

Evaluation of Releasing the Predator, *Hippodamia convergens* (Geur.) (Coleoptera: Coccinellidae) against the Cotton Aphid, *Aphis gossypii* Glover, Infesting Squash Plants under Semi-field Conditions

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ABSTRACT

After being reared in laboratory on artificial diet, efficacy of the predatory species *Hippodamia convergens* (Geur.) (Coleoptera: Coccinellidae) was evaluated against the cotton aphid *Aphis gossypii* Glover under semi-field conditions. Adults and 3rd instar larvae were released at two different rates (80 and 120 individuals) against *A. gossypii* on squash plants in a screen house. Results showed that the mean relative population densities of the aphid throughout the experiments were; 6761.75, 2114.71, 1765.08, 1614.58 and 1382.08 in case of control; release of 80 and 120 larvae and release of the same numbers of adults, respectively. Reduction percentages of aphid population in case of releasing 80 and 120 larvae and those of adults compared to the control were; 67.25, 72.45, 76.73, and 79.27%, respectively. Highest reduction percentage (79.27%) was obtained after releasing *H. convergens* at a rate of one adult predator : 73 prey individuals. The results indicated that *H. convergens* is a potential predator that can be, successfully, used for biological control of *A. gossypii*.

Key words: Coccinellidae, *Hippodamia convergens*, *Aphis gossypii*, Squash, release, semi-field conditions.

INTRODUCTION

Squash (*Cucurbita pepo* L.), is one of the most important vegetable crops in Egypt. Squash plants are attacked by many insect pest species, of which the cotton aphid, *Aphis gossypii* Glover was considered the most dominant pest species (El-Hosiny, 2001 and Nyoike and Liburd, 2010). It has a wide host range and its infestation leads to severe damages to the attacked host plants (Abou-Elhagag and Salman 2001, and Wu *et al.*, 2010). Damage may be caused directly by sucking plants' juice or indirectly by transmitting plant viruses.

Repeated use of pesticides has led to development of insects' resistance to insecticides and destruction of the natural balance between beneficial natural enemies and target pests. That directed the current researches toward development of integrated management strategies in which the biological control methods represent a main component (Schirmer *et al.*, 2006). Insect predators are of the major groups of biological control agents used for aphids' control. Those belong to family Coccinellidae, comprise one of the most active groups of predatory species, that feed during the larval and adult stages on different sap-sucking pests including aphids, whiteflies, jassids and mites as well as other small insects. This family gained an interested role as important group of predators in the biological control of insect pests attacking different crop plants. Many coccinellid species were recorded associated with the pests in fields of different economic crops (Shalaby *et al.*, 2008).

Many attempts have been made by releasing some biocontrol agents, particularly common coccinellid species for controlling aphid species. Vinson and Scarborough (1989) in USA demonstrated that releasing of adults and third instar larvae of the coccinellid predator, *Hippodamia convergens* (Geur.) were effective in reducing the population of *A. gossypii*, in cotton fields. In additions, Dreistadt and Flint (1996) found that releasing *H. convergens* adults for controlling *A. gossypii*, infesting outdoor potted chrysanthemum, at the rate of 34-42 adults / pot, provided 25-84% aphid control. Each beetle consumed 25-170 aphid individuals /day. There was a density-dependent functional response in predation, where predatory beetles consumed more aphids when released on plants with higher aphid densities. They concluded that, release of *H. convergens* could provide augmentative control of relatively high aphid densities on small potted plants.

The present study aimed to evaluate the efficacy of the predator *H. convergens*, reared on artificial diet, against the cotton aphid, *A. gossypii* under semi-field conditions.

MATERIALS AND METHODS

Stock culture of *A. gossypii*

A. gossypii individuals were collected from infested cotton fields at Qalubiya Governorate Egypt. A stock culture of the aphid species was maintained in the laboratory on squash young plants grown in plastic pots (no. 20). Pots were placed in a

screen cage (2.5 x 2.0 x 2.0 m.) kept under the natural weather conditions. The following technique was followed:-

- 1- Three grains of squash (Skandarany variety) were planted in the plastic pots containing clay and sand (1:1).
- 2- Three weeks after seeds' germination, squash plants were infested by 50 apterous adults of *A. gossypii* for each pot.
- 3- Nymphal instars of *A. gossypii* were used for infestation in the semi-field experiments.
- 4- Aphid individuals of the stock culture were used for the semi-field releasing predators' experiments.

