SEROLOGICAL STUDIES ON THE RELATIONSHIP BETWEEN SOME EGYPTIAN CLOVER INSECT PESTS AND THEIR PREDATORS

BORAEI,H.A. * YOUSSEF. ASMHAN E., * E.M. EL-KADY AND A.A. FARAG*****

* *Econ. Entoml. Dept. Fac. of Agric. Kafr El-Saheikh Tanta Univ., Egypt.,*
** *Agric Botany. Dept. Fac. of Agric. Kafr El-Saheikh Tanta Univ., Egypt.,*
*** *Plant protect. Res. Ins., Sakha, Agric. Res.*

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Abstract

In the present work prey-predator relationships had been studied by three methods; analyzing statistically the correlation between population fluctuations of both groups; evaluating the feeding efficiency of the predators in the laboratory and studying the possibility of using serology in this respect. Throughout the two years of the study (2001-2003), at the experimental farm and Econ. Entomol. Dept. of Kafr El-Sheikh Faculty of Agriculture, it was found that the lacewing predator *Chrysoperla carnea* Steph. swarmed twice a year, The first peak (22 individuals.) which occurred in Dec. followed the appearance of *Aphis gossypii* Golve and *Spodoptera littoralis* Boisd at the beginning of the season. The second peak which occurred in Feb. or Mar. (19 individuals.) synchronized with the peaks of the alfalfa weevil, the green bug, aphids and the leafhoppers. Highly significant correlation was detected between that predator and the alfalfa weevil.

The ladybird *Coccinella undicempunctata* L. was found to occur in two peaks. The first was observed only in Dec. (8 individuals.) coincided with *A. gossypii* and *S. littoralis.* However, the second peak occurred in Mar. (40 individuals.) coincided with the alfalfa weevil, aphids and the leafhoppers, with high positive correlation recorded for them.

The three larval instars of *Ch. carnea* required duration of 7.75, 8.35, 8.45, and 9.80 days to develop to pupation on the cotton leafworm larvae, cotton leafworm eggs, alfalfa weevil larvae and the aphids, respectively. During all the larval stage, it consumed 83.50, 425.5, 88.o5, and 336.50 individuals., not less than 50% of them were consumed by the 3rd instar solely (55.69, 77.44 57.99, and 50.89%), respectively.

The ladybird predator devoured 694.00 of the cotton leafworm eggs during the shortest duration of larval stage (8.25 days). However, the less individuals it consumed from other insect pests of clover field (329.75 aphids, 229.75 cotton leafworm larvae, and 149.oo alfalfa weevil larvae), the longer duration periods were (8.30, 11.75, 13.45 days), respectively. The 4th instar consumed relatively the highest percentages of individuals offered for the four mentioned insects, as it consumed 42.95, 37.76, 5o.05 and 49.83%, respectively.

Serological studies carried out to evaluate the relationship between the antiserum of two predators, *Ch. carnea* and *C. undecimpunctata* against insect pests antigens the degree of

reaction shown in the agar of the double diffusion test. Results of double diffusion test revealed strong reactions detected for the larvae of the cotton leafworm (6 lines) indicated that it might be the most acceptable prey by the predator, *Ch. carnea* followed by aphids (4 lines) and the alfalfa weevil (3lines). Sharp reaction observed between the predator, *C. undecimpunctata* antiserum and the aphids (6 precipitin lines) and the cotton leafworm (4 lines). Reactions were less detectable with the other heterologous antigens. It is worthily mentioning that both of the two antisera when tested with the antigens of the other predators, no reactions were detected.

Finally those results suggest that serological techniques might be used in detecting the relationship between the predators and the insect pests, the precise and quick method which now widely used in insect researches and rarely used in Egypt.

INTRODUCTION

Egyptian clover (*Trifolium alexandrinum* L.) is one of the most important crop in Egypt. Egyptian clover is usually used as good diet sours of proteins for animals. On the other hand, clover is good reservoir for natural enemies and pests.

The interactions between insects and their predators are essential ecological processes that contribute regulation of insect populations (Dent, 1995). Among the common predators in most field crops are the chrysopid, coccinellid, staphylinid beetles and certain dipterous species. The study of population density of predators occurring in clover field, have been determined by many authors, Boraei *et al.* (1993), El-Agamy, 1996, El-Dakhakhni *et al.* 1995, El-Hawary *et al.* 1995, and Talha, 2001, receded some insect pests on clover fields. *Hypera brunneipennis* Boh., *Aphis gossypii* Golv., *Spodoptera littoralis* Boisd., *Nezara viridula* L., and *Empoasca* spp., are the most dominant ones and most of damage in clover fields are due to them.

Predation efficiency of *Ch. carnea* and *C. undecimpunctata* on the immature stages of the main insect pests of clover were studied by Ahmed (2000), Salem (2002), El-Shafei (2003) and Khalifa (2005).

