Food consumption and immature growth of *Adalia bipunctata* (Coleoptera: Coccinellidae) on a natural prey and a factitious food

MOHAMMAD A. JALALI, LUC TIRRY and PATRICK DE CLERCQ*

¹Department of Crop Protection, Ghent University, Coupure Links 653, B-9000 Ghent, Belgium; e-mail: patrick.declercq@ugent.be

Key words. Adalia bipunctata, Myzus persicae, food consumption, food conversion, factitious food, biological control

Abstract. One factor limiting the adoption of aphidophagous coccinellids in augmentative biological control is cost-effective mass production. The use of factitious foods may lower production costs by reducing space and manpower requirements for mass rearing of the predator and its prey and by enhancing mechanization of rearing procedures. The objective of this study was to compare the efficiency of food conversion, consumption indices and growth rates of first to fourth instars of *Adalia bipunctata* when fed either a mixture of *Ephestia kuehniella* eggs and fresh bee pollen, or on the natural prey *Myzus persicae*. Larval survival did not differ among treatments, averaging 80 and 90% on the respective diets. Mean dry body weights of the coccinellid were significantly lower on *M. persicae* than on the factitious food only for second instars and the pre-pupal stage. Dry food consumption by the predator throughout larval development averaged 20.61 mg on factitious food and 14.82 mg (corresponding to an average of 284 third and fourth instars) on *M. persicae*. The efficiency of conversion of ingested food for total larval development averaged 25 and 30% on factitious food and aphids, respectively. The relative growth rate of the larval instars was higher on aphids, whereas the consumption index was higher on the mixture of *E. kuehniella* eggs and pollen. An additional experiment showed that rearing the previous instars on the factitious food had no substantial effect on aphid consumption in the fourth instar. The study suggests that this factitious diet may be an appropriate food source for mass rearing *A. bipunctata*.

INTRODUCTION

The green peach aphid *Myzus persicae* (Sulzer) is one of the most serious crop pests worldwide (Radcliffe, 1982). It has a very broad host range (more than 40 plant families) and transmits over 100 plant viruses (Blackman & Eastop, 1984). Due to an increasing number of difficulties and failures in the chemical control of the pest related in part to the development of insecticide resistance, research on its natural enemies and their field application is receiving growing attention.

The two-spot ladybird beetle *Adalia bipunctata* (L.) is a potential natural enemy of *M. persicae* and other aphids and has been used in augmentative biological control in Europe (Wyss et al., 1999; De Clercq et al., 2005). It is a polyphagous predator (Hodek, 1973), occurring in Europe, Central Asia and North America (Majerus, 1994). At present, it has a 6% share of the total world market for aphidophagous agents. In Western Europe, about 75% of the total commercial production of this predator is reserved for aphid control in avenue trees (R. Timmer, Koppert BV, the Netherlands, pers. commun.). However, some studies have indicated its potential for use against *M. persicae* in different greenhouse crops (Hämäläinen, 1977, 1980).

One of the factors limiting the augmentation of aphidophagous coccinellids (including *A. bipunctata*) is costeffective mass production. The use of factitious foods may lower production costs by reducing problems associated with space and manpower required for the mass rearing of live prey and by enhancing the mechanization

of the rearing procedures (Waage et al., 1985; De Clercq, 2002; De Clercq et al., 2005). One challenge in developing a factitious (or unnatural) food for a predator is to find a diet which can fully support the growth and reproduction of the natural enemy without deleterious effects on its fitness as a biological control agent. Predation rates of A. bipunctata have been determined on different aphid species (e.g. Ellingssen, 1969; Hämäläinen et al., 1975; Mills, 1981), but no studies have examined whether the rearing of the predator on an unnatural food affects its subsequent consumption of aphid prey. In biological control programmes against aphids, advanced larval stages of A. bipunctata are usually released (Wyss et al., 1999; J. Vermeulen, BioBest NV, pers. commun.). The diet experienced by early instars of the predator in the rearing environment may thus affect the efficiency of the later instars to suppress populations of target aphids in subsequent augmentative releases.

