Asian Journal of Biology

14(3): 14-23, 2022; Article no.AJOB.84633
ISSN: 2456-7124

Releasing Two Important Insect Predators to Control of Aphids under Open-field and Greenhouse Conditions

Mohamed Ahmed Gesraha [a*†] and Amany Ramadan Ebeid [a]

[a] Pests and Plant Protection Department, Agricultural and Biological Researches Institute, National Research Centre, Cairo, Egypt.

Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AJOB/2022/v14i330215

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/84633

Original Research Article

Received 12 January 2022
Accepted 16 March 2022
Published 22 March 2022

ABSTRACT

Background: Aphids are noxious and serious persistent insect pests in the open-fields and greenhouses worldwide. Many entomologists have studied the possibility of aphid control by applying ladybirds and green lacewings at different releasing rates either under open-field or under greenhouse conditions.
Study period: All experiments were conducted within June-July 2018 in two different fields.
Methodology: Releasing ladybird in open field and under greenhouse condition in the rates of 1:50, 1:100, and 1:200 (predator: aphid) to control aphid infestation. In addition, releasing lacewings in rates of 1:5, 1:10, and 1:20 (predator: aphid) to control aphis in open fields
Results: The outcome data clarified that releasing 3rd instar larvae of *Coccinella undecimpunctata* Linnaeus at the rate of 1 larva:50 aphid was more effective than 1:100 or 1:200 rates for controlling *Aphis gossypii* Glover population in Okra field; achieving more than 90% reduction in the aphid population within 15 days. Under the greenhouse conditions, releasing ladybird adults at 3 successive rates (200, 100 & 50 adults) for every 150 plants induced a significant reduction in *Aphis fabae* Scopoli,1763 infesting soybean; gaining 89.47% reduction within 2 weeks.
When *Chrysoperla carnea* Stephens 2nd instar larvae were released at rates of 1:5, 1:10, and 1:20 (predator: aphid), it was noticed that the first-rate was the most effective one, inducing 98.93%

†Professor;
**Corresponding author: E-mail: mgesraha@gmail.com;*

reduction in aphid's population; while the two other rates gave less reduction. Additionally, in the case of double releases of the same species, reduction percentage at the rate of 1:5 reached 99.63%, which emphasizes that it was the most effective ratio. The other rates induced 97.05 and 95.64% reduction. Generally, a double release was more effective in all tested rates than the single one, because of the cumulative existence of the predators in large numbers at the same period of the experiment.

Conclusion: It could be concluded that utilizing insect predators (*Coccinella undecimpunctata* and/or *Chrysoperla carnea*) at an early larval stage or *C. undecimpunctata* as newly emerged adults were sufficient to reduce the aphids' populations, both under open fields and greenhouses conditions.

Keywords: Releasing predators; lacewings; ladybird; aphid control; open fields; greenhouse.

1. INTRODUCTION

Aphids are serious and persistent pests in either open-fields and/or greenhouses. They are difficult to control or eradicate due to their high reproductive capability and short developmental time that helps them develop resistance to many different insecticidal groups. The presence of aphids facilitates the formation of honeydew, which can reduce the quality of a wide range of greenhouse crops, as well as its important role as a vector for viruses' transmission such as the cucumber mosaic virus and many different pot-viruses [1,2]. Efforts were developed to control aphids by using biological control methods such as releasing of insect predators and/or parasitoids. Coccinellids and Chrysopids predators were applied as means of control to such aphid infestation in open-fields and in greenhouses [3-14].

This study aims to evaluate the efficacy of releasing green lacewings, *Chrysoperla carnea* Stephens and ladybird, *Coccinella undecimpunctata* L. larvae to control *Aphis gossypii* Glover infestation in Okra plants at the open field. In addition, it also aims to evaluate releasing *Coccinella undecimpunctata* as newly emerged adults to control *Aphis fabae* Scopoli infesting Soybean plant under greenhouse conditions.

2. MATERIALS AND METHODS

2.1 Releasing of Insect Predators

2.1.1 *Coccinella undecimpunctata* Linnaeus, 1758

An experiment was conducted in June 2018, in the Belbace region, El-Sharkia Governorate, Egypt, in an already-cultivated Okra field, where regular agricultural practices were performed by their owner farmers. Four rectangular plots, heavily infested with *Aphis gossypii* were selected, with each plot denoted for a single rate of release. Each plot was divided into 4 equal plots ($60m^2$ each) to serve as replicates. Ladybird third instar larvae were applied in 3 rates of 1:50, 1:100, 1:200 as (predator: aphid), with each distinctive rate applied into its own distinctive lot. In every lot, the application covered the entire lot, i.e. the four subdivisions in every lot received the same equivalent treatment. The entire fourth lot was left out with no predator application, no predator was released; the experiment was designed as described by Long [15].

