double face scotch tap and stuck tightly to slide on the dorsal part. The slides were then dipped in the prepared insecticide aqueous solutions for ten seconds. Each insecticide was tested at five different concentrations. Three replicates of ten adults each were used for each concentration. Mortality was recorded two hours after treatment and all insects responded to touching with the fine brush were considered alive.

Abbott formula (Abbott, 1925) was adopted and data were then subjected to statistical analysis by the method of Busvine (1957). The rates of resistance were expressed as resistance ratio (RR) at the LC_{50} level of the field strains as compared with the laboratory strain which has been reared in condition laboratory for more than 15 generations without exposed to any insecticides.

$$\text{Resistance ratio (RR))} = \frac{LC_{50} \text{ of the field strains}}{LC_{50} \text{ of laboratory strain}}$$

Also the toxicity index of each insecticide was determined according to Sun (1950) as follow:

$$\text{Toxicity index (TI)} = \frac{LC_{50} \text{ of the most effective insecticide X 100}}{LC_{50} \text{ of the least effective insecticide}}$$

Tested insecticides include the following:

a - Organophosphorus insecticides: chlorpyrifos ethyl, Dursban 48% EC (Dow Agro Sciences); chlorpyrifos methyl, Reldan 50% EC, (Dow Agro Sciences); profenofos, Curacron 72% EC (Novartis); pirimiphos-methyl, Actellic 50% EC (Syngenta); dimethoate, Cygon 40% EC (Wilbur-Ellis); prothiofose, Tokuthion 50% EC (Bayer CropScience); malathion, Malathion 57% EC (Vapco); cyanofos, Cyanox 50% EC (Sumotomo).

b - Carbamate insecticides: carbosulfan Marshal 25% WP (FMC); methomyl, Lannate 90% WP (DuPont); pirimicarb, Aphox 50% EC (Syngenta); thiodicarb, Larvin 80% WP (Bayer CropScience).

RESULTS AND DISCUSSION

The resistance ratios of the organophosphorus (OP) and carbamate insecticides tested against the two field populations of cotton aphid, *A. gossypii* collected from the two Governorates representing Upper and Lower Egypt (Fayoum and Gharbia, respectively) are presented in Tables (1, 2). Very low levels of resistance to the carbamate carbosulfan (Marshal); the OP_S chlorpyrifos methyl (Reldan) and Chlorpyrifos ethyl (Dursban) were detected in both of the two strains during 2005-2007 seasons. The resistance ratios ranged from 1.0 to 4.4-fold, except for Gharbia in 2006 (6.5-fold). These results indicate that each of these insecticides exhibited high effect against cotton aphid. The low levels of resistance to chlorpyrifos methyl and chlorpyrifos ethyl might be attributed to restricting use of chlorpyrifos methyl against aphid pests in vegetable and chlorpyrifos ethyl against cotton leafworm and cotton bollworm only. As for carbosulfan, although it has been used for control of cotton aphid in cotton sine 1997 until now, it showed low and stable level of resistance which were 1.0-1.9-fold for Fayoum and 2.3-3.5-fold for Gharbia. Above results are agree with those findings in other countries which showed also low levels of resistance to chlorpyrifos methyl in Japan (Hama *et. al.*, 1995) ; to chlorpyrifos ethyl in USA (Grafton-Cardwell and Goodel, 1996) ; carbosulfan in Israel (Ishaaya and Mendelson, 1987) and in Taiwan (Hsu *et. al.*, 2005).

With regard to the OP profenofos (Curacron), it has been used for control both cotton leafworm and bollworms only since 1978 until now. However, both aphid populations had apparently developed high levels of resistance. The resistance levels were 24.2 and 36.5-fold in 2005 and increased to 66.9 and 93.6-fold in 2007, for Fayoum and Gharbia populations, respectively. The same trend was also observed for resistance to the OP_S dimethoate (Cygon) and cyanofos (Cyanox), where dimethoate resistance increased from 22.4 and 25-fold in 2005 to 34.7 and 86.2-fold in 2007, While cyanofos resistance increased from7.6 and 10.1-fold in 2005 to 33.6 and 29.3-fold in 2007 for Fayoum and Gharbia populations, respectively although the use of dimethoate and cyanofos was stopped for controlling cotton aphid in cotton in 1992 and 2000, respectively. From these results, it is suggested that the development of resistance to profenofos, dimethoate and cyanofos in field populations of cotton aphid might be occurred through the application of the other OP compounds against this pest. The cotton aphid, *A. gossypii* is reported to have developed high degree of resistance to profenofos (Kerns and Gaylor, 1992) and to dimethoate (Pan-Wenliang *et.al.*, 1996 and Gubran *et.al.*, 1992).

