Available online at www.sciencedirect.com
SCIENCE DIRECT®
Biological Control 31 (2004) 138–144
Biological Control
www.elsevier.com/locate/ybcon

Effect of *Aphis gossypii* Glover, *Brevicoryne brassicae* (L.), and *Megoura viciae* Buckton (Hemiptera: Aphidoidea) on the development of the predator *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae)

F.C. Tsaganou,[a] C.J. Hodgson,[b] C.G. Athanassiou,[c] N.G. Kavallieratos,[d,*] and Ž. Tomanović[e]

[a] *Department of Agricultural Sciences, Imperial at Wye, Wye, Ashford, Kent, TN25 5AH, UK*
[b] *Department of Biodiversity and Biological Systematics, The National Museum of Wales, Cardiff, CF1 3NP, Wales, UK*
[c] *Laboratory of Agricultural Zoology and Entomology, Agricultural University of Athens, 75 Iera Odos, 11855, Athens, Attica, Greece*
[d] *Laboratory of Agricultural Entomology, Department of Entomology and Agricultural Zoology, Benaki Phytopathological Institute, 8 Stefanou Delta, Kifissia, Attica, Greece*
[e] *Institute of Zoology, Faculty of Biology, University of Belgrade, Studentski trg 16, 11000, Belgrade, Serbia and Montenegro*

Received 10 November 2003; accepted 23 April 2004
Available online 9 June 2004

Abstract

Laboratory experiments were carried out to evaluate the influence of the aphids *Aphis gossypii* Glover, *Brevicoryne brassicae* (L.), and *Megoura viciae* Buckton on the development of the coccinellid *Harmonia axyridis* (Pallas). Adult *H. axyridis* consumed significantly more individuals of the smallest aphid, *A. gossypii*, than the two larger species, and females consumed twice as many of all three prey species as males. On the other hand, when aphid size (relative weights) was taken into account, it is likely that a greater weight of *M. viciae* was consumed. *H. axyridis* did not complete its development when fed exclusively on either *B. brassicae* or *M. viciae*, but when fed on *A. gossypii*, reproduction started about 8 days after emergence from the pupal stage. The 1st-instar larval stage was also shortest when fed on *A. gossypii* (1.8 days), but took 3.5 days and 4.7 days when fed on *M. viciae* and *B. brassicae*, respectively. In addition, larvae fed on *A. gossypii* were larger and heavier than those fed on the other two aphid species. When larvae were fed exclusively on *M. viciae*, all but one failed to reach the 3rd instar. However, when fed on *A. gossypii* during the 1st instar and then on *M. viciae* in subsequent instars, larval *H. axyridis* completed their development. It was concluded that *M. viciae* was either toxic to the 1st-instar larvae of *H. axyridis* or was an unsuitable host for their development.

Keywords: *Harmonia axyridis*; *Aphis gossypii*; *Brevicoryne brassicae*; *Megoura viciae*; Food consumption; Toxicity; Coccinellidae; Hemiptera; Aphidoidea

1. Introduction

Coccinellids are an important component of biological control programs under glass and in protected situations in UK. A lady beetle not yet used in UK is *Harmonia axyridis* Pallas. This is a large arboreal coccinellid whose fecundity seems to be superior to the majority of other coccinellids (Hukusima and Kamei, 1970). It appears to be more polyphagous than *Coccinella septempunctata* L., feeding on many aphid species (Hodek, 1973; Schanderl et al., 1985), scale insects (McClure, 1987), psyllids (Iablokoff-Khnzorian, 1982), and mites (Lucas et al., 1997), on a variety of crops. According to Freier and Triltsch (1995), *H. axyridis* is considered to be an excellent prospective biological control agent in glasshouses. *H. axyridis* originates from the far East

* Corresponding author. Fax: +3-2108077506.
E-mail address: nick_kaval@hotmail.com (N. Kavallieratos).

1049-9644/$ - see front matter © 2004 Elsevier Inc. All rights reserved.
doi:10.1016/j.biocontrol.2004.04.017

(Lucas et al., 1997) and has recently been introduced into Canada and USA.