The inspection of the aphid's number was executed at 4 different times, the 1st of which was on the same day of application (0-day) then at the 5th, then 10^{th,} and the 15th days, respectively post-release. The reduction in the percentage of an infestation then was calculated; the percentage of reduction between every 2 consecutive occasions of inspection represented a partial reduction. The net percentage of reduction was estimated using the data of the first inspection occasion, *i.e.* same day of application, as well as on the 15th day. The same pattern of inspection and calculations was applied to all rates of application across the 4 plots.

Another experiment was conducted at El-Manawat region, Giza Governorate, Egypt, under greenhouse conditions during July 2018. Newly emerged adults of the ladybird, *Coccinella undecimpunctata* were released in a greenhouse to control the legume aphid, *Aphis fabae* infesting soybean plants. Three successive releases at rates of 200, 100, and 50 adults for 150 potted plants were conducted at 8.00 am on day 0, day 5, and day 10, respectively. At the beginning of the experiment, each Soybean plant was cultivated in a pot, and when the plant was two months old, adult predators were applied. Natural aphid infestation was counted on samples of 50 treated plants and 25 for the control plants. Aphid counting was taken before release (0-day), on the 5th, then 10th, and the 15th day after release. Then the percentage of aphid reduction was calculated.

This exact experiment model was replicated thrice, potted plants patches were set apart from each other by about 100m, to minimize aphid or predators transformation. In addition, small plastic containers (1 cm^3 volume) supplied with honey droplets + yeast were distributed within each plant patch (1 container/5 plants) to encourage adult predators for an establishment or stilled down at the same plant area of release.

2.2 *Chrysoperla carnea* (Stephens, 1836)

Following the same pattern (in section 1.1), two experiments representing two types of releasing the green lacewings, were carried out in an already-cultivated Okra field at Belbace region, El-Sharkia Governorate, Egypt. The owner farmer customarily performed their regular agricultural practices. In both experiments, four rectangular lots heavily infested with aphid were chosen, each plot was assigned to the application of one of the tested release rates; then every lot was further subdivided into four areas of $60m^2$ each to serve as replicates. The lots were at a distance of about 150m apart from each other. Aphid lion, *Chrysoperla carnea* was released in three of the lots to control *Aphis gossypii* natural infestation. The 4th plot was used as a check plot.

2.2.1 The first experiment (single release)

Packages (A5 size - as a commercial container) containing *C. carnea* 2nd larval instar were transferred to the field at the releasing date [16]. The 2nd instar larvae of *C. carnea* were released at three different rates, ***i.e.,*** 1:5, 1:10, and 1:20 (predator: preys) as recommended by many authors [17-19]. Each rate was released in its distinct lot. Aphid population was counted just before release (0-day), at the 5th, then 10th, and the 15th days post-release for each treatment. Aphid reduction percentage of infestation was calculated between every 2 successive inspection dates for each rate of release, this output is the partial reduction. The net reduction was estimated at the end of the experiment and for each rate, it was calculated by using the initial number of aphid individuals at the beginning of the experiment and the number of aphid individuals found on the last day.

2.2.2 The second experiment (double release)

The same aforementioned procedures (section 2.2.1) were replicated but with one exception that, the 2nd release of the predator's larvae was carried out after 5 days of the first release. Aphid populations were counted after 0, 5, 10, and 15 days of the commencement of the experiments, the population inspection of Aphid on the 5th day was executed before the 2nd release. The reduction percentage in infestation was calculated according to Henderson and Tilton equation [20].

2.3 Statistical Analysis

Randomized Complete Block Design was applied for this experiment. Analysis of variance (ANOVA) F-test was applied using SPSS for Windows (Version 23.0). [21] Computer program was used. Duncan Multiple Range Test [22] was carried out to differentiate between means. The percentage of reduction was carried out using the Henderson and Tilton equation [20] as follow:

$$\text{Reduction }\% = \left(1 - \frac{\text{C before treatment} * \text{T after treatment}}{\text{C after treatment} * \text{T before treatment})}\right) * 100$$

Where: C= number of individuals in control

T= number of individuals in treatment

Student T-test was utilized to compare treated and control plots in the greenhouse experiment, and also to compare the efficiency of both predators at a single release in the open field.