The objective of this study was to compare the efficiency of food conversion, consumption indices and growth rates of all instars of *A. bipunctata* when fed either on a factitious food, consisting of a mixture of *Ephestia kuehniella* Zeller eggs and fresh bee pollen, or on the natural prey *M. persicae*. In order to assess the effect of previous diet on aphid consumption, predation rates of fourth instars of the predator on *M. persicae* were compared after having been reared on either food source from the first to the third instar. The findings may improve our understanding of the nutritional ecology of *A. bipunctata* and contribute to the optimisation of the

^{*} Corresponding author.

mass rearing of this predator for augmentative biological control purposes.

MATERIAL AND METHODS

Predator culture

Adalia bipunctata were originally purchased from Biobest NV (Westerlo, Belgium) as larvae. Prior to the experiments, the insect was reared for five generations at Ghent University on an ad libitum supply of a 50-50 mixture of frozen E. kuehniella eggs and bee pollen as a factitious food (De Clercq et al., 2005) at 23 \pm 1°C, 65 \pm 5% RH and a 16L : 8D photoperiod. Frozen eggs of E. kuehniella and frozen moist pollen, consisting of pollen pellets collected by honeybees, used in our study were supplied by Koppert BV (Berkel en Rodenrijs, The Netherlands) and stored for no longer than one month at -18°C. Myzus persicae used in the experiments were taken from a laboratory colony at Ghent University that was established using insects kindly provided by J.-M. Rabasse (INRA, Antibes, France). At our facilities, the aphid colony was maintained in a growth chamber on broad bean plants, Vicia faba L. var. major, at 26 ± 2° C, $60 \pm 20\%$ RH and a 16L : 8D photoperiod.

Experiments

All experiments were performed under constant conditions of $23 \pm 1^{\circ}\text{C}$, $65 \pm 5\%$ RH and a 16L: 8D photoperiod.

In a first experiment, food consumption and growth of the first to fourth instars of A. bipunctata were investigated on the factitious food (a 50-50 mixture of frozen E. kuehniella eggs and bee pollen) and on M. persicae. For the treatment using the factitious food, 20 newly hatched first instars (<12 h old) of A. bipunctata were randomly selected from different egg batches of uniform age harvested from the stock colony. These larvae thus originated from parents which had previously been reared on factitious food for five generations. Each larva was weighed on a Sartorius Genius ME215P balance (Sartorius, Göttingen, Germany), accurate to 0.001 mg, before being transferred into an individual 9-cm Petri dish lined with filter paper and containing a plastic cup (3.0 × 0.5 cm) as a food container. An amount of 20, 50, 50, and 80 mg of the factitious food was provided daily to individual first, second, third and fourth instars, respectively. The daily food consumption in each instar was determined individually for each larva from the fresh weight of food supplied and the dry weight of food remains (after 24 h drying at 60°C), based on a pre-determined relationship between dry and fresh weight (in mg) of the factitious food: dry wt = 0.4565 fresh wt + 0.0392 (n = 30, R^2 = 0.998, P < 0.001) (Mills, 1981; Isikber & Copland, 2001). The fresh weight of the larvae was recorded after each moult. To determine the dry weight of the different instars of A. bipunctata, 15 individuals of each stadium (<12h old) and pre-pupae were taken from the stock colony and their fresh weights were recorded. Subsequently, their dry weights were determined by placing them in an oven at 60°C for 24 h. The following relationship was observed between dry and fresh body weight (in mg) of the larvae: dry wt = 0.3217 fresh wt + 0.0122 (R² = 0.997, P < 0.001).

In a second treatment, food consumption and growth of A. bipunctata larvae was determined on M. persicae. Before starting the experiment, a group of about 150 newly hatched first instars of A. bipunctata originating from the stock colony were reared to adulthood on M. persicae. The resulting adults were transferred to communal rearing containers (30 \times 17 \times 9 cm), fed M. persicae and allowed to reproduce. Eggs of uniform age were harvested from the adult containers and upon hatching, 20 first instars (<12h old) were randomly collected from different egg batches. The larvae were weighed and transferred

