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Field evaluation of indigenous predacious insect, *Chrysoperla carnea* (Steph.) (Neuroptera: Chrysopidae), fitness in controlling aphids and whiteflies in two vegetable crops

A. Alghamdi¹, S. Al-Otaibi² and S. M. Sayed^{2,3*}

Abstract

The green lacewing, *Chrysoperla carnea* (Steph.) (Neuroptera: Chrysopidae), is a generalist predator in its larval stage of most species of soft bodied insect pests, especially aphids, whiteflies, thrips, coccids, and mealy bugs. This predator had been recorded in different regions in Saudi Arabia as indigenous species. The fitness of this indigenous predator for controlling the aphid, *Aphis gossypii* Glov., and the whitefly, *Bemisia tabaci* (Genn.), with five and ten releasing rates on sweet pepper and squash plants in the open field was evaluated. The experiments were carried out in Taif region, Saudi Arabia, during the summer of 2017. On squash plants, the reduction was more than 90% after the third predator release of ten larvae per plant for both pests and reached 100% only for the whitefly after six releases. On sweet pepper plants, reduction rates of the aphid and whitefly reached about 90 and 97%, after the second predator release of five and ten larvae per plant, respectively. A 100% reduction was achieved after four releases with five larvae/plant and three releases with ten larvae/plant. The present findings indicate that the releasing rates of five larvae/pepper plant and 10 larvae/squash plant were sufficient for suppressing both *B. tabaci* and *A. gossypii* populations.

Keywords: Green lacewing, Aphis gossypii, Bemisia tabaci, Squash, Sweet pepper, Fitness, Saudi Arabia

Background

The green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae), is a generalist predator. Adults feed only on nectar, pollen, and aphid honeydew, but its larvae are active predators. *C. carnea* occurs in a wide range of habitats (Henn and Weinzierl, 1990). It is considered as an effective generalist predator of most species of soft bodied insect pests, especially aphids, whiteflies, thrips, coccids, and mealy bugs (McEwen et al., 2001). *C. carnea* had been

³Department of Economic Entomology and pesticides, Faculty of Agriculture, Cairo University, Giza 12613, Egypt



C. carnea has the adaptability to different environmental conditions and food diversity. It has a high searching capacity and a higher potential to prey on about 200 aphid species and more than 80 species of other insect pests (Tauber et al., 2000). *C. carnea* has been widely used for biological control of aphids and other



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^{*} Correspondence: samy_mahmoud@hotmail.com

²Biotechnology Department, Faculty of Science, Taif University, Taif 888, Saudi Arabia

Full list of author information is available at the end of the article

insect pests because of its polyphagous habits and compatibility with selected chemical insecticides, microbial agents, and amenability for mass rearing (Uddin et al., 2005). It has been mass-reared and marketed commercially in the world, especially in North America and Europe (Tauber et al., 2000). Eggs of *E. kuehniella* are one of the most factitious preys for mass production of chrysopids' species because this food ensures quick growth and development, high survival rates, and higher fecundity (Specty et al., 2003).

Biological aphid control on sweet peppers (*Capsicum annuum* L.) includes applications of generalist predators and parasitoids (De Backer et al., 2015). The melon aphid, *Aphis gossypii* Glover (Homoptera: Aphididae), is one of the most common aphid species on sweet pepper plants (Sanchez et al., 2011). This pest species has a variation in its biological parameters and reproduction on different sweet pepper cultivars (Alizadeh et al., 2016).

Squash plants (*Cucurbita pepo* L.) are attacked by many insect pest species, especially *A. gossypii* (Nyoike and Liburd, 2010). Damage may be caused directly by sucking plants' juice or indirectly by transmitting plant viruses (Wu et al., 2010).

The whitefly, *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae), is one of the key pest species of sweet pepper and squash plants that causes direct damage by sucking the plant sap and indirect damage by transmitting of virus diseases (Banihashemi et al., 2017).

The present study aimed to evaluate the efficacy of releasing the indigenous *C. carnea* larvae for controlling *A. gossypii* and *B. tabaci* infesting sweet pepper and squash plants in open field trials.