3. RESULTS

3.1 Release of Predators to Control Aphids

The predators, *Coccinella undecimpunctata,* and *Chrysoperla carnea* were released separately to control *Aphis gossypii* at the Okra fields. Control of the aphids started when the mean infestation reached 10 individuals/plant [15].

3.2 Release of *Coccinella undecimpunctata* larvae

3.2.1 Open field release

Data in the Table (1) showed that, on 0-day the mean numbers of aphid populations in the treated plots of 1:50, 1:100, 1:200 predator/aphid rates, on one hand, and the check plot on the other, varied significantly ($F_{3,12}$=190.048**, *P*=0.000).

Data of the 5th-day post-release indicated that mean numbers of collected aphids were decreased compared to those at 0-day of release, except at the control treatment, where values varied even more significantly ($F_{3,12}$=29.701**, *P*=0.000) (Table 1).

In addition, at 1:50 treatment, the mean number of aphid individuals at each inspection day (0-day, 5th, 10th, and 15th day) was markedly reduced, reflecting a partial reduction of infestation. The partial reduction was calculated between each two successive inspection days. This reduction percentage ranged between ≈ 52 and 97%, for that recorded at 5th, 10th, and 15th inspection days; while the net reduction of 99.71% was recorded at the end of the experiment (between the initial number of aphid individuals at 0-day and the aphid's number at the end of the experiment) (Table 1).

A similar trend was recorded for the 10th day, where the mean number of counted aphids visibly diminished more than the counted at both 0-day and the 5th day of release, except in the check plot (Table 1). These mean numbers of aphid were insignificantly varied between each other, but all treatments varied significantly with that control treatment ($F_{3,12}$= 39.990**, *P*=0.000).

As for the 1:100 rate of release, the partial reduction was estimated too between each two successive inspection days, where it was 66.35, 52.91, and 65.89%, for the 5th, 10th, and 15th inspection days, respectively; the calculated net reduction was 94.60% (Table 1).

After 15 days of the release, another sharp decline in counted aphids was achieved, yielding 3.01, 44.65, 55.25, and 574.73 individuals, respectively for different release rates and control as well. Statistically, a significant difference was attained among all these treatments ($F_{3,12}$= 37.051**, *P*=0.000) (Table 1).
In the case of 1:200 treatment, it was clear that the number of counted aphid individuals at all the inspection dates declined sharply, reflecting the significant difference among the treatment rates ($F_{3,12}$= 55.201**), the partial reduction between every two successive inspections reached 49.74, 60.18, and 62.03%, respectively while the net reduction percentage was 92.40% (Table 1).

3.2.2 Greenhouse release

Data in the Table (2) demonstrates that applying newly emerged adults of ladybirds, *C. undecimpunctata* in 3 successive releases (200, 100 then 50 adults) on the 0-day, 5th, and 10th day, respectively, for the same number of plants (150 potted plants) induced a significant reduction in the mean number of *Aphis fabae* population infesting soybean plants.

Before releasing (at 0-day), there was a significant difference in the aphid's populations between tested pots and control ones (T=5.888**, *P*=0.000), then 200 beetles for 150 plants were just released.

On the 5th day post-treatment, where the 2nd release was carried out using (100 beetles/150 plants), the aphid population decreased significantly in treated pots from 218.21 to reach 160.12 aphids/plant. The calculated partial reduction reached 28.56% in the aphid population. However, aphid's population was increased from 233.85 to reach 240.21 aphids/ plant in control pots; being significantly different (T=17.509**, *P*=0.000)(Table 2).

Once again, on the 10th-day post-treatment, the 3rd release (50 beetles/150 plants) was applied. The aphid's population decreased significantly to reach 100.02 aphids/plant in treated pots; being significantly different from the control pots (235.30 aphids/plant) (T=29.881**, *P*=0.000), reflecting a 36.23% reduction in infestation (Table 2).

