COMPARISON OF FUNCTIONAL RESPONSE OF TWO AND ONE INDIVIDUAL FEMALE PREDATOR, *HIPPODAMIA VARIEGATA* GOEZE (COLEOPTERA: COCCINELLIDAE) TO DIFFERENT DENSITIES OF *APHIS GOSSYPII* GLOVER (HEMIPTERA: APHIDIDAE) UNDER LABORATORY CONDITIONS

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ABSTRACT: The functional response of two and one individual female predator, *Hippodamia variegata* Goeze (Col.: Coccinellidae) to different densities of third instar nymphs of *Aphis gossypii* Glover (Hem.: Aphididae) were studied in a growth chamber (25°C, 65 ± 5% RH and a photoperiod of 16L: 8D h) on the black eyed bean, *Vigna unguiculata*. Each experiment was replicated ten times. Using the logistic regression, a type II functional response was determined for two and one female ladybeetle. The searching efficiency (a') and handling times (T_h) of the female adults by using Nonlinear regression were estimated $0.003 \pm 0.000424^{h-1}$, 0.106 ± 0.00441^{h}, $0.122 \pm 0.021^{h-1}$ and 0.105 ± 0.048^{h} for two and one individual predator, respectively. The voracity of female *H. variegata* in both cases had a similar trend showing a direct dependence on prey density ($R^2= 0.9976$, 0.9991). This study revealed that two individual predator reduced the searching efficiency of *H. variegata*.

KEY WORDS: *Hippodamia variegata*, *Aphis gossypii*, functional response, two individual predator.

The cotton or melon aphid, *Aphis gossypii* Glover (Hemiptera: Aphididae) is a cosmopolitan, polyphagous species widely distributed in tropical, subtropical and temperate regions. This aphid is a pest of cotton, cucurbits and citrus, and in temperate zones principally attacks vegetables in fields and greenhouses (Leclant & Deguine, 1994). The palaearctic coccinellid species *H. variegata* is a widespread aphidophagous predator in Europe (Hodek & Honek, 1996). *H. variegata* has been cited as the most important predator of aphids on pepper in Bulgaria, on maize in Ukraine, on shrubs in Italy, on grain in India, and on cotton in Turkmenistan (Kontodimas & Stathas, 2005). It has been recorded feeding on 19 different aphid species in Turkey (Aslan & Uygun, 2005) and on 12 different aphid species and a psyllid on a variety of crops, weeds, and ornamental plants in Australia (Franzmann, 2002).

One of the criteria to evaluate the efficiency of a predator is its feeding response to changes in prey density, or its functional response, i.e., the number of prey that an individual predator kills as a function of prey density (Holling, 1966).

The functional response gives a quantitative description of the behavior of a predator when it encounters different densities of its prey. The term functional response shows the response of individual natural enemies to varying prey density (Solomon, 1949). Statistical methods for estimating the parameters of functional response models from experimental data are described in Livdahl & Stiven (1983), Houck & Strauss (1985), Williams & Juliano (1985) and Juliano & Williams (1987). When the number of prey killed is plotted against the number of prey available, a continuum of patterns may emerge from which ecologists delimit three main types of functional responses (Holling, 1966; Trexler et al., 1988). Functional response curves may represent linear increase (type I); an increase decelerating to a plateau (type II); sigmoid increase (type III) and a dome shaped response (type IV) (Pervez & Omkar, 2005).

An investigation on the functional response of the ladybeetle, *H. variegata* Goeze at different densities of the mealy plum aphid, *Hyalopterus pruni* Geoffroy showed that response of each larval stage and adults matched Holling's type II functional response (Atlihan & Guldal, 2009). The functional responses of female and male, third and fourth instars larvae of *Scymnus syriacus* Marseul to different densities of *Aphis spiraecola* Patch were found to be a type II (Emami, 1996). Functional response of *S. levaillanti* to the density of cotton aphid, *A. gossypii* and *S. loewii* to the different densities of bollworm eggs, *Helicoverpa zea* was studied and the results showed a type II functional response for the former (Isikber, 2005) and a type I functional response for the latter (Parajulee et al., 2006).

As all functional response experiments have been carried out using one single predator (parasitoid) in each patch to date, so the objective of this study was to compare the type of functional response shown by two and one individual female of *H. variegata* in a patch.

