



Suitability of *Bemisia tabaci* (Hemiptera: Aleyrodidae) biotype-B and *Myzus persicae* (Hemiptera: Aphididae) as prey for the ladybird beetle, *Serangium japonicum* (Coleoptera: Coccinellidae)

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Abstract. The ladybird, *Serangium japonicum* (Coleoptera: Coccinellidae), is one of the most important predators of whiteflies in China, however, the suitability of different kinds of prey and nutritional requirements of this predator are poorly studied. The objectives of the present study were to investigate the pre-imaginal development, percentage survival of immatures, adult longevity and fecundity of *S. japonicum* when reared on two different species of prey, mixed ages of the cotton whitefly *Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) biotype-B (i.e., eggs and nymphs) and green peach aphid *Myzus persicae* (Sulzer) (Hemiptera: Aphididae) in order to quantify their relative suitability as prey under laboratory conditions of $25 \pm 1^\circ\text{C}$, $50 \pm 10\%$ RH and a photoperiod of 16L : 8D. The results of this study indicate that the total developmental period (from egg to adult) was shorter when they were fed on *B. tabaci* (18.29 ± 0.13 days) than on *M. persicae* (19.85 ± 0.22 days). The percentage survival of the immatures, growth index, development rate, adult fresh weight, fecundity and percentage of eggs hatching were higher and the pre-oviposition period was significantly shorter when *S. japonicum* were fed on *B. tabaci* than on *M. persicae*. In contrast, there was no significant difference in their longevity and oviposition period when fed on *B. tabaci* and *M. persicae*. The results indicate that as a diet for *S. japonicum* *M. persicae* is inferior to *B. tabaci*. This is the first record of *S. japonicum* being able to complete its development feeding exclusively on *M. persicae*. Since *B. tabaci* and *M. persicae* often simultaneously occur in vegetable fields, these results indicate that it is likely that *S. japonicum* could be utilized to control mixed populations of aphids and whiteflies, and furthers our understanding of ladybird population dynamics in the field in relation to the availability of different species of prey.

1. INTRODUCTION

The cotton whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), occurs worldwide in tropical and subtropical climates as well as in protected agricultural systems in temperate regions. It is an important pest of more than 600 species of plants of numerous field and vegetable crops in many parts of the world (Naranjo et al., 2010). Whiteflies cause serious damage by feeding, producing honeydew and the resultant sooty mould, and disease transmission (Stansly & Natwick, 2010). Although known as a sporadic pest of cotton in China for many years, *Bemisia tabaci* Biotype-B did not become an important pest in northern China until 2000 (Wu et al., 2002), and is now widely distributed across China (Hu et al., 2011). Many attempts have been made to control *B. tabaci*. Because of its high reproductive rate, many generations per year and ability to rapidly develop resistance to insecticides, whiteflies

are difficult to control using conventional means (Castle et al., 2010). Thus, biological control becomes the primary method for suppressing whitefly populations in agroecosystems (Arno et al., 2010).

The green peach aphid, *Myzus persicae* (Sulzer) (Hemiptera: Aphididae), is a highly polyphagous insect pest, which feeds on more than 400 species of plants belonging to over 50 families including agricultural crops (e.g., potato, sugar beet and tobacco), horticultural crops (e.g., plants of Brassicaceae, Solanaceae and Cucurbitaceae) and stone fruit (e.g., peach, apricot and cherry) around the world. It causes enormous economic losses because it reduces crop yields by sucking plant sap, transmits over 100 plant viruses and secretes honeydew (Blackman & Eastop, 2000; Kim et al., 2005). Currently, control of *M. persicae* is mostly based on insecticides, and high levels of insecticide resistance are recorded in many parts of the world (Li et al.,

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2016). Effective and environmentally sound management of this whitefly and aphid is needed, and biological control using natural enemies is an attractive option.

