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Preference of two populations of *Propylea quatuordecimpunctata* (Coleoptera: Coccinellidae) for *Aphis fabae* and *Aphis gossypii* (Homoptera: Aphididae)

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Abstract. Prey preference of natural enemies is an important parameter used in studies on their efficiency. Feeding preferences of individuals from two populations of *Propylea quatuordecimpunctata* (L.) (Coleoptera: Coccinellidae) for the essential prey items, *Aphis fabae* Scopoli, 1763 and *Aphis gossypii* Glover, 1877 (Homoptera: Aphididae) were evaluated in the laboratory using Manly's β preference index (β). For the predator preference experiment, equal numbers of *A. fabae* and *A. gossypii* were offered to the predator. The experiment was conducted on a broad bean leaf disc with 20 replicates. After 24 h, the unconsumed aphids were counted. Individuals of *P. quatuordecimpunctata* from both populations consumed more *A. gossypii* [Karaj population (mean ± SE): 28.15 ± 1.04 and Sari population 34.35 ± 0.51] than *A. fabae* (Karaj population: 17.95 ± 0.64 and Sari population: 17.7 ± 1.01). The values of Manly's β preference index (β) for *A. gossypii* were 0.73 ± 0.01 and 0.77 ± 0.01 for the Karaj and Sari populations, respectively and for *A. fabae* were 0.26 ± 0.01 and 0.22 ± 0.01 for the Karaj and Sari populations, respectively. Comparison of the preference indices using a t-test revealed significant differences between the prey preferences of those from the Sari population ($t = 22.53$, $df = 38$, $P < 0.0001$) and the Karaj population ($t = 26$, $df = 38$, $P < 0.0001$). These results reveal that *P. quatuordecimpunctata* from the two populations prefer *A. gossypii* over *A. fabae*.

INTRODUCTION

Food quality is one of the most important factors determining the fitness of predatory ladybirds (Kalushkov & Hodek, 2005). Ladybirds (Coleoptera: Coccinellidae) are beneficial insects in crop ecosystems, as they feed on plant pests such as aphids, but not all aphid species are equally suitable for all ladybird species (Obrycki & Orr, 1990; Phoofolo & Obrycki, 1997; Kalushkov, 1998; Michaud, 2000; Kalushkov & Hodek, 2004; Mignault et al., 2006). Prey preference of coccinellid beetles should be considered because when they are given a choice between two different species of prey, they often prefer one of them. The prey of coccinellids is classified into one of four groups: essential, alternative (accepted but suboptimum), rejected and toxic (Majerus, 1994). Based on this classification, coccinellid beetles prefer essential prey because when fed this category of prey they perform best in terms of egg maturation, oviposition and development. Alternative and toxic prey is consumed at lower rates (Hodek, 1973; Majerus, 1994). Many factors can affect the prey preferences of coccinellids, including the mobility and defence of the prey (Provost et al., 2006), temperature (Al-Zyoud & Sengonca, 2004), learning ability and memory (Boivin et al., 2010), morphological character and previous feeding experience (Rahim Khan & Rafique Khan, 2002), predator and prey size (Thompson, 1975), abundance of prey (Soares et al., 2004) and genetic and environmental factors (Fujiyama & Katakura, 2001).

The melon aphid, *A. gossypii* Glover, 1877 and the black bean aphid, *A. fabae* Scopoli, 1763 are major insect pests of various crops in Iran. One of their common natural enemies is the aphidophagous ladybird *P. quatuordecimpunctata* (Coccinellidae), which is native and widespread in the Palearctic region (Pervez & Omkar, 2011). It is polymorphic, ranging in colour from cream to yellow to light orange and the 14 spots on the elytra may vary in shape and size (Pervez & Omkar, 2011). Hodek & Honek (1996) have recorded *P. quatuordecimpunctata* feeding upon nearly 20 species of aphids, which indicates that it is a polyphagous species (Kalushkov & Hodek, 2005). *P. quatuordecimpunctata* consumes a greater number of nymphs of *Macrosiphum albifrons* (Essig) (Aphididae) than either *Harmonia axyridis* (Pallas) or *Coccinella septempunctata* L. (Finlayson et al., 2010).