Inspection after 15 days of starting the experiment illustrated that the aphid population in treated pots decreased sharply and reached 10.00 aphids/plant; gaining 89.47% as a partial reduction in the infestation between the 10th and the 15th day. At the same time, the aphid population in the control pots reached 223.40 aphids/plant, being significantly different than the treated one (T=39.006**, *P*=0.000). The estimated net reduction in infestation was 95.20% at the end of the experiment (Table 2).

3.3 Releasing of *Chrysoperla carnea*

3.3.1 The single release of *C. carnea* larvae

Data illustrated in Table (3) summarizes the obtained results. Generally, at all inspection days, there were significant differences between the population numbers of aphid individuals at all tested rates of release.

At the 1st rate (1 larva: 5 aphids): the aphid's population on 0-day was 280.50 individuals/plant, it decreased to reach 140.34 individuals on the 5th-day post-release (first inspection); gaining 57.67% as a partial reduction in population. At the 2nd inspection time (10th day) the calculated partial reduction was 56.91%; while at the 3rd inspection time (15th day) the percentage of the partial reduction infestation reached 94.07%. The mean number of aphids in all inspection days was varied significantly between each other ($F_{3,12}$=10.997**). At the end of the experiment, the mean number of aphids reached 5.12 individuals/plant with a net rate of infestation reduction reached to 98.93% (Table 3).

At the 2nd rate (1 larva: 10 aphids): the mean number of aphid population at 0-day was 300.50 individuals and it decreased to reach 210.51 individuals at the 5th-day post-release reflecting a 40.78% partial reduction in the infestation. The mean number was 134.17 individuals after 10 days of the release, gaining 45.06% as a partial reduction compared to the previous inspection time. After 15 days of release, the mean number of aphid's population reached 81.32 individuals, achieving 51.23% as a partial reduction. The net infestation reduction reached 84.13%; being less than the recorded percentage at 1:5 releasing rate (98.93%). A considerable difference of the aphid's population was observed between each inspection time ($F_{3,12}$=38.780**, *P*=0.000), and with the previous treatment (1 larva: 5 aphids) ($F_{3,12}$=83.187**, *P*=0.000) (Table 3).

At the 3rd treatment (1 predator larva: 20 aphids): the mean number of aphid population was significantly different compared to either of the other inspection time ($F_{3,12}$=39.154**, *P*=0.000), it inducing a net reduction of 84.92% in the aphid population within 15 days. Variation in infestation was observed when compared to other release rates or with control treatment ($F_{3,12}$=83.187**, *P*=0.000)(Table 3).

3.3.2 Double Release of *C. carnea* larvae

In this experiment, 2nd instar larvae of the predator were released once at 0-day of the experiment, then after 5 days of the first release, using the same rates previously released, *i.e.,* 1:5, 1:10, and 1:20 (predator: aphids).

Data in Table (4) indicated that, at 0-day, the mean number of aphid ranged between about 345 to 400 individuals/plant; with significant differences among the 3 rates of release, while an insignificant difference was recorded between control and 1:5 rate ($F_{3,12}$= 18.681**, *P*=0.000). Moreover, the aphid population recorded in all inspection times showed significant variations ($F_{3,12}$=14.443**, *P*=0.000).

As for the 1st tested rate of release (1:5), the reduction percentages were recorded between every two successive release occasions (partial reduction) and at the end of the experiment. The estimated partial reduction percentages were 81.92, 80.49, 95.01, respectively for the 5th, 10th, and the 15th-day post-release, while the net rate of reduction was 99.63% (Table 4).

The figures after 5 days showed a significant difference between the control plot and all other plots ($F_{3,12}$=33.361**, *P*=0.000); but the aphid population at the rate of 1:10 & 1:20 treatments was insignificantly different between each other (Table 4). Also, it was observed that there was a significant difference in aphid populations ($F_{3,12}$=11.562**) at all inspection dates. The same figures of partial reduction percentages were recorded between every two successive intervals, were 50.05, 63.23, 83.94% at the 5th, 10th, and 15th days, respectively while the net reduction was 97.05% (Table 4).

After 10 days of the first release (5 days of the 2nd release), the mean number of aphid's population was 40.15, 90.17, 183.61, and 533.87 individuals, respectively for 1:5, 1:10, 1:20, and control plots, respectively, reflecting significant differences among all tested plots ($F_{3,12}$=14.023**, *P*=0.000) (Table 4).