into individual 9-cm Petri dishes. Each Petri dish was lined with filter paper and contained a 2-leaf seedling of broad bean, the stalks of which were inserted in an Eppendorf tube containing water. The broad bean seedling was infested with a predetermined number of third and fourth instar M. persicae (50, 70, 100 and 150 for first, second, third and fourth instars of the predator, respectively). Aphids were replenished after each daily observation. To determine the average fresh and dry weight of the prey offered, 500 third and fourth instars (in 10 replicates, each consisting of 50 nymphs) of M. persicae were weighed before and after 24 h drying in an oven at 60°C. The mean fresh and dry weight of an individual prey item from a mixture of third and fourth instars of M. persicae was determined to be 0.214 ± 0.017 and 0.052 ± 0.003 mg, respectively (mean \pm SE). Conversion from fresh to dry weight of *M. persicae* nymphs was done based on the relationship (in mg): dry wt = 0.2302 fresh wt + 0.0022 (n = 10, R² = 0.997, P < 0.001). The number of aphids consumed and larval moulting were monitored daily and fresh weight of the larvae was recorded after every moult. Dry weight gained by the different instars of the predator was determined according to the same procedure described in the first treatment. The following relationship between fresh and dry body weight of predator larvae fed on aphid prey was established (in mg): dry wt = 0.3484 fresh wt -0.0128 (R² = 0.993, P < 0.001).

A further experiment was set up to investigate aphid consumption and growth of fourth instars of A. bipunctata when the previous instars (L1–L3) were either reared on a factitious food or on M. persicae. Larvae of the first group were reared in a communal container ($30 \times 17 \times 9$ cm) on the factitious food up to the end of the third instar. From this group, 20 newly moulted fourth instars (<12h old) were then transferred to individual 9-cm Petri dishes. The larvae were supplied daily ad libitum with a mixture of third and fourth instars of M. persicae until they developed to pre-pupae. The aphid consumption and body weight gained by the predator larvae were monitored daily. Prey consumption, food conversion and growth rates of these fourth instars were compared with those of larvae that developed throughout on M. persicae (L1–L4).

The response of the ladybird to the tested foods was quantified using the following indices (Waldbauer, 1964):

The relative growth rate (RGR):

$$RGR = \frac{Dry\ weight\ gained\ (mg)}{Days\ feeding\ period\ \times\ Mean\ dry\ weight\ of\ insect\ during\ feeding\ period\ (mg)}$$

The consumption index (CI): $CI = \frac{F}{TA}$

F = dry weight of food ingested (mg)

T = duration of feeding period (days)

A = mean dry weight of insect during feeding period (mg)

The efficiency of conversion of ingested food to body matter (ECI) or the gross efficiency:

$$ECI = \frac{\text{Dry weight gained (mg)}}{\text{Dry weight of food ingested (mg)}}$$

The *consumption rate* (CR) of *A. bipunctata* when fed *M. persicae* was equally quantified using the following formula (Isikber & Copland, 2001):

$$CR = \frac{\text{The number of aphids eaten throughout the instar}}{\text{Duration of the instar (days)}}$$

Data analysis

Data were analysed by Student's t-test after first checking whether the data were distributed normally by the Kolmogorov-Smirnov test (K-S test). Levene's test was also performed for checking equality of variances (SPSS Inc, 2006). Proportions were normalized and variance equalized by arcsine of the square-root transformation before analysis.

TABLE 1. Initial dry body weight, dry weight of food consumed and dry body weight gained of different larval instars of *A. bipunctata* fed on *M. persicae* and a factitious food.