Alborz Mountains are located in northern Iran, adjacent to the southern margin of the Caspian Sea (Vernant et al., 2004). Its northern and southern boundaries, (Caspian Sea and Central Iran, respectively) differ in terms of temperature, humidity, altitude, geography and climate. The Alborz range of mountains is situated between Karaj and Sari (Alborz and Mazandaran Provinces, respectively). The purpose of this study was to determine the preferences of female *P. quatuordecimpunctata* from populations in Karaj and Sari for feeding on *A. gossypii* and *A. fabae* when kept under the same climatic conditions.

MATERIAL AND METHODS

Plants

Cucumber plants (*Cucumis sativus*) were reared in plastic pots (13 cm diameter, 10 cm high) in a climate-controlled room in a greenhouse kept at 25 ± 5°C, 50–70% RH and a 16L : 8D photoperiod. One-month-old plants with five leaves were used for rearing *A. gossypii*.

Broad bean plants (*Vicia faba*) were grown under the same greenhouse conditions in similar plastic pots. Plants that were approximately 8 cm tall were used for rearing *A. fabae*.

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Sources of aphids and predators

The two species of aphids used in this experiment were collected from a farm in the Karaj region, Alborz Province, Iran in June 2013. A colony of *Aphis fabae* was cultured on broad bean, *V. faba*, and *A. gossypii* on cucumber, *C. sativus* in a laboratory of the College of Science at the University of Tehran for 6 months. They were kept in a germinator at 23 ± 2°C, 55 ± 5% RH and under a 16L : 8D photoperiod. The ladybirds were collected as adults from alfalfa farms in Karaj City, Alborz Province (35°48′04.6″N, 50°57′39.6″E, at an altitude of 1315 m) in June 2013 and Sari City, Mazandaran Province (36°39′17.7″N, 53°10′21.5″E, at an altitude of 0 m), in August 2013, respectively. The colonies of *P. quatuordecimpunctata* were reared on *A. fabae* and *A. gossypii*. They were maintained in a germinator kept at 23 ± 2°C, 55 ± 5% RH and under a 16L : 8D photoperiod for one month before being used in the preference experiments. In order to avoid cannibalism, four larvae were placed in each Petri dish along with either *A. fabae* or *A. gossypii*. They were reared under same experimental conditions as described above.

Preference experiments

No choice test

Before evaluating their preference, equal numbers of each type of prey (n = 20) (4 to 5 days old) was placed on leaves of broad bean, which were individually placed in the center of Petri dishes (8-cm diameter) containing 2% agar. Subsequently, an adult female of the predator (5–6 days after emergence) was released in each arena and allowed to feed.

The unconsumed aphids were counted every 3 h. Eaten aphids were immediately replaced by additional aphids of the same species. There were 20 replicates of each experiment.

Preference test

A preference test was carried out using adults of *P. quatuordecimpunctata* from two populations. The aim of this test was to determine their preferred prey and whether this differed for the two populations studied.

For the choice preference trials, according to the previous experiment, the number of aphids offered was 70% of the potential consumption of each predator. In each replicate, aphids were individually offered on broad bean disks at a ratio of 40 : 40 3rd instar nymphs of each species (*A. fabae* : *A. gossypii*) to females of *P. quatuordecimpunctata* from the Karaj population and a ratio of 45 : 45 3rd instar nymphs of each species (*A. fabae* : *A. gossypii*) to females of *P. quatuordecimpunctata* from the Sari population. Each treatment was replicated 20 times. After 24 h, the number of individuals of each prey item consumed was recorded.

To determine the relationship between the number of prey consumed by *P. quatuordecimpunctata* and the body size of the prey the length of the prey was measured to an accuracy of 0.01 mm under a stereo microscope equipped with an ocular micrometer.

