Influence of different densities and nymphal instars of *Aphis gossypii* (Hemiptera: Aphididae) on developmental time and feeding rate of larvae of *Episyrphus balteatus* (Diptera: Syrphidae) under greenhouse conditions

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Abstract

Several aphidophagous insects such as predators and parasitoids are known to respond positively to aphid infested plants. This study was intended to evaluate the effect of different densities (40, 60, 80, 100) and nymphal instars (1st, 2nd, 3rd, 4th) of the cotton aphid *Aphis gossypii* Glover (Hemiptera: Aphididae) as prey on developmental time and feeding rate of the larvae of the syrphid fly *Episyrphus balteatus* DeGeer on cucumber leaves. The experiment was conducted at the greenhouse condition (22± 5°C, 55± 10% RH and 14L: 10 D h photoperiod) with 10 replications. Our study indicated that lower density and poor quality of prey caused higher larval mortality. Feeding on 3rd and 4th nymphal instars and higher density of prey (80 and 100 prey/day) caused shorter developmental time of *E. balteatus*. The larvae fed on densities of 40 and 60 prey, even though fed on higher nymphal instars, failed to complete their larval development. It is concluded that higher prey densities (80 and 100 prey) were highly suitable for predator's larval development and significantly reduced the developmental period of *E. balteatus*. These findings provide further evidence that *E. balteatus* has high predation capacity on aphids, and therefore can be used as a successful biocontrol agent against *A. gossypii*.

Key words: *Aphis gossypii, Episyrphus balteatus,* predation rate, nymphal instars, biological control

تاثیر تراکم و سنین مختلف پورگی شته (Aphis gossypii (Hemiptera: Aphididae بر دوره

رشدی و میزان تغذیه لارو مگس (Episyrphus balteatus (Diptera: Syrphidae

در شرایط گلخانه

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چکیده

بدیهی است چندین حشره شته‌خوار مانند شکارگرها و پارازیتوئیدها پاسخ مثبتی به گیاهان آلوده به شته نشان می‌دهند. در این مطالعه اثر تراکم‌های مختلف (٤٠، ٦٠، ٨٠ و ١٠٠) و سنین پورگی (١، ٢، ٣ و ٤) شته‌ی Aphis gossypii Glover (Hemiptera: Aphididae) به عنوان طعمه، بر دوره‌های رشدی و نرخ تغذیه‌ی لارو مگس شته‌خوار سیرفیده Episyrphus balteatus DeGeer (Diptera: Syrphidae)، روی برگ‌های گیاه خیار مورد بررسی قرار گرفت. آزمایش در شرایط گلخانه‌ای (٥ ±٢٢ درجه سلسیوس، رطوبت نسبی ١٠ ±٥٥ درصد و دوره نوری ١٤ ساعت روشنایی و ١٠ ساعت تاریکی) با ١٠ تکرار انجام شد. تراکم‌های مختلف طعمه اثر قابل توجهی بر میزان تغذیه شکارگر داشتند. مطالعه ما نشان داد که تراکم کم تر و کیفیت پایین طعمه، موجب تلفات زیاد لاروهای شکارگر می‌شود. از سوی دیگر، نشان داده شد که تغذیه از سنین پورگی بالاتر (٣ و ٤) و تراکم بالاتر شکار (٨٠ و ١٠٠ شکار در روز) باعث کوتاه‌تر شدن دوره رشد مگس *E. balteatus* شد. لاروهای تغذیه کرده از تراکم‌های ٤٠ و ٦٠ شکار، با وجود تغذیه از پوره‌های سنین بالاتر، موفق به تکمیل دوره لاروی خود نشدند. نتیجه‌گیری شد که تراکم‌های بالاتر (٨٠ و ١٠٠ طعمه) برای رشد لاروهای شکارگر بسیار مناسب بوده و طول دوره رشد لارو مگس *E. balteatus* را به طور معنی‌داری کاهش می‌دهند. این یافته‌ها، شواهد

بیش‌تری را ارائه داد که مگس *E. balteatus* ظرفیت بالایی در شکار شته‌ها داشته و در نتیجه می‌تواند به عنوان یک عامل بیوکنترل برای شته *A. gossypii* مورد استفاده قرار گیرد.

واژگان کلیدی: *Episyrphus balteatus, Aphis gossypii*، نرخ شکارگری، سنین پورگی، کنترل زیستی

Introduction

Aphids are major agricultural and horticultural pests throughout the world. They cause damage to crops directly through feeding on phloem tissue and also can contribute in severe indirect damage by acting as primary vectors of many plant viruses. Cotton aphid or melon aphid, *Aphis gossypii* Glover is one of the most serious greenhouse pests having a broad host range. It attacks more than 700 host plant species and transmits important viruses like potyviruses (Blackman & Eastop, 2000).