However, during the ecological study on the alfalfa weevil, Boraei (1984) was able by using the agar double diffusion test to differentiate between different genera *Sitona Humeralis* F. and *Phytonomus variabilis,* collected from Hungary and *Phytonomus* collected from Egypt. He differentiated also between various defined physiological phases within one and the same species (newly emerged active adults and diapausing ones).

Many outhers suggest that serological techniques might be used in detecting the relationship between the predators and the insect pests. Pettrsson 1972, Ashby 1974, Mollet and Armabrust 1977. In the present work we are dealing with the

prey-predator relationships, so in the following we will throw a glance on serological studies carried out to investigate such interesting relationships among predators and their preys.

In Egypt, serology was seldom followed in entomological researches. Recently (1999) El-Kordy *et al.* used serology in this respect.

The objective of the present work was studying the prey-predator relationships in clover fields by the following methods :

- Population fluctuations of certain common predators associated with the insect pests in clover fields.
- Predation efficiency of the chrysopid, *Ch. carnea* Steph. The coccinellid, *C. udecimpunctata* L. and
- Serological studies on the relationship between some insect pests and their predators.

MATERIALS AND METHODS

1. Population fluctuations of two predators, *Ch. carnea*, *C. undecimpunctata* and associated insect pests :

Fields were planted in Kafr El-Sheikh on Oct. The experimental area was about half feddan. Sampling took place weekly all the season round, from Nov. till May, by the aid of sweeping net. Catch of each sample (50 double strokes) transferred to the laboratory in a plastic bag, where anaesthetized before identification and counting.

2. Predation efficiency of *Ch. carnea* and *C. undecimpunctata*

The two predators; the lacewing, *Ch. carnea* and the ladybird beetle, *C. undecimpunctata* were reared under laboratory conditions (26-31C and 65-75% R.H.), their feeding efficiency on three insect pests collected from clover fields (*A. gossypii, S. littoralis* eggs, larvae and *H. brunneipennis* larvae) were studied. Adults of the predators were collected from the clover fields, transferred to the laboratory. Each couple was confined in glass jar covered with black muslin sheet as site for oviposition, and furnished with paper to provide enough humidity. Eggs were collected and incubated until hatching. A piece of cotton soaked in sugar solution (10%) in small plastic container was placed inside for feeding.

Newly hatched larvae of *Ch. carnea* and *C. undecimpunctata* (64) were divided into four replicates and moved individually to Petri-dishes (10 cm) with filter

paper at their bottoms. Each replicate was provided daily with know number of the tested insect pests collected from clover fields . As the predatory instars progressed, number of each group introduced hosts were also increased. Number of devoured prey, by each larval instar of the predator, was calculated daily until pupation occurred.

3- Serological studies

Samples of the prevailing insect pests and associated predator which were used in the serological tests, were collected from clover fields. Insects of each sample were anesthetized with ether and was transferred to polyethylene bag (about 20 mg of fresh insects), well tied by rubber band; labeled and preserved shortly after collection, in -20C freezing unit.

The samples were crushed and ground in mortar to obtain fine powders. The powders were suspended in saline solution (1% Nacl). The suspensions (10 mg/ml) were transferred to beakers with few drops of sodium azide (0.2 ml). The solutions were refrigerated for at least 24 hr to dissolve soluble protein and then the homogenate was centrifuged for 15 min at 2500 rpm and the supernate poured through a millipore-sterilizing filter. The supernatants were obtained and used for serology tests or injection after mixing them with the adjuvant.

The antigens were prepared antigen A-*Ch. carnea* larvae and adults, collected from Jan. till May, antigen B-*C. undecimpunctata* larvae and adults, collected from Mar. to May, antigen 3-*Empoasca* spp. nymphs and adults, collected from Mar. till May, antigen 4-*N. viridula* nymphs and adults, collected from Feb. till May, antigen 6- *H. brunneipennis* larvae, collected in Feb. and Mar., antigen 14- *S. littoralis* larvae, collected during Nov. and May, antigen 18-*A. gossypii,* nymphs and adults, collected from Feb. to Apr.

Mature male New Zealand white rabbits were used for injection program and antisera preparation. Each rabbit received 10 injections, first one subcutaneous followed by a second interamuscular and so on, as follows: 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0, 5.0 and 6.0 ml, respectively. In the first injection, a mixture of complete adjuvant and insect extract (antigens A, B,) in the ratio of 1:1 was thoroughly emulsified in a vortex stirrer before injection into rabbits.

The rabbits were bled one week after the last injection, blood samples were incubated at 37C for two hr, then transferred to a refrigerator at 4C for 24 hr, to allow clot to separate from the serum. Obtained antiserum was centrifuged for 15

minutes at 2500 rpm in order to separate blood cells. Obtained clear antiserum was mixed with sodium azide. 0.2% and kept at -20C until using.

Two different antisera were antiserum A-*Ch. carnea* larvae and adults collected from Jan. till May, antiserum B-*C. undecimpunctata* larvae and adults, collected from Mar. till May.

