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RESEARCH PAPER

The use of actometry and electronic nose devices to assess the locomotor activity of two species of Coccinellidae (Coleoptera)

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Abstract

G. Heit, V. Messina, G. Mareggiani, R. Cohen, and P. Carrizo. The use of actometry and electronic nose devices to assess the locomotor activity of two species of Coccinellidae (Coleoptera). Cien. Inv. Agr. 38(2):265-275. The effect of odor signals on the locomotor activity of Cycloneda sanguinea and Harmonia axyridis was evaluated using two different actometric devices. The purpose of the analysis was to differentiate between the responses elicited by direct contact with the substrates and the responses that occurred if the coccinellids were exposed only to volatiles and had no direct substrate contact. The odor profile of the environmental volatiles (semiochemicals) in the bioassays was analyzed using an electronic nose (Modular Sensor System Moses II). The treatments used included volatiles emitted by pepper leaves (healthy or infested with Myzus persicae). M. persicae molts and honeydew, and healthy leaves treated with an artificial inducer of the systemic acquired response to herbivory (AISAR). The results suggest that a specific stimulus linked to aphid-host interaction would trigger a distinctive locomotor activity pattern in H. axvridis, with or without any contact with the aphids or the host plant. In contrast, C. sanguinea was not able to differentiate between volatiles from infested or uninfected pepper leaves or to distinguish these volatiles from clean filter paper (control). If C. sanguinea was in contact with aphid-infested pepper leaves (regardless of the previous prey density), it moved slowly and scanned the area exhaustively; H. axvridis showed significantly reduced locomotor activity when it was in contact with aphid-infested pepper leaves or aphid molts and honeydew, all reliable signals of the presence of a food source. In both species, treatment with Actigard (AISAR: Acibenzolar-S-methyl) failed to evoke the behavioral response observed in the treatment with aphid-infested pepper plants. The study also demonstrated that the E-nose can differentiate among blends of volatiles released from cut healthy or aphid-infested pepper leaves.

Key words: Acibenzolar-S-methyl, Coccinellidae, electronic nose, locomotor activity, semiochemicals.

Introduction

As important members of a guild of arthropods that prey on agricultural pest species, coccinellids can be used against aphids to provide a significant degree of biological control (Dixon, 2000; Kehrli and Wyss, 2001). In fact, they are widely used in integrated pest management programs designed to maintain aphid populations below economic damage thresholds (Dixon, 2000).

The predatory efficiency of coccinellids is determined by their search strategies. Adult ladybirds are highly capable of tracking aphid popu-

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lations in space and time (Osawa, 2000); in fact, the peak in the arrival of many coccinellids at a prey patch generally occurs before or just at the density peak of the aphid population (Evans, 2003). This observation suggests that the search pattern and ovipositing behavior of adult coccinellids could be associated with semiochemicals. The use of chemical signals could allow the coccinellids to assess the age of an aphid colony. This ability would be advantageous because the offspring would probably starve to death if the prey colony were waning (Dixon, 2000; Tschanz *et al.*, 2005).

Semiochemicals released from a diverse group of plants and insects mediate key processes in the behavior of insects (Paré and Tumlinson, 1999). Leaves normally release small quantities of volatile chemicals. Plants damaged by herbivorous insects react with the systemic production of several novel compounds and produce a complex volatile blend composed terpenoids, fatty acids, methyl salicylate, and other compounds (Paré and Tumlinson, 1999; Pichersky and Gershenzon, 2002; Arimura et al., 2005). Furthermore, the emission of herbivore-induced plant volatiles (HIPVs) is inducible by ozone and fungal pathogens. Mold infection might even increase the attraction of parasitoids to infected plants (Tumlinson et al., 2003; Vuorinen et al., 2004; Holopainen, 2004).

Compared with chewing insects, aphids are phloem-sap feeders and cause only limited tissue damage. However, they cause the activation of genes involved in defense responses within 2 to 6 hours after they have penetrated plant tissues with their stylets. Through gene activation, they, and trigger the systemic emission of variable blends of de novo-synthesized volatiles (Zhu-Salzman et al., 2004; Couldridge et al., 2007). As a result, induced plant volatiles can cause a damaged plant to appear different from its undamaged neighbors. This difference is detectable by the parasites and the herbivore predators that feed on the plant (Visser, 1986; Turlings et al., 1995; Takabayashi and Dicke, 1996; Dicke and Vet, 1999; Wegener et al., 2000; Van Poecke et al., 2001).

HIPVs have been proposed for use in agricultural pest control because they might repel herbivores

and serve to attract the enemies of the herbivores. ethyl salicylate, appears to be a key factor that can mediate the attraction of several natural enemies of agricultural pests (Farag and Paré, 2002; James, 2003; De Boer and Dicke, 2004). Chemicals such as salicylic and 2,6-dichloro-isonicotinic acids can also act as inducers of systemic resistance in plants (ISR). However, they have not been exploited commercially because of their phytotoxicity to most crops (Romero *et al.*, 2001).