On the 15th day of the first release (10 days of the 2nd release): the mean number of aphid's populations diminished sharply to reach 2.12, 15.32, and 26.25 individuals, for rates of 1:5, 1:10, and 1:20 respectively, at the treated plots, but it increased in case of the control plot to reach 564.73 individuals; showing a significant difference between control plot and all other plots. Also, the 1:10 and 1:20 treatment plots showed insignificant variation between each other, but they significantly differed with 1:5 plots ($F_{3,12}$=58.746**) (Table 4). The net percentage of reduction in infestation after 15 days of the first release was 99.63, 97.05, and 95.64% for treated plots, across rates of 1:5, 1:10, and 1:20, respectively (Table 4).

Table 1. Mean numbers of aphid population throughout the release of *Coccinella undecimpunctata* 3rd instar larvae in Okra field

Release rate Predator: Aphids	Mean number of aphid ± SE Days of check 0-day	5th -day	10th -day	15th-day	$F_{3,12}$-value	Net Infestation Reduction (%) (end of experiment)
1:50 (Partial Reduction)	680.45±1.37a A	415.34±0.95 b B (52.22%)	114.15±2.07 b C (74.73%)	03.01±0.14 d D (97.58%)	528.718**	99.71
1:100(Partial Reduction)	545.87±3.62 b A	234.62±3.34 d B (66.35%)	120.17±0.84 b C (52.91%)	44.65±1.61 c D (65.89%)	706.102**	94.60
1:200 (Partial Reductio)	480.53±2.70 c A	308.51±2.87 c B (49.74%)	133.61±2.61 b C (60.18%)	55.25±1.92 b D (62.03%)	55.201**	92.40
Control	379.81±3.34 d D	485.18±1.80 a C	527.67±25.94 a B	574.73±1.56 a A	40.113**	
$F_{3,12}$-value	190.048**	29.701**	39.990**	37.051**		
P-value	0.000	0.000	0.000	0.000		

***= Highly Significant*

In horizontal rows (inspection time), means followed with different Capital letter are statistically different (P>0.5)

In vertical columns (release rate), means followed with different small letter are statistically different (P>0.5)

Table 2. Release *Coccinella undecimpunctata* adults on *Aphis fabae* under greenhouse conditions

Treatment (inspection time)	Mean number of aphid ± SE Release rate (Beetles/150 plant)	Experiment plots	Control plots	T-value df=6
0-day (1st release)	200	218.21±2.51 a B	233.85±0.87 a A	5.888**
5-day (2nd release) Partial reduction (%)	100	160.12±3.97 b B (28.56%)	240.21±2.27 a A	17.509**
10-day (3rd release) Partial reduction (%)	50	100.02±3.07 c B (36.23%)	235.30±3.32 a A	29.881**
15-day (end of experiment) Partial reduction (%)	0	10.00±1.84 d B (89.47%)	223.40±5.15 b A	39.006**
Total Reduction (end of the experiment)		95.20%		
$F_{3,12}$-value		9.067**	4.598*	
P-value		0.000	0.023	

***= Highly Significant* **= Significant*

In horizontal rows (T-value), means followed with different Capital letter are statistically different (P>0.5).

In vertical columns (F-value), means followed with different small letter are statistically different (P>0.5).

Total reduction = The compare between the initial number of aphid population at the beginning of the experiment and the last number at the end of the experiment

Table 3. Mean numbers of aphid population throughout the Single Release- *Chrysoperla carnea* 2nd larval instar in Okra field

Release rate Predator: Aphids	Mean number of aphid ± SE Days of check				$F_{3,12}$-value	(%) Final Infestation Reduction
	0-day	5th -day	10th -day	15th-day		
1st (1:5) (Partial Reduction %)	280.50±16.36 b A	140.34±5.93 d B (57.67%)	70.15±2.23 d C (56.91%)	05.12±0.30 d D (94.07%)	10.997**	98.93
2nd (1:10) (Partial Reduction %)	300.50±23.74 b A	210.51±1.31 c B (40.78%)	134.17±4.48 c C (45.06%)	81.32±2.61 c D (51.23%)	38.780**	84.13
3rd (1:20) (Partial Reduction %)	374.75±12.62 a A	254.62±3.08 b B (48.50%)	183.61±1.32 b C (37.84%)	96.25±3.09 b D (57.81%)	39.154**	84.92
Control	300.00±9.13 b D	355.68±3.09 a C	412.62±5.99a B	512.73±2.97 a A	49.218**	
$F_{3,12}$-value	6.523**	58.257**	14.174**	83.187**		
P value	0.007	0.000	0.000	0.000		

***= Highly Significant*

In horizontal rows (inspection time), means followed with different Capital letter are statistically different (P>0.5).