Developmental stage	n	Diet	Dry body weight at the start of instar (mg) ^a	Dry weight of food consumed during the instar (mg) ^a	Dry body weight gained during the instar (mg) ^a
L1	20 20	Factitious food <i>M. persicae</i>	$\begin{array}{c} 0.045 \pm 0.005a \\ 0.036 \pm 0.002a \end{array}$	$\begin{array}{c} 1.051 \pm 0.086b \\ 0.557 \pm 0.011a \end{array}$	$0.160 \pm 0.014b$ $0.103 \pm 0.007a$
L2	19 19	Factitious food <i>M. persicae</i>	$\begin{array}{c} 0.205 \pm 0.014b \\ 0.139 \pm 0.007a \end{array}$	$\begin{array}{c} 1.639 \pm 0.103b \\ 1.291 \pm 0.021a \end{array}$	$0.351 \pm 0.042a$ $0.353 \pm 0.021a$
L3	18 19	Factitious food <i>M. persicae</i>	$\begin{array}{c} 0.556 \pm 0.042a \\ 0.492 \pm 0.021a \end{array}$	$4.059 \pm 0.463b$ $2.125 \pm 0.179a$	$\begin{array}{c} 1.056 \pm 0.154a \\ 0.908 \pm 0.039a \end{array}$
L4	16 18	Factitious food <i>M. persicae</i>	$\begin{array}{c} 1.612 {\pm}~0.039a \\ 1.401 \pm 0.054a \end{array}$	$13.893 \pm 0.370b$ $10.754 \pm 0.350a$	$3.580 \pm 0.250a$ $3.110 \pm 0.132a$
Pre-pupa	16 18	Factitious food <i>M. persicae</i>	$\begin{array}{c} 5.192 \pm 0.250b \\ 4.511 \pm 0.132a \end{array}$	_ _	_ _

^aMeans (\pm SE) within the same instar and column followed by the same letter are statistically equivalent (Student's t-test, $\alpha = 0.05$); n = number of individuals surviving (initial number of insects was 20).

RESULTS

Food consumption, food conversion and growth on aphids and factitious food

Larval survival did not differ among treatments, averaging 80 and 90% on the factitious food and M. persicae, respectively. Overall, the mean dry body weights of different larval instars of A. bipunctata were somewhat lower on M. persicae than on the factitious food (Table 1). These differences were significant for second instar (t = -7.97, df = 36, P < 0.001) and for the pre-pupal stage (t=-4.18, df=32, P=0.016). The effect of food on the dry weight gained during the different instars (L1–L4) of A. bipunctata was only significant for the first instar (t= -7.27, df = 38, P = 0.001). The proportion of contribution of a larval instar in the total dry body weight gain continuously increased from the first to the last instar on both tested diets. For instance, about 2.30 and 3.11% of total dry body weight gain occurred during the first instar on M. persicae and factitious food, respectively, whereas this was 69.50 and 69.55% for the fourth instar.

Dry food consumption by A. bipunctata on either diet increased with progressive larval development (Table 1). On both foods, 65–70% of total consumption occurred in the fourth instar. The total dry food consumption of A. bipunctata throughout larval development (L1–L4) was 14.82 ± 0.25 mg and 20.61 ± 0.37 mg on M. persicae and factitious food, respectively. Dry weight of ingested food was significantly higher for the factitious food in all larval instars (t-tests, P < 0.05). The consumption rate (i.e. number of M. persicae consumed per day) by A. bipunctata increased exponentially from the first to the fourth instar. Whereas first instars of the predator killed about four third and fourth instars of M. persicae per day, this number reached about 67 in the final instar (Table 2).

The successive larval instars contributed 3.8, 8.7, 14.7 and 72.8% of total number of aphids consumed, respectively.

The efficiency of conversion of ingested food (ECI) increased as the predator developed from the first to the third instar on both diets (Fig. 1a). However, it decreased (on *M. persicae*) or remained constant (on factitious food) in the fourth instar. For the second and third instar, and also for the total larval period, ECI was significantly higher on M. persicae than on factitious food (t = 4.03, df = 36, P = 0.048; t = 8.61, df = 35, P < 0.001; and t = 5.73, df = 32, P = 0.002, respectively). ECI of the different larval instars of A. bipunctata ranged from a minimum of 18.5 ± 1.2 and $15.2 \pm 1.3\%$ to a maximum of 42.7 ± 1.8 and $26.0 \pm 3.8\%$ on *M. persicae* and factitious food, respectively. The consumption index (CI) of A. bipunctata ranged from 1.18 ± 0.04 to 2.52 ± 0.04 on M. persicae and from 1.37 ± 0.04 to 3.08 ± 0.25 on factitious food. On both diets, the CI showed a decreasing trend with successive larval instars (Fig. 1b). The CI was significantly higher on the factitious food than on M. persicae for the first and fourth instar and for the total larval period (t = -19.63, df = 38, P = 0.007; t = -6.54, df = 32, P = 0.005; and t = -6.11, df = 32, P = 0.009, respectively). The mean relative growth rate (RGR) of A. bipunctata ranged from a minimum of 0.35 ± 0.02 and 0.34 ± 0.01 to a maximum of 0.48 ± 0.06 and 0.69 ± 0.04 on factitious food and M. persicae, respectively (Fig. 1c). The RGR of the second and third instars of A. bipunctata was significantly lower when provided with the factitious food than when given M. persicae (t = 5.98, df = 36, P =0.008; and t = 6.14, df = 35, P = 0.005 for the second and third instar, respectively).

