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Functional response of the coccinellid predator *Adalia fasciatopunctata revelierei* to walnut aphid (*Callaphis juglandis*)

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Abstract The functional response types and parameters of 3rd and 4th instar larvae, and adult females and males of a coccinellid predator, *Adalia fasciatopunctata revelierei* (Mulsant) (Col.: Coccinellidae), were evaluated at five different densities of *Callaphis juglandis* (Goeze) (Hemiptera: Aphididae) in order to understand their role for the aphid's biological control. Experiments were carried out in petri dishes at 25±1°C, 60±10% r.h. and 16L:8D photoperiod in a controlled temperature room. All tested stages exhibited a Type II response determined by a logistic regression model. The attack rate (α) and handling time (T_h) coefficients of a Type II response were estimated by fitting a "random-predator" equation to the data. Although the estimates of α for all stages of *A. fasciatopunctata revelierei* tested were similar, the longest T_h was obtained for 3rd instar larva because of the lower consumption rate at densities above 40 prey/day. Results indicated that the adult female has the highest predation of *C. juglandis* followed by 4th instar larvae, adult males and 3rd instar larvae. However, further field-based studies are needed to draw firm conclusions.

Keywords Biocontrol · Pest control · Predation · Predator-prey systems

Introduction

Predation is assumed to be one of the significant biotic mortality factors reducing insect pest populations, and using them in insect pest management programs has been receiving increased attention because of the current need to reduce the exclusive use of insecticides for pest control (DeBach and Rosen 1991; Luck 1984; Riudavents and Castane 1998; Sarmento et al. 2007; Wiedenmann and Smith 1997). Functional response of a predator is one of the important factors regulating population dynamics of predator–prey systems, and functional response curves can be used to infer basic mechanisms underlying predator–prey interactions, clarify coevolutionary relationships, and enhance biological control (Houck and Strauss 1985).

Coccinellids are one of the important groups of predatory insects because of their biocontrol potential and they have been receiving attention as biological contol agents mainly of hemipterous species (Atlıhan and Özgökçe 2002; Isikber and Copland 2002; Obrycki 1998; Oliveira et al. 2004; Pervez and Omkar 2005). Although they generally show a Type II

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functional response (Kawauchi 1991; Ofuya and Akingbohungbe 1988; Uygun and Atlihan 2000), the other types of functional responses are also reported for the group (Hassel et al. 1977; Hu et al. 1989; Lou 1987). On the other hand, the response of a single species of coccinellid predator can differ between various types of prey (He et al. 1994; Hu et al. 1989; Lou 1987) and the response of different stages of a predator can differ within a single type of prey (Koch et al. 2003).

Callaphis juglandis (Goeze) (Hemiptera: Aphididae) is an important pest that attacks walnut trees in Turkey, especially in the Lake Van basin. This species is common throughout Europe (Barbagallo et al. 1995; Heie 1992; Nieto Nafria and Mier Durante 1998; Petrovič 1998), Central Asia and the USA (Blackman and Eastop 2000). The aphid reduces tree vigor and causes quality and yield losses by consuming the phloem content, and by excreting honeydew that may harm the husks of the walnut or allow sooty mold to develop.

The aphidophagous coccinellid *Adalia fasciatopunctata revelierei* (Mulsant) (Col.: Coccinellidae) is one of the most abundant natural enemies of *C. juglandis* in the walnut orchards of the Lake Van basin (Erol and Atlıhan 1995). In spite of its high population in walnut orchards, there is no informaton on its consumption capacity and efficacy. The present study was thus carried out to determine the functional response type of larva, adult female and male of *A. fasciatopunctata revelierei* using different densities of *C. juglandis* in the laboratory.

Materials and methods

Insect rearing Adults of *A. fasciatopunctata revelierei* (*Afr*) were collected from walnut orchards and brought to the laboratory. They were kept in a glass jar (100×150 mm) with slightly moist filter paper on which females could oviposit. The glass jars were covered with muslin and predators were reared on walnut leaves infested with *C. juglandis*. The eggs were collected from the filter paper and reared individually from the hatching of the egg to the stages that were used for the experiments. Thus, individuals of the same age and size as well as with normal response were used to standardize individuals used in experiments.

Stock cultures of the predator were maintained at 25±2°C, 65±5% r.h. and 16 h of artificial light of ~4,000 Lux in a controlled environment room.