In vertical columns (release rate), means followed with different small letter are statistically different (P>0.5)

Table 4. Mean numbers of aphid population throughout the Double Releases – *Chrysoperla carnea* 2nd larval instar in Okra field

Release rate Predator: Aphids	Mean number of aphid ± SE Day of check				$F_{3,12}$-value	(%) Final Infestation Reduction
	(I) 0-day	(II) 5th day	10th day	15th day		
1:5 (Partial Reduction %)	380.45±1.37 b A	183.34±1.68 c B (81.92%)	40.15±1.85 d C (80.49%)	02.12±0.10c D (95.01%)	14.443**	99.63
1:10 (Partial Reduction %)	345.67±1.92 c A	218.51±0.96 b B (50.05%)	90.17±1.63 c C (63.23%)	15.32±0.25b D (83.94%)	11.562**	97.05
1:20 (Partial Reduction %)	400.53±0.80 a A	214.62±2.11 b B (57.66%)	183.61±2.74 b C (23.77%)	26.25±1.37b D (86.48%)	65.382**	95.64
Control	375.84±2.19 b D	475.68±3.74 a C	533.87±0.68 a B	564.73±7.05a A	39.748**	
$F_{3,12}$-value	18.681**	33.361**	14.023**	58.746**		
P value	0.000	0.000	0.000	0.000		

***= Highly Significant* *(I)= First release* *(II)= Second release*

In horizontal rows (inspection time), means followed with different Capital letter are statistically different (P>0.5)

In vertical columns (release rate), means followed with different small letter are statistically different (P>0.5)

4. DISCUSSION

4.1 Release of *Coccinella undecimpunctata*

4.1.1 Open field release

Our obtained results confirmed the results that were obtained by many researchers such as Long [15] and Zaki et al. [7] when they carried out a single release of *C. undecimpunctata* to control *A. gossypii* and obtained a 99.97% reduction in infestation within 15 days. The results found by Karaman et al. [11] were in accordance with our results when they released *C. septempunctata* to control corn leaf aphid, *Rhopalosiphum maidis* in Upper Egypt. The obtained results were in harmony with the findings of Mushtaq et al. [23], who examined the larval instars (1^{st} to 3^{rd}) of the ladybird to control aphid, *Chaitophorus* spp under laboratory conditions.

4.1.2 Greenhouse release

Under greenhouse conditions, results of releasing *Coccinella* adults agreed with Farag [6] and Zaki et al. [7], in controlling *Aphis fabae*. The results also matched with those of Snyder et al. [24] on using ladybird beetle, *Harmonia axyridis* Pallas to control the aphid, *Macrosiphum euphorbiae* Thomas that attacks greenhouse-grown roses *Rosa hybrida* L. Also, our obtained results matched with those of Bale et al. [25] who discuss the relationship between the biological control agents and the GM-crops**.** The obtained findings were in harmony with that reported by Shannag and Obeidat [26] when they released *C. septempunctata* to control *A. fabae*; they reported that release of a newly hatched *C. septempunctata* larva onto each plant significantly reduced aphid density to 32.8 and 57.2% on *A. faba* within 14 days. Also, matched with Seko et al. [27], who conducted a release experiment to assess the effectiveness in controlling two aphid species, *A. gossypii* and *Aulacorthum solani*, using second instars of a flightless strain of *H. axyridis*, they observed that the number of *A. gossypii* was suppressed in greenhouses that contained the flightless strain compared with the greenhouses that contained the wild-type strain. Our obtained results are in harmony with Riddick [28] who stated that ladybirds are effective aphid predators in greenhouses. The results were in accordance with Abd-Allah et al. [14] who also applied different coccinellids rates (30, 60, and 90 eggs/plant) to control *A. gossypii* under greenhouse conditions, gaining a 74.4% reduction in aphid's infestation.

4.2 Releasing of *Chrysoperla carnea*

According to the obtained results, it could be concluded that the release rate of 1 predator/ 5 aphids) in both treatments (Single and Double releases) exhibited the best results and was the most effective tested rate, where it yielded about 100% reduction in the infestation within 15 days.