Table 2. Consumption rate and total number of M. persicae consumed by different larval instars and during total larval period of A. bipunctata (means \pm SE).

Developmental stages	L_1	L_2	L_3	L_4	L_1 – L_4
Consumption rate (no. of aphids/day)	4.04 ± 0.08	15.28 ± 0.25	23.81 ± 2.00	67.08 ± 2.18	30.98 ± 0.52
Total no. of aphids consumed	10.67 ± 0.21	24.83 ± 0.40	41.67 ± 3.51	206.83 ± 6.73	284.01 ± 4.81

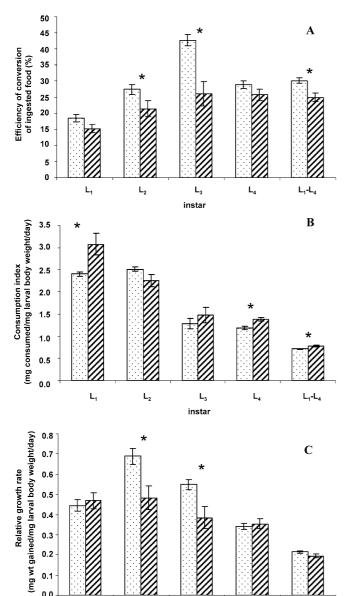


Fig. 1. Efficiency of conversion of ingested food (A), consumption index (B) and relative growth rate (C) during different instars and total larval development of *Adalia bipunctata* when fed on a factitious food (hatched) or *Myzus persicae* (dotted). Within the same instar, bars (\pm SE) with an asterisk indicate a significant difference among means according to Student's t-test ($\alpha=0.05$). Conversion efficiency data (%) were arcsine squareroot transformed before analyses.

L₃

instar

Effect of previous diet on prey consumption by fourth instars

The results from the experiment assessing the effect of the diet experienced by younger instars (L1-L3) of A. bipunctata on aphid prey consumption by the last instar larvae (L4) are presented in Table 3. Larvae that were fed from the first to the third instar on a mixture of E. kuehniella eggs and pollen and offered aphids from the fourth instar on were heavier at the end of fourth instar than those reared throughout on aphids (t = -6.39, df = 38, P < 0.001), but dry body weight gain was similar for both groups. Also, differences between RGR and CI of these two groups were significant with higher values for larvae of the former group (t = 4.12, df = 38, P = 0.004; and t =6.33, df = 38, P = 0.011 for RGR and CI, respectively). However, aphid consumption rates, food conversion indices and developmental times did not differ among treatments.

DISCUSSION

Overall, larvae of A. bipunctata had similar survival and body weights when fed on M. persicae or on a factitious food consisting of E. kuehniella eggs mixed with moist bee pollen, indicating the equivalence of these foods in terms of nutritional content for larval development. In an earlier study, De Clercq et al. (2005) investigated the value of the same factitious food to support development and reproduction of A. bipunctata as compared with pea aphids, Acyrthosiphon pisum (Harris). Latter authors showed that larvae of the coccinellid fed *E*. kuehniella eggs and pollen became adults that were heavier and more fecund than those fed pea aphids, whereas egg hatch on the factitious food was similar to that on pea aphids. When comparing the nutritional value of natural and unnatural foods for A. bipunctata, it should be considered that even though many aphids can be categorized as natural prey for the coccinellid, not all aphid species will be equivalent in nutritional terms (Hodek, 1996). The green peach aphid may not be a highly suitable prey for A. bipunctata and the predator's growth and reproductive rates on other aphid species may thus be superior.