Of the aphid species used here, *Megoura viciae* Buckton has been claimed to be toxic to several coccinellids, such as *Adalia decempunctata* (L.), *Adalia bipunctata* (L.), and *Exochomus quadripustulatus* (L.) (Coleoptera: Coccinellidae) (Blackman, 1965, 1967; Dixon, 1958; Radwan and Lövei, 1983). Dixon (1958) reported that the rapid distress of *A. bipunctata* and *A. decempunctata* when fed on *M. viciae* was due to a substance which the aphid had sequestered from its host plant. Later, however, Dixon et al. (1965) conducted a thorough analysis of *M. viciae*, searching for physiologically active compounds, but found nothing to suggest any toxicity.

An understanding of the interactions between predator and prey (in terms of growth rate and voracity) and the range of possible hosts of a predator is an important step in assessing the potential of a biological control agent. The work described below was part of a larger study on the growth rate and voracity of *H. axyridis* when fed on various aphid species in the laboratory.

2. Materials and methods

2.1. Aphid cultures

Three aphids species were used, namely *Megoura viciae* Buckton, *Brevicoryne brassicae* (L.), and *Aphis gossypii* Glover (Hemiptera: Aphidoidea), which had been maintained as clones at Wye for some years. Seedlings of broad bean (*Vicia faba* L.), turnip (*Brassica rapa* L.), and cotton (*Gossypium hirsutum* L.) were used as their respective food plants and each culture was kept in a $60 \times 40 \times 52$ cm perspex cage. Each cage had a 31×43 cm opening for access and ventilation, over which a gauze cover was fastened with Velcro. The cages were kept in a growth room at about 26 °C, 10–50% RH, and a light regime of 16:8 (L:D) h.

Each culture was maintained by the addition of suitable seedlings at weekly intervals. Extra plants were also grown to provide additional leaves for the petri dish experiments.

2.2. Coccinellid culture

Harmonia axyridis was imported as 2nd- and 3rd-instar larvae from Belgium a few weeks prior to the commencement of the experiments. These larvae were then reared on the eggs of *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) to build up the culture. Because *H. axyridis* was cannibalistic, the larvae had to be reared separately.

Cultures of the ladybird were set up in the laboratory at 26 °C where they were fed on a variety of aphid species [*Aphis fabae* Scopoli, *M. viciae*, *Myzus persicae* (Sulzer), *B. brassicae*, or *Aphis sambuci* L. (Hemiptera: Aphidoidea)], collected from field populations. Adult beetles were occasionally stored in an incubator at 15 °C and 12:12 (L:D) h on a weak honey solution for later use with no apparent ill effects on their survivorship or fecundity (McClure, 1987). All experiments used similarly aged larvae or adults.

2.3. Sexing adult beetles

According to Majerus (1994), *H. axyridis* can be sexed based on size (females tend to be larger), by color (males are paler), and by examination of the IX and X abdominal segments. The shape of the latter, however, was not found to be very reliable. At the end of each experiment, all specimens were dissected to confirm their sex.

2.4. Food consumption by adult H. axyridis

After the adult beetles had emerged, they were fed on one of the three aphid species for 4 days, but were then starved for 24 h to ensure that they were hungry before being used in the experiments. Ten replicates (i.e., of 5 individual males and 5 individual mated females) were used for each of the three aphid species. Each adult *H. axyridis* was placed separately in a 8.5 cm petri dish with a hole cut in the lid, covered with a piece of gauze to allow ventilation. The bottom of each petri dish had a thin covering of 3.5% agar gel. A leaf of the correct host plant for the aphid species, cut to fit the petri dish, was placed underside upwards on top of the agar. A known number of aphids of mixed ages were carefully introduced onto the leaf using a paint brush. Mortality during this process was considered to be low, less than 5%. The number of each aphid species consumed was counted daily when new aphids were introduced into the dish. The number of aphids needed was determined from preliminary tests. Twenty adult apterae of each species were also weighed using a Sartorius microbalance to determine their approximate relative sizes.

2.5. Food consumption and development rates of larval H. axyridis

Preliminary tests examined the aphid instar and species on which the beetle larvae would feed. These tests showed that whilst all larval instars would consume all aphid species as adults, the youngest beetle larvae had trouble with the largest aphid instars (particular those of *M. persicae*). Therefore, the experiments with larvae were basically similar to those for the adults except that the stage and number of each aphid species provided varied. Thus, all stages of *A. gossypii* were offered to the youngest beetles but, in the *M. viciae* replicates, only the

smallest instars were fed to the youngest beetles; 3rd instar and adults were fed to the oldest larvae. Newly-hatched ladybird larvae were introduced, one per petri dish. There were 10 replicates for each of the three aphid species.