In general, double releases were more effective in all tested rates of release than those recorded in a single release, because of the aggregation of the predators in large numbers at the same period of the experiment. These results nearly matched with that reported by many authors such as Uschchekov [29,30], Beglyarov & Ushakov [18]; Hassan [3,31]; Shuvakina [4]; Farag [6] and Zaki et al. [7] when they released *C carnea* at the rate of (1 predator larva/5 aphids) and achieved 100% reduction in aphid's population within 12 days. Dey [32] reported that three families of Neuroptera (Chrysopidae, Coniopterygidae, Hemerobiidae) were found to be active predators in biological control programs. Also, results were in accordance with that reported by Karaman et al. [11] who released *C. carnea* to control corn-leaf aphid, where the percentage reduction reached ≈ 94% in the two experimental seasons. Alghamdi et al. [13] released *C. carnea* at rates ranging between 1:5 and 1:30 (predator: aphids) to control *A. gossypii* population within 2-5 weeks; while rates of 1:40 or 1:50 (predator: aphid) showed insignificant differences with the check treatment.

5. CONCLUSION

Utilizing insect predators at an early larval stage or as newly emerged adults were sufficient to reduce the aphid's population, infesting several crops or vegetables in open-fields and/or under greenhouses conditions. This presented study demonstrated that *Coccinella* larvae were more efficient than *Chrysoperla* larvae in all tested rates of release to control *A. gossypii* under open-field conditions.

6. SIGNIFICANT STATEMENT

This study is important to keep the environment clean and free of chemical insecticide pollution as much as possible, exhibit the role of the natural enemies' to utilize as a biological control method of IPM program, and decrease build up resistance strains of the insect.

AVAILABILITY OF DATA AND MATERIALS

All data and materials are available.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Kishaba AN, Castle SJ, Coudriet DL, Mccreight J, Bohn GW. Virus Transmission by *Aphis gossypii* Glover to Aphid-resistant and Susceptible Muskmelons. J Amer Soc Horti Sci,. 1992; 117(2): 248-254.
2. Pinto ZV, Rezende JAM, Yuki VA, Piedade SMS. Ability of *Aphis gossypii* and *Myzus persicae* to Transmit Cucumber mosaic virus in Single and Mixed Infection with Two Potyviruses to Zucchini Squash. Summa Phytopathol Botucatu. 2008;34(2):183-185.
3. Hassan SA Investigations on the use of the predator *Chrysopa carnea* for the control of the green peach aphid, *Myzus persicae* on peppers in the greenhouse. Zeitschrift fuer Angewandte Entomologie. 1977;82:234-239.
4. Shuvakhina EY. *Chrysopa stoics* an effective natural enemy. Zaslch Rast Moskva. 1983;9:29.
5. Liu H, Oin L. Mass rearing of *Coccinella septempunctata* to control *Aphis gossypii* in Hebei province, China. Chinese J Biol Cont. 1987;3:138-139.
6. Farag NA. Studies on the biological control of whiteflies and Aphids on some vegetable crops. Ph D. Thesis, Environmental Science- Agricultural Department, Ain Shams University, Cairo, Egypt. 1995; 262.
7. Zaki FN, Shaarawy MF, Farag NA. Release of two predators and two parasitoids to control aphids and whiteflies. J Pest Sci. 1999;72(1):19–20.
8. Van Driesche RG, Heinz KM. An overview of biological control in protected culture. In Biological Control in Protected Culture; Heinz KM, Van Driesche, RG, Parrella, MP, Eds; Ball Publ: Batavia, IL, USA. 2004;1–24.
9. Van Lenteren JC, Loomans AJM, Babendreier D, Bigler F. *Harmonia axyridis*: An environmental risk assessment for Northwest Europe. In: Biological control to invasion: The ladybird *Harmonia axyridis* as a model species. Ed. by Roy H, Wajnberg E, Springer, Netherlands. 2008;37–54.
10. Wu XH, Zhou XR, Ping PB. Influence of five host plants of *Aphis gossypii* Glover on some population parameters of *Hippodamia variegata* (Goeze). J Pest Sci. 2010;83(2):77–83.
11. Karaman GA, Gharib AH, Elmandarawy MBR, Gadallah NS, Elgepaly HMKH. Release of two Predators to Control Aphid with Special References to native Aphidophagous and their Parasitoids in Sorghum. Middle East J Agric Res. 2014;3(2):302-310.
12. Alghamdi A, Sayed SM. Biological characteristics of indigenous Chrysoperla carnea (Neuroptera: Chrysopidae) fed on a natural and an alternative prey. Asian J Biol. 2017;2(2):1–6.
13. Alghamdi A, Al-Otaibi S, Sayed SM. Field evaluation of indigenous predacious insect, Chrysoperla carnea (Steph.) (Neuroptera: Chrysopidae), fitness in controlling aphids and whiteflies in two vegetable crops. Egypt J Biol Pest Cont. 2018;28:20.
14. Abd-Allah MAM, Emam AS, Mahmoud SA El-Z, El-Rahman AA. Biological Control of *Aphis gossypii* Glover on Cucumber Plants by Release *Coccinella septempunctata* L. under Glasshouse Conditions. Egypt Acad J Biol Sci. 2019;11(2):95-68.
15. Long PU. Principles and techniques of biological control of insects. Science Publ House, Beijing, 1984;318.
16. El-Arnaouty SA, Gaber N, Tawfik MFS. Biological control of the green peach aphid *Myzus persicae* by Chrysoperla carnea (Neuroptera: Chrysopidae). Egypt J Biol Pest Cont. 2000; 10(12):109–116.
17. Beglyarov GA, Ushchenkov AT. Biological control of aphids on green crops. Zaschita Rastenii. 1977;2:25-27.
18. Beglyarov GA, Ushchenkov AT, Lychkia VV. Biological protection of green vegetable crops on Moscow Sovkhoz. Zaschita Rastenii. 1980;2:35
19. Pruszynski S, Domagal T, Piatkowski J, Kosmalski W. New elements in integrated programmes of greenhouse crop protection. Materiali Sesji Instytutu Ochrony Roslin. 1989;29(1):175-183.
20. Henderson CF, Tilton EW. Tests with acaricides against the brown wheat mite. J Econ Entomol. 1955;48:157-161.
21. SPSS. IBM SPSS Statistics for Windows