Functional response studies The experiments were conducted with densities of 10, 20, 40, 80 and 160 *C. juglandis* in petri dishes (140×18 mm). The aphids were transferred into the petri dishes with a fine soft brush. A single *Afr* larva of 3rd and 4th instars, adult female and male were starved for 24 h and released into the dishes 2 h after the aphids were introduced. This interval ensured that aphids had dispersed throughout the dish. Only nymphs (3rd and 4th instar) of *C. juglandis* were used to prevent aphid reproduction during the experiment and to standardize consumption. After 24 h, the predators were removed and the number of consumed and/or killed prey was counted (n=10–12).

Treatments were carried out at 25±1°C, 60±10% r.h. and 16L:8D photoperiod in a climate cabinet.

Data analysis The logistic regression of the proportion of aphids consumed (N_e/N_0) as a function of initial density (N_0) was used to determine the shape of the functional response of *Afr* stages tested because of difficulties in discriminating between Holling's (1966) Type II and Type III (Trexler et al. 1988; Juliano 2001).

The polynomial function that describes the relationship between the proportion of aphids consumed and initial density was determined by fitting data (Juliano 1993):

$$\frac{N_e}{N_0} = \frac{\exp\left(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3\right)}{1 + \exp\left(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3\right)} \qquad (1)$$

where (N_e/N_0) is the probability an aphid will be consumed, and P_0, P_1, P_2 and P_3 are the maximum likelihood estimates as being intercept, linear, quadratic and cubic coefficients, respectively.

The type of functional response was determined by fitting data to the model (1). The signs of P_1 and P_2 were used to distinguish the shape of the curves. When the function is negative (linear coefficient <0), the predator displays a Type II functional response that indicates the proportion of prey consumed declines monotonically with the initial number of prey. When a positive density-dependent result for the proportion of prey consumed (linear coefficient >0 and quadratic coefficient <0) is obtained, the predator displays a Type III functional response (Juliano 2001).

Logistic regression analysis indicated that our data fit the Type II response for all stages of predator; for this reason, further analysis was restricted to the Type II response. The handling time and attack rate coefficients of a Type II response were estimated with nonlinear least square regression as described by Juliano (1993). Because the experiment was carried out without prey replacement during the course of the experiment, the appropriate model for a type II functional response is the "random-predator" equation (Rogers 1972):

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

in which N_e = number of prey attacked, T = exposure time (24 h), N_0 = inital prey density, α = attack rate, a constant rate of successful search, and T_h = handling time.

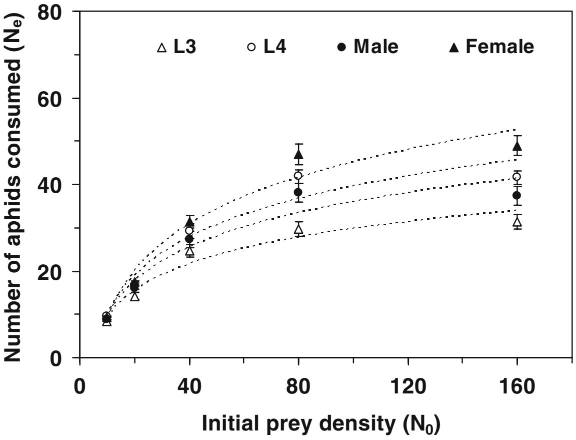
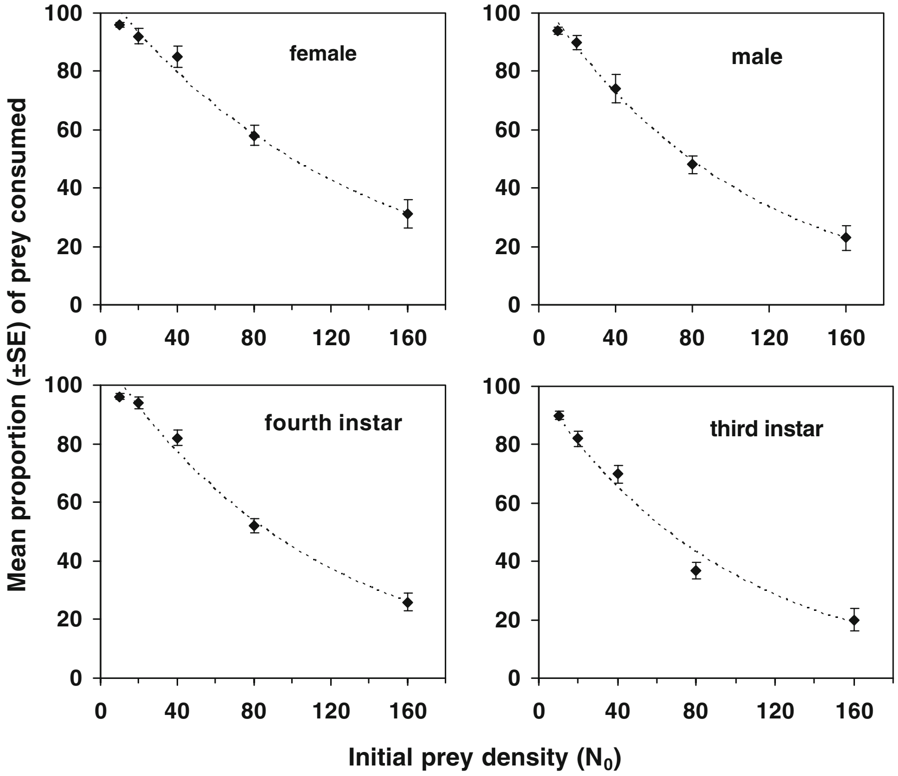