Materials and methods

Predator collection and mass rearing

Individuals of C. carnea males and females were collected, using a 30-cm-diameter sweep net, from clover fields at Taif in west Saudi Arabia. The adults were maintained in a cylindrical plastic vessel (10 cm diameter × 8 cm height) with a hole (4 cm in diameter) in the lid and covered with gauze for allowing proper ventilation inside the vessel. The adults were fed on an artificial diet as described by Sayed and Alghamdi (2017). Eggs laid in the vessels were carefully harvested, using forceps and breaking the stalk beneath the egg, and transferred to other vessels with the larval food. Eggs of E. kuehniella, sterilized with UV, were used, as a factitious prey, for C. carnea larvae. The rearing was carried out under the controlled conditions of 25 °C, 60% R.H., and 16:8 photoperiods (L:D). The newly hatched larvae of the predator were used in continuing mass rearing for three generations, while the cocoons were collected daily and transferred to the vessels of adult rearing. The rearing of larvae was undertaken in the same plastic vessels described above and maintained with paper clips to reduce cannibalism. The rearing was continued during the whole experiment period in order to get the second instar larvae for releasing dates. The second instar larvae were selected and collected by a fine camel brush and transferred to releasing envelopes.

Predator release in the field

The experiment was carried out in Al Hada (Taif Governorate, Saudi Arabia) on two vegetable crops sweet pepper and squash during their vegetative and fruitage growth stages.

Randomized complete block design (RCBD) was practiced in an area of $(100 \times 200 \text{ m})$ for each crop. Five blocks were used for each crop. Each block contained three plots (two treatments + control). Each plot was about $(2 \times 2 \text{ m})$ and kept free from any pesticidal treatment. One plant was selected from each plot for the experiment. The distance between treatments and/or the control was about 50 m. The *C. carnea* second instar larvae were released on the plants, at two rates (five and ten larvae per plant) as treatments. Thus, both treatments and the control were replicated five times (no. of plants sampled). The control was run without release of the predator.

Twenty envelopes (A5 size) containing the second larval instar of *C. carnea* were transferred to the field every releasing date (El-Arnaouty et al., 2000). Ten envelopes contained ten larvae and ten contained five larvae for each one. A paper sheet displaying *E. kuehniella* eggs was inserted in each envelope in order to avoid the cannibalism, until larval dispatching on the plants. Releasing of *C. carnea* second instar larvae started on July 28, 2017, and continued till September 16th (10-day intervals), with a total of six releases at the same rate on the same tested plant in both crops.

Pests' count

On the same day of the first predator release, aphid and whitefly nymphs/leaf were counted on each investigated plant and also on the same day of each subsequent release and in 10 days after the last release. Randomized three leaves from each treated and untreated plants were selected from different height of the plant and examined on both surfaces to count aphid and whitefly populations. These inspections were carried out using a ×4 magnification lens in the field.

Statistical analysis

Duncan multiple range test, through one-way ANOVA (SPSS, 2015), was used to estimate the significance between the infestation rates of both pests. The

reduction percentages in aphid and whitefly counts were calculated per plant leaf as a mean from each plant (five plants for each treatment) compared to the control. These percentages were estimated according to Abbott's (1925) formula because there were no significant differences among the treatments and the control for each pest density on both crops in the beginning of the experiment.

$$Reduction\% = \frac{Control \ count-Treatment \ count}{Control \ count} \times 100$$

The pest reductions (%) were analyzed through six inspections, started 10 days after the first release. Reduction percentages of each pest were compared at the same inspection date for both pests, using t test with a probability of 5% (SPSS, 2015).

Results and discussion

According to the General Authority of Meteorology and Environmental Protection (KSA), the minimum temperature during the experiment period ranged between 19 and 23 °C, while the maximum temperature ranged between 33 and 35 °C. The average humidity was 31% at daytime and 36% at night.

It was, generally, observed that the aphid and whitefly densities increased gradually from the end of July till the end of August on both investigated plants in the control (untreated), and then, the densities decreased gradually during September (Tables 1, 2, 3, and 4). This may be because the leaves of both plants became old with few vegetative growth.