(Version 23.0). Armonk, NY: IBM Corp. Chicago, IL;2015.
22. Duncan DB. Multiple ranges and multiple F-test. Biometrics. 1955;11:1-42.
23. Mushtaq S, Nazeer A, Imtiaz AK, Bismillah S, Ashraf K, Murad A, Muhammad TR, Muhammad A, Khwaja J, Arqam BZ, Saeed A. Study on the efficacy of ladybird beetle as a biological control agent against aphids (*Chaitophorus* spp.). J Entomol Zool Stud. 2015;3(6): 117-119.
24. Snyder WE, Ballard SN, Yang S, Clevenger GM, Miller TD, Ahn JJ, Hatten TD, Berryman AA. Complementary biocontrol of aphids by the ladybird beetle *Harmonia axyridis* and the parasitoid Aphelinus asychis on greenhouse roses. Biol Cont. 2004;30:229-235.
25. Bale JS, Van Lentern JC, Bigler F. Biological control and sustainable food production. Philosophical Transactions of the Royal Society, B: Biological Sciences. 2008;363:761–776.
26. Shannag HK, Obeidat WM. Interaction between plant resistance and predation of Aphis fabae (Homoptera: Aphididae) by *Coccinella septempunctata* (Coleoptera: Coccinellidae). Ann Appl Biol. 2008;152(3):331-337.
27. Seko T, Sumi A, Nakano A, Kameshiro M, Kaneda T, Miura K. Suppression of aphids by augmentative release of larvae of flightless *Harmonia axyridis*. J Appl Entomol. 2013;138:326–337.
28. Riddick EW. Identification of Conditions for Successful Aphid Control by Ladybirds in Greenhouses. Insects. 2017;8(38): 1-17.
29. Uschchekov AT *Coccinella septempunctata* Wesm in greenhouses. Zaslch Rast Moskva. 1976;10:16-17.
30. Uschchekov AT. Crysopa perla for aphid control. Zaslch Rast Moskva. 1989;11:20-22.
31. Hassan SA. Release of *Chrysopa carnea* to control *Myzus persicae* on eggplant in small greenhouse plots. Ziteschrift für Pflanzen und Pflanz. 1978;85(2): 118-123.
32. Dey SR. Aphid pest management with neuroptera (Insecta). The Beats of Natural Sciences. 2014;6(1):1-5.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/84633