Rearing of *H. convergens*

H. convergens adults were collected from cotton fields at Qalubia Governorate. They were mass reared as described by Bahy El-Din (2013) on a selected artificial diet (out of other three experimentally tested artificial diets). This diet consisted of : dried yolk of eggs (20.48%) , dry powdered aphids (3.18%), fresh homogenized aphids (0.76%), sucrose (59.52%), powdered yeast (3.62%), pollen grains (5.13%), maize oil (1.16%), royal Jelly capsules (4.21%), multi-vitamins (1.63%) and streptophenicol (1.47%). The predator individuals were released after approximately 7 days from adults' emergence (this period represents the pre-ovipositional period during which the feeding capacity of females was found higher than during the pre-ovipositional and post ovipositional periods (El-Heneidy *et al.*, 2008).

Semi field experiment of *A. gossypii*

A semi field experiment (using screen houses 2.5 x 2.0 x 2.0 m) was carried out at the Department of Biological Control, Plant Protection Research Institute, Agricultural Research Center, Giza, Egypt in season 2011. The experiment started on May, 25th 2011 (planting date) and continued till July, 27th 2011. *H. convergens* (3rd instar larvae or 7 days old adults) were released once for controlling the aphid on squash plants. Experiments were carried out as follow:-

- 1- Fifteen screen houses were used, represented five different applications at three replicates each.
- 2- Five rows contained 30 squash plants (=one replicate). A distance of 30 cm. was left between plants, while that between rows was 50 cm.
- 3- All agricultural practices were followed regularly, except for the absence of any chemical insecticidal treatment.
- 4- An artificial infestation with *A. gossypii* was made on June, 24th 2011. A total of 1800 aphid individuals were handily distributed, by the aid of a fine camel brush, on the 30 plants in each experimental plot (60 aphid individuals/ plant).

- 5- Four release applications of the predatory species *H. convergens* ; (80 and 120 third instar larvae and 80 & 120) adults (sex ratio of adults was 1: 1) were carried out at each experimental plot.
- 6- Control treatment was kept free from any release.
- 7-The predatory stages (larvae or adults) of *H. convergens*, were released only once on July, 3rd 2011 (*i.e.*, 12 days after artificial infestation of the squash plants with aphids in all of the fifteen experimental tested plots).
- 8- Inspection of 10 plants/plot was carried out at three day intervals (3 leaves/ plant; *i.e.*, 30 leaves for each of the fifteen experimental plots).
- 9- Total counts of aphid individuals on the examined leaves were recorded, and percentage of reduction in the aphid population was estimated, compared to the control.
- 10-Total numbers of the *H. convergens* feeding stages (larvae or adults), were also recorded.
- 11- Differences among data obtained from the four releasing rates of the predator were statistically analyzed, using ANOVA and SAS (1988) comparing to the control.

RESULTS AND DISCUSSION

Data presented in tables (1 and 2) summarize the semi-field experimental results carried out in 2011 summer plantation of squash, at Giza Governorate.

Population density of the cotton aphid *A. gossypii*

As indicated in table (1), the artificial infestation with *A. gossypii* adults and nymphs was made on June, 24th 2011, one month after sowing, with 60 aphid individuals/squash plant. The population increased gradually until it reached 8819; 9076; 8742; 8763 and 9097 individuals in the plots of releasing at 80 and 120 larvae; 80 and 120 adults and control, respectively on July, 3rd 2011 (just before predator's release).

Average numbers of *A. gossypii* during the period extended from June, 24th 2011 (just after release) till July, 27th 2011 (at the end of the experiment) are presented in table (1). Overall means of total numbers of the aphid populations were; 2114.17±752 and 1765.08±739.8 for larval treatments, 1614.58±713.8 and 1382.08±717 for adult treatments and 6761.75±1557.5 aphid individuals/ 10 plants for the control, in case of releasing 80 and 120 larvae, 80 and 120 adults of *H. convergens* and control, respectively.

The releasing rate was expressed as ratio between the total numbers of released predator's stage: the total number of counted aphids at the time of release. Accordingly, releasing the predatory adults at the rate of 80 individuals was expressed as

Table (1): *A. gossypii* counts /10 squash plants (30 leaves) before and after releasing *H. convergens* adults and larvae, at two releasing rates (80 and 120 individuals) at Giza in summer 2011