MATERIALS AND METHODS

Predator and prey stocks

Adults of *H. variegata* were collected from aphid (*A. gossypii*) colony on an infested *Hibiscus syriacus* at the College of Agriculture in the University of Guilan (North of Iran). They were reared on the black eyed bean, *Vigna unguiculata* infested with *A. gossypii* Glover. The predator was reared for one generation on different nymph stages of the cotton aphid, *A gossypii*, before starting the experiments. The nymphs of *A. gossypii* were collected from *Hibiscus syriacus* and reared on black eyed bean. All aphids and predators stocks were kept in a growth chamber at 25 ± 1°C, 65 ± 5% RH and a photoperiod of 16: 8 (L: D) h.

Functional response

To study the functional response of *H. variegata,* two individual female adults of the predator were presented to different densities of third instar nymphs of *A. gossypii* (20, 40, 80, 120, 160 and 200) in a transparent plastic container (15×13×3 cm) on a leaf of black eyed bean. In the case of one predator, female was presented to a different densities of prey (10, 20, 40, 60 and 80) as well.

Lady beetle female was starved 24 h. Aphids were gently transferred by a fine brush from host plant to the host plant leaves in experimental arenas. Each experiment was replicated five times. After 24 h, the number of aphids eaten by two and one female ladybeetles was recorded.

Data analysis

Data analysis for functional responses includes two steps. In the first step, the shape (type) of functional response is determined. A logistic regression of proportion of eaten prey versus initial number of prey offered is the most effective way to make this determination (Juliano, 2001). To do this, a polynomial logistic function was fitted to data:

$$N_e/N_o = \exp(P_o+P_1N_o + P_2N_o^{2} + P_3N_o^{3})/(1+\exp(P_o+P_1N_o+P_2N_o^2+P_3N_o^3)) \qquad [1]$$

where $P0$, $P1$, $P2$, and $P3$ are the parameters to be estimated using the CATMOD procedure in SAS software (Juliano, 2001, SAS Institute, 2003). The two data sets were fitted individually to equation 1 and types of functional responses were determined by examining the signs of $P1$ and $P2$. A positive linear parameter ($P1$) together with a negative quadratic parameter ($P2$) would indicate a type III functional response, whereas if the linear parameter is negative, a type II functional response is indicated (Juliano, 2001). In the second step, nonlinear least squares regression (PROC NLIN; SAS Institute Inc. 2003) was used to fit Rogers' type II random predator equation (Rogers, 1972) to data and to estimate the functional response parameters. Because prey were depleted during the experiment, this model, which does not assume constant prey density, is appropriate for this experiment (Rogers, 1972, Juliano, 2001). Holling's disc equation, in contrast, is based on an assumption of unchanging prey density (Rogers, 1972), and is thus inappropriate for this experiment (Juliano, 2001). The form of Rogers' type II random predator model is:

$$N_a = N_o\,[1-\exp(a\,(T_h N_o - T))] \qquad [2]$$

where N_a is the number of prey eaten, N_o is the number of prey offered, T is the total time available for the predator, a is the searching efficiency, and T_h is the handling time. Statistical analysis of the functional response was performed using the SAS software (SAS Institute, 2001). In order to estimate handling time and searching efficiency, a nonlinear regression (the least square technique with DUD initialization) was used.

In order to evaluate the ratio of natural mortality of prey, control treatments were performed in the absence of predators with the above-mentioned prey densities. Laboratory conditions were the same as previously mentioned. Voracity (Vo) of *H. variegata* females on *A. gossypii* was determined using the following equation (Soares et al., 2003):

$$V_o = (A - a_{24})\ ra_{24}$$

where Vo is the quantification of voracity (the number of prey eaten), A is the number of prey available, a24 is the number of prey alive after 24 h and ra24 is the ratio of prey alive after 24 h in initial number of aphids. Curves were drawn by Excel.

RESULTS

The number of prey eaten increased with increasing the prey density. Parameter estimates for logistic regressions of proportion of prey killed (N_a/N_o) against number of prey offered (N_o) for two and one individual female adults of *H. variegata* for 24 h are presented in Table 1. The logistic regression for female predator had a negative linear parameter (a type II functional response) and the proportion of prey consumed by female ladybeetle declined with increasing prey density (Fig. 1, 2). The coefficient of attack rates (a') and handling time (T_h) (estimated by Rogers random attack equation) for this predator are presented in Table 2.

The Voracity (number of prey eaten) of adult females increased significantly with prey density ($R^2 = 0.9976, 0.9991$) in both cases (Fig. 3).