The intensive use of different chemical pesticides causes negative effects on the environment. Therefore, using natural enemies in biological control of insect pests become an important component of integrated pest management (Hodek & Honek, 1996; Atlihan & Kaydan, 2010). In order to increase or augment the effect of natural enemies in a prey population, it is necessary to understand the interaction between natural enemies and their preys (Rabb, 1974). In addition, it is important to measure the effectiveness of natural enemies prior to use them in a biological control program (Bazzocchi & Burgio, 2001; Mushtaq *et al.*, 2013).

Aphid communities are subject to predation by a broad range of specialist and generalist arthropod predators and parasitoids. Predators such as hoverflies (Gilbert, 1986), coccinellids (Hodek & Honek, 1996), lacewings (Principi & Canard, 1984), cecidomyiid midges (Madahi *et al.*, 2013), spiders (Sunderland *et al.*, 1986) and parasitoids (Stáry, 1970) are major components of the natural enemy guild associated with aphid colonies. Among these, hoverflies are efficient aphidophagous predators (Alhmedi *et al.*, 2008). *Episyrphus balteatus* De Geer (Diptera: Syrphidae) is an economically important syrphid species feeding on a broad range of aphid species in the field and greenhouses (e.g., Völkl *et al.*, 2007; van Lenteren, 2012). Adults of *E. balteatus* feed on nectar, pollen, plant saps, and aphids' honeydew, while, their larvae are voracious predators of more than 100 species of aphids (Sadeghi and Gilbert, 2000). Its predation has been shown on some species like *A. gossypii* (Poligui *et al.*, 2011), *Myzus persicae* Sulzer (Verheggen *et al.*, 2009), *Acyrthosiphon pisum* Harris (Putra and Yasuda, 2006) and *Brevicoryne brassicae* L. (Hindayana, 2001). Some studies have revealed that poor prey quality often results in high larval mortality (Sugiura and Takada, 1998; Sadeghi and Gilbert, 2000). However, its potency, for use in biological control, has not yet been studied on different prey densities and nymphal instars.

Understanding the response of the predator to varying prey densities and nymphal instars that each individual feeds on is essential for proper approach to the modeling of prey-predator interactions (Huffaker *et al.*, 1971). The limitation in their use in biocontrol may be accounted for the problem of rearing the adults (Schneider, 1969; Frazer, 1972). Therefore, egg production of females exposed to different prey types may be used as an indicator of prey quality and a correlation between preference and prey quality is to be expected. The aim of this study was to find out the effect of different nymphal densities and instars of *A. gossypii* on the biological parameters of *E. balteatus*.

Materials and methods

Host plant

The cucumber *Cucumis sativus* L. var. Guilan was grown in plastic pots (30 × 20cm) filled with a mixture of compost and rice flakes (3: 1). All plants were grown in a greenhouse at 22± 2 °C, 50± 10% RH and 14L: 10hD photoperiod.

Prey and predator stocks

The nymphs of *A. gossypii* were collected from rose mallow, *Hibiscus syriacus* (L.), *Cucumis sativus*, eggplant (*Solanum melongena*), watermelon *Citrullus lanatus* (Thunb.) Matsum. & Nakai and cucurbit fields in Guilan Province, Rasht (from Gil Sq 37° 21' 15.48" N,49° 25' 3.30" E to Manzarieh Blvd (37° 15' 35.46" N,49° 36' 11.34" E), fields around Pir Bazaar villages; Sheykh Mahalleh (37° 21' 16.08" N,49° 25' 54.72" E) and Lakesar (37° 21' 15.48" N,49° 25' 3.30" E). They were reared for several generations on cucumber in separate controlled muslin cages (2× 1.20× 1.80 cm).

Larvae of *E. balteatus* were collected from the colony of *A. gossypii* on infested rose mallow, cucumber, eggplant, watermelon and cucurbit fields, and the colony of *Aphis pomi* on Japanese quince, *Chaenomeles speciosa* Nakai at the Faculty of Agricultural Sciences, University of Guilan, Northern Iran. The predators were reared for four generations on *A. gossypii* before the start of the experiments.

The collected syrphid larvae were fed on *A. gossypii* on cucumber in muslin net covered containers (5 × 16 × 14 cm) daily. Pupae were also placed in the same containers. After emerging adults, they were transferred in muslin net covered containers (50 × 70 × 100 cm) where fed on *Gaillardia aristata*, canola pollen on disposable containers hanging at different heights supplied with a cotton piece soaked with 10% honey-water solution as supplementary diet. The diet was refreshed every 2-3 days. Females were stimulated to lay more eggs by placing cucumber seedlings infested with cotton aphids.