The ladybird, *Serangium japonicum* Chapin (Coleoptera: Coccinellidae), is an important predator of whiteflies and scale insects (Ren et al., 2009). It occurs in China and Japan, where *S. japonicum* is potentially a good biological control agent of *B. tabaci* in various crop systems in China, especially vegetables and cotton (Ren et al., 2001). *Serangium japonicum* larvae and adults feed on the immature stages of *B. tabaci* and can effectively suppress its population growth (Li et al., 2015). However, the nutritional ecology, prey preference and food requirements for development and reproduction of *S. japonicum* are not well studied.

Food quality is one of the most important factors influencing the vigour of predators because not all species of prey are equally suitable as food. These influences range from slight effects such as longer developmental times, or a decrease in percentage larval survival to dramatic effects such as toxicity of certain species of prey resulting in high mortality (Obrycki & Kring, 1998). Given that there may be a number of alternative prey available to some predatory insects in an agroecosystem, preference for and the suitability of prey for development are important in determining the potential of predators as biological control agents of a specific pest, which may determine the success or failure of predators in biological control systems (Thompson, 1999). Furthermore, many predatory insects are opportunistic predators in the field and feed on a wide range of species of prey and plant products, but this behaviour may not clearly indicate prey suitability and predator nutritional requirements (Strohmeyer et al., 1998).

In developing an effective IPM program for controlling *B. tabaci* it would be advantageous to maximize the effectiveness of the chosen natural enemy. During a preliminary investigation, we found that *S. japonicum* frequently fed not only whiteflies, but also on the green peach aphid in vegetable fields in north western China. This prompted us to speculate that *S. japonicum* could prey on both whiteflies and aphids. Since *S. japonicum* is known as a promising predator of whiteflies, we therefore hypothesized that different species of prey might vary in their suitability as prey for *S. japonicum*. The objective of this study was to determine whether *B. tabaci* and *M. persicae* were equally suitable for the development, survival, longevity and fecundity of *S. japonicum*.

2. MATERIALS AND METHODS

2.1. Insects

Serangium japonicum adults were originally collected from the experimental farm of the Northwest A&F University (Yangling, Shaanxi, China; 34°17'09.35"N and 108°04'17.12"E). They were maintained in the laboratory on potted cabbage (*Brassica oleracea* L.) infested with immature *B. tabaci*. All cultures were kept in nylon mesh covered cages (60 × 60 × 60 cm) in a greenhouse maintained at 20–30°C under a natural light regime.

Myzus persicae used as prey for the ladybird was cultured on sweet pepper (*Capsicum annuum* L.) plants in the laboratory for

3 years. *Bemisia tabaci* B biotype was collected from tomato (*Solanum lycopersicum* L.) in a greenhouse in Yangling, Shaanxi, China (34°17'37.01"N and 108°01'03.34"E). They were reared on cabbage in mesh covered cages (60 × 60 × 60 cm) in the same greenhouse. The whiteflies were identified as *B. tabaci* B-biotype by RAPD-PCR using the mitochondrial COI gene (Wu et al., 2002). To produced mixed ages of *B. tabaci* (eggs and nymphs), cabbage plants were exposed to *B. tabaci* adults in the stock culture cages for 48 h, and then incubated in growth chambers under the same conditions as the ladybirds and aphids. To avoid food adaptation, populations of the adult beetles were established in the same greenhouse and supplied with a mixture of *B. tabaci* and *M. persicae* on their respective host plants in mesh covered cages (60 × 60 × 60 cm) for several generations before they were used in this study.

