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Effect of Three Species of Predatory Ladybirds on Oviposition of Aphid Parasitoids

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Abstract. The effect of three species of aphidophagous ladybirds, *Coccinella septempunctata*, *Harmonia axyridis*, and *Propylea japonica*, on oviposition of an aphid parasitoid, *Aphidius colemani*, was assessed in the bean aphid, *Aphis craccivora*, in relation to physical presence, visual cues or larval tracks of ladybirds. Female parasitoids responded to the presence of a ladybird larva by reducing the number of eggs laid in the patch, but visual cues of larvae did not affect the oviposition. Effect of larval tracks on parasitism was selective among species; *C. septempunctata* larval tracks significantly reduced parasitism, but the same was not true for the larval tracks of *H. axyridis* and *P. japonica*. As parasitized aphids are also at risk of predation by predatory ladybirds, therefore, it is advantageous for female parasitoids to avoid aphid patches which contain these predators. Significance of such an oviposition response is also discussed.

Key words: Aphid parasitoids, oviposition, predatory ladybirds.

Introduction

Several species of predatory and/or parasitoid arthropods are often attracted to a number of plants with aphid colonies and, thus, aphidophagous arthropod guilds are constructed in the field (e.g. Sakuratani, 1977; Aalbersberg *et al.*, 1988; Winder *et al.*, 1994; Wyss, 1995). Recently, incidences of intraguild predation (IGP), which is the killing and eating of species that use similar resources and are potential competitors (Polis *et al.*, 1989; Polis & Holt, 1992), have been widely reported in aphidophagous arthropods (Rosenheim *et al.*, 1993; Cisneros & Rosenheim, 1997; Yasuda & Shinya, 1997; Ferguson & Stiling, 1996; Agarwala *et al.*, 1998; Lucas *et al.*, 1998; Yasuda & Ohnuma, 1999; Yasuda & Kimura, 2000). In addition, studies have shown that aphidophagous insects like ladybirds, lacewings, and nabid and mirid bugs readily eat aphids parasitized by wasps (Wheeler *et al.*, 1968; Wheeler 1974, 1977; Takizawa *et al.*, 2000). Under the circumstance, it might be expected that female parasitoids should avoid oviposition in aphid patches which contain predators or were recently visited by them. Taylor *et al.* (1998) revealed that female parasitoids spent less time in aphid patches that included predatory ladybirds. Despite this, the observed proportions of ovipositions were found to be highest in patches with adult predators. This showed that the responses by parasitoids to an aphidophagous ladybird may not be simple and further experiments are needed to understand the oviposition behaviour of parasitoids.

Female aphidiid parasitoids of several species are capable of using honeydew and/or aphid-related volatiles as cues for assessment of the number of aphids (Bouchard & Cloutier, 1985; Budenberg, 1990; Shaltiel & Ayal, 1998). Battaglia *et al.* (1995) found that visual cues were also important in the host recognition of aphidiid parasitoids. Therefore, it is predicted that female parasitoids might use these capabilities in selecting suitable aphid patches for oviposition which are safe from predators.

In this study we tested this prediction by assessing the effect of three species of predatory ladybirds on the oviposition of an aphidiid parasitoid, *Aphidius colemani*, in relation to physical presence of a predator or presence of larval volatile and visual cues in aphid patches.

Materials and Methods

The bean aphid, *Aphis craccivora*, and adults of the three ladybirds, *Harmonia axyridis*, *Coccinella septem-*

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Table 1. Number (mean±SE) of non-parasitized and dead aphids due to unknown factors when ten aphids were supplied in each treatment.