Biological parameters of *E. balteatus*

E. balteatus eggs were placed individually on cucumber fresh leaves in aerated petri dishes (10 cm in diameter and 1 cm in height). First larval instars of *E. balteatus* were daily exposed to different densities (40, 60, 80 and 100) of different (1st, 2nd, 3rd and 4th) nymphal instars of *A. gossypii* separately on a cucumber fresh leaf that placed upside down on moistened filter paper and cotton wool to maintain leaf freshness. Aphids were gently transferred with a fine brush from the host plant on the leaves. After 24 hours, the number of aphids consumed by the larval syrphid were recorded until pupal stage. In order to determine the effects of prey densities on the predator developmental time, only data of prey densities, 60, 80 and 100, were used. Larval development was not observed at density of 40 nymphal instar. Syrphid larval instars were differentiated by larval shells. Each treatment was replicated 10 times.

Statistical analysis

The feeding rate and developmental times of the larvae of *E. balteatus* fed on *A. gossypii* nymphal instars were subjected to the one-way analysis of variance (ANOVA) and t-test using the statistical software of Minitab 16.0 (Minitab Inc. 1994). Statistical differences among means were compared using Tukey Honestly Significant Difference (HSD) test at P < 0.05. Before analysis, all data was tested for normality by Kolmogorov-Smirnov method.

Results

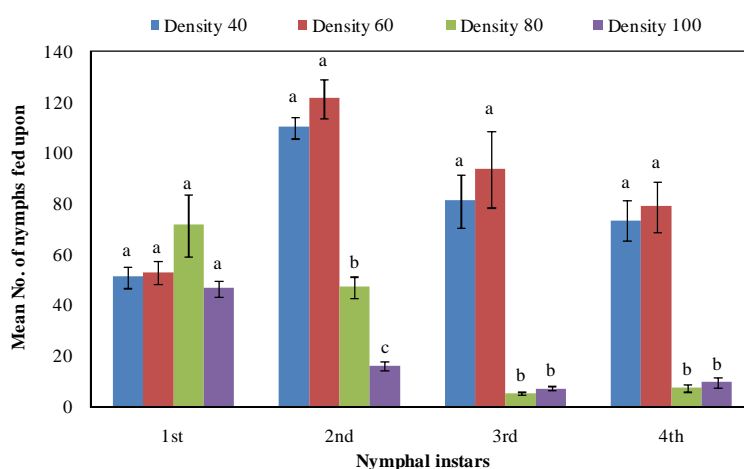**Feeding rate of *E. balteatus* larvae on different nymphal instars and densities of *A. gossypii***

The highest feeding rate was observed in 3rd larval instar of *E. balteatus* (467.2± 67.2) at density of 100 2nd instar nymphs and the lowest (5.333± 0.422) was observed in the 1st instar at the density of 80 3rd instar nymphs.

Effect of prey densities on predator feeding rate of *E. balteatus*

The feeding rate of 1st instar larvae of *E. balteatus* on different prey densities is shown in Fig. 1. First nymphal instar had no significant effect on mean feeding rate of predator larvae (F=2.17, df= 3, 36, P= 0.110). However, there was significant difference (F= 100.02, df=3,36, p=0.000), (F=16.94, df=3,32, p=0.000), (F=32.70, df=3,37, p=0.000) in feeding rate of 1st instar larvae of *E. balteatus* on different prey densities of 2nd, 3rd and 4th instar nymphs, respectively. The highest and lowest mean number of prey were at densities of 60 and 100 of 2nd instar nymphs, at 60 and 80 of 3rd nymphal instars, and at 60 and 80 of 4th instar nymphs.

Fig. 1. Effect of different densities of *Aphis gossypii* on feeding rate of 1st
larval instar *Episyrphus balteatus* (Mean± SE).
Same alphabets are showing non-significant differences for each nymphal
instar by Tukey post-hoc test (*P*< 0.05).

The feeding rate of 2nd larval instar of *E. balteatus* on different prey densities is given

in Table 1. It was also found that there was no significant variation in feeding rate on

different prey densities of 1st nymphal instars (F= 1.03; df=2, 24; *P*= 0.375). However,

there was significant differences (F= 14.45, df=2, 23, *P*= 0.000), (F= 34.52, df=2, 24,

P= 0.000) in feeding rate of 2nd larval instar of *E. balteatus* on different prey densities of 3rd

and 4th nymphal instars, respectively.

Results suggest that 3rd instar larvae of *E. balteatus* were unable to complete their

larval development when fed on different densities of 1st and 2nd instar nymphs. In fact, 3rd

instar larvae of *E. balteatus* failed to complete their development at density of 40 and 60

of third nymphal instar.

Table 1. Effect of different densities of *Aphis gossypii* on feeding rate of 2nd larval instar
Episyrphus balteatus (Mean± SE).