Aphid densities did not differ significantly, 10 days after the first release, between the releasing rates of five second instar larvae (88.27 and 4.4 nymphs/ squash and pepper leaves, respectively) and the ten one (75.6 and 2.47 nymphs/squash and pepper leaves, respectively) (Tables 1 and 3). Meanwhile, whitefly densities, at this period, were significantly different between the releasing rates of five larvae (10.87 and 2.47 nymphs/squash and pepper leaves, respectively) and the ten one (6.2 and 0.6 nymphs/ squash and pepper leaves, respectively) (Tables 2 and 4). Moreover, the whitefly reduction on squash plants in this period was significantly different between both releasing rates (Table 1), while the reduction of aphid counts was insignificant (Table 2). This reduction reached more than 90% after the third release of ten larvae per plant for both pests and increased to 100% in the case of the whitefly only after six releases. The maximum reduction in the releasing rate of five larvae/plant was achieved after six releases (74% for aphid and 64% for whitefly). Abrams and Matsuda (1996) indicated that sharing of two prey species to a predator may affect each other's densities positively because an increase in the populations of one pest species resulting in decreased predation on the other pest species. However, Messelink et al. (2008) stated that whitefly control of each of two predators in the absence of the thrips was not sufficient, while whitefly densities in the presence of thrips were reduced significantly.

On sweet pepper plants, the reduction in densities of both pests and after 10 days of the first release were significantly different (Tables 3 and 4). The aphid reduction reached about 90 and 97% after the second release of five and ten larvae per plant, respectively. A 100% reduction was achieved after four releases with five larvae/plant and three releases with ten larvae/plant (Table 3). The whitefly reductions seemed to be similar to those of aphids (Table 4). El-Arnaouty et al. (2000) obtained the best results from M. persicae control by releasing C. carnea second instar larvae on green pepper plants under greenhouse conditions, while the lower aphid suppression was achieved after releasing the combination of eggs and second instar larvae or eggs only of the predator.

The present findings indicate that the releasing rate of five larvae/pepper plant was sufficient for obtaining a good suppression of both *B. tabaci* and *A. gossypii*. Meanwhile, the releasing rate of ten larvae/squash plant was found adequate for better suppression of both pests. This result coincides with Zaki et al. (1999) who found that *C. carnea* induced highly significant reduction of *A. gossypii* and *B. tabaci* at different releasing rates on various vegetable crops.

Moreover, researchers such as Easterbrook et al. (2006) indicated that *C. carnea* is effective in open field than in protected crops and found that the aphid, *Chaetosiphon fragaefolii* infestation, was significantly reduced at the releasing rate of eight larvae/ strawberry plant in open field experiments but did not give a significant reduction in aphids under protected crops, even at a releasing rate of 25 larvae/ plant.

Conclusions

In conclusion, the obtained data showed that the aphid and whitefly populations on sweet pepper and squash plants, in open fields, were successfully suppressed by the releases of *C. carnea*. Control of two sap-sucking pests under investigation differed according to the releasing rate of the predator, at five and ten larvae per pepper and squash plant, respectively. The predatory action could be characterized as both curative and preventive.