Statistical analysis

In order to determine whether each population of predator preferred *A. fabae* or *A. gossypii*, Manly's preference index (β) was calculated based on the number of prey consumed (Manly, 1974):

$$\beta = \frac{\log \bar{P}_i}{\sum_{j=1}^{m} \log \bar{P}_j}$$

where β = Manly's β for the two types of prey (*A. fabae*, *A. gossypii*) i; P_i = proportion of prey *I* remaining at the end of the experiment relative to the number at the beginning (i = 1, 2, 3, 4,…, m); P_j = proportion of all types of prey remaining at the end of the experiment relative to the number at the beginning (j = 1, 2, 3, 4,…, m); and m = number of different kinds of prey, Manly's β can vary between zero and unity. For two prey, *A. fabae* and *A. gossypii* (m = 2), a value of 0.5 represents no preference, a value larger than 0.5 indicates a preference for prey *A* and smaller than 0.5 indicates a preference for prey *B*. This method takes into account the depletion in prey density due to predation during the experiment (Sherratt & Harvey, 1993). Results for the different treatments were compared using Student's t-tests or Mann-Whitney U tests.

RESULTS

In the first experiment, *A. fabae* and *A. gossypii* were offered separately to adult females of *P. quatuordecimpunctata*. In 24 h, those from the Karaj population consumed (mean ± SE): 34.70 ± 2.029 and 23.00 ± 0.423, and those from the Sari population: 44.05 ± 1.037 and 22.15 ± 1.139, respectively, of the *A. gossypii* and *A. fabae* offered. In 24 h, *P. quatuordecimpunctata* females from the Karaj population consumed significantly more 3rd instar nymphs of *A. gossypii* than of *A. fabae* (t = –5.645, df = 20.649, P = 0.0001). In addition, female from the Sari population showed a stronger tendency to feed on 3rd instar nymphs of *A. gossypii* than of *A. fabae* (Mann-Whitney, U = –5.431, P = 0.0001).

In the second experiment the greater consumption of *A. gossypii* by female *P. quatuordecimpunctata* from the two populations indicates a significant preference for this prey, with those from the Karaj population consuming 28.15 ± 1.04 and from the Sari population 34.35 ± 0.51, respectively, which are greater than the numbers of *A. fabae* consumed by *P. quatuordecimpunctata* from the Karaj population, 17.95 ± 0.64, and Sari population, 17.7 ± 1.01, respectively. Manly's β index for *A. gossypii* is 0.73 ± 0.01 and 0.77 ± 0.01 for the Karaj and Sari populations, respectively. Manly's β index (β) for *A. fabae* is 0.26 ± 0.01 and 0.22 ± 0.01 for the Karaj and Sari populations, respectively. Comparing the preference indices using t-tests indicate a significant preference of the predator from both the Sari (t = 22.53, df = 38, $P < 0.0001$) and Karaj populations (t = 26, df = 38, $P < 0.0001$) for *A. gossypii*. These results indicate that *P. quatuordecimpunctata* from both populations prefer *A. gossypii* over *A. fabae* (Fig. 1A and B).

DISCUSSION

Prey items differ in nutritional content, energy content and costs associated with capture and consumption; therefore predators should select the most suitable (Hodek 1993; Hodek & Honek, 1996; Roger et al., 2000; Soares et al., 2004). Suitability of aphids as food depends on various factors such as the physiological status of the host plant, the aphid morph and the food requirements, enzyme equipment and nutritional budgets required for the development and reproduction of the ladybirds (Klingauf, 1988; Srivastava, 1988; Dixon, 1998; Soares et al., 2004). In addition, the number of prey ingested depends on morphological, physiological and nutritional requirements of the predators (Eubanks & Denno, 2000). Ladybirds consume different quantities of different aphids because they differ in morphology, behaviour and chemical composition. (Okamoto, 1966; Liepert & Dettner, 1996; Dixon, 2000).