	No. of nymphal instars fed upon			
Prey density	**1st**	**2nd**	**3rd**	**4th**
40	-	-	-	136.20± 25.50[b] (77.00-225.00)
60	216.00± 22.50[a] * (141.00-275)	334.50± 53.90[a] (95.00-456.00)	276.40± 46.00[a] (177.00-441.00)	259.30± 43.50[a] (108.00-40.9.00)
80	174.60± 16.59[a] (112.00-252.00)	182.80± 9.32[b] (135.00-223.00)	52.90± 14.80[b] (10.00-150.00)	50.90± 10.10[b] (16.00-100.00)
100	171.80± 22.20[a] (60.00-336.00)	112.50± 23.30[b] (18.00-226.00)	36.90± 8.59[b] (10.00-80.00)	58.30± 15.50[b] (14.00-170.00)

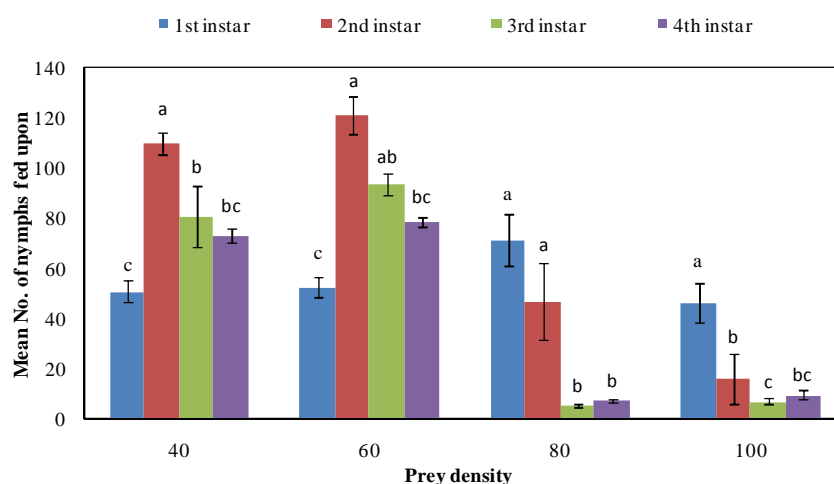Same alphabets in columns are showing non-significance by Tukey post-hoc test (*p* < 0.05).
 * Ranges of prey nymphs fed upon.

There was also no significant difference in larval feeding rate on different prey densities of 3rd instar nymphs (T-test, df= 17, t = -0.32; P = 0.752), as the feeding rate on 3rd instar nymphs at density of 80 and 100 were 310.40± 17.00; and 301.60 ± 21.50, respectively. However, there was significant difference (F= 4.19, df= 2.22., p= 0.030) in feeding rate of 3rd instar larvae of *E. balteatus* on different prey densities of 4th nymphal instars. The feeding rate of 3rd instar larvae on 4th nymphal instars were (359.70± 12.5 at density of 60 (ranged 337.00 - 380.00) and 252.50± 11.30 at density of 80 (ranged 218.00-342.00), respectively.

Effect of prey nymphal instars on feeding rate of *E. balteatus*

There was significant difference between feeding rate of 1st larvae of the predator on 1st and 2nd nymphal instars at densities of 40 and 60 (F= 16.93, df=3,31, P= 0.000) and (F=74.59, df=3,32, P=0.000), respectively.

The highest and lowest mean number of prey consumed were 110.00± 4.16, 50.80± 4.36, 121.30± 7.53 and 52.80± 4.45 at densities of 40 and 60 of 2nd and 1st instar nymphs, respectively (Fig. 2). There were significant differences between feeding rate on 2nd and 4th nymphal instars at prey density of 80 (F= 31.65, df= 3, 39, P= 0.000). Second larval instars fed on the highest and lowest mean number of 1st and 3rd instar nymphs at density of 100 (Table 2). There was significant difference (df= 15, t= -2.83, P= 0.013) in feeding rate of 3rd instar larvae of *E. balteatus* when fed on 80 prey densities of 3rd and 4th nymphal instars and mean number of feeding rate was 310± 17.00 and 252.50± 11.30, respectively. The highest and lowest feeding rate was shown by 3rd larval instar of *E. balteatus* (467.2± 67.2) and (260± 24.60) at density of 100 of 2nd and 4th instar nymphs.

Fig. 2. Effect of different nymphal instars of *Aphis gossypii* on feeding rate of 1st larval instar *Episyrphus balteatus* (Mean± SE).
Same alphabets are showing non-significant differences for each prey density by Tukey post-hoc test (P< 0.05).

Table 2. Effect of different nymphal instars of *Aphis gossypii* on feeding rate of 2nd larval instar *Episyrphus balteatus* (Mean± SE).

Nymphal instars of prey	Prey density			
	40	60	80	100
1st	-	216.00 ± 22.5^{a} *(141.00-257.00)	174.60 ± 16.50^{a} (112.00-260.00)	171.80 ± 22.2^{a} (60.00-336.00)
2nd	-	334.50 ± 53.90^{a} (95.00-456.00)	182.80 ± 9.32^{a} (135.00-223.00)	112.50 ± 23.3^{ab} (18.00-226.00)
3rd	-	276.40 ± 46.00^{a} (177.00-441.00)	52.90 ± 14.80^{b} (10.00-150.00)	36.90 ± 8.59^{c} (10.00-80.00)
4th	-	259.30 ± 43.50^{a} (108.00-409.00)	50.90 ± 10.10^{b} (16.00-100.00)	58.30 ± 15.50^{bc} (14.00-170.00)

Same alphabets in columns are showing non-significance in column by Tukey post-hoc test ($p< 0.05$).
* Ranges of prey nymphs fed upon.