Because of the higher water content of M. persicae (75.9%) as compared to the factitious food (54.3%), A. bipunctata larvae ingested a higher fresh weight of aphids (60.9 \pm 2.5 mg) than of the factitious food (45.0 \pm 2.8 mg) during their larval development. Nevertheless, dry food consumption by larval instars on M. persicae was

Table 3. Consumption of *Myzus persicae* by fourth instars of *A. bipunctata* when the previous instars (L1–L3) were reared on a factitious food (group 1) or on *M. persicae* (group 2).

L₁-L₄

Treatment	n of a	nn no. phids sumed	Develop- mental time (days)	Consumption rate (aphids/day)	Final dry weight (mg)	Dry body weight gain during instar (mg)	Conversion efficiency (%)	Consumption index	Relative growth rate
Group 1	20 219.28	\pm 5.60a	$3.36 \pm 0.17a$	$65.31\pm1.79a$	$4.98 \pm 0.13a$	$3.27 \pm 0.13a$	$28.85\pm1.12\;a$	$1.01 \pm 0.28a$	$0.29 \pm 0.01a$
Group 2	18 206.83	\pm 6.73 a	$3.08 \pm 0.33 a$	$67.08\pm2.18a$	$4.51 \pm 0.13b$	$3.11 \pm 0.13a$	$28.92\pm1.23a$	$1.18 \pm 0.38b$	$0.34 \pm 0.01b$

Means (\pm SE) were compared using Student's t-test (α = 0.05). Conversion efficiency data (%) were subjected to arcsine of the square-root transformation before analysis. n = number of replications.

consistently lower than that on the factitious food suggesting a lower nutritional quality of the factitious food and the need to consume larger amounts in order to obtain the necessary nutrients (Cabral et al., 2006). However, this difference may also be related to the fact that ingested *M. persicae* are converted to body mass more efficiently than the factitious food (Fig. 1a).

Blackman (1967) noted a fresh biomass consumption of 69.1 ± 11.2 mg for *A. bipunctata* during larval development on *M. persicae* at 20°C, whereas Ferran et al. (1984) in Hodek (1996) reported a figure of 55 mg. However, comparisons among studies are complicated by variability in experimental conditions as related to dissimilarity in prey species, prey stages, temperature and the host plants on which the aphids were reared; in addition, genetic variability among *A. bipunctata* populations may also account for differences between studies.

Gurney & Hussey (1970) state that it is difficult to obtain reliable figures for the total number of aphids consumed during the larval life of a coccinellid, because there is a difference between the minimum number required for the predator larva to mature and the number consumed from a continuous abundance of food; i.e., the larva may eat more than it needs to mature. These workers noted that the total larval consumption of *A. bipunctata* on *M. persicae* amounted to 206 aphids at 21°C (prey stage not specified). From their data, the consumption rate would be 15.85 aphids per day, which is lower than the figure we obtained at 23°C (284 aphids and 30.98 aphids per day, respectively).

Our results showed that the food conversion efficiency (ECI) increased and consumption index decreased on either diet as larval development of A. bipunctata progressed. This suggests that younger instars compensated for poorer efficiency of conversion by increasing consumption. Furthermore, when fed aphids, the ECI of the ladybird markedly increased as the predator developed from the first to the third instar. This increase may be explained by the fact that smaller sized younger instars spent more time searching, capturing and processing the prey. The energy cost associated with these activities may decrease as the larvae develop through successive instars. As a consequence, a greater proportion of energy intake may contribute to body weight gain, increasing the ECI of older larval instars. On the other hand, the ECI of fourth instars was lower than that of the third instars when fed on M. persicae. Isikber & Copland (2001) observed a similar result on Scymnus levaillanti Mulsant and Cycloneda sanguinea (L.) fed Aphis gossypii Glover. Latter authors assumed that this may be related to the greater metabolic costs of the last instar needed in preparation for the process of pupation. However, on the factitious food, the ECI values of the third and fourth instars of A. bipunctata were similar, suggesting that specific factors related to the type of food ingested may be important. In a study by Mills (1981) the greatest ECI of A. bipunctata larvae preying on A. pisum was observed during the first instar (53.3%). In contrast, we calculated an ECI of 18.5% for the first instar of A. bipunctata when