The number of aphids consumed each day by each larval stage, the duration of each larval instar, and all details of larval development were recorded. In addition, on moulting to adulthood, each was weighed and sexed. This was repeated for each larva fed on each aphid species.

2.6. Suitability of *M. viciae*

From the data obtained in the above experiments, it appeared that, when 1st-instar larvae of *H. axyridis* were fed on *M. viciae*, there was high mortality. Further tests were therefore conducted to check the suitability of this aphid as food for the other larval stages.

Twenty 1st-instar larvae were placed separately in petri dishes as above and fed on *A. gossypii* until they had moulted into the 2nd instar. Five of these larvae were then removed and placed in separate dishes with *M. viciae*, while the rest were allowed to continue feeding on *A. gossypii.* When the remaining 15 2nd-instar larvae reached the 3rd instar, five more larvae were removed and fed separately on *M. viciae*. This was repeated at the 4th instar and the last five larvae were used as the control. The effect of *M. viciae* on the mean developmental period of each larval stage and on the body length and weight of the 3rd and 4th instars were then compared with the data when reared on *A. gossypii* alone and with the data from the previous experiments.

2.7. Statistical analyses

Data were submitted to analysis of variance. Means were separated by using Duncan's multiple range test (DMRT) at $P = 0.05$ (Duncan, 1955).

3. Results

3.1. Consumption by adult *H. axyridis*

The mean number of each of the three aphid species consumed daily by adult male and female *H. axyridis* during a period of 7 days is shown in Table 1. ANOVA showed significant differences ($P < 0.001$) in consumption rates among days, between sexes, and among prey species. Thus, *H. axyridis* females ate more of all three prey species than males, and both males and females ate significantly more *A. gossypii* than *M. viciae* or *B. brassicae*.

It was not possible to know exactly what weight of each aphid was eaten because, whilst all parts of *A. gossypii* were consumed, some parts (legs, head, and shrunken cuticle) of both *B. brassicae* and (more particularly) *M. viciae* sometimes remained afterwards. In the latter case, there was no way of knowing how much weight loss was due to consumption and how much to dehydration, irrespective of how much of the body had actually been eaten. For this reason, the weight of aphid eaten was ignored as a possible parameter for comparison. Nonetheless, the weight ratios of the three aphid species (based on mean adult female weight) was approximately 28:11:1 (*M. viciae*:*B. brassicae*:*A. gossypii*). Thus, assuming that this ratio is reasonably constant for all aphid instars and that most of each individual was consumed, it can be estimated (using the data in Table 1) that each female *H. axyridis* could have consumed, on each day of the experiment, about 280 μg *A. gossypii*, 375 μg *B. brassicae*, and 1 g *M. viciae*. Therefore, it is possible that about 1.3-fold more *B. brassicae* and 3.5-fold more *M. viciae* could have been consumed (by weight) than *A. gossypii*. Nonetheless, Table 1 clearly shows that adult *H. axyridis* killed far more *A. gossypii* than the other two aphids during the 7 days of the experiment.

Harmonia axyridis laid its first eggs after about 8 days when fed on *A. gossypii*, but no eggs were laid within 22 days after adult emergence when the coccinellid fed on either *B. brassicae* or *M. viciae*.

Table 1
Mean number of each aphid species eaten (±SE) each day by adult *H. axyridis* ($n = 10$)

Day	*A. gossypii*		*B. brassicae*		*M. viciae*	
	Female	Male	Female	Male	Female	Male
1	254.2 ± 8.4	143.0 ± 7.7	24.8 ± 1.1	25.0 ± 1.1	33.2 ± 1.3	21.2 ± 0.6
2	246.6 ± 8.3	155.8 ± 4.1	32.2 ± 1.7	21.0 ± 1.1	35.0 ± 1.4	23.6 ± 0.6
3	265.8 ± 8.0	157.8 ± 2.3	31.8 ± 1.1	26.8 ± 0.7	33.0 ± 1.8	23.6 ± 1.2
4	268.8 ± 8.6	162.6 ± 1.6	33.2 ± 0.9	26.6 ± 0.7	34.0 ± 1.4	26.2 ± 0.6
5	274.8 ± 5.8	150.8 ± 5.0	35.0 ± 2.6	24.6 ± 1.6	37.8 ± 0.6	23.2 ± 1.3
6	329.0 ± 8.0	115.4 ± 5.3	39.0 ± 3.6	24.4 ± 1.1	40.2 ± 1.7	21.2 ± 0.8
7	327.0 ± 5.2	116.0 ± 6.7	42.4 ± 2.5	24.2 ± 0.9	42.0 ± 0.9	22.0 ± 1.1
P value						
Among days	0.010		0.0001		0.003	
Between sexes	0.0001		0.0001		0.0001	
Interaction						
Day + sex	0.0001		0.003		0.0001	