Fig. 2 Functional response of larvae and adults of *Adalia fasciatopunctata revelierei* feeding on *Callaphis juglandis*. Points show average number of aphids consumed and/or killed by the predator at each level of initial prey density. Error bars show standard errors

Fig. 1 Mean proportion (± SE) of prey consumed by *Adalia fasciatopunctata revelierei* larvae and adults at five different densities of *Callaphis juglandis*



The NLIN procedure in SAS was used to estimate parameters of Eq. 2 (SAS 2007). After α and T_h were determined for the original data (m_t), the differences among α values, as well as T_h values, were tested for significance by estimating the variance using the jackknife technique (Meyer et al. 1986). The jack-

Table 1 Maximum likelihood estimates (±S.E.) from logistic regression (as a function of initial prey densities) of proportion of prey consumed by larvae and adults of *Adalia fasciatopunctata revelierei*

Stage	Parameters	Estimates	χ^2	P
3rd instar	intercept	4.1357 (0.5328)	60.2466	< 0.001
	linear	−0.1021 (0.0268)	14.5325	0.0001
	quadratic	0.0007 (0.00037)	3.3151	0.0686
4th instar	intercept	8.3621 (1.6727)	24.9932	< 0.001
	linear	−0.2440 (0.0757)	10.3844	0.0013
	quadratic	0.0024 (0.00097)	5.9151	0.015
Female	intercept	8.2508 (1.4551)	32.1509	< 0.001
	linear	−0.2533 (0.0661)	14.6849	0.0001
	quadratic	0.0026 (0.00085)	9.5871	0.0020
Male	intercept	6.4974 (0.8120)	64.0275	< 0.001
	linear	−0.2172 (0.0381)	32.4668	< 0.001
	quadratic	0.0023 (0.00050)	21.3336	< 0.001

knife pseudo-value (m_j) was calculated for the n samples using the following equations:

$$\begin{aligned} m_{j_\alpha} &= n.m_{t\alpha} - (n-1)m_{i\alpha} \\ m_{j_{T_h}} &= n.m_{tT_h} - (n-1)m_{i_{T_h}} \end{aligned} \tag{3}$$

The mean values of (n–1) jackknife pseudo-values for α and T_h for each prey stage were subjected to analysis of variance followed by Duncan's Multiple Range Test ($P \le 0.01$) (SAS 2007). The effect of prey density on the proportion of aphids consumed was analyzed by one-way ANOVA followed by Duncan's Multiple Range Test ($P \le 0.05$) (SAS 2007). The maximum predation rates (attack rates) as T/T_h (Hassel 2000), which represent the maximal number of aphids that can be consumed during the time interval considered, were calculated by using estimates of T_h obtained by non-linear least squares regression. The standard errors of the maximum predation rates (T/T_h) of all stages were estimated by using the jackknife technique (Meyer et al. 1986).

Results

The percentage of prey consumed by each predatory stage in the petri dishes declined monotonically with the inital prey density (Fig. 1) and the logistic regression suggested Type II functional response for all stages (Fig. 2). This was because the estimate of the linear coefficient was significantly <0 and the quadratic coefficient was positive (Table 1). Therefore, the "random-predator" equation for Type II was used to estimate the attack rate coefficient (α) and the handling time (T_h) (Table 2). This model fits the observed data reasonably well.