DISCUSSION

A type II functional response was shown by two and one individual female of *H. variegata*. In this study the declining proportion of prey consumption with the increasing prey density indicates that the functional response data were described well by a Type II asymptotic curve. This type of functional response has been reported for many different coccinellids including: *Aphidecta obliterata* (L.) and *Adalia bipunctata* (L.) preying on *Elatobium abietinum* (Walker) (Timms et al., 2008), larvae and adults of *Propylea quatuordecimpunctata* preying on *Diuraphis noxia* (Mordvilko) (Messina & Hanks, 1998), *Coccinella undecimpunctata* (L.) preying on *A. fabae* and *Aleyrodes proletella* (Moura et al., 2006), male adults of *Cheilomenes sulfurea* preying on *A. fabae* (Hodek et al., 1984), adult females of *Cheilomenes sexmaxulata, Propylea dissecta,* and *Coccinella transversalis* preying on *Aphis craccivora* (Koch) or *Myzus persicae* (Sulzer) (Pervez & Omkar, 2005), and adults of *Curinus coeruleu* preying on *Heteropsylla cubana* (Silva et al., 1992). In contrast, type III functional responses appear to be relatively rare among coccinellids. Isikber (2005) determined functional responses of two coccinellids species under three temperature regimes, and found that only *Cycloneda sanguine* exhibited a type III functional response, and only at 25°C. Sarmento et al. (2007) showed that *Eriopis connexa* exhibits different types of functional responses to *Macrosiphum euphorbiae* (type III) and *Tetranychus evansi* (type II).

The success of coccinellids in biological control (Obrycki & Kring, 1998) coupled with the prevalence of type II functional responses, suggesting that successful biological control is possible with type II functional responses. Fernandez-Arhex & Corley (2003) reviewed the literature on functional responses of parasitoids used in biological control and showed that only about a quarter of studies showed type III functional responses, providing further evidence that although functional responses are an important element in selection of efficient biocontrol agents, there is no general relationship between success in biocontrol and the type of functional response. Other aspects of prey and predator biology, including: prey preference, switching, intrinsic rate of increase of prey and predator, killing rate of predator, host patchiness, predator patch allocation time, effect of host plant, effect of abiotic factors, and intra- and inter-specific predator competition, can all have important effects on the efficiency of a biocontrol agent (Murdoch & Briggs, 1996; Obrycki & Kring, 1998). Functional response experiments are routinely done in controlled conditions, like those used in our experiments. Clearly, in nature, the predator will encounter uncontrolled and highly variable conditions. Changes in temperature, humidity, and other aspects of weather could radically change the functional responses of predators to change in prey density. It was found that the density (two individuals) of predator influenced the searching efficiency of *H. variegata* , as it declined to over 40 times. It can be assumed that the increase of predator (parasitoid) density in patches of functional response experiments would decrease their searching efficiency. This may be similar to the behavior of predators in mutual interference experiments, as inverse density dependence in searching efficiency is known as parasitoid interference or mutual interference (Beddington, 1975). However, the voracity of the predator in both cases (two and one individual predator) had similar trends resulting in direct prey density dependence.

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Table 1. Maximum likelihood estimates from logistic regression of proportion of prey eaten as a function of initial prey densities by two and one female of *Hippodamia variegata.*

	Parameters	Estimate	SE	X^2	P
Two individual predator	Constant	13.7816	1.5423	79.85	<.0001
	Linear	-0.1348	0.0195	43.76	<.0001
	Quadratic	0.000398	0.000060	47.96	<.0001
One individual predator	Constant	4.1601	1.0137	16.84	<.0001
	Linear	-0.0960	0.0764	1.58	0.2088
	Quadratic	0.00162	0.00169	0.92	0.3381

Table 2. Coefficient of attack rate (a') and handling time (T_h) of one and two individuals female of *Hippodamia variegata* fed on *Aphis gossypii* (Rogers's model).

Model	Parameter	Estimate	SE	95% CI Lower	95% CI upper	R^2
Two individual Predator	a‘	0.003	0.000424	0.00215	0.00384	0.996
	T_h	0.106	0.00441	0.0977	0.01153	
One individual predator	a'	0.122	0.021	0.08	0.165	0.981
	T_h	0.105	0.048	0.008	0.201	