Effect of different prey nymphal densities on predator's developmental time

First instar nymphs of prey significantly influenced the developmental times of 1st and 2nd larval instars of *E. balteatus* (F= 18.58, df= 3, 24, *p*= 0.000) and (F= 93.05, df=2, 18, *p*=0.000), respectively. Longer and shorter developmental times was recorded for 1st larval instars 4.143± 0.261 and 1.500± 0.289 days at prey densities of 40 and 100, and for 2nd larval instar (10.25± 0.629) and (2.00± 0.548) days at prey densities of 60 and 100, respectively. First and 2nd larval instars developmental times were significantly affected by 2nd instar nymphs of prey (F= 18.44, df=3, 17, *P*=0.000) and (F= 48.14, df=2,16, *P*=0.000), respectively. Longer and shorter developmental times for 1st instar larvae were 3.500± 0.289 and 1.500± 0.289 days at prey density of 40, 80 and 100, respectively. Longer and shorter 2nd larval development times were 8.333± 0.760 and 2.00± 0.316 days at densities of 60 and 100 of 2nd instar nymphs, respectively.

There was significant differences among 1st, 2nd and 3rd predator's larval developmental times when feeding on 4th nymphal instars (F= 5.30, df=3, 25,*P*=0.007), (F= 10.07, df=3, 25, *P*=0.000) and (F= 30.74, df=2, 18, *P*=0.000), respectively (Table 3).

There were significant differences between 2nd instar larval developmental time on 3rd nymphal instars at prey densities of 60, 80 and 100 (F= 28. 95, df= 2, 16, *p*= 0.000). Mean developmental time was 3.50± 0.224 days at density of 60 and 1.50± 0.289 days at densities of 80 and 100 preys. Only 3rd instar larvae successfully completed their development by feeding on density of 80 and 100 of 3rd nymphal instars (Tables 4 and 5).

Influence of different nymphal instars on developmental time of predator

Results showed that only 1st instar larvae completed their developmental time at density of 40 of different nymphal instars while only a few number of larvae were able to

complete 2nd larval instar on 4th instar nymphs in 3.80.± 0.663 days. There were significant differences among developmental times of 1st larval instars on 1st and 3rd nymphal instars at density of 40 (F= 6.63, df=3, 29, *p*= 0.002). The longest and shortest developmental times of *E. balteatus* larvae were 4.143± 0.261 and 2.600± 0.306 days on 1st and 3rd instar nymphs at density of 40 preys, respectively. There was also significant difference among

developmental times of 1st and 2nd larval instars feeding on different nymphal instars of prey at density of 60 (F= 8, df=3, 30, *p*=0.001) and (F= 7.22, df=2, 17, *p*= 0.006), respectively. The longest and shortest larval developmental times at density of 60 were 3.875± 0.39 and 2.22± 0.278 days on 1st and 3rd instar nymphs of 1st larval instars (8.33± 0.76) and (4.60± 0.510) days on 2nd and 3rd nymphal instars of 2nd larval instars, respectively. Only a few number of larvae were able to complete 3rd larval instar when fed on 4th instar nymphs at 8.33± 0.333 days.

Table 3. Effect of different densities of Aphis gossypii 4th nymphal instar on developmental times of Episyrphus balteatus larvae (Mean± SE).

Prey density	Larval instars		
	1st	2nd	3rd
40	2.667± 0.236ᵃ (2.00-4.00)	3.80± 0.663ᵃᵇ (2.00-6.00)	-
60	2.250± 0.250ᵃᵇ (1.00-3.00)	5.143± 0.800ᵃ (3.00-8.00)	8.33± 0.333ᵃ (8.00-9.00)
80	1.50± 0.224ᵇ (1.00-2.00)	1.75± 0.36ᵇ (1.00-4.00)	3.857± 0.261ᵇ (3.00-5.00)
100	1.33± 0.333ᵇ (1.00-2.00)	1.50± 0.224ᵇ (1.00-2.00)	4.11± 0.324ᵇ (3.00-6.00)

Same alphabets in columns are showing non-significant differences by Tukey post-hoc test (*p*< 0.05).
* Ranges of prey nymphs fed upon

There was no significant difference between 1st and 3rd larval development times at density of 80 (df= 11, t= -0.67, *P*= 0.519), but it was significant for 2nd larval instar developmental time (F= 4.68, df=3, 29, *P*= 0.010) (Table 4). There was a non-significant difference between 1st, 2rd and 3rd larval instars feeding on different nymphal instars at density of 100 prey. Third larval instar failed to complete its development when fed on 1st nymphal instars. Although there was no significant difference in 2nd larval instar developmental time on different nymphal instars (Table 5), the longest and shortest development times of 2nd instar larvae were obtained on 2nd, 3rd and 4th instar nymphs of prey, respectively.