High levels of resistance was also observed to the carbamate thiodicarb (Larvin) during 2005-2007 ranging between 33.0-39.3-fold except for Gharbia in 2006 which was 25.1-fold although this compound was used against cotton bollworm only and its use was stopped in 2004. This result might be due to the existence of cross-resistance between thiodicarb and the other OP and carbamate insecticides used against this pest.

In respect of insecticides which have been only used for control of aphid pests in vegetable in general, a moderate to relatively high resistance was found during 2005-2007. Field population of cotton aphid exhibited moderate levels of resistance to the OP pirimiphos methyl (Actellic) ranging between 5.9-12.1-fold. The same trend was also observed for the OP malathion (Malathion) and the carbamate methomyl (Lannate) but levels was higher and fluctuated from one season to another which ranged between 5.2-19.9-fold and 9.8-19.0-fold for malathion and methomyl, respectively. As for the carbamate pirimicarb (Aphox), cotton aphids also showed relatively high resistance levels but tended to increase from one year to another recording a maximum value of 20.4 and 17.9-fold in Fayoum and Gharbia strains, respectively. Also, Fayoum strain exhibited further increase in resistance to the OP prothiofose during three cotton seasons to reach a value of 15.9-fold, but Gharbia showed decline in resistance to a level of 6.4-fold. From these results, it may be assumed that there is a limited degree of cross-resistance between these insecticides and other OP and carbamate insecticides. The comparison of the present results with these of other reports on *A. gossypii* indicated the presence of resistance to pirimicarb (Nauen and Elbert, 2003; Delorme *et.al.*, 1997; Silver *et.al.*, 1995; Hama *et.al*., 1995); to methomyl (Hama *et.al.*, 1995). Saito *et.al.,* (1995) indicated that all six clones of *A. gossypii* showed moderate resistance to the OP insecticides malathion, fenitrothion, diazinon and dichlorvos (max., 45-fold) and the carbamates carbaryl, methomyl (max. 70-fold) except pirimicarb to which extremely high resistance was observed (1600-fold).

In comparing the levels of resistance to insecticides tested in Lower Egypt (Gharbia strain) to upper Egypt (Fayoum strain), results presented in Tables (1, 2) showed that the Gharbia strain recorded higher resistance to profenofos, dimethoate and malathion than the Fayoum strain, while the resistance levels of prothiofose in Fayoum were higher than in Gharbia strain. No significant difference in levels of resistance between the two stains was observed to the insecticides chlorpyrifos ethyl, chlorprifos methyl, pirimiphos methyl, cyanofos, pirimicarb, methomyl and thiodicarb.

With regard to OP compounds, chlorpyrifos methyl was the most toxic followed by chlorpyrifos ethyl and then pirimiphos methyl, while the other OP compounds were the least toxic against both strains during 2005-2007.

In general, the relative toxicity factors ranged between 61-87% for chlorpyrifos ethyl, 20-49% for pirimiphos methyl, while the other OP_S were less toxic which ranged between 3-26% during 2005- 2006, but in 2007 became far less toxic (relative toxicity ranged between 1-5%) except for profenofos (8%) and prothiofose (15%) as compared with the toxicity of chlorpyrifos methyl. Similar results were also reported by Ayad *et.al.*, 1991-1992 who found that chlorpyrifos ethyl showed the most toxic effect on Sharkia field strain of *A. gossypii* followed by pirimiphos methyl, malathion and dimethoate. Recently, Khalid *et.al.*, (2005) also found the same results on *A. gossypii* collected from eight Egyptian Governorates in cotton season 2004. In general, they indicated that chlorpyrifos ethyl was the most toxic insecticide followed by profenofos, dimethoate, pirimiphos methyl, prothiofose, malathion and cyanofos.