3.2. Consumption and development of larval H. axyridis

The duration of each *H. axyridis* instar when fed on the three aphid species is shown in Table 2. All larvae survived and managed to complete their life cycle when fed on *A. gossypii*. However, larvae fed on *B. brassicae* or *M. viciae* had high mortalities, with 60 and 90% dying in the first instar when fed on *B. brassicae* or *M. viciae*, respectively, despite having been fed the smallest available individuals of the two species. Thus, all larvae had died by 2 days after the first moult when fed on *B. brassicae*, while the one remaining larva died in the 3rd instar when fed on *M. viciae*. In addition, the duration of the 1st instar was significantly shorter when fed on *A. gossypii* (1.8 days) than when fed on *M. viciae* (3.5 days) or *B. brassicae* (4.7 days) (Table 2).

With regard to some growth parameters of *H. axyridis*, the head-widths of all 1st-, 2nd-, and 3rd-instar larvae grew by approximately 0.2 mm during each instar (Table 3), and this appeared to be independent of prey species. In addition, total body length and weight also increased significantly at each moult. Interestingly, the total body length of the 2nd-instar larvae appeared to be unaffected by the prey species fed upon. On the other hand, larval weight was significantly reduced when fed on either *B. brassicae* or *M. viciae* in comparison with those fed on *A. gossypii* (Table 3).

The number of each aphid species eaten by each instar is shown in Table 4. The number eaten increased significantly at each subsequent instar when fed on *A. gossypii*, but was very much less (in the 1st and 2nd instars) when fed on *B. brassicae* and *M. viciae* and did not increase significantly after the first moult.

Table 2
Mean duration (± SE) in days for each instar of *H. axyridis* when fed on three different aphid species (*n* in brackets)

Prey species	L1	L2	L3	L4	Prepupa	Pupa	Total development time
A. gossypii	1.8 ± 0.1 (10)	3.0 (10)	3.0 (10)	4.0 (10)	1.0 ± 0.2 (10)	3.1 (10)	15.9
B. brassicae	4.7 ± 0.2 (10)	1.5 ± 0.3 (4)	—	—	—	—	—
M. viciae	3.5 ± 0.4 (10)	5.0 (1)	6.0 (1)				
P	0.0001	—	—	—	—	—	—

Where —, all larvae dead.

Table 3
Growth parameters of each larval instar of *H. axyridis* when fed on three species of aphid (means ± SE) (*n* in brackets)

Species	Instar (reps)	Head (mm)	Length (mm)	Weight (mg)
A. gossypii	1st instar (10)	0.3	1.91 ± 0.1^{d}	0.19 ± 0.00^{d}
	2nd instar (10)	0.5	3.59 ± 0.0^{c}	1.43 ± 0.01^{c}
	3rd instar (10)	0.7	4.77 ± 0.1^{b}	2.45 ± 0.02^{b}
	4th instar (10)	1.1	9.89 ± 0.1^{a}	6.30 ± 0.03^{a}
	P value	ns	⩽0.0001	⩽0.0001
B. brassicae	1st instar (10)	0.3	1.62 ± 0.1^{b}	0.17 ± 0.01^{b}
	2nd instar (4)	0.5	3.60 ± 0.1^{a}	1.33 ± 0.03^{a}
	P value	ns	⩽0.0001	⩽0.0001
M. viciae	1st instar (10)	0.3	1.62 ± 0.01	0.17 ± 0.001
	2nd instar (1)	0.5	3.40	0.98

Data not sharing the same letters in each block differ significantly at $P < 0.0001$ (Duncan multiple range test).

Table 4
Mean number (± SE) of each of three aphid species eaten by each instar of *H. axyridis* (*n* in brackets)

Aphid species (reps)	Instar (reps)	Means ± SE	*P* value
A. gossypii	1st instar (10)	58.1 ± 0.7^{d}	⩽0.0001
	2nd instar (10)	64.5 ± 0.6^{c}	
	3rd instar (10)	101.0 ± 4.3^{b}	
	4th instar (10)	232.7 ± 7.5^{a}	
B. brassicae	1st instar (10)	26.0 ± 0.4	0.39
	2nd instar (4)	24.5 ± 0.5	
M. viciae	1st instar (10)	18.0 ± 0.5	ns
	2nd instar (1)	27.6	

Data not sharing the same letter differ significantly at $P \leqslant 0.0001$ (Duncan multiple range test).