feeding on M. persicae. This difference may be due in part to an effect of prey body size. Blackman (1967) stated that, for small first instar A. bipunctata, it is very difficult to prey on a relatively large sized A. pisum, so the larvae do not wholly ingest the prey and resort to extra-oral digestion. In the case of the smaller sized green peach aphid, M. persicae, mostly the whole body (including non-digestible cuticle) is ingested. Schüder et al. (2004) noted an ECI of 29.4% for the total larval stage of A. bipunctata on Sitobion avenae (Fabricius) at 25°C, which is similar to our results on M. persicae (30.2%) and factitious food (24.9%). However, their data were based on the fresh weights of the prey and predator. ECI-values observed for our coccinellid predator were much lower than those reported by Zheng et al. (1993) (46-60%) for the total larval stage of Chrysoperla carnea (Stephens) supplied with various densities of E. kuehniella. Cohen (1984) states that predaceous insects with piercingsucking mouthparts enabling pre-oral digestion of food are able to obtain a larger proportion of highly digestible materials from their prey than chewing predaceous insects, and therefore have higher ECI values. However, extra-oral digestion has also been observed in A. bipunctata larvae (Blackman, 1967) and other coccinellids (Hodek, 1996).

The relative growth rate (RGR) of insects tends to decrease as larvae age (Scriber & Slansky, 1981). In the current study, there was a similar overall trend when the predator was provided with *M. persicae*, but the first instars showed a lower RGR than the second and third instars. We speculate that the lower growth rate of the first instars may partially be associated with their prolonged development (in comparison with the following two instars), requiring a larger proportion of ingested food to be metabolized. Alternatively, it may also be the result of their increased searching and handling time as compared with larger instars feeding on the same size aphids in a similar arena.

There were no major effects of previous diet on aphid consumption by fourth instar larvae of *A. bipunctata*. The observations showed only a marginal increase in aphid consumption of 6% when the younger instars were provided with factitious food instead of aphid prey. The lower CI and RGR of larvae fed previously on *E. kuehniella* eggs and pollen may be linked to the greater body weight of these individuals and to a lesser degree to the prolongation of development of the fourth instar.

The findings of this study may have bearing on the practical use of *A. bipunctata* in augmentative biological control of aphid pests. For this purpose, usually advanced larval stages of the coccinellid are released. The predation rates of *A. bipunctata* larvae measured in this and previous studies (Jalali et al., unpubl.) indicate their potential for impact on populations of *M. persicae* under suitable conditions. Our results further indicate that no deleterious effects on the predation capacity of advanced *A. bipunctata* larvae on aphids have to be expected when the previous stages of the predator have been reared on an inanimate factitious food. In conclusion, a comparison of

the feeding behaviour of *A. bipunctata* larvae on one of its natural prey, *M. persicae*, and a factitious food consisting of a mixture of lepidopteran eggs and pollen corroborates the findings of an earlier study (De Clercq et al., 2005) suggesting that the factitious food may be adequate to support the mass rearing of *A. bipunctata*.

