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Aphid aqueous-extract as a source of host searching kairomones for the aphidophagous predator *Coccinella septempunctata* L. (Col., Coccinellidae)

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Abstract

Adult Coccinella septempunctata, a coccinellid predator of the bean aphid Aphis fabae Scop., responded to aqueous extract containing kairomones of the prey. The crude aphidextract was topically dropped onto either clean leaves of Vicia faba or clean Petri-dishes. After application of aqueous-extract at different aphid densities, the predator showed variations in the attractivity and searching behaviour in comparison with control. The variations are summarized as follows: (1) the aqueous-extract of aphids attract or direct C. septempunctata adults to the treated vials at all aphid densities. The number of attracted or directed coccinellid adults was directly proportional with aphid concentrations; (2) the predator C. septempunctata adult intensified its searching behaviour by increasing the total path length walked; (3) the number of turns exhibited is significantly higher as compared with those of control; and (4) the arrested time spent by coccinellid adults with treated plant is much more than that with clean plant at all aphid densities. The present results show that the aphid aqueous-extract contains the active ingredients (kairomone) which induce considerable changes in the searching pattern of the predator coccinellid adults.

1 Introduction

There have been numerous studies of kairomones as stated by BROWN et al. (1970). Earlier investigations have shown that the behaviour of Trioxys indicus may be modified greatly by the water soluble kairomones present in the haemolymph of its host (SINGH et al., 1979; SINGH and SRIVASTAVA, 1987; SRIVASTAVA and SINGH, 1988). The same has been found in other parasitoids species (LEWIS et al., 1975; WESELOH, 1977; LEWIS and NORDLUND, 1984). NORDLUND et al. (1981) have considered that kairomones can be used to manipulate the host searching behaviour of the parasitoids in order to maintain their active populations in the target area, particularly early in the season. Therefore, kairomones have been reported to have powerful biological activity and offer great potential as effective and environmentally sound insect pest management tools (NORDLUND, 1984). SHONOUDA (1996, 1998a,b) found that the aphid-kairomone could be extracted from the odour and the body of Aphis fabae by using the solvent n-pentane. The n-pentane fractions showed kairomonal effect on the searching pattern of the syrphid females and coccinellid adults.

The present study suggests that the aqueous-extract of *A. fabae* contains the prey-seeking stimulants (kairomones), hence, the effect of aphid-extract on the behaviour of *C. septempunctata* adults at different densities of its prey *A. fabae* were studied in the laboratory.

2 Material and methods

The bean aphid *A. fabae* and its predator *C. septempunctata* were reared as shown by SHONOUDA (1996). The coccinellid adults were starved for 24 h, before use in biological experiments.

Different initial numbers, that is 10, 50, 100 and 200 individuals of aphids were homogenized in 2 ml distilled water and centrifuged. The supernatants were used as the different concentrations of aphid extracts, given the symbols AC1, AC2, AC3 and AC4, respectively. The first experiment was designed to show the effect of different concentrations of aphid extracts, in addition to distilled water as control on the attractivity of the coccinellid adults. The experiment was conducted by preparing 5 vials with growing clean plants of V. faba. They introduced into a glass box ($70 \times 30 \times 20$) with wire mesh lid. The first vial was spraved with distilled water, as control, while the other four vials were sprayed with the different concentrations of aphid extract. respectively. The 5 vials were sprayed with a constant volume of water (20 ml) and were left until the water dried. After that 60 starved coccinellid adults was put inside the glass box and were left for 2 h. Five replicates were performed. At the end of experiment, the number of coccinellid adults were counted on either treated vials or control vial.

The second experiment was conducted by preparing 5 Petridishes with clean plant-cuttings of V. faba. By using a micropipette, 200 µl distilled water were dropped on the plant-cuttings in the first dish as control. while 200 µl of each of the 4 concentrations of aphid extract were dropped on the plant-cuttings in the other 4 dishes, respectively. One starved coccinellid adult was introduced into each dish. In this experiment the walking pattern of the coccinellid adult was traced for 60 sec on the covered dish by using a black pen. Also, the number of turnsexhibited by the coccinellid adult was recorded during the same time. The walking pattern was measured by a map counter and was transformed into centimeters. Ten replicates were performed.

The third experiment was conducted by preparing a glass Petri-dish divided into two equal halves by lining with a black pen. A clean plant-cutting was placed in each half. 200 µl of the first concentration of aphid-extract (AC_1) were dropped on the clean plant-cutting in one half while the plant-cutting in the other half was left clean. A control glass Petri-dish was prepared in the same manner with 200 µl distilled water dropped in one half instead of the aphid-extract. One coccinellid adult was released at the middle line and was allowed to walk for 20 min. The arrestment time (in min) spent by the coccinellid adult in each half was recorded by using a stop watch. Ten replicates were performed. The experiment was repeated using all different concentrations of aphid extract and utilizing different coccinellid adults.

All the experiments were conducted under laboratory conditions of 25 ± 5 °C and 40–50 % R. H. Wilcoxon's signed rank test and unpaired t-test were applied to test the significance of difference between the values of the different experiments.