3.3. Comparison of toxicity of *M. viciae* compared with *A. gossypii* on *H. axyridis*

The previous experiment had shown very high mortality of the 1st-instar larvae of the predator when fed on either *B. brassicae* or *M. viciae*. A further experiment examined the effect of *M. viciae* on *H. axyridis* growth rate by introducing *M. viciae* as food at the start of either the 2nd, 3rd or 4th instar, having been fed on *A. gossypii* during previous instars. The results were then compared with those from the previous experiment for the same larval stage (Table 5).

All instars fed on *M. viciae* developed more slowly than when the same instar had been fed on *A. gossypii*, although this difference was only significant in the 3rd and 4th instars. In addition, the period of development for each instar raised on *M. viciae* did not differ significantly, whether it had just completed one instar or three instars on this aphid (Table 5). This suggests that, whilst 2nd-, 3rd-, and 4th-instar larvae of *H. axyridis* are mildly affected by having to feed on *M. viciae* (with a longer developmental time), the 1st instar is seriously affected and most die. The period spent as a prepupa and pupa was not significantly affected by the food quality of the previous instars (Table 5).

Table 7
Comparison of the means (± SE) weight (mg) of adult *H. axyridis* when fed on either *A. gossypii* or on *M. viciae*, having been reared previously on *A. gossypii*

Feeding treatment	Mean weight	*P* value
A. gossypii only	22.35 ± 0.53^{a}	0.008
M. viciae from 2nd instar	18.23 ± 0.92^{b}	
M. viciae from 3rd instar	18.96 ± 2.41^{b}	
M. viciae from 4th instar	21.10 ± 2.61^{a}	

Where data not sharing the same letter differ significantly (Duncan multiple range test).

Table 5
Comparison of the mean development (days) of certain immature stages of *H. axyridis* when fed on either *A. gossypii* or *M. viciae*

Instar	Aphid prey	*n*	Mean time ± SE in days	*P* value
2nd	*A. gossypii* only	15	2.8 ± 0.24	0.267
	M. viciae only	5	3.1 ± 0.41	
3rd	*A. gossypii* only	10	3.0 ± 0.28^{a}	0.0001
	M. viciae from 2nd instar	5	4.2 ± 0.32^{b}	
	M. viciae from 3rd instar	5	4.5 ± 0.35^{b}	
4th	*A. gossypii* only	5	4.0 ± 0.44^{a}	0.0001
	M. viciae from 2nd instar	5	6.2 ± 0.25^{b}	
	M. viciae from 3rd instar	5	5.7 ± 0.18^{b}	
	M. viciae from 4th instar	5	5.5 ± 0.20^{b}	
Prepupa	*A. gossypii* only	5	0.9 ± 0.41	0.939
	M. viciae from 2nd instar	5	0.8 ± 0.51	
	M. viciae from 3rd instar	5	0.9 ± 0.33	
	M. viciae from 4th instar	5	1.0 ± 0.45	
Pupa	*A. gossypii* only	5	3.0 ± 0.62	0.047
	M. viciae from 2nd instar	5	3.8 ± 0.45	
	M. viciae from 3rd instar	5	3.6 ± 0.37	
	M. viciae from 4th instar	5	3.5 ± 0.28	

Data in a given group not sharing the same letter differ significantly (Duncan multiple range test).

Table 6
Comparison of the means (± SE) total-body length (mm) and weight (mg) of 3rd- and 4th-instar larvae of *H. axyridis* when fed on either *A. gossypii* or on *M. viciae*, having been reared previously on *A. gossypii*

Instar	Aphid prey	*n*	Mean total-body length (mm) ±SE	*P* value	Mean weight (mg) ±SE	*P* value
3rd	*A. gossypii* only	10	4.82 ± 0.29	0.785	2.57 ± 0.22	0.849
	M. viciae from 2nd instar	5	4.55 ± 0.26		2.41 ± 0.21	
4th	*A. gossypii* only	5	9.63 ± 0.35	0.03	6.28 ± 0.38^{a}	0.009
	M. viciae from 2nd instar	5	9.00 ± 0.44		5.50 ± 0.11^{b}	
	M. viciae from 3rd instar	5	9.26 ± 0.45		5.85 ± 0.19^{b}	

Data sharing the same letter do not differ significantly (Duncan multiple range test).