As for carbamate compounds, carbosulfan was the most toxic followed by methomyl, while thiodicarb and pirimicarb were less toxic against both stains during 2005-2007. The relative toxicity factor ranged between 12-27% for methomyl, 0.2-0.9% for pirimicarb and 0.1-0.3% for thiodicarb as compared with the toxicity of carbosulfan. Ayad *et.al*., 1991-1992 reported that methomyl was higher toxic than pirimiphos methyl on field strain of the cotton aphid. Also, Khalid *et.al*., 2005 found that carbosulfan was the most toxic followed by methomyl, while pirimicarb and thiodicarb were less toxic on field strain of *A. gossypii*.

The present study, here, revealed that the OP_S chlorpyrifos methyl and chlorpyrifos ethyl and the carbamate carbosulfan are more potent against cotton aphid, recording very low levels of resistance and high toxic action. In contrast, the OP_S profenofos, dimethoate, cyanofos and the carbamate thiodicarb are less potent, where they showed very high levels of resistance and very low toxic effect. On the other hand, the OP pirimiphos methyl and methomyl have moderate resistance as well as toxic action. The other insecticides (the OP_S malathion and prothiofose, the carbamates pirimicarb and thiodicarb) exhibited moderate levels of resistance associated with less toxic action. Therefore, the use of chlorpyrifos methyl, chlorpyrifos ethyl, and carbosulfan could be recommended for control of cotton aphid in resistance management programs.

Finely, careful evaluations of resistance monitoring data on field population of pest were needed to provide an adequate data base that would allow more flexibility in choosing an appropriate insecticide.

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الملخص العربى

تقصى مستويات المقاومة للعديد من المبيدات الفوسفورية والكرباماتية فى" من القطن"

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قدرت حساسية ثمانية مبيدات فوسفورية وأربعة مبيدات كرباماتية باستخدام طريقة غمرالشرائح على سلالتين حقليتين لحشرة "من القطن" والتى جمعت من حقول القطن فى محافظتى الفيوم والغربية واللتان تمثلا الوجة القبلى والوجة البحرى خلال مواسم زراعة القطن فى الفترة من 2005 الى 2007.

أوضحت النتائج فى كلا من السلالتين ظهورمستويات مقاومة صغيرة لمركبات كلوربيريفوس الميثيل ، كلوربيريفوس الايثيل وكاربوسلفان ، وفى مقابل ذلك ظهرت مستويات مقاومة عالية جدا لمركبات بروفينوفوس - ديمثويت - سيانوفوس وثيودايكارب ، كما لوحظت مقاومة متوسطة لمركبات بريميفوس ميثيل - ملاثيون- بروثيوفوس - ميثوميل وبريميكارب. ومن ناحية أخرى كانت سلالة محافظة الغربية أعلى فى المقاومة لمركبات بروفينوفوس- دايمثويت وملاثيون عن سلالة محافظة الفيوم ، بينما كان مستوى المقاومة لمركب بروثيوفوس فى سلالة الفيوم أعلى عن مثيلة فى سلالة الغربية ، ولكن لا توجد اختلافات معنوية فى مستويات المقاومة بين السلالتين فى بقية المبيدات الأخرى المختبرة

وقدرت أيضا معاملات السمية لهذه المبيدات على هاتين السلالتين الحقليتين خلال الفترة من 2005 الى 2007. ففى حالة المركبات الفوسفورية كان مركب كلوربيريفوس الميثيل هو الأكثر سمية يلية مركب كلوربيريفوس الايثيل وبعد ذلك مركب بريميفوس الميثيل بينما كانت بقية المركبا ت أقل سمية. وفى حالة المركبات الكرباماتية كان مركب كاربوسلفان هو الأكثر سمية يلية مركب ميثوميل ، بينما كان مركبى الثيودايكارب والبريميكارب أقل سمية فى كلتا السلالتين.

نستنتج من هذه الدراسة أن المبيدات الفوسفورية كلوربيريفوس الايثيل وكلوربيريفوس الميثيل والمبيد الكارباماتى كاربوسلفان ظهرت لها أقل مستويات مقاومة وعلى ذلك يمكن تفضيلها فى مكافحة من القطن عن بقية المبيدات المختبرة.