Acibenzolar-S-methyl (ASM - Actigard 50WG), an artificial inducer of systemic acquired resistance to herbivory (AISAR), does not have the disadvantage of phytotoxicity. It activates the same defense responses that are produced by the natural signal molecule, salicylic acid, in the biological activation of induced systemic resistance (Ryals et al., 1994). Shimoda et al. (2002) have reported that the application of artificial inducers of systemic acquired resistance to herbivory in lima bean leaves caused the emission of volatile compounds that attracted spider mites, which are predators. The logical next step in such investigations may be to test whether the application of ASM to healthy pepper plants could produce the same modification of the search behavior of coccinellids that is produced by aphid-infested pepper plants.

Electronic noses (E-noses) represent a practical approach for determining the presence of significant differences between one blend and others. The differences in sensitivity between devices of this type and odorant receptors do not appear to be critical (Berna et al., 2009). Enoses are designed to mimic the performance of animal olfactory organs by using an array of diverse chemical sensors. These sensors concurrently respond to the volatiles present in a gas sample. The main assumption of this approach is that such an array of sensors can obtain a digital fingerprint (odor profile) from the sample headspace (Ridgway et al., 1999). These authors have demonstrated that an E-nose can differentiate between uninfested wheat and wheat infested by mites at levels that are commercially threatening in the cereal industry. Furthermore, McKellar et al. (2005) have demonstrated that an oxide sensor-based E-nose might serve as a useful screening tool for discriminating among

chrysanthemum cultivars having varying degrees of resistance to western flower thrips (*Frankliniella occidentalis*). These discriminations were based solely on the presence of volatile chemicals released from cut leaves.

The aim of this work was to study the locomotor activity of adults of two aphidophagous coccinellids, *Cycloneda sanguinea* Linnaeus and *Harmonia axyridis* Pallas, when exposed to volatiles or in direct contact with different substrates from which the volatiles would be released. *C. sanguinea* is native to Central and South America (Iskber and Copland, 2002), whereas *H. axyridis* is a cosmopolitan species. The latter species was introduced to Argentina by the end of the 1990s and dispersed rapidly throughout the country. It has become an interesting case study of an exotic generalist predator (Koch, 2003, Saini, 2004).

Materials and methods

Rearing of predator and prey. Culture of host plant

Healthy adults of *Cycloneda sanguinea* and *Harmonia axyridis* (Coleoptera: Coccinellidae) were collected in the field and used as parents. Adults of the first generation produced by mass rearing were used in the behavioral bioassays. Both species were fed with nymphs and adults of the green peach aphid, *Myzus persicae* (Hemiptera: Aphididae). The aphids were reared on pepper plants, *Capsicum annuum* CV. California Wonder, growing in 350 cc polyethylene pots. Predators, prey and host plants were maintained under standardized environmental conditions: 24 ± 2 °C, $65 \pm 10\%$ RH, and 16 H photophase (60 watts).

Volatile sources

The following volatile sources were evaluated. I) Health-pl, 3 g of healthy pepper leaves placed on filter paper; II) High-inf, 3 g of high-level aphid-infested pepper leaves placed on filter paper. A high level of infection was obtained by infesting plants with *M. persicae* one month before the bioassays; III) Low-inf, 3 g of low-level aphid-infested pepper leaves placed on fil-

ter paper. A low level of infection was obtained by infesting plants with *M. persicae* one week before the bioassays. IV) Hd&Mo, honeydew and molts of *M. persicae* placed on filter paper. These sources were collected from 3 g of highly infested leaves using a brush and scalpel and placed on filter paper. V) AISAR, 3 g of leaves from healthy pepper plants sprayed with 3 mL of an artificial inducer of systemic acquired resistance (Acibenzolar-S-methyl= Actigard®); and VI) Ctrl, clean filter paper as a control.

Actometry analysis

Standardized environmental conditions of 24 ± 2 °C, $65 \pm 10\%$ RH, and 16 H photophase (60 watts) were used. *C. sanguinea* and *H. axyridis* adults, chosen randomly using a randomized sample and starved for 24 H, were tested. Each bioassay was performed twice a day, in the morning (9-12 H) and in the afternoon (13-16 H), to avoid the possible confounding effect produced by any tendency of different behaviors to be performed at different hours of the day (Omkar *et al.*, 2004). The odor sources evaluated as treatments were those described previously.

Two experimental series were conducted with two different devices.