The body length of the larvae and of the subsequent adults fed on *M. viciae* were measured and the latter were also weighed (Table 6). Both the 3rd- and 4th-instar larvae fed on *M. viciae* were smaller and weighed less than those fed on *A. gossypii*, but this was only significant in the 4th instar. The effect on adult weight was more subtle (Table 7). The mean weight of adults raised from their 2nd and 3rd instars on *M. viciae* (18.2 ± 0.9 and 19.0 ± 2.4 mg, respectively) was significantly less than when reared entirely on *A. gossypii* (22.4 ± 0.5 mg). However, when fed on *M. viciae* only during the 4th instar, their mean weight (21.1 ± 2.6 mg) was not significantly different from those fed on *A. gossypii* only.

4. Discussion

In our experiments, female *H. axyridis* tended to eat almost twice the number of aphids eaten by males, whichever of the three species they fed upon (Table 1). Moreover, the number eaten per female increased each day during the 7 day observation period, perhaps because their eggs were developing. Male consumption remained reasonably constant throughout the observation period. That female coccinellids eat more than males has been noted in many other studies, such as with *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae) feeding on the eggs and crawlers of the mealybug *Maconellicoccus hirsutus* Green (Hemiptera: Pseudococcidae) (Babu and Azam, 1988); with *Coccinella septempunctata* L. (Coleoptera: Coccinellidae) feeding on the aphid *Lipaphis erysimi* (Kaltenbach) (Hemiptera: Aphidoidea) (Singh and Singh, 1994), and with *Pullus mediterraneus* Fabre (Coleoptera: Coccinellidae) when feeding on the eggs of the soft scale, *Saissetia oleae* Olivier (Hemiptera: Coccidae) (Ba M'Hamed and Chemseddine, 2001).

In these experiments, both adults and larvae ate more *A. gossypii* than *B. brassicae* or *M. viciae* (Tables 1 and 4). Elliott et al. (1994) suggested that differences in the number of aphids consumed by *Cycloneda ancoralis* (Germar) (Coleoptera: Coccinellidae) larvae might be related to prey size, with the predator tending to consume more *A. gossypii* than *Aphis helianthi* Monell (Hemiptera: Aphidoidea), *L. erysimi*, and *Diuraphis noxia* (Kurdjumov) (Hemiptera: Aphidoidea) because it was the smallest aphid species offered. It is highly probable that the greater number of *A. gossypii* consumed in our experiments was also due to its small size. Similar results have been found with *H. axyridis* by Hukusima and Kamei (1970), who reported that it consumed 1.5-times more *M. persicae* than *Amphorophora oleracea* Van der Goot (Hemiptera: Aphidoidea) due to the smaller body size of the former species. However, in our experiments, when the relative size (as shown by mean weight) of the three aphid species was taken into consideration, it is likely that a greater weight of *M. viciae* was consumed than of the other two species. Our results showed that larval *H. axyridis* were able to complete development on *A. gossypii*, suggesting that this aphid species constitutes "essential" food (see Elliott et al., 1994; Hodek, 1973; Hodek and Honćk, 1996) (i.e., this aphid seemed to satisfy the nutritional requirements of both the larvae and adults of this coccinellid). The potential for using *H. axyridis* for the control of *A. gossypii* looks good. Orlandini and Martellucci (1997) reported that *H. axyridis* gave excellent control of *A. gossypii* on melon in the field in Italy, and Dong (1988) showed that *H. axyridis* could successfully control *A. gossypii* at predator:prey ratios of 1:100–200 without the need to apply pesticides.