2.2. Pre-adult development and immature survival

Effect of prey species on the pre-adult development and immature survival of *S. japonicum* was evaluated. Leaves with beetle eggs (< 2 h old, about eighty eggs) were excised from plants and placed in two plastic boxes (length × width × depth = 8 × 6 × 3 cm, covered with fine muslin cloth for ventilation) until they hatched. A piece of filter paper was placed at the bottom of the boxes, and a few drops of water were added as needed to maintain humidity. The boxes with the insects were kept in a climatic chamber (25 ± 1°C, 50 ± 10% RH under a photoperiod of 16L:8D). The *S. japonicum* eggs were inspected carefully every 6 h and the number that hatched were recorded. Newly hatched first instars of *S. japonicum* (< 6 h old) were individually transferred to Petri dishes (9.0 cm in diameter and 3.0 cm in height) using a soft camel hair brush and placed in the close vicinity of the prey. A piece of filter paper was placed at the bottom of the Petri dish and a few drops of water were added as needed to maintain humidity. Our preliminary test showed that the first instar larvae of *S. japonicum* did not feed on the adult aphids, so only first instar nymphs of *M. persicae* (about 20–30) were fed to first instar larvae of *S. japonicum*. A mixed age group of aphids (about 60–80) were selected for testing after the first instar ladybirds moulted to the second instar. The sweet pepper leaf with aphids was replaced every 12 h to avoid microbial contamination.

To provide whitefly prey, a cabbage leaf infested with mixed ages of *B. tabaci* was put into a Petri dish (9.0 cm in diameter and 3.0 cm in height). One first instar larva of *S. japonicum* (< 6 h old) was added to the arena. Afterwards, each *S. japonicum* larva was transferred daily to a new arena containing fresh prey.

All experiments were conducted in climatic chambers at 25 ± 1°C, 50 ± 10% RH under a photoperiod of 16L:8D. Forty newly hatched *S. japonicum* larvae (< 6 h old) were reared on each category of prey. *S. japonicum* larvae were checked for moulting or death every 6 h. The duration of development of each immature stage of *S. japonicum* was recorded and the initial body mass of newly-emerged adults was determined by weighing them individually on an electronic balance (Mettler-Toledo XS64, Greifensee, Switzerland) with a precision of 0.1 mg.

Percentage immature survival, adult emergence and development rate (1/developmental period) were recorded using the following formulae, and the growth index is that proposed by Dubey et al. (1981). Survival was calculated as: Percentage survival (%) = Number surviving in an assessed instar / Number surviving in the previous instar × 100. Growth index was calculated as: Growth index = Percentage pupation / Developmental time of larvae × 100. Percentage pupation (%) = Number of pupae / Number of fourth instar larvae × 100. Percentage adult emergence (%) = Number of adults emerged / Number of pupae × 100. Percentage

survival to the adult stage (%) = Number of adults that emerged/Initial number of first instar larvae \times 100.

2.3. Longevity and reproductive performance

To evaluate the longevity and reproductive performance of *S. japonicum*, newly emerged adults were sexed and paired. A total of fifteen randomly selected pairs were used. Each pair was kept in a Petri dish (9.0 cm in diameter and 3.0 cm in height). A piece of filter paper was placed at the bottom of the Petri dish and a few drops of water added as needed to maintain humidity. Petri dishes were placed in climatic chambers with the same settings as above. Fresh leaf disks with *B. tabaci* or *M. persicae* were provided daily as a source of food. Each couple was observed daily to record longevity, pre-oviposition period, oviposition period and fecundity (number of eggs laid) until all beetles died.

2.4. Data analysis

Duration of development of immature stages, developmental rate, longevity, preoviposition period, oviposition period, fecundity and adult body weight of *S. japonicum* were analyzed using one-way analysis of variance (ANOVA); percentage survival was analyzed using the survival analysis (Log Rank test). The experimental data were analyzed using the statistical package SPSS 10.0 (2000; Chicago, IL, USA).

3. RESULTS

3.1. Development and survival of immature *S. japonicum*

The duration of development of the eggs, third instar and fourth instar larvae of *S. japonicum* were shorter when they fed on *B. tabaci* than on *M. persicae* (Table 1). When larvae of *S. japonicum* fed on *B. tabaci*, the developmental time of the pupa increased by 17.1% compared to those fed on *M. persicae* (Table 1). The total developmental period (from egg to adult) was significantly shorter when they fed on *B. tabaci* than on *M. persicae* (Table 1). The body mass of newly emerged adults differed significantly after feeding on *M. persicae* compared to *B. tabaci*, and was higher when fed on *B. tabaci* (Table 1). The percentage survival of first, second, third and fourth instar larvae and the pupal stage did not vary significantly between *S. japonicum* larvae fed on *B. tabaci* and *M. persicae* ($\chi^2 = 1.459$, $P = 0.227$) (Fig. 1). The percentage of larvae reaching the adult stage was much higher (95%) when fed on *B. tabaci* than on *M. persicae* (67.5%) ($\chi_{0.05}^2 = 9.928$, $P < 0.005$).