Table (1): Toxicity index and resistance to several organophosphorus insecticides in the cotton aphid, *Aphis gossypii* Glover collected from Fayoum and Gharbia Governorates during 2005 - 2007 growing seasons

		Fayoum					
Insecticide	**season**	**Slope ± EC**	**LC_{50} ppm (95 % FL.)**	**RR***	**TI****	**Slope ± EC**	**LC**
Chlorpyrifos methyl Reldan 50 % EC	Lab. strain	2.11± 0.33	69.5 (50.5 - 89.9)	----	----	2.11± 0.33	69.5
	2005	1.52 ± 0.28	177.7 (118.7 - 247.5)	2.6	100 %	2.64 ± 0.37	158
	2006	2.08 ± 0.39	132.1 (93.9 - 173.8)	1.9	100 %	2.07 ± 0.44	259
	2007	1.45 ± 0.28	95.6 (70.7- 130.2)	1.4	100 %	1.43 ± 0.56	199
Chlorpyrifos ethyl Dursban 48% EC	Lab. strain	1.69 ± 0.31	51.4 (31.6 - 71.2)	----	----	1.69 ± 0.31	51.4
	2005	1.88 ± 0.31	212..8 (149.9 - 281.5)	4.1	84 %	2.92 ± 0.99	198
	2006	1.49 ± 0.43	218.2 (67.9 - 340.5)	4.2	61 %	1.20 ± 0.26	333
	2007	1.43 ± 0.27	131.4 (84.9 - 191.6)	2.6	73 %	2.85 ± 0.38	228
Pirimiphos-methyl Actellic 50 % EC	Lab. strain	1.10 ± 0.27	71.5 (35.5 - 112.7)	----	----	1.10 ± 0.27	71.5
	2005	1.14 ± 0.37	861.7 (538.8 - 2544.4)	12.1	21 %	1.16 ± 0.89	421
	2006	1.83 ± 0.39	645.3 (455.9 - 968.3)	9.0	21 %	1.70 ± 0.38	527
	2007	1.98 ± 0.31	483.6 (352.1 - 631.7)	6.8	20 %	2.08 ± 0.31	548
Profenofos Curacron72% EC	Lab. strain	0.62 ± 0.11	28.1(12.0-60.1)	----	----	0.62 ± 0.11	28.1
	2005	0.93 ± 0.27	680 (393.7 - 1875.2)	24.2	26 %	3.48 ± 0.53	102
	2006	1.63 ± 0.41	781.5 (491 - 1154.2)	27.8	17 %	1.19 ± 0.26	902
	2007	0.70 ± 0.18	1879.8 (970.3 - 4993.6)	66.9	5 %	1.44 ± 0.29	263

RR * (Resistance ratio) = LC_{50} of the field strain / LC_{50} of the laboratory strain

TI** (Toxicity index) = (LC_{50} of the most effective insecticide / LC_{50} of the least effective insecticide) x 100

Cont. 1 table (1);

		Fayoum					
Insecticide	**season**	**Slope ± EC**	**LC_{50} ppm (95 % FL.)**	**RR***	**TI****	**Slope ± EC**	**LC**
Dimethoate Cygon 40 % EC	Lab. strain	0.51 ± 0.22	53.1 (23.9 - 120.7)	----	----	0.51 ± 0.22	53.1
	2005	1.28 ± 0.39	1191.6 (575.1 - 1878.7)	22.4	15 %	2.56 ± 0.43	137
	2006	1.45 ± 0.41	622.9 (282.5 - 942)	11.7	21 %	2.12 ± 0.41	222
	2007	2.10 ± 0.31	1840 (1409. - 2384.6)	34.7	5 %	1.73 ± 0.29	457
Prothiofose Tokuthion 50% EC	Lab. strain	1.39 ± 0.37	211.6 (138.2 - 370.8)	----	----	1.39 ± 0.37	211
	2005	1.34 ± 0.27	1569.8 (1068.4 - 2511.9)	7.4	11 %	1.28 ± 0.24	222
	2006	1.59 ± 0.56	2969 (1929 - 6053.3)	14.0	5 %	1.65 ± 0.28	126
	2007	1.87 ± 0.29	3359.1 (2542.5 -4606.7)	15.9	3 %	0.64 ± 0.19	135
Malathion	Lab. strain	0.91 ± 0.20	246.4 (85.6 - 466.3)	----	----	0.91 ± 0.20	246.4