Our results also suggest that *B. brassicae*, and perhaps even more, *M. viciae* might be either toxic or unsuitable as food to *H. axyridis*, particularly the 1st-instar larvae. Why certain prey are harmful to coccinellids is not yet fully understood (Hodek and Honćk, 1996), but similar observations have been noted by other workers. Thus, Hukusima and Kamei (1970), also working with *H. axyridis*, found that, of 10 aphid species, *Aphis pomi* De Geer (Hemiptera: Aphidoidea), *B. brassicae*, and *Hyalopterus pruni* (Geoffroy) (Hemiptera: Aphidoidea) seemed to be least suitable as food for the larvae, prolonging their development by a third in comparison with the six most suitable species. However, in Hukusima and Kamei's experiments, the larvae did succeed in completing their development on all 10 species, including *B. brassicae*. In our experiments, the larvae only developed to the 2nd instar when fed exclusively on *B. brassicae* and just to the 3rd instar on *M. viciae*. Our results with *B. brassicae* agree with the observations of several other workers. Telenga and Bogunova (1936) found that *H. axyridis* refused *B. brassicae* in the field; Hodek and Honćk (1996) found that when adult *H. axyridis* were transferred from *H. pruni* to *B. brassicae* in the laboratory, the predator ate 90% less and ceased ovipositing; and Garcia et al. (1999) reported high mortality when *H. axyridis* was fed on *B. brassicae*. In addition, Blackman (1967) found *B. brassicae* to be unsuitable as prey for *A. bipunctata*, with a third of the larvae dying within a week. Possible reasons why *B. brassicae* may be unsuitable was thought to be the waxy powder which covers the aphids body, possibly making it unpalatable and difficult to handle (Hodek and Honćk, 1996; Rotheray, 1989). However, it has recently been shown that *B. brassicae* is able to take up and sequester glycosinolates from its cruciferous host plants (Bridges et al., 2002). Glucosinolates are considered to be the major cause of rejection of Cruciferae by most herbivores and their sequestration in *B. brassicae* is the most likely explanation of their unsuitability to *H. axyridis*.

When larval *H. axyridis* were fed exclusively on *M. viciae*, the larvae died approx. 3.5 days after their

emergence (Table 2). Although *M. viciae* has been claimed to be toxic to other coccinellids (see Section 1) and Dixon (1958) had reported rapid distress of *A. bipunctata* and *A. decempunctata* when fed on *M. viciae*, he later (Dixon et al., 1965) conducted a thorough analysis of *M. viciae*, but found nothing to suggest any toxicity. Our results show that larval *H. axyridis* fed exclusively on *M. viciae* during their 2nd, 3rd, and 4th instar did complete their development, although at a slower rate than on *A. gossypii* (Table 5) and that the resultant adults weighed less (Table 7) when fed on *M. viciae* from the 2nd or 3rd instar, but not when fed on it only from the 4th instar (Table 7). Olszak (1986, 1988) also obtained smaller *A. bipunctata*, *Propylea quatuordecimpunctata* (L.) (Coleoptera: Coccinellidae), and *C. septempunctata* when these species were fed on non-preferred aphids such as *A. pomi*.

Thus, it is unlikely that *H. axyridis* will be useful as a biological control agent for either *B. brassicae* or *M. viciae*. In addition, it seems possible that *M. viciae* could sequester allelochemicals, as has now been shown for *B. brassicae*. The role of these chemicals in the tri–trophic interactions involving the leguminous host plants, *M. viciae* and its coccinellid predators needs further study.

Acknowledgments

The authors would particularly like to thank Biological Crop Protection, Wye, for supplying the *H. axyridis*. Also Dr. M.J.W. Copland (Imperial at Wye) for his critical reading of the manuscript and valuable discussion, Professor Dr. Fotios T. Gravanis (Technological Educational Institute of Larissa, Greece) for his assistance, and Dr. Alan Clewer (Imperial at Wye) for his guidance in the statistical analysis. We would also like to thank three anonymous reviewers for their helpful suggestions.

References

Babu, T.R., Azam, K.M., 1988. Predation potential of *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae) in relation to temperature. J. Res. APAU 16, 108–110.

Ba M'Hamed, T., Chemseddine, M., 2001. Assessment of temperature effects on the development and fecundity of *Pullus mediterraneus* Fabre (Coleoptera: Coccinellidae) and consumption of *Saissetia oleae* Olivier (Hemiptera: Coccidae) eggs. J. Appl. Entomol. 125, 527–531.

Blackman, R.L., 1965. Studies on specificity in Coccinellidae. Ann. Appl. Biol. 56, 336–338.

Blackman, R.L., 1967. The effects of different aphid foods on *Adalia bipunctata* Linnaeus and *Coccinella septempunctata* Linnaeus. Ann. Appl. Biol. 59, 207–219.

Bridges, M., Jones, A.M.E., Bones, A.B., Hodgson, C., Cole, R., Bartlett, E., Wallsgrove, R., Karapapa, V.K., Watts, N., Rossiter, J.T., 2002. Spatial organisation of the glucosinolate-myrosinase system in brassica specialist aphids is similar to that of the host plant. Proc. R. Soc. Lond. B 269, 187–191.