REFERENCES

- BLACKMAN R.L. 1967: The effects of different aphid foods on Adalia bipunctata L. and Coccinella 7-punctata L. *Ann. Appl. Biol.* **59**: 207–219.
- BLACKMAN R.L. & EASTOP V.F. 1984: Aphids on the World's Crops: An Identification and Information Guide. John Wiley & Sons, Chichester, 414 pp.
- CABRAL S., SOARES A.O., MOURA R. & GARCIA P. 2006: Suitability of Aphis fabae, Myzus persicae (Homoptera: Aphididae) and Aleyrodes proletella (Homoptera: Aleyrodidae) as prey for Coccinella undecimpunctata (Coleoptera: Coccinellidae). *Biol. Control* 39: 434–440.
- Cohen A.C. 1984: Food consumption, food utilisation, and metabolic rates of Geocoris punctipes (Heteroptera: Lygaeidae) fed Heliothis virescens (Lepidoptera: Noctuidae) eggs. *Entomophaga* **29**: 361–367.
- DE CLERCO P. 2002: Dark clouds and their silver linings: exotic generalist predators in augmentative biological control. *Neotrop. Entomol.* **31**: 169–176.
- DE CLERCQ P., BONTE M., VAN SPEYBROECK K., BOLCKMANS K. & DEFORCE K. 2005: Development and reproduction of Adalia bipunctata (Coleoptera: Coccinellidae) on eggs of Ephestia kuehniella (Lepidoptera: Phycitidae) and pollen. *Pest Manag. Sci.* **61**: 1129–1132.
- ELLINGSEN I.-J. 1969: Fecundity, aphid consumption and survival of the aphid predator Adalia bipunctata L. (Coleoptera: Coccinellidae). *Norsk. Entomol. Tidsskr.* **16**: 91–95.
- Gurney B. & Hussey N.W. 1970: Evaluation of some coccinellid species for the biological control of aphids in protected cropping. *Ann. Appl. Biol.* **65**: 451–458.
- Hämäläinen M. 1977: Control of aphids on sweet peppers, chrysanthemums and roses in small greenhouses using the ladybeetles Coccinella septempunctata and Adalia bipunctata (Coleoptera: Coccinellidae). *Ann. Agric. Fenn.* **16**: 117–131.
- Hamalainen M. 1980: Evaluation of two native coccinellids for aphid control in glasshouses. *IOBC/WPRS Bull.* 3: 59–63.

- Hämäläinen M., Markkula M. & Raij T. 1975: Fecundity and larval voracity of four ladybeetle species (Coleoptera: Coccinellidae). Ann. Entomol. Fenn. 41: 124–127.
- HODEK I. 1973: *Biology of Coccinellidae*. Dr. W. Junk, The Hague, 260 pp.
- HODEK I. 1996: Food relationships. In Hodek I. & Honěk A. (eds): *Ecology of Coccinellidae*. Kluwer, Dordrecht, pp. 143–238.
- ISIKBER A.A. & COPLAND M.J.W. 2001: Food consumption and utilization by larvae of two coccinellid predators, Scymnus levaillanti, and Cycloneda sanguinea, on cotton aphid, Aphis gossypii. *BioControl* **46**: 455–467.
- MAJERUS M.E.N. 1994: *Ladybirds*. Harper Collins, London, 367 pp.
- MILLS N.J. 1981: Some aspects of the rate of increase of a coccinellid. *Ecol. Entomol.* **6**: 293–299.
- RADCLIFFE E.B. 1982: Insect pest of potatoes. *Annu. Rev. Ento-mol.* 27: 173–204.
- SCHÜDER I., HOMMES M. & LARINK O. 2004: The influence of temperature and food supply on the development of Adalia bipunctata (Coleoptera: Coccinellidae). *Eur. J. Entomol.* 101: 379–384.
- Scriber J.M. & Slansky F. 1981: The nutritional ecology of immature insects. *Annu. Rev. Entomol.* **26**: 183–211.
- SPSS Inc. 2006: SPSS Base 15.0 for Windows, User's Guide. SPSS Inc., Chicago, IL.
- WAAGE J.K., CARL K.P., MILLS N.J. & GREATHEAD D.J. 1985: Rearing entomophagous insects. In Singh P. & Moore R.F. (eds): Handbook of Insect Rearing. Vol. 1. Elsevier, New York, pp. 45–66.
- WALDBAUER G.P. 1964: The consumption, digestion and utilization of solanaceous and non-solanaceous plants by larvae of the tobacco hornworm, Protoparce sexta (Johan) (Lepidoptera: Sphingidae). *Entomol. Exp. Appl.* 7: 253–269.
- Wyss E., VILLIGER M., HEMPTINNE J.-L. & MÜLLER-SCHÄRER H. 1999: Effects of augmentative releases of eggs and larvae of the two-spot ladybird beetle, Adalia bipunctata, on the abundance of the rosy apple aphid, Disaphis plantaginea, in organic apple orchards. *Entomol. Exp. Appl.* 90: 167–173.
- ZHENG Y., HAGEN K.S., DAANE K.M. & MITTLER T.E. 1993: Influence of larval dietary supply on the food consumption, food utilisation efficiency, growth and development of the lacewing Chrysoperla carnea. *Entomol. Exp. Appl.* 67: 1–7.

Received January 9, 2009; revised and accepted March 13, 2009