Treatment	Non-parasitized	Dead	N
Control (1)*	4.4±0.6	1.8±0.2	20
H. axyridis			
Larva in a tube	5.6±0.6	1.2±0.1	20
Larva removed	5.9±0.4	1.5±0.2	38
Larva	6.3±0.5	1.2±0.3	34
C. septempunctata			
Larva in a tube	4.4±0.4	1.4±0.3	20
Larva removed	6.0±0.4	1.5±0.2	41
Larva	5.8±0.4	2.0±0.2	32
P. japonica			
Larva in a tube	4.7±0.6	1.3±0.2	19
Larva removed	5.9±0.4	1.1±0.2	44
Larva	6.3±0.4	1.7±0.2	28
Control (2) without a parasitoid	8.4±0.2	1.6±0.2	24
A tube without a ladybird larva	4.6±0.6	1.6±0.3	17

* A parasitoid and ten aphids only in Petri dishes; details provided in Materials and Methods.

punctata, and *Propylea japonica*, were collected from several species of plants in fields at the Yamagata University. Adult ladybirds were fed on bean aphids in plastic containers (15 cm in diameter and 9 cm in height) and eggs laid by the females were collected every day. Egg batches were kept individually into 9 cm Petri dishes and larvae reared to the fourth instar on bean aphids. Pupae of the parasitoid, *A. colemani*, were obtained from Koppert B. V., the Netherlands, through Tomen Corporation, Japan.

In the experiments, an apical twig of a bean plant, *Vicia faba*, infested with ten second or third instar aphids was taken in a 9 cm Petri dish that was lined with a filter paper. The cut end of the twig was applied with cotton and kept wet during the experiment. Aphids were exposed to a 2-day-old previously mated female parasitoid for 30 minutes. Parasitism of aphids was determined after 7 days by dissecting the aphids for larvae of parasitoids. The experiments consisted of six treatments: (1) a parasitoid, (2) a parasitoid + a filter paper containing larval tracks, (3) a parasitoid + a glass capillary with an enclosed ladybird larva, (4) a parasitoid + a filter paper containing larval tracks + a ladybird larva, (5) no parasitoid, (6) a parasitoid + a glass capillary without a ladybird larva. Treatment (5) was designed to show mortality of aphids, if any, without parasitism for 7 days. Treatment (6) was included in the experiment to examine the effect of a glass capillary on oviposition by parasitoids.

Fourth instar larvae developed on excess supply of aphids and preparing for prepupation were used as predators in this experiment. These larvae did not eat aphids during the experiments. Filter papers containing larval tracks of the ladybirds were produced by releasing an active fourth instar larva into a 9 cm Petri dish lined with a filter paper for 24 hours. Larvae were kept without food. A glass capillary (0.9 cm in diameter and 3.6 cm in length) enclosing a larva with an end corked was used in order to evaluate the role of visual cues of the predator. Replications of different treatments were shown in Table 1.

All the treatments of the experiment were done at a constant temperature of 20±1.0℃ and a photoperiod of 16L : 8D, but rearing of insects was done in an incubator at 25±1.0℃ and 16L : 8D photoperiod. Means of parasitism in the treatments were compared with the control by Dunnett multiple comparison test. Data of parasitism were arcsine-square-root transformed before analysis.

Results

The number of aphids that died due to unknown factors during 7 days of the experiment was less than two through all treatments, and there was no difference in the mortality between treatments either with or without a parasitoid (Kruskal–Wallis, $H_{cor}=18.38$, $P>0.05$; Table 1). In the control (1), which included a parasitoid and ten aphids, 46% of aphids were parasitized (Fig. 1). There were also no significant differences in the extent of parasitism between the control and the treatments where a larva was presented in a glass capillary (Dunnett test, for *H. axyridis*, $d=\text{Abs (Dif)}-\text{LSD}=-0.14$, $P>0.05$; for *C. septempunctata*, $d=-0.22$, $P>0.05$; for *P. japonica*, $d=-0.22$, $P>0.05$; Fig. 1). The parasitoids exhibited similar responses to the three species of ladybirds in terms of visual cues (ANOVA, $F=0.88$, $P>0.05$) which suggested that the parasitoids did not recognize or appreciate the visual cues. No effect of using the glass capillary on oviposition by the parasitoids was observed as there was no difference in parasitism between the control and the treatment without a larva in the tube (ANOVA, $F=0.17$, $P>0.05$; Table 1). However, parasitism was significantly lower in treatments with a predator as compared to the control. This was found to be true for the three species of ladybirds tested in this study (Dunnett test, for *H.*