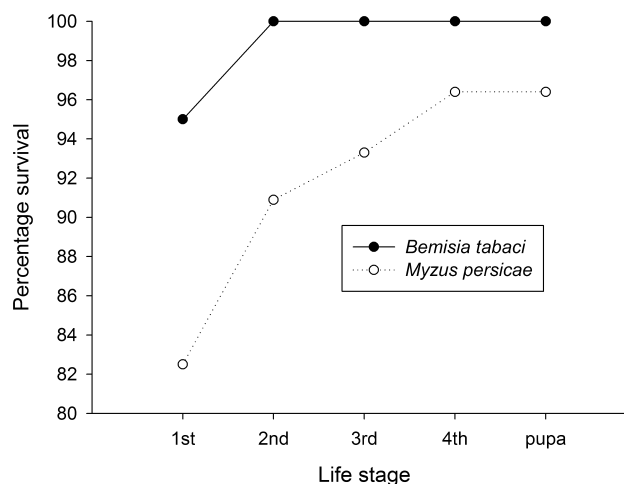


Fig. 1. Percentage survival of the immature stages of *Serangium japonicum* fed on *Myzus persicae* and *Bemisia tabaci*.

3.2. Reproductive potential and longevity on different species of prey

The growth index of *S. japonicum* was higher when fed on *B. tabaci* than on *M. persicae* (Table 2). The developmental rate of the ladybirds was similar on the two species of prey (Table 2). When *S. japonicum* fed on *B. tabaci*, the preoviposition period markedly decreased by 55.5% compared to when fed on *M. persicae* (Table 2), but the percentage of eggs that hatched significantly increased (Table 2). The oviposition period and longevity did not differ significantly when *S. japonicum* was fed on *B. tabaci* or *M. persicae*, although they were shorter when the ladybird fed on *B. tabaci* (Table 2). The fecundity of the ladybird adults fed *B. tabaci* was significantly increased by 59.3% compared to on *M. persicae* (Table 2).

Over most of the oviposition period, daily fecundity of *S. japonicum* fed on *B. tabaci* was higher than on *M. persicae* (Fig. 2). Daily fecundity of the ladybird was more than 10 eggs per female in the periods 14 to 19 days, 27 to 29 days and 33 to 36 days after the onset of oviposition. When fed on *B. tabaci* a maximum daily fecundity of 46 eggs was recorded on day 17. Mean total fecundity per female was 294.5 eggs when fed on *B. tabaci* (Table 2) and a single female laid 489 eggs. When *S. japonicum* fed on *M. persicae*, the daily fecundity was a relatively constant (around 5 eggs) for 53 days (Fig. 2) and the maximum daily fecundity recorded was 17 eggs on day 42. Mean fecundity per female was 184.9 eggs and a single female laid 372 eggs.

Table 1. Duration of development (days) of the different immature stages and adult body mass of *Serangium japonicum* fed on *Myzus persicae* and *Bemisia tabaci*.