Gel double diffusion test difico agar 1% was introduced on blaze for 30 min and poured in Petri-dishes (5.5 cm). Sodium azide (0.2%) was added to the gel before pouring. After solidification, four peripheral wells and one central were made in the gel. The central well was filled with the antiserum, whereas the others filled with the tested antigens. Each well was filled with 0.2 ml of the sample. Petri-dishes were kept carefully at 4C for one week, then the reactions were examined for detecting and counting the resulting precipitin (Boraei 1984).

Throughout the period of this study, chemical control was not applied. Data obtained were statistically analyzed using Duncan's Multiple test (Duncan, 1955).

RESULTS AND DISCUSSION

1. Population fluctuations of two predators, *Ch. carnea*, *C. undecimpunctata* and associated insect pests :

Data in Table (1) presented the monthly records of the two predators and associated insect pests in clover fields .

***Ch. carnea* and associated insect pests :**

The chrysopid, *Ch. carnea* started to appear by the beginning of Nov. in the season 2001-2002. Then a sudden increase occurred in Dec. declaring the first peak, represented by 22 individuals per 50 double strokes. This peak, which followed by relatively high numbers of the two pests, *A. gossypii* (19 individuals) and *S. littoralis* (20 individuals.) which recorded at the beginning of the season (Table 1).

The second peak of the predator was recorded in Feb. represented by 19 individuals, synchronized with the peaks of the two pests; *H. brunneipennis* (681 individuals.) and *N. viridula*, (18 individuals.), then occurred the second peak of *A. gossypii* (192 individuals.) in Mar. However, results of the second season took a different trend, the population of the predator *Ch. carnea* didn't appear significantly until Feb., with only one peak recorded in Mar., represented by 19 individuals. That peak was associated with the peaks of the three main pests; *H. brunneipennis*, (653 individuals.), *A. gossypii* (223

individuals.) and the leafhopper, *Empoasca* spp. (257 individuals.) (Table 1) in the same month.

As shown in the present study, the lacewing predator appeared in clover fields in Nov. and increased gradually to reach its highest peak in Mar., but sometimes another peak occurred early at the beginning of the season as happened in the first season (2001/2002) in Dec. Similar findings were recorded by many authors (Ali *et al.* 1982, Mesbah 1991, and El-Mezayyen 1998).

Statistical analysis in Table (2) showed that highly significant correlation was found between the lacewing and the alfalfa weevil in both seasons, as will as the leafhopper but only in the second season and negative correlation was found between the lacewing and the green stung bug in the second season. In this respect, the former authors (Ali *et al.* 1982) sustained that result, since they found that the high population density of the lacewing was associated with the highest peak of abundance recorded for the alfalfa weevil larvae. On the other hand, Moawed *et al.* in 1985 mentioned that the peak of that predator occurred at the end of the season in May coinciding with *S. littoralis*. Moreover El-Mezayyen (1998), recorded also two peaks of abundance, and showed that there was a high significant correlation between the predator and the green stink bug, *N. viridula.*

***C. undecimpunctata* and associated insect pests :**

Data of Table (1) indicated that ladybird insects were not found in clover fields during early season and if appeared, just in low density as occurred in 2002/2003 season, whenever only eight individuals were recorded in Nov. It began to appear in Feb. but in low densities, and by Mar. it reached abruptly to a distinct peaks represented by 41 and 40 individuals per 50 double strokes in the first and second season, respectively. However population densities didn't decline significantly until the season came to an end. The present results are similar to those recorded by many authors (Abd El-Galil *et al.* 1982 and El-Mezayyen 1993). In general, they recorded that important predator in clover fields were found increasing exponentially from Mar. or Apr., and reaching its peak in May in El-Tahrir and Kafr El-Sheikh area.

As far as relationships between the coccinellid predator and the associated pests are concerned, it was noticed that peaks of the predator were generally coincided with the occurrence of the peaks of the insect pests; *H. brunneipennis, A. gossypii, S. littoralis* and *Empoasca* spp. in the two successive studied seasons (Tables 1). Ali *et al.* (1982) found that the highest population of the ladybird was associated

with the highest peak of *H. brunneipennis* during Mar. However, high positive correlation values were recorded between the predator and both of the later two ones (*A. gossypii* and *Empoasca* spp.) and it was also positive for the alfalfa weevil but not significant. (Table 2). Negative correlation was found between *C. undecimpunctata* and population of *S. littoralis* and *N. viridula* in 2001/2002 season. In 2002/2003, positive correlation with between ladybird and all the considered pests, except the green stink bug. In this regard, Ali *et al.* (1982), found that the coccinellid population, as in the present work, was correlated with the population of the alfalfa weevil larvae. However it is worthily to mention that El-Mezayyen (1993) found that correlation was negative and insignificant, was found between *C. undecimpnctata* and *H. brunneipennis* Larvae.

2- Predation efficiency of *Ch. carnea* and *C. undacimpuntata*.