Discussion

Biological control was basically defined as the action of parasitoids, predators or pathogens in maintaining another organism's population at a lower average than would occur in their absence (Coll & Ridgeway, 1995). It has been shown that members of the subfamily Syrphinae are specialized predators and found to be voracious feeders on aphids (Alhmedi *et al*., 2008; Mushtaq *et al*., 2014;). Among them, *E. balteatus* is an economically important syrphid consuming a broad range of aphid species in the fields and greenhouses (e.g., Völkl *et al*., 2007; van Lenteren, 2012).

Table 4. Effect of *Aphis gossypii* nymphal instars at density of 80 on developmental times (days) of *Episyrphus balteatus* larvae (Mean± SE).

Nymphal instars	Larval instars		
	1st	2nd	3rd
1st	1.500± 0.224[a] (1.00-2.00)	2.800± 0.249[a] (2.00-4.00)	-
2nd	1.500± 0.289[a] (1.00-2.00)	2.500± 0.224[ab] (2.00-3.00)	-
3rd	1.500± 0.289[a] (1.00-2.00)	1.500± 0.224[b] (1.00-2.00)	4.143± 0.340[a] (3.00-5.00)
4th	1.500± 0.224[a] (1.00-2.00)	1.750± 0.366[b] (1.00-4.00)	3.875± 0.340[a] (3.00-5.00)

Same alphabets in columns are showing non-significant differences by Tukey test ($p < 0.05$) and in 4[th]column paired *t*-test ($p < 0.05$).
* Ranges of prey nymphs fed upon.

We found that the densities of *A. gossypii* significantly affected developmental times of different larval instars of *E. balteatus.* At low prey density (40 prey/day), none of the larvae developed into adults and only 1st larval instar completed its development. Similar results were found on different prey densities. At low prey density (5 prey/day) only 30% of larvae pupated, of which less than 35% emerged as adults. The larval and pupal mortality decreased with increasing number of prey eaten. For example, larvae of *Ischiodon scutellaris* (Fabricus) consumed at least 25 or 30 aphids/day to properly complete their development. A newly hatched larva destroyed only a few aphids and as the development progressed the number of consumed prey increased. Threshold number of prey needed to complete development (50% success) appears to be 20 aphids/day. Even at the lowest density (5 aphid/day), about 10% of larvae went through adult stage (Singh & Mishra, 1988). However, Agarwala and Sara (1986) observed that the larvae of *E. balteatus* fed up to 618 cotton aphids on cotton.

The larvae of *E. balteatus* fed on different nymphal instars showed significantly different feeding rates. The feeding rate of *E. balteatus* larvae on prey nymphs increased until 1-2 days before pupation. First and 2nd nymphal intars at different densities were not

suitable for larval development of the predator. Only 3rd and 4th instar nymphs at densities of 80 and 100 provided essential nutrients to complete different larval developmental times. It was shown that at density of 40, only 1st larval instar completed its developmental time by feeding on 1st to 4th nymphal instars of *A. gossypii*. This suggests that density of 40 is not suitable for rearing *E. balteatus*. The mean number of different nymphal instars of *M. persicae* eaten by $1^{st,}$ 2nd and 3rd instar larvae of *E. balteatus* were 25.20 ± 2.23, 125.20 ± 7.04 and 315.20 ± 16.27 preys, respectively (Fathipour *et al.*, 2005). The highest rate of 2nd and 3rd larval feeding of *E. corollae*, on 1st and 2nd instar nymphs of *B. brassicae* were 25.00±2.61 and 38.2± 0.58, respectively (Arabiyan, 2010). Accordingly, Jalilian *et al.*

(2011) found that 1st instar larvae of *E. balteatus* preferred younger nymphs of *M. persicae*, and for the 2nd instar larvae, the most preferred nymphs were younger ones. In the case of 3rd instar larvae, no significant differences were found in nymphal instars. A single maggot of *I. scutellaris* could consume about 260 aphids (3rd instar nymphs of *Rhopalosiphum maidis* (Fitch)) till pupation (Singh & Mishra,1988). Developmental time of *E. balteatus* larvae fed on different densities of prey was longer as the density of nymphal instars decreased. This study showed that larval instars fed on densities of 40 and 60 of nymphal instars (especially 1st and 2nd) had a significantly longer developmental time than those fed on densities of 80 and 100 of 3rd and 4th nymphal instars. Similar results were found on *E. balteatus* and *Eupeodes corolla* Fabricius that the total developmental time of both species tended to be shorter when fed on higher prey densities (Putra & Yasuda, 2006). The larval