Inspection date	No. of predator's larvae		No. of predator's adult		Control <i>A. gossypii</i>	L.S.D.
	80	120	80	120		
24/6/2011	600	600	600	600	600	-
27/6	1362	1407	1379	1349	1418	-
30/6	3809	3938	3801	3816	3944	-
3/7 *	8819 A	9076 A	8742 A	8763 A	9097 A	155.90
6/7	5452 B	3600 C	3212 C	1736 D	11177 A	77.69
9/7	1467 B	1038 B	921B	221 C	14073 A	68.82
12/7	2793B	1387 C	570 D	0 D	15852 A	78.92
15/7	945 B	115 C	150 C	0 C	11229 A	65.34
18/7	109 B	20 B	0 B	0 B	9004 A	31.61
21/7	14 B	0 B	0 B	0 B	3937 A	23.57
24/7	0 B	0 B	0 B	0 B	563 A	4.43
27/7	0 B	0 B	0 B	0 B	247 A	4.21
Total	25370	21181	19375	16485	81141	-
Seasonal mean	2114.17 ± 752.05	1765.08±739.79	1614.58±713.76	1382.08±717.01	6761.75±1557.52	-
% Reduction	67.25	72.45	76.73	79.27	0	-

* Date of release Means in the same row not followed by the same letter are significantly different (P< 0.05) using L.S.D. in SAS.

1 predatory adult: 109 aphid individuals, while releasing of 120 adults was expressed as 1: 73. Similar ratio for releasing *H. convergens* larvae at two rates (80 larvae; i.e. 1 larva : 110 aphid individuals, and 120 larvae; i.e. 1 larvae: 76 aphids) were followed.

After releasing 80 and 120 adult stage on July, 3rd 2011, the population of aphids decreased gradually until the infestation disappeared (zero) on July, 18th 2011, 15 days post of the release of at 80 adults. While the infestation disappeared (zero) on July, 12th 2011, 9 days post of the release of 120 adults.

The previous data showed that the feeding capacity of releasing 120 adults was greater than 80 ones. Statistical analysis revealed highly significant differences between seasonal mean numbers of *A. gossypii* in the control and each of the release of 80 and 120 adults. While, there were significant differences in aphids' seasonal means between the two releasing treatments.

After releasing the 3rd instar larvae, at the two different rates, the population of aphids reached 8819 and 9076 individuals /10 plants at the time of release, decreased successively until disappeared (zero) on July, 24th and 21st 2011, 21 and 18 days post release of 80 and 120 larvae, respectively. Data presented in table (1) showed that the number of devoured aphids by *H. convergens* larvae increased as the number of released larvae was increased, and consequently aphids' infestation disappeared earlier in the plot which received the higher number of larvae (120) than the other plot. Thus it was

confirmed that decreasing the predator: prey ratio (increasing number of released larvae) led to better control of *A. gossypii*. Statistical analysis among the five treatments showed highly significant differences between the seasonal mean numbers of aphid in the control and those in the releasing plot (80 and 120 larvae and adults). Also, there were significant differences between the seasonal mean numbers of aphid in the two releasing areas. In addition, there were significant differences between releasing 80 adults and 80 larvae. While, the difference between seasonal mean aphid counts in releasing plot of 80 adults and that of 120 larvae was non-significant.

The mean percentages of reduction of aphid population in case of releasing 80 and 120 larvae; 80 and 120 adults compared to the control were, 67.25; 72.45; 76.73; and 79.27%, respectively. These results indicated that highest reduction percentage was obtained in case of releasing 120 adults (one adult predator: 73 aphid individuals).

Also, obtained data in table (1) revealed that, by increasing the total numbers of released *H. convergens* individuals (larvae or adults), the percentages of aphid reduction were also increased. i.e.; a direct relationship was obtained between the total numbers of released predator (larvae or adults) and the obtained percentages of aphid reduction. The obtained data showed, also, that *H. convergens* adults were of higher potential than larvae in controlling *A. gossypii*.

In agreement with the present results, El-Heneidy *et al.* (2008) stated that, feeding capacity of the larval stage of *H. convergens* was; 1107.43±9.61,

Table (2): Population density of *H. convergens* (adults and larvae) / 10 plants (30 leaves) post releasing of the predator against *A. gossypii* on squash plants at Giza, season 2011