During the larval stage, which lasted for 9.80 days, the chrysopid predator consumed 336.50 individuals of aphids. Daily consumed were 12.64, 42.78 and 42.81 individuals in the first, second and third instars, respectively. Table (3). Many authors pointed to the efficiency of that predator on aphids, as El-Shafei who mentioned (2003) that it might consumed greater numbers of aphids (766.69 individuals.). Durations and feeding capacity of *C. undecimpunctata* larvae fed on *A. gossypii* nymphs are presented in Table (3). Throughout the total larval duration (8.30 days), the predator consumed 329.75 individuals. of aphids, compared with Salem (2002) who recorded lower number 323. Daily consumed aphids were 23.50, 28.88, 44.67 and 60.73 nymphs for the fourth instars, respectively. In similar work, the larval stage of *C. undecimpunctata* required an average of 234 aphids to complete its development, that was completed in about 6 days (Ahmed, 2000), Khalifa(2005) mentioned that it consumed greater number of aphid nymphs however, he Just counted 271.8 individuals.

As far as eggs of the cotton leaf-worm was offered to the predator, Ch. carnea, it was found that, 19.00, 77.00 and 329.50 eggs were consumed by the three larval instars which lasted for 2.25, 2.05, and 4.05 days, respectively (Table 4). The third instar was the most efficient as it consumed 77.44% of the total number of egg consumed. In this respect, Megahed (1982) counted 538.20 eggs, of S. littoralis.

However, C. *undecimpunctata* consumed more eggs (694.00) during shorter period (8.25 days) (Table 4). Daily consumed S. littoralis eggs were 41.14, 48.89, 107.00 and 132.44 eggs in the first, second, third and fourth instars, respectively.

Data presented in Table (5) showed duration of *Ch. carnea* larval instars and number of consumed the cotton leaf-worm 1st instar larvae by each of those instars, 1st lasted for 2.00 days during which, it consumed 12.0 larvae of the prey, 2nd lasted 2.50 days and consumed 25.0 larvae, while 3rd consumed throughout 3.25 days 46.50 larvae, which represented more than half (55.69%) of all larvae devoured by the predator.

As seen in Table (5), 229.75 of 1st instar larvae of the cotton leaf worm were consumed by of *C. undecimpunctata* larvae during 11.75 days. The 4th instar, solely, devoured 50.05% of them. El-Maghraby *et al.,* (1993) found similar results, as he concluded that 16.00 individuals were quite enough to feed on such number of egg and newly hatched larvae.

Data presented in Table (6) showed duration of the predator *Ch. carnea* larval instars. The three instars, which collectively lasted for 8.45 days, consumed 88.05 larvae; most of them were consumed by the third instar (51.05 larvae of the alfalfa weevil, 57.99% of all consumed larvae). Daily consumed of the alfalfa weevil were 6.00, 11.11 and 12.15 larvae for the three instars, respectively.

Durations and feeding capacity of *C. undecimpunctata* larvae fed on larvae of the Egyptian alfalfa weevil are shown in Table (6). Through out the total larval duration (13.25 days), the predator consumed 149.00 individuals, nearly three quarters (49.83%) of them were consumed by the fourth instar. Daily consumed weevil were 3.91, 6.55, 13.14 and 17.47 larval for the fourth instars, respectively.

3. Serological studies

Serological studies carried out to evaluate the relationship between the antiserum of two predators against insect pests antigens the degree of reaction shown in the agar of the double diffusion test.

Relationship between the predator, *Ch. carnea* and associated insect pests:

When antiserum of the predator *Ch. carnea* was tested against the antigen of the insect pests collected from clover fields positive reactions were obtained. Most acceptable insect might be the larvae of the cotton leafworm, hence strong reaction (6 precipitin lines) were detected, followed by aphids, the leafhopper, the alfalfa weevil and the green stink bug (5, 4, 3 and 2 lines, respectively). On the other hand, it gave negative reactions with the other heterologous antigens (*C. undecimpunctata, I. senegalensis, M. corollae.* Table (7) and Fig (1-2).

Relationship between the predator *C. undecimpuntata* and associated insect pests:

Data presented in Table (7) and illustrated in Figs (3-4) obviously indicated that the same trend was observed with antiserum of *C. undecimpunctata*, the sharp reaction, the clearest band (7 precipitin lines), was detected for the homologous antigen of *C. undecimpunctata,* (Fig. 29-30). Strong reactions between the ladybird predator and the two insect pests; *A. gossypii* and *S. littoralis*, represented by 6 and 4 precipitin lines, respectively. Indicated that they are preferred by the predator. Reactions were less detectable with the other heterologous antigen of the leafhopper and the alfalfa weevil, as only one single line was observed in the agar between antigen of each of them and the antiserum of the predator. These results agree with the findings of Pickavance (1972) who showed that the predators arthropods were feeding on aphids in the field gave positive precipitin reaction when tested against aphids. It was also recorded by Leathwick and Winterbourne in (1984) they found that the aphid antigen when tested with the antiserum *C. undecimpunctata* gave positive reaction.