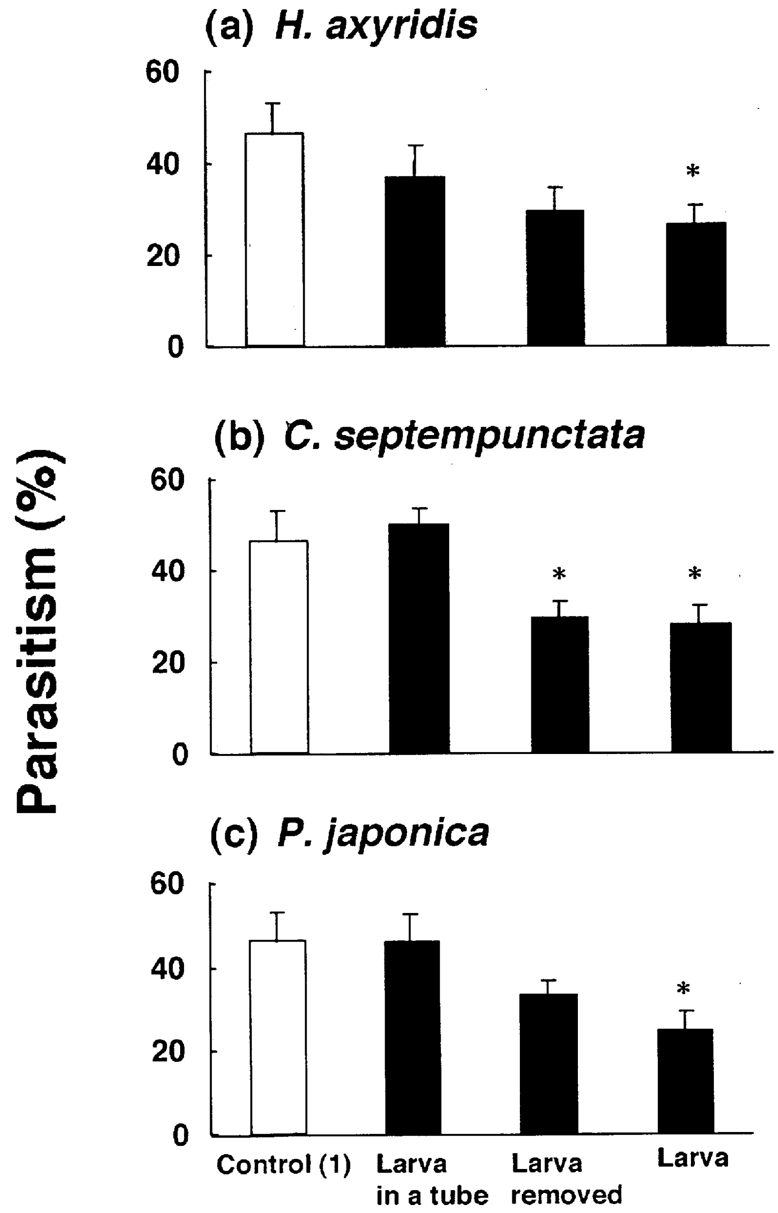

Fig. 1. Aphids parasitized (mean±SE) by *A. colemani* in four treatments. Means were compared by Dunnett multiple comparison test. Bars with an asterisk show significant difference with the control (Dunnett test, $P<0.05$).

axyridis, $d=0.01$, $P<0.05$; for *C. septempunctata*, $d=0.02$, $P<0.05$; for *P. japonica*, $d=0.05$, $P<0.05$; Fig. 1). Effect of larval tracks on parasitism was not uniform among species; in *C. septempunctata*, larval tracks significantly reduced parasitism (Dunnett test, for *C. septempunctata*, $d=0.01$, $P<0.05$), but not in *H. axyridis* and *P. japonica* where the differences in parasitism between the control and treatments were not significant (Dunnett test, for *H. axyridis*, $d=-0.03$, $P>0.05$; for *C. septempunctata*, $d=-0.03$, $P>0.05$).