Malathion 57% EC	2005	1.20 ± 0.30	2450 (1273.8 - 3900.4)	9.9	7 %	1.39 ± 0.29	475
	2006	0.94 ± 0.26	4026.7 (2311.4 - 15148)	16.3	3 %	1.97 ± 0.29	127
	2007	1.37 ± 0.27	2091.4 (1339.1 - 3007.2)	8.5	5 %	1.45 ± 0.28	490
Cyanofos Cyanox 50 % EC	Lab. strain	1.62 ± 0.28	240.4 (161.9 - 328.8)	----	----	1.62 ± 0.28	240.4
	2005	3.26 ± 0.49	1831.8 (1464.6 -2228.4)	7.6	10 %	1.43 ± 0.28	243
	2006	1.36 ± 39	6381.9 (3988.7 -10953)	26.5	2 %	1.51 ± 0.27	469
	2007	1.37 ± 0.27	8071.1 (5210.6 - 12847)	33.6	1 %	1.51 ±0.27	704

RR * (Resistance ratio) = LC_{50} of the field strain / LC_{50} of the laboratory strain

TI** (Toxicity index) = (LC_{50} of the most effective insecticide / LC_{50} of the least effective insecticide) x 100

Table (2): Toxicity index and resistance to several carbamate insecticides in the cotton aphid, *Aphis gossypii* Glover collected from Fayoum and Gharbia Governorates during 2005 - 2007 growing seasons

Insecticide	season	Fayoum					
		Slope ± EC	**LC_{50} ppm (95 % FL.)**	**RR***	**TI****	**Slope ± EC**	**LC**
Carbosulfan Marshal 25 % WP	Lab. strain	2.24 ± 0.39	12.0 (8.0 - 5.8)	----	----	2.24 ± 0.39	12.
	2005	1.24 ± 0.27	21.4 (15.0 -29.6)	1.8	100 %	1.74 ± 0.29	28.
	2006	2.24 ± 0.39	12 (8 - 15.8)	1.0	100 %	1.44 ± 0.28	41.
	2007	1.35 ± 0.29	22.8 (10.9 -34.4)	1.9	100 %	1.89 ± 0.39	36.
Methomyl Lannate 90% WP	Lab. strain	1.82 ± 0.34	10.2 (7.5 - 13.6)	----	----	1.82 ± 0.34	10.
	2005	1.50 ± 0.30	132.3 (78.1 - 188.1)	13	16 %	2.95 ± 0.69	193
	2006	1.82 ± 0.34	99.8 (51.4 - 151.5)	9.8	12 %	1.96 ± 0.39	163
	2007	1.30 ± 0.26	123.1 (154.3 - 348.7)	12.1	19 %	1.70 ± 0.29	133
Pirimicarb Aphox 50% EC	Lab. strain	1.14 ± 0.33	354.6 (108.4 - 645.3)	----	----	1.14 ± 0.33	354
	2005	1.16 ± 0.26	6066.3 (3895.8 - 10488)	17.1	0.4 %	1.55 ± 0.28	345
	2006	2.26 ± 0.60	5556.1 (3895 - 7545)	15.7	0.2 %	1.80 ± 0.38	479
	2007	1.80 ± 0.30	7235.2 (5593 - 10921)	20.4	0.3 %	1.79 ± 0.31	635
Thiodicarb Larvin 80% wp	Lab. strain	1.41 ± 0.28	484.3 (305.2 -709.4)	----	----	1.41 ± 0.28	484.
	2005	1.36 ± 0.37	15979 (10622.6 - 37068.3)	33	0.1 %	2.27 ± 0.61	185
	2006	2.08 ± 0.34	18211 (11293 -25137)	37.6	0.1 %	1.42 ± 0.57	121
	2007	1.36 ± 0.26	19019.4 (12340 - 43258)	39.3	0.1 %	1.23 ± 0.27	172

RR * (Resistance ratio) = LC_{50} of the field strain / LC_{50} of the laboratory strain

TI** (Toxicity index) = (LC_{50} of the most effective insecticide / LC_{50} of the least effective insecticide) x 100