Dixon, A.F.G., 1958. The escape responses shown by certain aphids to the presence of the coccinellid *Adalia decempunctata* (Linnaeus). Trans. R. Entomol. Soc. 110, 319–334.

Dixon, A.F.G., Martin-Smith, M., Subramaniam, G., 1965. Constituents of *Megoura viciae* Buckton. J. Chem. Ecol. 1965, 1562–1564.

Dong, Y.G., 1988. Trials on the control of *Aphis gossypii* Glover with *Coccinella axyridis* Pallas. Zhejiang Agric. Sci. 3, 135–139.

Duncan, D.B., 1955. Multiple range and multiple F tests. Biometrics 11, 1–42.

Elliott, N.C., French, B.W., Michels Jr., G.J., Reed, D.K., 1994. Influence of four aphid prey species on development, survival and adult size of *Cycloneda ancoralis* (Germar) (Coleoptera: Coccinellidae). Southwest. Entomol. 19, 57–61.

Freier, B., Triltsch, H., 1995. *Harmonia axyridis* Pallas—ein interessanter Marienkafer fur den biologischen Pflanzenschutz. Gesunde Pflanzen 47, 269–271.

Garcia, M.F., Becerra, V.C., Reising, C.E., 1999. *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae) biological study. Rev. Facul. Cien. Agrar. Univ. Nac. Cuyo 31, 85–91.

Hodek, I., 1973. Biology of Coccinellidae. Dr W. Junk, The Hague.

Hodek, I., Honćk, A., 1996. Ecology of Coccinellidae. Kluwer Academic Publishers, Dordrecht.

Hukusima, S., Kamei, M., 1970. Effects of various species of aphids as food on development, fecundity and longevity of *Harmonia axyridis* Pallas. Res. Bull. Fac. Agric. Gifu Univ. 29, 53–66.

Iablokoff-Khnzorian, S.M, 1982. Les coccinelles: Coléoptères—Coccinellidae. Sociétè Nouvelle des Éditions Boubée, Paris.

Lucas, E., Coderre, D., Vincent, C., 1997. Voracity and feeding preferences of two aphidophagous coccinellids on *Aphis citricola* van der Goot and *Tetranychus urticae* (Koch). Entomol. Exp. Appl. 85, 151–159.

Majerus, M.E.N., 1994. Ladybirds. Harper Collins, London.

McClure, M.S., 1987. Potential of the Asian predator, *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae), to control *Matsucoccus matsumurae* (Kuwana) (Homoptera: Margarodidae) in the United States. Environ. Entomol. 16, 224–230.

Olszak, R.W., 1986. Suitability of three aphid species as prey for *Propylea quatuordecimpunctata* (Linnaeus). In: Hodek, I. (Ed.), Ecology of Aphidophaga. Academia, Prague and Dr. W. Junk, Dordrecht, pp. 51–55.

Olszak, R.W., 1988. Voracity and development of three species of Coccinellidae, preying upon different species of aphids. In: Niemczyk, E., Dixon, A.F.G. (Eds.), Ecology and effectiveness of aphidophaga. SPB Academic Publishers, The Hague, pp. 47–53.

Orlandini, G., Martellucci, R., 1997. Melone: lotta biologica all *Aphis gossypii* Glover. Colture Protette 6, 33–36.

Radwan, Z., Lövei, G.L., 1983. Aphids as prey for the coccinellid *Exochomus quadripustulatus* (Linnaeus). Entomol. Exp. Appl. 34, 283–286.

Rotheray, G.E., 1989. Aphid Predators. Richmond, Slough.

Schanderl, H., Ferran, A., Larroque, M.M., 1985. Les besoins trophiques et thermiques des larves de la coccinelle *Harmonia axyridis* Pallas. Agronomie 5, 417–421.

Singh, D., Singh, H., 1994. Predatory potentiality of coccinellids, *Coccinella septempunctata* Linnaeus and *Hippodamia variegata* (Goetze) over mustard aphid, *Lipaphis erysimi* (Kaltenbach). Crop Res. Hisar 7, 120–124.

Telenga, N.A., Bogunova, M.V., 1936. The most important predators of coccids and aphids in the Ussuri region of Far East and their utilization. Zashch. Rast. 1939 (10), 75–87 (in Russian).