Stage	Duration of development, days \pm SE		df	F	P
	<i>Bemisia tabaci</i>	<i>Myzus persicae</i>			
Egg	3.83 \pm 0.01	4.94 \pm 0.04	1,79	652.552	0.0001
First instar	2.16 \pm 0.06	2.33 \pm 0.09	1,67	0.935	0.337
Second instar	1.77 \pm 0.03	1.93 \pm 0.12	1,64	1.546	0.218
Third instar	1.52 \pm 0.06	2.08 \pm 0.04	1,62	59.652	0.0001
Fourth instar	4.18 \pm 0.04	4.43 \pm 0.13	1,62	4.714	0.034
Pupa	4.83 \pm 0.06	4.14 \pm 0.08	1,62	60.053	0.0001
Egg–adult	18.29 \pm 0.13	19.85 \pm 0.22	1,61	33.030	0.0001
Adult body weight (mg)	1.53 \pm 0.02	1.43 \pm 0.03	1,61	6.065	0.017

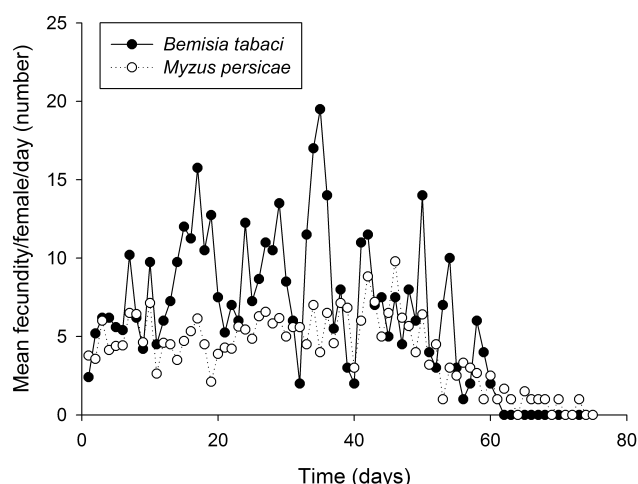


Fig. 2. Mean fecundity of *Serangium japonicum* fed on *Bemisia tabaci* and *Myzus persicae*. Note: for clarity the error bars of each data point were not included.

4. DISCUSSION

The quality of prey affect the development, survival and reproduction of coccinellids (Thompson, 1999). Hodek (1996) categorized prey into essential, alternative and rejected based on their effect on the developmental rate, survival and reproductive capacity of coccinellid predators. Our study indicates that both *B. tabaci* and *M. persicae* are suitable prey for the development of *S. japonicum*, but the percentage survival of immatures of *S. japonicum* fed on *M. persicae* was relatively low (67.5%). Zhang et al. (2012) document *Acyrtosiphon pisum* as a suitable prey for *Propylea japonica*, however, the larvae of this ladybird fed on *A. pisum* had a lower percentage survival than those fed on other species of aphid. Osman & Selman (1993) also report that *M. persicae*, *A. pisum* and *Aphis fabae* are suitable prey for the lacewing, *Chrysoperla carnea*, but those fed on *A. fabae* suffered a higher juvenile mortality than those fed on *M. persicae*. The enhanced performance of predatory coccinellid larvae on certain prey may be related to their higher protein levels or higher palatability (Omkar & Srivastava, 2003).

There may be different levels of suitability of individual essential prey. In the present study the different species of prey did not significantly affect the percentage survival of the coccinellid larvae, but each larval instar survived less well when fed on *M. persicae* than on *B. tabaci*. However, significantly fewer of the *S. japonicum* fed on *M. persicae* survived to the adult stage and were lighter in weight, had a lower fecundity and a lower percentage of their eggs

hatched. Osman & Selman (1993) report that although *A. fabae* is a suitable prey for the lacewing *C. carnea* it results in this predator producing smaller cocoons and having a lower fecundity. The fertility of *Propylea japonica* reared on *Ephestia kuehniella* eggs is significantly lower than when reared on *A. pisum*, though both are regarded as essential prey (Hamasaki & Matsui, 2006). These studies reveal that suppression of growth and developmental rate of larvae is likely to affect the size and maturation of adults, which in turn may result in reduced reproductive success or survival. More interestingly, we found that when *S. japonicum* fed on *M. persicae*, the preoviposition period was significantly prolonged but the oviposition period and longevity were not significantly different. When adults of *S. japonicum* are fed on the less nutritious food (i.e., *M. persicae*) it may affect the development of their testes and sperm and/or oocytes and ovarian development that could result in the extension in pre-oviposition period. On the other hand, these females could allocate more resources to longevity at the cost of a reduction in reproduction so as to maintain population development. Further investigations are needed to elucidate the physiological mechanisms that underlie the effect of feeding on less nutritious prey on the subsequent fitness and life history characteristics of *S. japonicum*.