Inspection date	No. of predator's larvae						No. of predator's adults					
	80			120			80			120		
	L	A	Total	L	A	Total	L	A	Total	L	A	Total
3/7 *	25	0	25	34	0	34	0	33	33	0	51	51
6/7	23	0	23	26	0	29	0	27	27	0	40	40
9/7	4	0	4	7	0	7	37	17	54	83	28	111
12/7	0	3	3	0	6	6	3	5	8	8	15	23
15/7	0	7	7	0	18	18	0	0	0	0	8	8
18/7	0	7	7	0	11	11	0	0	0	0	0	0
21/7	0	6	6	0	9	9	0	0	0	0	0	0
24/7	3	0	3	3	0	0	0	0	0	0	0	0
27/7	0	0	0	0	0	0	0	0	0	0	0	0
Total/season	55	23	78	70	44	114	40	82	122	91	142	233
Seasonal mean	5.50 ± 2.96	2.30 ± 0.95	7.80 ± 2.67	7.00 ± 3.74	4.40 ± 1.92	11.40 ± 3.62	4.00 ± 3.49	8.20 ± 3.82	12.20 ± 5.75	9.10 ± 7.83	14.20 ± 5.71	23.30 ± 10.81
L.S.D.	3.09											

* Date of release of the predator *H. convergens*, L (larvae), A (adults).

786.33±8.60, 533.20±5.65 and 370.25±3.79 of *A. craccivora* nymphs. Total feeding capacity of adults was the highest during the ovipositional period; it ranged between 207-263 nymphs/adult/day. Dong (1988) reported that on cotton plants, aphids could be successfully controlled by the predator *Harmonia oxyridis* Pall., at a predator: prey ratio of 1: (100-120), without the need to apply the chemical insecticides. Ghanim and El-Adl (1991) studied the predation of *C. undecimpunctata* on *Schizaphis graminum* in field cages in Egyptian wheat fields. For 1 larva of the predator to 80-100 aphids, aphid numbers were reduced by 31.75-32.80% and economic injury level to the crop averaged 17.04-18.88%. The introduction of predator larvae at 1: every 40-50 aphids gave corresponding figures of 89.71-91.28 and 2.14-3.06%, while introduction at 1 predator: 20- 25 aphids gave 88.19-89.77 and 3.21-4.08%. El-Arnaouty *et al.* (2000) studied the effect of releasing *H. axyridis* (larvae and adults), for the biological control of the cowpea aphid *A. craccivora*. Larvae were released as third instar at two different rates of predator: prey; *i.e.*, 1 / 30 and 1 /50 aphid individuals in a faba bean field. Results showed that the predator *H. axyridis* was able to control the aphid species and the best rate of release was 1 predator / 30 aphids /plant. Zibai and Hatami (2001) determined the efficacy of third larval instar of the predator *H. variegata* against *A. gossypii*, on cucumber plants under greenhouse conditions. The predator: prey ratios of 1:30 and 1:90 reduced significantly the population of the aphid.

Numbers of predatory stages through the experiment

Data in table (2) represent total numbers of *H. convergens* larvae and adults during the period July, 3 to 27 2011. Total numbers were; 55, 70, 40 and 91 larvae and 23, 44, 82 and 142 adults, in case of the release 80 and 120 larvae and 80 and 120 adults, respectively. The numbers of larvae counted/10

plants at the same day of release were 25 and 34 larvae after the release of 80 and 120 larvae respectively. Correspondent numbers of adults were 33 and 51adults. The numbers decreased in the subsequent counts until no adult was found on July, 15th and 18th in the plots of the release at 80 and 120 adults, respectively. Absence of larvae was, mostly, attributed to their develop to pupal stage, while absence of adults was attributed to the absence of their prey (*A. gossypii*). In the plot of releasing larvae, *H. convergens* adults appeared by 3 and 6 individuals on July, 12th in plots of releasing 80 and 120 larvae, respectively. These adults remained on squash plants until July, 21st and disappeared on July, 24th when larvae of the predator started to appear again.

Also, in the plot of releasing adults, *H. convergens* larvae were 37 and 83 larvae on July, 9th. These larvae were, mostly, resulted from eggs deposited by the released adults. Presence of larvae continued up to July, 12th (3 and 8 larvae/ 10 plants). While larvae disappeared on July 15th when *A. gossypii* individuals were consumed by the released predator.

Statistical analysis among the five treatments showed that, highly significant differences existed between the control and each of the releasing rate (80 and 120) larvae or adults. There were significant differences also between the release of 120 adults and other treatments. There were significant differences between releasing of 80 adults and 80 larvae. While, the difference between releasing of 80 adults and 120 larvae was insignificant.

These results also indicated that, releasing the predator *H. convergens* at 120 adult individuals gave higher control rate against the cotton aphid than in the other treatments. This may be due to that the adult females deposited eggs that hatched to larvae

consumed high numbers of the prey.

Generally, the predator *H. convergens* seemed to be a potential predator that can be used for controlling the cotton aphid *A. gossypii* in squash fields or any other fields that are subjected to be attacked by this pest. The recommended ratio is 1 predator: 73 aphid individuals.

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