developmental times of *E. corolla* decreased with progressing nymphal instars of prey. The longest developmental times of 2nd and 3rd larval instars of *E. corolla* feeding on 1st and 2nd nymphal instars of *Brevicoryne brassicae* L., were 25.00 ±2.61 and 38.20± 0.58days, respectively (Arabian, 2010). The longer developmental time of *E. balteatus* on *A. gossypii* is likely due to the smaller size and less energy of younger nymphs of aphids. Size and the amount of energy reserves of nymphs can influence the length of life. In some studies, it has been shown that the rate of collision with prey can be a function of the size of prey. Therefore, larger prey is favorable for providing higher nutritional value for predator (Charnov, 1976).

The short life time and high feeding rate of syrphid larvae are useful feature that improves their efficiency in biological control programs (Hart *et al.*, 1997). It can be concluded that higher densities (80 and 100) of prey/day and 3rd and 4th nymphal instars of prey are highly favorable to rearing *E. balteatus*. It is believed that *E. balteatus* serves as an effective natural enemy against aphid species including *A. gossypii*.

Table 5. Effect of *Aphis gossypii* nymphal instars at density of 100 on development times of *Episyrphus balteatus* larvae (Mean± SE).

Nymphal instars	Larval instars		
	1st	2nd	3rd
1st	1.500± 0.289[a] *(1.00-4.00)	2.00± 0.548[a] (1.00-4.00)	-
2nd	1.500± 0.289[a] (1.00-3.00)	2.00± 0.316[a] (1.00-3.00)	5.00± 0.837[a] (3.00-7.00)
3rd	1.333± 0.333[a] (1.00-2.00)	1.500± 0.224[a] (1.00-2.00)	4.50± 0.423[a] (3.00-7.00)
4th	1.333± 0.333[a] (1.00-2.00)	1.500± 0.224[a] (1.00-2.00)	4.11± 0.351[a] (3.00-6.00)

Same alphabets in columns are showing non-significant differences by Tukey post-hoc test ($p< 0.05$).
* Ranges of prey nymphs fed upon

Acknowledgments

References

Agarwala, B. K. & Sara, J. L. (1986) Larval voracity, development and relative abundance of predators of *Aphis gossypii* on cotton in India. In: *Ecology of Aphidophaga* (ed. Hodek, I.), 339–344.

Alhmedi, A., Haubruge, E. & Francis, F. (2008) Role of prey-host plant associations on *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) and *Episyrphus balteatus* DeGeer (Diptera: Syrphidae) reproduction and predatory efficiency. *Entomologia Experimentalis et Applicata* 128, 49–56.

Arabiyan, M. (2010) Biological characteristics and thermal requirements of *Eupeodes corolla* F. on cabbage aphid, *Brevicoryne brassicae* L. Msc. thesis, Faculty of Agriculture, Razi University. pp85.

Atlihan, R. & Kaydan, B. (2010) Functional response of the coccinellid predator, *Adalia fasciato-punctata revelierei* to walnut aphid (*Callaphis juglandis*). *Phytoparasitica* 38, 23–29.

Bazzocchi, G. G. & Burgio, G. (2001) Functional response of *Lysiphlebus testaceipes* (Cresson) (Hymenoptera: Braconidae) against *Aphis gossypii* Glover (Homoptera: Aphididae) at two constant temperatures. *Bulletin of Insectology* 54, 13–21.

Blackman, R. L., & Eastop, V. F. (2000) Aphids on the worlds crops: an identification and information guide, 2 ed. Wiley, London, United Kingdom.

Charnov, E. L. (1976) Optimal foraging, the marginal value theorem. *Theoretical Population Biology* 9, 129–136.

Coll, M., & Ridgeway, R. L. (1995) Functional and numerical responses of *Orius insidious* (Heteroptera: Anthocoridae) to its prey in different vegetable crops. *Annals of the Entomological Society of America* 88, 732–738.

Fathipour, Y. Jalilian, F., Talebi, A. A. & Moharramipour, S. (2005) Voracity of larvae of three hoverfly species (Dip.: Syrphidae) as potential biocontrol agents of *Myzus persicae* (Hom., Aphididae) on greenhouse crops. *Integrated Control in Protected Crops, Temperate Climate IOBC/wprs Bulletin* 28(1), 91–94.

Frazer, B., D. (1972) A simple and efficient method of rearing aphidophagous hoverflies (Diptera: Syrphidae). *Journal of Entomological Society of British Columbia* 69, 23–24.

Gilbert, F. (1986) Hoverflies. Naturalists' Handbook 5.76 pp. Cambridge University Press, Cambridge, UK.

Hart, A. J., Bale, J. S. & Fenlon, J. S. (1997) Developmental threshold, day- degree requirements and voltinism of the aphid predator *Episyrphus balteatus* (Diptera: Syrphidae). *Annals of Applied Biology* 130, 427–437.