S. japonicum has considerable potential as a biocontrol agent of *B. tabaci* in China (Lin et al., 2008). However, the general biology of this ladybird is not well studied. To date, most information on *S. japonicum* is limited to its compatibility with entomopathogenic fungi (Fatiha et al., 2008; Hu et al., 2009) or insecticides (He et al., 2012; Li et al., 2015; Yao et al., 2015). Only two studies assess the development, survival and reproduction of this ladybird when fed on *B. tabaci* on different host plants, which resulted in the longevity and fecundity of the females differing significantly (Jing et al., 2003; Yao et al., 2011). In general, host plants on which prey feed affect the nutrition of the prey, and in turn, affect the development and survival of the predator (Giles et al., 2002). Therefore, it is necessary to evaluate the effects of different species and cultivars of host plants on the development, survival and reproduction of *S. japonicum* in order to improve the efficiency of the control of whiteflies.

Currently more than 40 species of coccinellid are described as predators of *B. tabaci*; however, recent work has only focused on *Serangium parcesetosum* and *Delphastus catalinae*. Detailed evaluation and utilization of most ladybeetles is lacking (Arno et al., 2010). We think this is due to the poor knowledge of their food ecology

Table 2. Growth and reproductive parameters of *Serangium japonicum* fed on *Myzus persicae* and *Bemisia tabaci*.

Parameters	<i>Bemisia tabaci</i>	<i>Myzus persicae</i>	df	F	P
Growth index	10.31	9.01			
Developmental rate (day ⁻¹)	0.069 ± 0.001	0.067 ± 0.001	1,61	2.056	0.157
Preoviposition period (days)	9.3 ± 0.3	20.9 ± 1.2	1,29	84.599	0.001
Oviposition period (days)	61.4 ± 5.2	71.2 ± 2.5	1,29	2.944	0.097
Longevity (days)	79.8 ± 3.9	89.3 ± 3.8	1,59	3.078	0.085
Fecundity	294.5 ± 38.5	184.9 ± 26.5	1,29	5.510	0.026
Percentage of eggs that hatched (%)	96.7 ± 0.8	72.9 ± 3.1	1,29	59.294	0.001

and behaviour. In the present study, we firstly found that *S. japonicum* can complete its development and growth when fed exclusively on *M. persicae*, although this aphid is not the best quality prey for this predator. In northern China, both *B. tabaci* and *M. persicae* often coexist in the field and on greenhouse grown vegetables, though their relative abundance varies over time (Zhang et al., 2007). Since *S. japonicum* is an effective predator of *B. tabaci* (Ren et al., 2001), the possibility of utilizing a single natural enemy to control mixed populations of aphids and whiteflies may reduce costs when *S. japonicum* is used for whitefly control in greenhouses. In addition, our results demonstrate that *S. japonicum* can complete its life history when only feeding on *M. persicae*. Therefore, this ladybeetle can survive throughout the season in the absence of whiteflies. Mixed diets consisting of several species of prey (e.g. different species of aphids) and other non-prey food (e.g. nectar or pollen) are known to favour the development and survival of *Chrysoperla rufilabris* (Legaspi et al., 1994), *Chrysoperla carnea* (Patt et al., 2003) and *Coccinella septempunctata* (Hauge et al., 1998; Nielsen et al., 2002), but the effects of mixed diets consisting of *B. tabaci* and aphids or other alternative food on the demographic parameters of predatory ladybirds have not been evaluated. Therefore, further investigation of the effects of mixed *B. tabaci* and aphid diets on *S. japonicum* would be valuable in terms of using ladybirds as biological control agents of whiteflies and/or aphids and whether the presence of both prey enhance or reduce the biocontrol of each of them.

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