Moreover, in case of compatibility (feeding), number of common antigens were detected between the predator and prey, and vise versa, no common antigens were observed in the case of incompatibility (no feeding). In our study, rabbits were immunized with 10 injections. However, Mollet and Armabrust (1977) used only 2 injections to produce antiserum against the alfalfa weevil adult and was able to detect strong reaction enough for a detectable reaction with a very high degree of specificity but as he found, it was not useful in field predation studies for the lack of reaction.

As a conclusion, we may suggest that agar double diffusion test, using unabsorbed antisera is quite efficient to differentiate between various insect species and in detecting the relationship between the predators and the insect pests, the precise and quick method which now widely used in insect researches and rarely used in Egypt.

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Table 1. Population fluctuations of two predators, *Ch. carnea, C. undecimpunctata* and associated insect pests in clover fields during the seasons, 2001/002 & 2002-003.

Date	PREDATORS		PESTS				
	Ch. carnea	*C. undecim-punctata*	*H. Brunneipennis*	*A. gossypii*	*S. littoralis*	*N. viridula*	*Empoasca* spp.
	L+A	L+A	L+A	A+N	L	A+N	A+N
Nov.	1	0	0	19	20	7	7
Dec.	22	0	7	15	6	10	13
Jan.	13	0	211	33	2	10	16
Feb.	19	8	681	152	6	18	45
Mar.	14	41	472	192	14	10	100
Apr.	5	38	85	80	0	15	300
May	2	25	10	7	0	4	70
Total	76	7112	1466	498	48	74	551
2002-2003							
Nov.	0	8	0	1o4	23	5	10
Dec.	3	0	6	354	15	5	50
Jan.	5	0	56	44	9	12	90
Feb.	10	4	592	82	10	4	209
Mar.	19	40	653	223	5	8	257
Apr.	18	35	145	18	0	14	30
May	0	31	3	9	0	20	1
Total	55	118	1455	834	62	68	647

N. Nymphs L . Larvae A. Adults

Table 2. Simple correlation coefficients between *Ch. carnea*, *C. undecimpunctata* and associated insect pests in clover fields during two successive seasons.

Pests / Predator	*H. brunneipennis*	*A. gossypii*	*S. littoralis*	*N. viridula*	*Empoasca* spp.
	2001-002				
Ch. carnea	+.496**	+.109	+.233	+.269	-.288
C. undecimpunctata	+.054	+.491**	-155	-.094	+.710**
2002/003					
Ch. carnea	+.528**	+.257	+.281	-.032	+.466**
C. undecimpunctata	+.259	+.203	+527*	+.290	+.106

* Significant (P= 0.05) **Highly significant (P=0.01)

Table 3. Larval duration and predation efficiency of *Ch. carnea* and *C. undecimpuctata* larvae fed on *A. gossypii* nymphs under laboratory conditions

Larval instar	Duration (days ±SD)	Consumed aphids (No. ±SD)	Consumed aphids %	Daily no. consumed
Ch. carnea				
1st	2.75 ± 0.96	34.75 ± 1.71	10.33	12.64
2nd	3.05 ± 1.29	130.50 ± 1.29	38.78	42.78
3rd	4.00 ± 0.82	171.25 ± 0.96	50.89	42.81
Total	9.80	336.50		
C. undecimpuctata				
1st	2.00 ± 0.82	47.00 ± 2.16	14.25	23.20
2nd	2.00 ± 0.82	57.75 ± 2.22	17.51	28.88
3rd	2.25 ± 0.46	100.50 ± 4.20	30.48	44.67
4th	2.05 ± 1.29	124.50 ± 4.20	37.76	60.73
Total	8.30	329.75		

Table 4. Larval duration and predation efficiency of *Ch. carnea* and *C. ndecimpuctata* reared on *S. littoralis* eggs under laboratory conditions.

Larval instar	Duration (days ±SD)	Consumed eggs (No. ± SD)	Consumed eggs %	Daily no. consumed
Ch. carnea				
1st	2.25 ± 0.50	19.00 ± 0.82	04.47	8.44
2nd	2.05 ± 1.29	77.00 ± 0.82	18.09	37.56
3rd	4.05 ± 0.58	329.50 ± 1.41	77.44	81.35
Total	8.35	425.50		
C. undecimpuctata				
1st	1.75 ± 0.95	72.00 ± 1.63	10.37	41.15
2nd	2.25 ± 1.50	110.00 ± 3.56	15.83	48.89
3rd	2.00 ± 0.82	214.00 ± 3.70	30.88	107.00
4th	2.25 ± 1.70	298.00 ± 4.32	42.92	132.44
Total	8.25	694.00		

Table 5. Larval duration and predation efficiency of *Ch. carnea* and *C. undecimpuctata* larvae fed on the cotton leaf worm 1st instar larvae under laboratory conditions.