Releasing rate per plantInfestationInfestationReductionInfestationReductionInfestationReductionInfestationControl52.13103.00-179.80-179.80-192.93-26.47-68.93-Control $\pm 13.7^{A}$ $\pm 21.9^{A}$ $\pm 52.2^{A}$ $\pm 53.3^{A}$ $\pm 33.60^{A}$ $\pm 23.3^{A}$ $\pm 23.3^{A}$ 5 47.87 88.27 14.32 118.47 34.08 106.20 44.96 66.20 47.68 19.33 6 47.87 88.27 14.32 118.47 34.08 106.20 44.96 66.20 47.68 19.33 7 47.87 28.74 $\pm 29.1^{B}$ ± 7.6 $\pm 2.01^{B}$ ± 7.6 $\pm 7.9^{B}$ $\pm 7.9^{B}$ 10 49.47 75.60 26.56 49.13 72.68 17.87 20.07 8.73 23.10 360 7 413.0^{A} $\pm 16.2^{C}$ ± 6.7 $\pm 17.9^{B}$ ± 7.6 $\pm 2.9^{B}$ ± 7.6 $\pm 7.9^{B}$ 10 49.47 75.60 26.56 49.13 72.68 17.87 20.7^{A} 26.7^{C} $\pm 7.6^{A}$ $\pm 7.6^{A}$ $\pm 7.9^{B}$ 10 49.47 75.60 26.56 49.13 72.68 17.87 20.7^{A} $\pm 5.1^{C}$ $\pm 2.36^{A}$ $\pm 3.0^{C}$ 10 49.47 75.60 $2.14.3^{A}$ $ 50.237^{A}$ $ 105.03^{A}$ $ 8.73^{A}$ $ 1$		28 Jul.	7 Aug.		17 Aug.		27 Aug.		6 Sep.		16 Sep.		26 Sep.	
Control52.13103.00-179.80-192.93-126.47-68.93.(untreated plants) $\pm 13.7^h$ $\pm 21.9^h$ $\pm 25.2^h$ $\pm 37.3^h$ $\pm 3360^h$ $\pm 233.3^h$ $\pm 233.3^h$ 5 47.87 88.27 14.32 118.47 34.08 106.20 44.96 66.20 47.68 1933 6 10 49.47 75.60 26.56 49.13 72.68 17.87 92.00 8.73 93.10 3.60 9 10 $\pm 13.0^h$ $\pm 14.3^B$ ± 10.6 $\pm 16.2^c$ ± 6.5 $\pm 7.3^c$ ± 0.7 $\pm 5.1^c$ ± 2.88 $\pm 3.0^c$ 10 $\pm 31.30^h$ $\pm 113.0^h$ $\pm 114.3^B$ ± 10.6 $\pm 16.2^c$ ± 6.5 $\pm 7.3^c$ ± 0.7 $\pm 2.1^2$ $\pm 3.0^c$ 10 49.47 75.60 26.56 49.13 72.68 17.87 92.00 8.73 93.10 3.60 9.66 6.0432 8.730 $= 16.2^c$ ± 6.5 $\pm 7.3^c$ ± 0.7 $\pm 2.1^2$ ± 2.8 $\pm 3.0^c$ 3.60 6.0462 $= 2.42$ $= = 2.125$ $= = 105.02$ $= 105.003$ $=$ 85.274 $= 7.values$ $=$ $= = 2.125$ $=$ $= 10.542$ $=$ 15.722 $=$ 12.482 $=$	leasing rate per plant	nfestation	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	ontrol 5 ntreated plants) 4	52.13 ± 13.7 ^A	103.00 ± 21.9 ^A	1	179.80 ±52.2 ^A	I	192.93 ± 37.3 ^A	1	126.47 ± 36.0 ^A	I	68.93 ± 23.3 ^A	I	24.47 ± 8.4 [≜]	1
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	TI V	47.87 ± 11.3 ^A	88.27 ±17.0 ^B	14.32 ± 7.4	118.47 ± 29.1 ^B	34.08 ± 5.0	106.20 ± 20.1 ^B	44.96 ±6.7	66.20 ± 12.9 ^B	47.68 ± 7.6	19.33 ± 7.9 ^B	67.84 ± 14.2	6.40 ± 2.9 ⁸	73.88 ±4.5
F values 0.432 8.730 $ 50.237$ $ 186.907$ $ 105.003$ $ 85.274$ \cdot (with $df = 2.42$)T values $ 2.125$ $ 10.542$ $ 15.572$ $ 12.482$ $-$ (with $df = 1.8$) $ 2.125$ $ 10.542$ $ 15.572$ $ 12.482$ $-$	7 1	49.47 ± 13.0 ^A	75.60 ±14.3 ^B	26.56 ± 10.6	49.13 ± 16.2 ^C	72.68 ± 6.5	17.87 ± 7.3 ^C	92.00 ± 0.7	8.73 ±5.1 ^C	93.10 ± 2.8	3.60 ± 3.0 [⊂]	94.74 ± 2.0	1.93 ± 1.8 ^C	92.08 ± 2.1
T values 2.125 - 10.542 - 15.572 - 12.482 - (with $df = 1.8$)	values () () () () () () () () () () () () ()	0.432	8.730	I	50.237	I	186.907	I	105.003	I	85.274	I	77.561	I
	values vith <i>df</i> = 1.8)	I	I	2.125	I	10.542	I	15.572	I	12.482	I	4.198	I	8.147
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Releasing rate per plant	Infestation	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction
Control (untreated plants)	6.13 ± 4.1 ^A	14.60 ±6.6 ^Å	I	21.13 ± 7.6 ^A	1	26.33 ± 8.5 ^A	I	24.73 ± 7.8 ^A	I	15.93 ±6.6 ^A	I	10.67 ± 4.5 ^A	1
5	5.60 ± 3.4 ^A	10.87 ± 3.5 ^B	25.62 ± 12.9	14.00 ± 6.0 ^B	33.68 ± 5.4	15.27 ± 6.5 ^B	41.22 ±16.0	12.60 ± 7.0 ^B	49.00 ± 27.2	7.73 ±5.5 ^B	51.54 ± 11.8	3.87 ± 3.3 ^B	63.76 ±19.2
10	6.33 ± 3.1 ^A	6.20 ±4.5 ^C	57.54 ± 17.9	4.13 ± 3.0 ^C	80.50 ± 10.3	1.40 ± 1.5 ^C	90.98 ±4.7	0.87 ± 1.1 ^C	93.32 ± 4.3	0.27 ± 0.8 ^C	96.62 ± 5.6	0.00 ^C	100
F values (with $df = 2.42$)	0.173	10.451	I	31.849	I	60.179	I	58.327	I	37.313	I	43.018	I
T values (with $df = 1.8$)	I	I	3.237	I	9.001	I	6.658	I	3.600	I	7.72	I	4.215
Ρ	0.842	< 0.001	0.012	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.007	< 0.001	< 0.001	< 0.001	0.003
Means within each columr reduction comparing to co	thearing differe Introl (mean ± S	ent superscript D)	letters are signi	ificantly differe	nt according to	Duncan test (/	^o = 0.05). Infest	ation = no. of v	vhitefly nymph	is per leaf (mea	in ± SD). Reduc	tion = percent	age (%) of