Hindayana, D. (2001) Resource exploitation by *Episyrphus balteatus* DeGeer (Diptera: Syrphidae) and intraguild predation. Ph.D. Dissertation, University of Hannover, Hannover. 95 pp.

Hodek, I. & Honek, A. (1996) *Ecology of Coccinellidae*. 464 pp. Dordrecht Boston London: Kluwer Academic.

Huffaker, C. B., Messenger, P. S. & DeBach, P. (1971) The natural enemy component is natural control and the theory of biological control. pp. 16–27. In: *Biological Control* (ed. Huffaker, C. B.). Plenum Press, New York.

Jalilian, F., Fathipour, Y., Talebi, A. A. & Sedaratian, A. (2011) Functional response and mutual interference of *Episyrphus balteatus* and *Scaeva albomaculata* (Diptera: Syrphidae) fed on *Myzus persicae* (Homoptera: Aphididae). *Applied Entomology and Phytopathology* 78(2), 257–273.

Madahi, Kh., Sahragard, A. & Hosseini, R. (2013) Influence of *Aphis gossypii* Glover (Hemiptera: Aphididae) density on life table parameters of *Aphidoletes aphidimyza* Rondani (Diptera: Cecidomyiidae) under laboratory conditions. *Journal of Crop Protection* 2(3), 355–368.

Minitab lnc. (1994) Philadelphia, PA, USA.

Mushtaq, S., Rana, S. A., Khan, H. A. & Ashfaq, M. (2013) Diversity and abundance of family Aphididae from selected crops of Faisalabad, Pakistan. *Pakistan Journal of Agricultural Sciences* 50, 103–109.

Mushtaq, S., Rana S. A., Rana, N., Maalik, S. & Ehsan, N. (2014) Developmental duration and predatory efficiency of *Episyrphus balteatus* on four aphid species in

Pakistan. *International Journal of Agricultural and Biological Engineering* 16, 614-618.

Poligui, R.N., Haubruge, E., & Francis, F. (2011) Predominant effects of host plant species on *Aphis gossypii* aphid and *Episyrphus balteatus* hoverfly in a tritrophic approach. *Communications in Agricultural and Applied Biological Sciences* 76, 445–456.

Principi, M. M & Canard, M. (1984) Feeding habits. Biology of Chrysopidae (ed. by Canard, M., Semeria, Y & New, T. R.), pp. 76–92. Junk Publishers, The Hague, The Netherlands.

Putra, N. S. and Yasuda, H. (2006) Effects of prey species and its density on larval performance of two species of hoverfly larvae, *Episyrphus balteatus* De Geer and *Eupeodes corolla* Fabricius (Diptera: Syrphidae). *Applied Entomology and Zoology* 41, 389–397.

Rabb, R. L. (1974) Naturally occurring biological control in the eastern United States, with particular reference to tobacco insects. pp. 294-309 in Huffaker, C. B. (Ed.) *Biological control*. 511 pp. Plenum Press, New York.

Sadeghi, H. & Gilbert, F. (2000) Aphid suitability and its relationship to oviposition preference in predatory hoverfly. *Journal of Animal Ecology* 69, 771–784.

Schneider, F. (1969) Bionomics and physiology of aphidophagous Syrphidae. *Annual Review of Entomol*ogy 14, 103–124.

Singh, P. & Mishra, S. (1988) Development of a syrphid fly, *Ischiodon scutellaris* (Fabricus) on *Rhopalosiphum maidis* (Fitch). *Journal of Aphidology* 2, 28–34.

Stáry, P. (1970) Biology of Aphid Parasites, with Respect to Integrated Control, Series Entomologica 6. Junk Publishers, The Hague, The Netherlands.

Sugiura, K. & Takada, H. (1998) Suitability of seven aphid species as prey of *Cheilomenes sexmaculata* (Fabricius) (Coleoptera: Coccinellidae). *Japanese Journal of Applied Entomology and Zoology* 42, 7–14 (in Japanese with English summary).

Sunderland, K. D, Fraser, A. M. & Dixon, A. F. G. (1986) Field and laboratory studies on money spiders (Linyphiidae) as predators of cereal aphids. *Journal of Applied Ecology* 23, 433–447.

van Lenteren, J. C. (2012) The state of commercial augmentative biological control: plenty of natural enemies, but a frustrating lack of uptake. *Biocontrol* 57, 1–20.

Verheggen, F. J., Haubruge, E., De Moraes, C. M., & Mescher, M. C. (2009) Social environment influences aphid production of alarm pheromone. *Behavioural Ecololgy* 20, 283–288.

Völkl, W., Mackauer, M., Pell, J. K, & Brodeur, J. (2007) Predators, parasitoids and pathogens. In: van Emden, H., Harrington, R. (eds) Aphids as crop pests. CAB International, Wallingford, pp 187–234.