Larval instar	Duration (days ± SD)	Consumed larvae		Daily no. consumed
		No. ± SD	%	
Ch. carnea				
1st	2.00 ± 0.82	12.00 ± 2.16	14.37	6.00
2nd	2.50 ± 1.29	25.00 ± 2.16	29.94	10.00
3rd	3.25 ± 0.96	46.50 ± 2.65	55.69	14.31
Total	7.75	83.50		
C. undecimpuctata				
1st	2.75 ± 0.96	12.25 ± 2.16	05.34	04.45
2nd	3.50 ± 0.58	31.50 ± 2.29	13.71	09.00
3rd	2.50 ± 1.29	71.00 ± 0.82	30.90	28.40
4th	3.00 ± 0.82	115.00 ± 3.56	50.05	38.33
Total	11.75	229.75		

Table 6. Larval duration and predation efficiency of *Ch. carnea* and *C. ndecimpunctat* reared on alfalfa weevil larvae .

Larval instar	Duration (days ± SD)	Consumed larvae		Daily no. consumed
		No. ± SD	%	
Ch. carnea				
1st	2.00 ± 0.00	12.00 ± 0.82	13.62	6.00
2nd	2.25 ± 0.69	25.00 ± 1.41	28.39	11.11
3rd	4.20 ± 0.50	51.05 ± 0.00	57.99	12.15
Total	8.45	88.05		
C. undecimpunctata				
1st	2.75 ± 0.92	10.75 ± 1.71	7.22	3.91
2nd	2.75 ± 1.71	18.00 ± 0.82	12.17	6.55
3rd	3.50 ± 1.29	46.00 ± 2.16	30.87	13.14
4th	4.25 ± 0.50	74.25 ± 1.71	49.83	17.47
Total	13.25	149.00		

Table 7. Precipitin lines detected with double diffusion test between *Ch. carnea, C. undecimpunctata* intserum and antigens of some clover insect pests .

Insect antigens	No. of Precipitin lines	
	Ch. carnea	*C. undecimpunctata*
Ch. carnea (A)*	8	0
C. undecimpunctata (B)	0	7
I. senegalensis (D)	0	0
Empoasca spp. (3)	4	1
N. viridula (4)	2	0
H. brunneipennis (6)	3	1
M. corollae (10)	0	0
S. littoralis (14)	6	4
A. gossypii (18)	5	6

*Refer to materials and methods to know the characters of a special antigen or antiserum expressed here, in the following figures, by a letter or number.

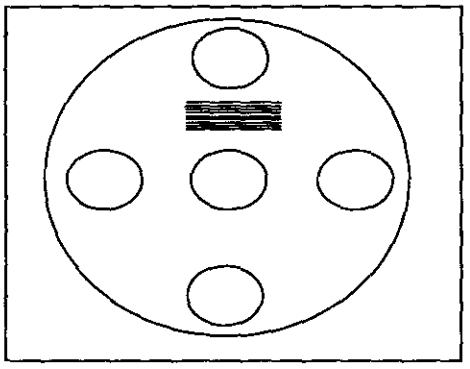

Fig.(1). Antiserum of *Ch. Carnea* (A) against antigens; *Ch.* carnea (a), *C. undecimpunctata* (b), *I. senegalensis* (d), and *M. corollae* (1)

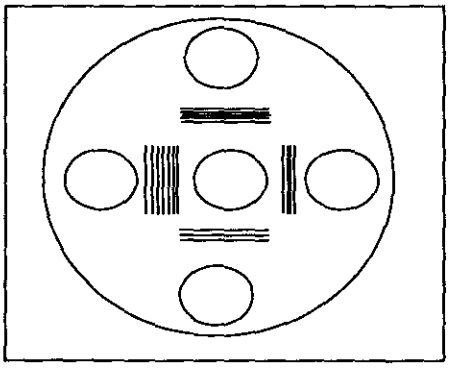

Fig.(2).Antiserum of *Ch. carnea* (A) against antigens; A. gossypii (18), *S. littoralis* (14), *H. brunneipennis* (6)and *Empoasca* spp. (3).

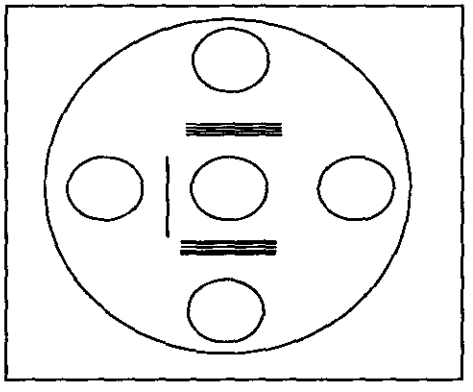

Fig. (3). antiserum of *C. undecimpunctata* (B) was diffused against the antigens; *S. littoralis*, (14), *A. gossypii* (18), and *N. viridula* (4).