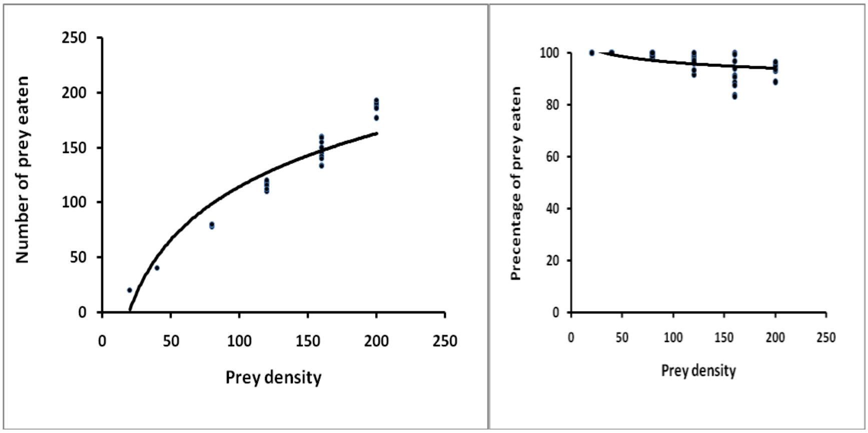

Figure 1. Functional responses (up) and percentage of predation (down) of two individual female *Hippodamia variegata* to different densities of *Aphis gossypii*.

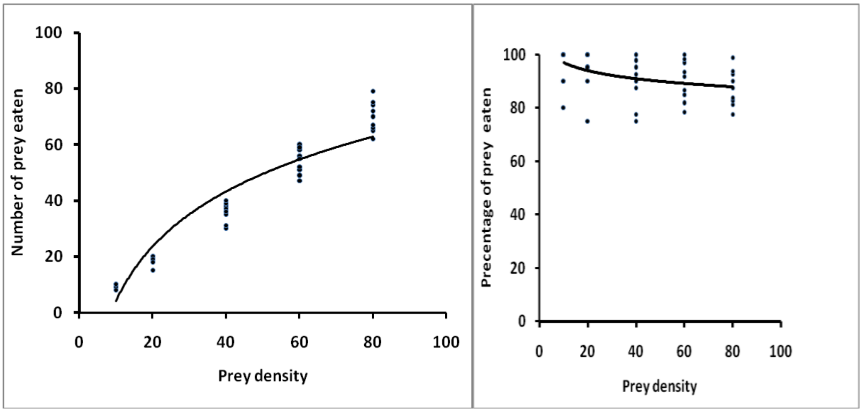

Figure 2. Functional responses (up) and percentage of predation (down) of one individual female, *Hippodamia variegata* to different densities of *Aphis gossypii*.

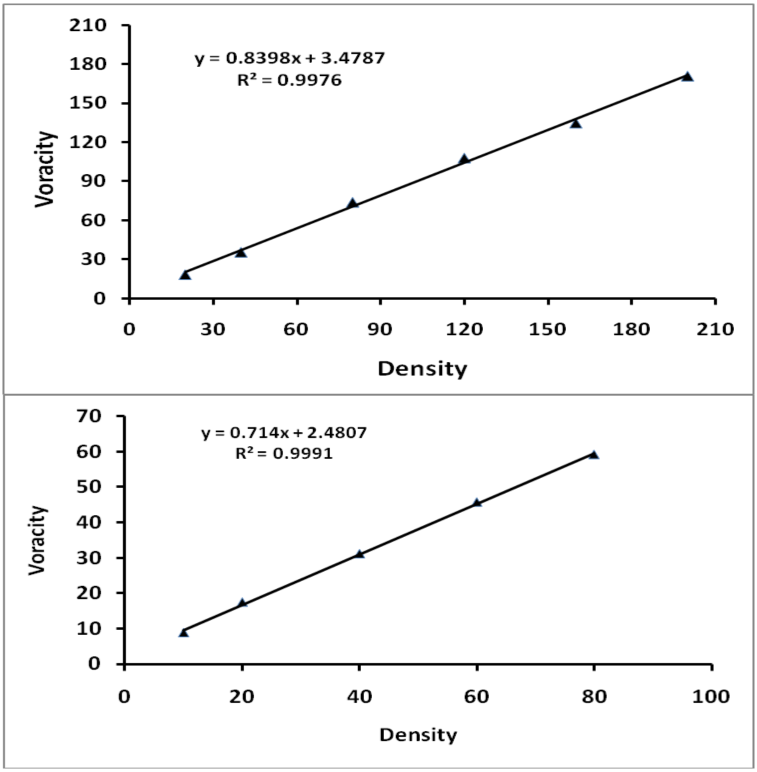

Figure 3.Voracity of two (up) and one (down) individual female *Hippodamia variegata* on different densities of *Aphis gossypii*.