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Table 3 Population de	ensities and	reduction (%) of the aph	iid, A. gossyp	ii, on sweet	pepper plan	its received t	wo releasing	g rates of th	e second lar	val instar of	C. carnea	
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Releasing rate per plant	Infestation	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction
Control (untreated plants)	3.87 ±2.4 ^A	11.53 ±4.1 ^A	I	15.20 ± 4.8 ^A	I	16.47 ± 5.4 ^A	I	14.60 ±5.8 ^A	I	12.47 ± 5.3 ^A		7.67 ± 3.7 ^A	1
2	4.73 ± 3.2 ^A	4.40 ±1.9 ^B	61.54 ± 7.2	1.47 ± 1.5 ^B	90.24 ± 4.4	0.40 ± 0.7 ^B	97.60 ±1.8	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100
10	3.53 ± 2.9 ^A	2.47 ±2.4 ⁸	78.78 ± 11.5	0.47 ± 0.7 ^B	96.96 ± 2.6	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100
F values (with $df = 2.42$)	0.710	38.718	I	116.077	I	134.497	I	96.557	I	82.893	I	65.194	I
T values (with $df = 1.8$)	I	I	2.837	I	2.936		2.981	I		I		I	
Ρ	0.497	< 0.001	0.022	< 0.001	0.019	< 0.001	0.018	< 0.001		< 0.001		< 0.001	
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Releasing rate per plant	Infestation	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction	Infestation	Reduction
Control (untreated plants)	32.73 ± 2.5 ^A	5.33 ± 2.8 ^A		6.73 ± 3.9 ^A		6.93 ± 3.7 ^A		5.93 ± 3.7 ^A		7.07 ±4.6 ^A		7.00 ± 4.2 ^A	1
2	3.13 ± 3.0 ^A	2.47 ±2.5 ^B	53.60 ± 27.4	0.60 ± 1.1 ^B	91.06 ± 6.6	0.13 ± 0.5 ^B	97.98 ±4.5	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100
10	2.60 ± 2.8 ^A	0.60 ±1.1 [⊂]	89.06 ± 12.6	0.13 ± 0.5 ^B	97.92 ± 4.7	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100	0.00 ^B	100
F values (with $df = 2.42$)	0.151	16.839	I	35.870	I	51.947	I	39.549	I	34.848	I	40.833	I
T values (with $df = 1.8$)	I	I	2.633	I	1.905	I	1.000	I	I	I	I	I	
μ	0.861	< 0.001	0:030	< 0.001	0.093	< 0.001	0.347	< 0.001	I	< 0.001	I	< 0.001	I
Means within each columr reduction comparing to co	n bearing differ ontrol (mean ± 5	ent superscript l 5D)	etters are signi	ificantly differe	nt according to	Duncan test (/	^o = 0.05). Infest _i	ation = no. of v	whitefly nympl	rs per leaf (me:	an ± SD). Reduc	tion = percent	age (%) of

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Authors' contributions

All authors read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

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Author details

¹Biology Department, Faculty of Science, Taif University, Taif 888, Saudi Arabia. ²Biotechnology Department, Faculty of Science, Taif University, Taif 888, Saudi Arabia. ³Department of Economic Entomology and pesticides, Faculty of Agriculture, Cairo University, Giza 12613, Egypt.

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