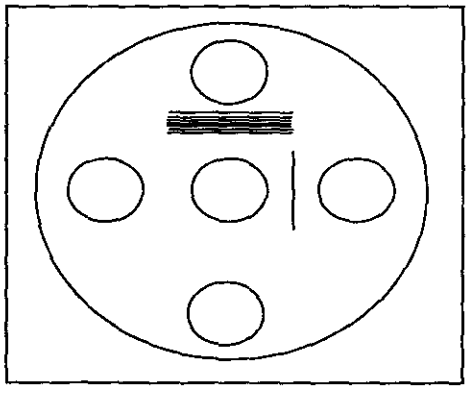

Fig. (4). antiserum of *C. undecimpunctata* (B) was diffused against the antigens; *C. undecimpunctata* (B), *I. senegalensis* (D), *Ch. carnea* (A), and *H. brunneipennis* (6).

دراسات سيرولوجية على العلاقة بين بعض آفات البرسيم المصري الحشرية ومفترساتها

حسين عبدالمنعم برعى * ، اسمهان السعيد يوسف* ، السيد مصباح القاضى ، عبدالسلام عبدالسلام فرج*****

* *قسم الحشرات الاقتصادية ، كلية الزراعة بكفرالشيخ ، جامعة طنطا*

** *قسم النبات الزراعى ، كلية الزراعة بكفرالشيخ ، جامعة طنطا*

*** *معهد بحوث وقاية النبات ، مركز البحوث الزراعية ، محطة سخا*

أجريت هذه الدراسة على مدى موسمين متتاليين ٢٠٠١/٢٠٠٢ و ٢٠٠٢/٢٠٠٣م فى كل من المزرعة البحثية وقسم الحشرات الاقتصادية بكلية الزراعة بكفرالشيخ وكان الهدف من البحث هو دراسة علاقة المفترسات بالآفات الحشرية فى حقول البرسيم المصرى وقد درست هذه العلاقة بثلاث طرق منها : دراسة التغيرات العددية لبعض المفترسات الحشرية وأهم الآفات الحشرية المصاحبة لها ، دراسة الكفاءة الافتراسية لنوعين من المفترسات الحشرية (أسد المن – أبو العيد ١١ نقطة) على بعض آفات البرسيم الحشرية ودراسة العلاقة السيرولوجية بين بعض المفترسات والآفات الحشرية المصاحبة لها. وقد تم الحصول على النتائج الآتية :-

١- التغيرات العددية لبعض المفترسات والآفات الحشرية المصاحبة لها والعلاقة بينهم .

تمت هذه الدراسة باستخدام شبكة الكنس ٥٠ ضربة مزدوجة أسبوعيا على نوعين من المفترسات الحشرية الهامة ، وخمسة من الآفات الحشرية المصاحبة لها والمفترسات التى تم اختيارها (*Chrysoperla. carnea, Coccinella. undecimpunctata,*) أما بالنسبة للآفات الحشرية الخمسة فهى كالآتى :

(*Hypera. brunneipennis, Aphis. gossypii, Spodoptera. littoralis, Nezara. viridula* and *Empoasca* spp.)

يظهر أسد المن فى بداية شهر نوفمبر وكان له ذروتان خلال الموسم الأول من الدراسة الذروة الأولى فى ديسمبر بتعداد ٢٢ فرد والذروة الثانية فى فبراير بتعداد ١٩ فرداً عقب أعلى زيادة فى أعداد سوسة ورقة البرسيم والبقة الخضراء فى فبراير والمن فى مارس . كما بلغ التعداد أقصاه فى الموسم الثانى فى مارس حيث كان ١٩ فرد فى نفس وقت زيادة سوسة ورق البرسيم والمن والجاسيد ، وقد وجد ارتباط معنوى عالى بين المفترس ويرقات سوسة ورق البرسيم ، وعلاقة موجبة مع المن خلال موسمى الدراسة وسالبة مع البقة الخضراء فى الموسم الثانى.

ظهر أبو العيد ١١ نقطة فى بداية شهر فبراير الى مايو ، وكان أعلى تعداد له فى شهر مارس ٤١ فرداً ، وجد ان زيادة أعداد مفترس أبو العيد ١١ نقطة ارتبطت مع زيادة أعداد المن فى نفس الشهر فى الموسم الأول من الدراسة . أما فى الموسم الثانى من الدراسة فقد وجد أن له ذروة واحدة فى شهر مارس تتزامن مع سوسة ورق البرسيم والمن والجاسيد . وقد لوحظ ارتباط معنوى قوى جداً بين أبو العيد والمن

والجاسيد وعلاقة سالبة مع البقة الخضراء فى الموسم الأول أما فى الموسم الثانى العلاقة تكون موجبة مع جميع الآفات ما عدا دودة ورق القطن كان الارتباط معنوى.

٢- الكفاءة الافتراسية لمفترس أسد المن وأبى العيد ١١ نقطة على آفات البرسيم المصرى الحشرية .