Fig. 1. The percentage of numbers of *C. septempunctata* adults directed or attracted to the different concentrations of aphidextract (AC), in addition to control

3 Results and discussion

By applying the different concentrations of aqueous extract from A. fabae, the predator C. septempunctata adult showed different responses. In the first experiment, the coccinellid adults responded to the supernatant of the aqueous extract of aphids by increasing the number of coccinellid adults which attract or direct towards the treated plants. The number of coccinellid adults is directly proportional to the concentrations of the aphid extract (fig. 1). The mean numbers of attracted coccinellid adults to AC_4 (x[\] = 24.5), AC_3 (x[\] = 15.5), AC_2 (x[\] = 11) and AC_1 $(x^{\setminus} = 5)$ were higher than the control $(x^{\setminus} = 4)$. The percentages of attracted adults to each concentration of aphid extract were 40.8 %, 25.8 %, 18.4 % and 8.3 %, while the percentage of coccinellid adult attracted to distilled water as control was lower and equal to 6.7 %. The results suggested that the aqueous fractions of aphid extract contain the chemical cues (kairomones) which stimulate and direct the predator coccinellid adults to the treated plants.

The second experiment showed that the coccinellid adults moved vigorously under the effect of different concentrations of aphid extract as compared with the control. The mean path length walked by coccinellid adults at the 4 concentrations of aphid extract were longer than that with the control (fig. 2). The path lengths walked by coccinellid adults under the effect of AC_4 (t = 3.2, p < 0.01), AC₃ (t=2.9, p<0.01) and AC₂ (t = 2.08, p<0.05) were significantly higher than that in the control, while the path length walked under the effect of AC_1 (t = 0.4, p > 0.05) was insignificantly high when compared with control. Also, the turning rate of coccinellid adults was affected by the different concentrations of aphid extract. The mean number of turns exhibited by coccinellid adult with the 4 concentrations of aphid extract were higher than that in the control (fig. 2). The number of turns walked by coccinellid adults under the effect of AC_4 (t = 2.5, p < 0.01), AC₃ (t = 2.3, p < 0.05) and AC₂ (t = 2.0, p < 0.05) were significantly higher than that in the control, while the number of turns exhibited under the effect of AC1 (t=0.8, p>0.05) was insignificantly high as compared to the control. It is clearly obvious that the different concentrations of aphid extract had their effect on the search-



Fig. 2. The mean path length (cm) and the mean turning rate (turns/min) exhibited by C. septempunctata under the effect of different concentrations of aphid-extract (AC) in addition to control.

With hatching: path length; without hatching: turning rate

ing behaviour of coccinellid adults such as the path length and the number of turns exhibited. The changes in the searching behaviour of the different predators and parasites of aphids had been observed by CARTER and DIXON (1982) on the coccinellid larvae and by BOUCHARD and CLOUTIER (1984) on the aphid parasitoid *Aphidius nigripes*. They found that klinokinetic and orthokinetic behaviour of insects were affected by decreasing the walking speed and increasing the turning rate after encounter of the aphid kairomone or aphid host itself.

Concerning the third experiment, the aphid extract retained the coccinellid adults much longer than the control. The mean of time spent by a group of coccinellid adults within half Petri-dishes treated with different concentrations of aphid extract were much more than that with clean plant cuttings in the other halves (fig. 3). The mean differences of the time spent with AC4 (t = 3.9, p < 0.01), AC₃ (t = 3.6, p < 0.01) and AC₂ (t = 2.2, p < 0.05) were significantly higher than that with the clean plant. While, the mean of time spent by coccinellid adults within half Petri-dishes treated with either AC₁ or with distilled water (control dish) were more-or-less similar to



Fig. 3. The mean of time arrested by *C. septempunctata* adults inside the half Petri-dish treated with aphid-extract (AC) or distilled water (control)

those with the clean plant-cuttings in the other halves. The mean differences of the time spent showed no significant change with either AC₁ or with the control (p > 0.05). SRIVASTAVA and SINGH (1988) found that the parasitic females of *Trioxys indicus* respond to aqueous extract from its host *Aphis craccivora* by showing the following responses: (i) the host searching time significantly decreased curvilinearly; (ii) the frequency of host contact, prick and oviposition increased exponentially and (iii) the retention period with the hosts significantly increased linearly.

SHONOUDA et al. (1996, 1998a,b) found that the n-pentane fractions of the extract from Aphis fabae contain the kairomonal stimulants which increase significantly the oath length, number of turns and the spent time exhibited by either the predator M. corollae female or the coccinellid adult. Furthermore, the present results suggested that there are different kairomones that could be extracted from aphids and one of these kairomones was extracted by the aqueous solution. Also, the present results clearly show that the aqueous extract of A. fabae contains the chemical cues (kairomones) which affect and maniplate the behaviour patterns of the predator C. septemvunctata adults. Further studies on the active ingredients of the aqueous extract of aphids may furnish information about the role of kairomones in aphid biological control n the course of insect management programs.

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