Journal of Asia-Pacific Entomology 18 (2015) 297–302

Contents lists available at ScienceDirect

Journal of Asia-Pacific Entomology

journal homepage: www.elsevier.com/locate/jape

Predation activity of *Chrysoperla carnea* (Neuroptera: Chrysopidae) upon *Aphis fabae* (Hemiptera: Aphididae): Effect of different hunger levels

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ARTICLE INFO

Article history:
Received 14 July 2014
Revised 16 February 2015
Accepted 17 March 2015
Available online 23 March 2015

Keywords:
Chrysoperla carnea
Aphis fabae
Functional response
Starvation
Predator–prey interactions

ABSTRACT

This research investigated the effect of different hunger levels on the functional response of three larval instars of *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae) to a mixture of third and fourth nymphal instars of *Aphis fabae* Scopoli (Homoptera: Aphididae). Hunger levels for the first and second larval instars of the predator were 4, 12 and 24 h. For the third instar, the hunger levels were 2, 4 and 8 h. At 4, 12 and 24 h, the functional responses for the first instar larvae of the predator were from types II, II and III, whereas those for second instar were from types III, III and II, respectively. The last instar of the predator showed the functional response types II, II and III at 2, 4 and 8 h hunger levels, respectively. The handling times estimated for the first instar larvae of the predator were not significantly different between hunger levels. However, there was a significant difference between the handling times estimated for the second and third larval instar of the predator at different hunger levels.

Introduction

The black bean aphid, *Aphis fabae* Scopoli (Hemiptera: Aphididae), is a polyphagous cosmopolitan pest (Blackman and Eastop, 2000). It is one of the most important pests of different crops throughout the world (Minks and Harrewijn, 1989; Völkl and Stechmann, 1998).

The common green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae), is a voracious predator of a wide range of soft-bodied arthropods, including aphids, scales, caterpillars, mealy bugs, psyllids, whiteflies, leafhoppers, thrips and eggs and small larvae of lepidopteran pests, mites and others (e.g., Principi and Canard, 1984; Pappas et al., 2007; Yadav and Pathak, 2010). This species has received much attention as a commercial biological control agent due to its high voracity, searching ability and ease of laboratory and commercial rearing (Principi and Canard, 1984; Tolstova, 1986).

The functional response refers to the changes in the predation rate of the predator in response to the changes in prey densities (Solomon, 1949; Holling, 1959, 1965). The functional response is an important characteristic of predator–prey dynamics. Understanding the functional response of predators is important to improve the effectiveness of the biological control of insect and mite pests (Jeschke et al., 2004). The functional response of a natural enemy is influenced by abiotic factors, e.g., temperature (McCaffrey and Horsburgh, 1986; Enkegaard, 1994; Mohaghegh et al., 2001; Skirvin and Fenlon, 2003) and relative humidity (Svendsen et al., 1999). However, it is also affected by characteristics of the natural enemy itself such as life stage and sex (Shipp and Whitfield, 1991; Omkar and Pervez, 2004; Ding-Xu et al., 2007; Seko and Miura, 2008; Cabral et al., 2009). An additional predator characteristic that may influence the functional response is the predator's hunger level. This may affect their activity as well as their searching and capturing efficiency.

Several studies have been carried out on the functional response of *C. carnea* to different prey types (Fathipour and Jaafari, 2003; Atlihan et al., 2004; Hassanpour et al., 2009; Abd El-Gawad et al., 2010; Hassanpour et al., 2011). However, there are a few studies regarding the influence of different hunger levels of arthropod predators on functional responses or other elements of predation activity (Hénaut et al., 2002; Rossi et al., 2006). Hénaut et al. (2002) found that starvation changed the search path characteristics of *Orius majusculus* (Reuter). They assumed that this could be related to the physiological states of hunger and energy availability. In other study, both well-fed and hungry spiders, *Nesticodes rufipes* (Lucas), showed a type II functional response to the adults of *Musca domestica* (L.) over a 168 h period (Rossi et al., 2006). They additionally found that the higher number of prey killed by well-fed spiders over a 24 h period most likely occurred due to their greater weight, compared to that of the hungry spiders.

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http://dx.doi.org/10.1016/j.aspen.2015.03.005

The aim of this study was to evaluate the effect of different hunger levels of three larval instars of *C. carnea* on their functional responses to *A. fabae* nymphs.

Materials and methods

Both the rearing of insects and experiments were carried out in a growth chamber at 26 ± 2 °C, 55 ± 5 % RH and L16:D8 photoperiod.

Insect culture

C. carnea was obtained from a laboratory culture at the Iranian Research Institute of Plant Protection (IRIPP) in Tehran, Iran. The procedure for rearing the predator was similar to that described by a previous study by Hassanpour et al. (2011). A stock culture was established by introducing approximately 30–40 *C. carnea* adults of both sexes to plastic containers (11.5 cm diameter and 22.5 cm height). These were covered by gauze for ventilation, as well as a substrate for egg-laying on the top. The adults were fed on an artificial diet consisting of brewer's yeast, honey and distilled water in a 4:7:5 ratio. This was smeared as a paste on transparent plastic tapes inserted in the containers. Extra water was provided by placing a wet sponge on the top of the containers. Eggs laid on the inner surface of the containers as well as on the gauze were transferred daily to other plastic containers (17.5 cm diameter and 7.5 cm height) with screen-covered holes (1 cm in diameter) in the lids for ventilation. The hatching predator larvae were in the latter containers with abundant food (frozen eggs of *Anagasta kuehniella* (Zeller) (Lep., Pyralidae) were provided every day). Several layers of paper towels were added to the containers to reduce cannibalism among the *C. carnea* larvae.