عند تغذية مفترس أسد المن على حوريات من القطن معملياً اكتمل الطور اليرقى للمفترس فى خلال ٩,٨ يوماً استهلك خلالها ٣٣٦,٥ فرداً ، وأن الطور اليرقى الثالث بلغت مدته ٤ يوم استهلك خلالها ٥٠,٨٩% من جملة أعداد المن المفترسة. أما عند تغذيته على بيض ويرقات دودة ورق القطن وجد له ثلاثة أعمار يرقية استغرقت ٨,٣٥ و ٧,٧٥ يوماً افترس خلالها ٤٢٥,٥ بيضة ٨٣,٥ يرقة ، ووجد أن العمر اليرقى الثالث ومدته ٤,٥ و ٤,٢ يوماً يستهلك حوالى ٧٧,٤٤ و ٥٧,٩٩% من جملة عدد البيض واليرقات المفترسة.

عند تغذية يرقات مفترس أبى العيد على حوريات من القطن معملياً اكتمل الطور اليرقى للمفترس فى خلال ٨,٣ يوم استهلك خلالها ٣٢٩,٧٥ فرد بلغ العمر اليرقى الرابع ٢,٠٥ يوم استهلك خلاله ٣٧,٧٦ % من مجموع أعداد المن المستهلكة عند تغذيته على بيض ويرقات دودة ورق القطن اكتمل العمر اليرقى للمفترس فى ٨,٢٥ و ١١,٧٥ يوم استهلك خلالها ٦٩٤ بيضة و ٢٢٩,٧٥ يرقة على التوالى .

عند تغذية يرقات مفترس أبو العيد على الفقس الحديث لسوسة ورق البرسيم اكتمل العمر اليرقى للمفترس فى ١٣,٢٥ يوماً استهلك خلالها ١٤٩ يرقة وبلغ العمر اليرقى الرابع للمفترس ٤,٢٥ يوماً استهلك خلاله ٤٩,٨٣% من مجموع أعداد اليرقات المستهلكة.

٣- الدراسات السيرولوجية

تم دراسة العلاقة السيرولوجية بين نوعين من المفترسات الحشرية والآفات المصاحبة لها عن طريق بعض من الاختبارات السيرولوجية الهامة الشائعة (اختبار الانتشار فى الأجار). ويتم ذلك بصب آجار نقى ساخن فى طبق بترى (٥,٥مم) ، وبعد التصلب يتم عمل حفرة واحدة فى منتصف الطبق يوضع بها أنتسيرم الحشرة ، أربع حفر على جوانب الطبق يوضع بها أنتيجين الحشرات أى حوالى ٠,٢ مم فى كل حفرة سواء للأنتسيرم أو الأنتيجين بحيث تكون المسافة بين كل حفرة وأخرى متساوية ، وحفظة فى الثلاجة لمدة ٧-١٠ يوماً فيظهر التفاعل فى صورة خطوط ترسيبية وتظهر النتائج كالآتى:

١- العلاقة بين مفترس أسد المن وأبى العبد ١١ نقطة وبعض آفات البرسيم المصرى الحشرية:

فى حالة اختبار انتسيرم أسد المن ***(A)*** مع أنتجينات آفات البرسيم (المن ، نطاطات الأوراق ، البقة الخضراء ، سوسة ورق البرسيم ، دودة ورق القطن) أظهر التفاعل ترسيب موجب ويتكون عدد من الخطوط الترسيبية (٥ ، ٤، ٢ ، ٣ ، ٦ خطوط ترسيبية على التوالى) وهذا يدل على أن المفترس يتغذى على آفات البرسيم تلك الموجودة فى الحقل . أما عند اختبار انتسيرم أبو العيد ١١ نقطة مع أنتجينات نفس الآفات (المن ، نطاطات الأوراق ، البقة الخضراء ، سوسة ورق البرسيم ، دودة ورق القطن) يتكون عدد من الخطوط الترسيبية ٦ ، ١ ، ٠، ١ ، ٤ خط ترسيبى على

التوالى دليل على وجود علاقة بين هذا المفترس والآفات بمعنى أن ابوالعيد ١١ نقطة يتغذى على هذه الآفات فى الطبيعة ، ولكن وجد علاقة ضعيفة مع هذا المفترس والبقة الخضراء ونطاطات الأوراق. ومن هذه الدراسة نستنتج الاتى : المفترسات الحشرية هامة جداً فى برامج المكافحة الحيوية دليل ارتباطها القوى مع بعض آفات البرسيم المصرى فى الحقل والتغذية عليه بكفاءة فى المعمل. دراسة العلاقة بين الآفات الحشرية ومفترساتها بالاختبارات السيرولوجية تعتبر طريقة سريعة وسهلة لمعرفة علاقة المفترس بالآفة عند مقارنتها بالطرق الأخرى. ولذلك يوصى باستخدامها عند دراسة هذه العلاقة لدقتها وسرعة إنجازها عند أخذ المفترسات فى الاعتبار فى برامج المكافحة المتكاملة للآفات.