Females of *Afr* responded more vigorously at prey densities higher than 40 aphids/day with an elevation

Table 2 Mean estimated values (±S.E.) of attack rate (α), handling time (T_h), and 95% confidence intervals (CL) for *Adalia fasciatopunctata revelierei* feeding on *Callaphis juglandis* nymphs

Stage	Attack rate (α)	95% CL		Handling time (T_h)	95% CL	
		Lower	Upper		Lower	Upper
Female	6.653±1.9381 a*	6.2557	13.0906	0.0197±0.00118 b	0.0193	0.0241
Male	5.071±2.0321 a	−2.5789	10.7206	0.0238±0.00310 ab	0.0139	0.0336
4th instar	5.514±2.4345 a	−4.9641	18.9917	0.0225±0.00204 ab	0.0160	0.0290
3rd instar	5.209±2.2368 a	−1.0923	12.5600	0.0304±0.00113 a	0.0268	0.0340

* Within columns, values followed by a common letter do not differ significantly (DMRT; $P \le 0.01$)

in the curve over other stages tested. The functional response curve of 3rd instar larva was significantly lower than the curves of the other predatory stages at prey densities higher than 20 aphids/day (Fig. 2).

The handling time (T_h), which is sometimes a good indicator of the predation rate, was the shortest for adult females and the longest for the 3rd instar larva (F=2.596; df=3; P=0.0603) (Table 2). Although differences among the handling times of adult females, males and 4th instar larvae were not statistically significant, the general trend usually remained towards higher performance for adult females. The estimate of handling time (T_h) value for females was 17%, 12% and 35% lower than those of males, 4th instar larvae and 3rd instar larvae, respectively. This was because adult females consumed considerably more aphids at densities of 80 and 160 aphids/day (Fig. 2). The attack rate coefficients (α) obtained for all stages were similar (F= 4.343; df=3; P=0.8726) (Table 2).

The maximum predation rate (T/T_h) was the highest for adult females (50.76±4.23), followed by 4th instar larvae (44.44±3.52), adult males (42.02±2.74) and 3rd instar larvae (32.89±3.25), (F=3.224; df=3; P=0.0576).

Discussion

The declining proportion of prey consumption with the increasing prey density for all predatory stages indicates that the functional response data were described well by a Type II asymptotic curve. The negative values obtained for the linear parameters ($P_1 \leq 0$) were a good indicator of a Type II response, as well as inverse density dependence relationship between the proportion of prey consumed and the initial prey density for all predatory stages tested. A Type II functional response was reported for other coccinellids (Atlıhan and Özgökçe 2002; Isikber and Copland 2002; Kawauchi 1991; Pervez and Omkar 2005). An inverse density-dependence relationship between the proportion of prey consumed and the initial prey density with Type II functional responses indicates the demographic destabilization of the aphid population. The persistence of the predator and prey populations in such a relationship might be due to factors such as metapopulation dynamics that seem likely to account for long-term population persistence in orchard ecosystems (Lester et al. 2005) as well as alternative prey. *Afr* is an aphidophagous predator, and its attack rate on *C. juglandis* might be lowered by attacking the other aphid, *Cromaphis juglandicola* Kalt., which may be another prey of predators in the walnut ecosystem. On the other hand, patch dispersal of both prey and predators might be another factor that allows predator and prey populations to display such a relationship (Lester et al. 2005). It should be noted, however, that none of these possible reasons mentioned was investigated in this study.

The higher proportion of prey consumption at lower densities for all stages of *Afr* tested was a typical result for a Type II response, and it indicates that this predator would be more effective at controlling the *C. juglandis* population at lower densities. The higher functional response curve at prey densities higher than 40 aphids/day (depending on higher consumption rate) for adult females may be due to a higher nutrient requirement which, in turn, may be a result of egg production, delayed satiation (Mills 1982), or a possible faster digestive rate of adult females (Pervez and Omkar 2005).

The attack rate coefficients obtained for the different stages tested did not differ statistically. This indicates that the attack rate coefficient did not change between different predator stages. Other reports on the attack rate coefficients of coccinellids are in agreement with the results described here (Atlıhan and Özgökçe 2002; Pervez and Omkar 2005). The handling time is a good indicator of consumption rate and effectiveness of a predator because it reflects the cumulative effect of time taken during capturing, killing, subduing, and digesting the prey (Veeravel and Baskaran 1997). The higher handling time value obtained for 3rd instar larvae was due to a lower consumption rate, especially at densities of 80 and 160 aphids/day. Because of lower consumption by 3rd instar larvae, any estimate of the voracity of *Afr* should consider the life stage.

Controlled laboratory studies provide insights into the development and population dynamics of insects, and therefore the results obtained in this study can be useful for the further study of population dynamics and the development of management tactics for control of *C. juglandis*. The relevance of the laboratory experiments on prey consumption is, however, limited because densities used in the laboratory may be unrealistically high. In addition, the time used by *Afr* in searching for food may reduce its prey consumption capacity under natural conditions.

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