The colony of *A. fabae* was obtained from a laboratory culture reared on bean plants in the University of Tehran, Karaj, Iran. They were subsequently reared on bean plants (*Vicia faba* L. *cv.* Barakat) in a growth chamber.

Experimental design

The experimental arenas were plastic Petri dishes (9 cm in diameter) with a mesh-covered hole in the lid for ventilation. Furthermore, the floor was lined with a thin layer of solidified agar solution (3%) to prevent bean leaves from desiccation. In each Petri dish, two bean leaves (approximately 35 cm^2) were centred upside down on the agar solution. Each setup was infested with one of 5–6 densities (2, 4, 8, 16, 24 for the first instar (L1); 4, 8, 16, 32, 64, 96 for the second instar (L2); and 10, 25, 50, 100, 150, 200 for the third instar (L3) lacewings) of a mixture of third and fourth instars nymphs of *A. fabae*. Preliminary experiments showed that the survival of the aphid nymphs during the experimental period was more than 97%. Thus, there was no need to control treatment in the experiments. The maximum prey densities for each predator instar were determined based on a preliminary feeding study. The predator larvae used for the experiment had been less than 24 h in the actual instar stage. For obtaining the second and third larval instars of the lacewing for use in the experiments, the first instar larvae (<12 h old) were reared singly in Petri dishes (9 cm in diameter with a screen-covered hole in the lid for ventilation) until they reached the desired stage. The hunger levels for L1 and L2 were 4, 12 and 24 h and for L3 were 2, 4 and 8 h. The predator larvae were starved for the above-mentioned durations by placing them singly in Petri dishes devoid of prey. Each predator was only tested once. Ten replicates of each prey density were performed for all larval instars.

The predators were introduced to the experimental arenas 1 h after the aphids were transferred to the leaves. After 24 h for L1 and L2 and after 4 h for L3, the predators were removed and the numbers of prey consumed (total minus alive) were evaluated. Consumed preys were not replaced during the experiments.

Processing of data

The type of functional response was determined by a logistic regression analysis [SAS/STAT, CATMOD procedure (SAS Institute, 2001)] of the proportion of killed prey (N_e) in relation to initial prey density (N_0) (Trexler et al., 1988; Trexler and Travis, 1993). The data were fitted to a function that describes the relationship between N_0 and N_e/N_0 (model 1) (Juliano, 2001):

$$\frac{N_e}{N_0} = \frac{\exp\left(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3\right)}{1 + \exp\left(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3\right)} \tag{1}$$

where N_e is the number of aphid eaten, N_0 is the initial prey density and P_0, P_1, P_2 and P_3 are the intercept, linear, quadratic and cubic coefficients, respectively, related to the slope of the curve, and estimated using the method of maximum likelihood. The slope for the type II response is declining (i.e., P_1 is negative), whereas for the type III response is accelerating (i.e., P_1 is positive and P_2 is negative) (Juliano, 2001). Because the values estimated for the linear coefficient in the original cubic model were not significant ($P_1 > 0.05$) in some cases (first instar, 4 and 12 h hunger levels; second instar, 24 h hunger level; third instar, 8 h hunger level), the model was reduced by omitting the cubic terms until all of the remaining coefficients were significant (see Juliano, 2001).

Once the type of functional response was determined, the random attack equation (Royama, 1971; Rogers, 1972) was used to describe the functional responses. The random attack equation allows for prey depletion during the course of the experiment. For the types II and III functional responses, the following models were used to fit the data:

$$N_e = N_0\{1 - \exp[a(T_h N_e - T)]\} \tag{2}$$

$$N_e = N_0\{1 - \exp[(d + bN_0)(T_h N_e - T)/(1 + cN_0)]\} \tag{3}$$

where T is the total time available for attack; a is the attack rate; T_h is the time spent by the predator for handling, killing and consuming each prey; and the coefficients of b, c and d are constants from the function that relates a and N_0 in the type III functional response: $a = (d + bN_0)/(1 + cN_0)$ (Hassell et al., 1977).

In the type III functional response, since the two constants (c and d; model 3) were not significantly different from 0 (i.e., 95% confidence interval [CI] included 0), a reduced version of the model (model 4) (see Juliano, 2001) was applied to estimate the parameters:

$$N_e = N_0\{1 - \exp[bN_0(T_h N_e - T)]\} \tag{4}$$

Parameters were estimated using the non-linear least squares regression procedure (PROC NLIN; SAS Institute, 2001) and compared based on their 95% CI. Differences between the parameters were considered significant if their CIs did not overlap (e.g., Zamani et al., 2006; Li et al., 2007; Madadi et al., 2007; Mahdian et al., 2007; Hassanpour et al., 2011).

An analysis of variance (one-way ANOVA; PROC GLM; SAS Institute, 2001) was used to compare the consumption of *A. fabae* nymphs by each larval instars of *C. carnea* at the different hunger levels. The means were compared using Tukey's test at $P < 0.05$ (SAS Institute, 2001).

Results

The functional responses of the three larval instars of *C. carnea* to *A. fabae* are illustrated in Fig. 1. The results of the logistic regression analyses (full or reduced model of Eq. (1)) indicated that in treatments with hunger levels of 4 and 12 h starvation, the functional response of *C. carnea* L1 to *A. fabae* nymphs was of type II (significant negative linear coefficient, Table 1). However, the response after 24 h starvation was of type III (significant positive linear coefficient, Table 1). In contrast, for *C. carnea* L2, at the same hunger levels, functional responses of types