Discussion

This study revealed that female parasitoids of *A. colemani* have the abilities to recognize aphid patches which contained a ladybird larva or contained chemical tracks left by the *C. septempunctata* larva.

It is well known that aphidiid parasitoids are able to use a very wide variety of chemical stimuli in finding their hosts. For instance, honeydew is used by several species of aphid parasitoids as a cue for assessment of the number of aphids in a colony (Bouchard & Cloutier, 1985; Budenberg, 1990; Shaltiel & Ayal, 1998), and aphid-related volatiles involved in host locations are also detectable by wasps (Kitt & Keller, 1998). In addition, aphid parasitoids also utilize visual cues in host recognition (Battaglia *et al.*, 1995). In view of these results, it is reasonable to assume that some parasitoids might perceive chemical or visual cues caused by their predators and/or intraguild predators in order to regulate their oviposition.

Taylor *et al.* (1998) showed that an aphid parasitoid responded to the presence or recent presence of a predator by reducing the time it spent in a patch. Our results demonstrated that parasitoids responded to presence of a ladybird larva by reducing the number of eggs laid in such aphid patches; this might be attributable to reduced residence time in a patch. Visual cues of predators failed to elicit similar response in oviposition even though the larvae of three ladybird species used in this study had different colour patterns on the dorsal surface of their abdomen. This suggested that visual cues were not effective in location of suitable aphid patches for oviposition in this parasitoid species. In terms of chemical cues, although the parasitoid generally reduced oviposition in all treatments with larval tracks of ladybirds, the reduction was found to be significant in *C. septempunctata* only. Larval tracks of ladybirds are known to elicit selective responses in aphids; green and red clones of the pea aphid, *Acyrthosiphum pisum*, responded by producing higher proportions of winged forms but not by the black bean aphid, *A. fabae* and vetch aphid, *Megoura viciae* (Dixon & Agarwala, 1999). Such a difference in responses among aphid species depended on the degree of risks of predation from ladybirds. Similar relationship between parasitoids and predatory ladybirds of aphids is not denied in the present context.

In addition, the outcome of IGP depends on relative risks of predation among the interacting species of predators (Rosenheim *et al.*, 1993; Cisneros & Rosenheim 1997; Yasuda & Shinya, 1997; Ferguson & Stiling 1996; Lucas *et al.*, 1998; Yasuda & Ohnuma, 1999; Yasuda & Kimura, 2000). It has been shown that generalist predators usually dominate over specialist predators (*e.g.*, Ninomiya, 1968; Lucas *et al.*, 1998). As a result, specialist predators like hoverflies and wax producing coccinellids forage in patches with low prey densities which usually do not attract generalist predators like neuropterans and many aphidophagous ladybirds (*e.g.*, Kan & Sasakawa, 1986; Kan, 1988; Agarwala unpublished). This might be also done in aphid parasitoids as parasitized aphids are vulnerable to predation by ladybird larvae (Takizawa *et al.*, 2000). By reducing oviposition in aphid patches that contained ladybird larvae or larval tracks, in some species, female parasitoids reduce risks of predation of its larvae and, therefore, increase chances of oviposition in predator-free aphid patches.

The aphidiid parasitoid, *A. colemani*, is a commercialized parasitoid, and is widely used to reduce aphid abundance (e.g. Grasswitz, 1998). Chances of success in biological control of aphids by introduced parasitoid species may improve if inter-specific relationships such as competition, predation or IGP between introduced and indigenous species are clearly understood. Of these interactions in aphidophagous insects in relation to aphid parasitoids, ant and parasitoid relationships have been reported. For instance, there were negative effects of ants on survival of aphid parasitoids (Volkl & Kroupa, 1997) and species specific ant-parasitoid interactions (Volkl & Mackauer, 1993). In addition, IGP by aphidophagous insects to aphid parasitoids is now well established (*e.g.*, Muller & Godfray, 1999). However, effect of aphidophagous insects on oviposition behaviour of parasitoids is still poorly understood. Therefore, future studies are needed to generalize the phenomenon.

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