Biological features such as the distribution and diversity of prey, season, weather conditions and the use of different agricultural methods may affect plant growth and change the prey preference of ladybirds in the field (Omkar & Mishra, 2005). The results of preference experiments indicate that females of *P. quatuordecimpunctata* from both populations eat both *A. gossypii* and *A. fabae*, but the Manly's β indices (β) of females from both populations for *A. gossypii* were significantly greater than for *A. fabae*. This preference for particular species of prey may mean

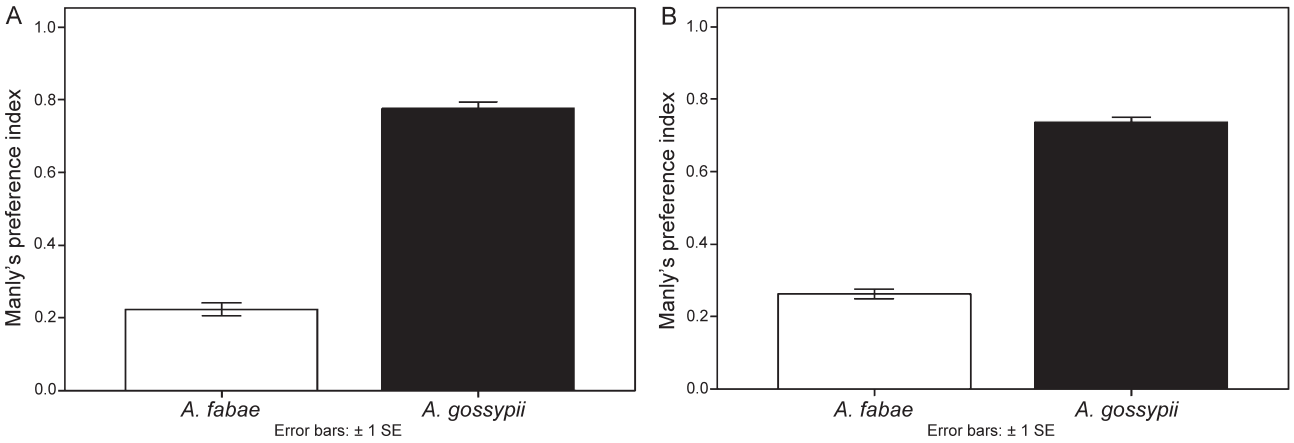

Fig. 1. The Manly's preference index for *Propylea quatuordecimpunctata* from the Sari (A) and Karaj populations (B) when offered equal numbers of 3rd instar nymphs of two species of aphids.

that predators feed on particular species of prey independently of their abundance or accessibility. Females from both populations significantly preferred eating *A. gossypii* to *A. fabae*. As predicted by optimal foraging theory, animals tend to hunt efficiently for food (Soares et al., 2004). Predators feed on the different types of prey available so as to maximize the nutritional gain while minimizing the costs and risks associated with predation, thus, when a predator encounters two types of prey it selects the one most likely to maximize its net energy gain (Stephens & Krebs, 1986).

Large individuals of prey contain more energy than small prey; therefore predators should prefer large prey (Soares et al., 2004). In our experiments, we used two species of aphids (4–5 days old) that differed in size (*A. gossypii* = 1.12 ± 0.01 mm, *A. fabae* = 1.55 ± 0.02 mm). That is the *A. gossypii* were smaller than the *A. fabae*. If size was the only factor involved in prey preference, *A. fabae* would have been the preferred prey. In our experiments, *A. gossypii* appeared to move less than *A. fabae.* High prey mobility generally reduces the encounter rate, increases prey handling time, reduces attack success and results in lower prey profitability (Crawley, 1992). Physical structures such as thickness of the cuticle, length of appendages and overall body size may affect the acceptance of particular prey by ladybirds (Nedved & Salvucci, 2008). An easy way of avoiding being eaten is to be toxic, unpalatable and defended chemically. Siphuncular wax is the most obvious chemical defence of many aphids against predators, including ladybird larvae (Pasteels, 2007). The two species of aphid used in our experiments differed in terms of their physical structure, size, chemical defence, behaviour and ability to escape capture. Each of these features can affect the selection of particular prey by ladybirds in the laboratory. There is now a need to determine whether and to what extent the physical structure, chemical defence, behaviour and ability of the prey to escape capture determined *P. quatuordecimpunctata* preference for *A. gossypii* over *A. fabae* in this study.

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