First series: exposure to the odors of volatile semiochemicals. Individual locomotor activity was evaluated using an actometer (modified from Manrique et al., 2006), as described in Heit et al. (2007). The actometer consisted on two pieces: a) an experimental *arena*, where the insects were placed, and b) a stimulus delivery chamber, where the odour sources were placed. The experimental arena consisted on a transparent 12-cm-diameter-cylindrical box, made of acrylic plastic and composed by two horizontal compartments, of equal height (2 cm), separated by wire mesh. The arena, the upper compartment where the insects were placed had a removable cover on which a squares grid was drawn (1.5 cm ruling). On the wire mesh, individuals were placed on a piece of perforated filter paper. All the insects, paper pieces and leaves were used only once and then discarded. The lower compartment (the chamber used for the dispersal of the volatile compounds) had a hole in its floor. This hole led to the chamber containing the odor sources or substrates (the stimulus delivery chamber). This design was used to prevent direct contact between the insect and the volatile source or substrate. Four min were allowed for acclimatization before the start of the experiment to allow the uniform diffusion of the volatiles after the introduction of each individual into the arena. N=288 replicates for *C. sanguinea* and N=336 replicates for *H. axyridis* were performed for the entire series.

Second series: exposure of individuals to direct contact with the odor source. The experimental arena consisted only of a Petri dish, 9 cm in diameter, with a grid of squares drawn on its cover (1.5 cm). In these tests, each individual was placed in the experimental arena in direct contact with the substrate to be evaluated. In this series of bioassays, the aphids were removed from the infested leaves with a brush. This procedure prevented the predators from stopping to eat the aphids and thus prevented alterations in the search behavior. N=360 replicates for *C. sanguinea* and N=288 replicates for *H. axyridis* were performed.

For both series, the behavioral response, evaluated as the amount of locomotor activity, was estimated by recording the number of grid squares crossed by each individual during the first 60 seconds subsequent to the acclimatization period following its placement in the arena. The bioassays were analyzed using a factorial design in which the sex of the individuals (male or female), the time of the bioassay (morning or afternoon) and the volatile source or substrate (see above) represented the three factors. The data were square-root transformed to satisfy the assumptions of the tests and then analyzed with three-way ANOVA and Tukey multiple comparison tests (α =0.05) (Sokal and Rohlf, 1995; Zar, 1999) by means of InfoStat software (Di Rienzo et al., 2009).

Electronic nose evaluation

A MOSES II (Modular Sensor System) electronic nose was used to analyze the odor profile (Baby *et al.*, 2000). MOSES II contains two gas-sensor modules, one of which consists of eight quartz microbalance sensors (OMB). This type of sensor consists of vibrating quartz crystals covered with polymeric selective coatings on which gases are adsorbed. The initial vibration frequency (v.) of the crystals decreases in response to the mass increase resulting from the adsorption of the gases. The resulting difference between v_{1} and the final frequency (v_{0}) is proportional to the concentration of the adsorbed gas. Eight pure and doped semiconductive SnO₂ sensors comprise the other module of sensors (SnO₂). Doping with different elements increases the selectivity of SnO₂ for different gases. The SnO₂ surface conductivity changes as the semiconductor adsorbs oxidizing or reducing gases (Messina et al., 2009). E-noses generally provides data in the form of two-dimensional plots constructed using the PCA (principal component analysis) statistical method. The PCA algorithm takes advantage of the fact that the sensors are relatively nonspecific by combining the signals of all the sensors to produce a unique signal. This method tends to group similar odors in clusters. In the resulting two-dimensional plot, the axes are the components that make the greatest percentage contribution to the expression of the odor.

Samples of 3 ± 0.005 g for each odor source described were placed in a 10 mL glass vial with a screw cap and silicon septum. The samples were stabilized at 40°C for 10 min (incubation time) in an 86.50 Dani Headspace sampler and introduced into the MOSES II. Synthetic air was employed as a carrier gas, with a flow of 30 mL/min. The data were analyzed using the Infostat (Di Rienzo *et al.*, 2009) Principal Component Analysis. MANOVA (multivariate analysis of variance) was used to cross validate the PCA results.

Results

Actometry analysis

Figure 1 shows the values of locomotor activity observed in *C. sanguinea* adults exposed only to the volatile semiochemicals emitted by the five substrates evaluated and the values for adults in direct contact with the same substrates.



Figure 1. The locomotor activity of *Cycloneda sanguinea* when exposed to volatile semiochemicals or in direct contact with the substrates evaluated in this study. Locomotor activity was estimated by counting the number of squares crossed by the insect during 60 seconds. Health-pl, healthy pepper leaves. High-pl, high-level infested pepper leaves. Low-inf, low-level infested pepper leaves. Hd&Mo, honeydew and molts on filter paper. AISAR, healthy pepper leaves sprayed with Actigard[®]Ctrl, clean filter paper control. Mean locomotor activity and standard error are shown. Different letters indicate significant differences (α =0.05).

No interaction among the factors was observed in the factorial ANOVA (P>0.05). The locomotor activity of C. sanguinea did not differ significantly among the treatments ($F_{5,264,0.05}=0.791$; P=0.557) that involved exposure to the volatile products of the different substrates evaluated. No differences were found between morning and afternoon locomotor activity $(F_{1.264,0.05}=1.537; P=0.216)$ or between the sexes (F_{1.264.0.05}=0.046; P=0.829). However, statistically significant differences among treatments were found ($F_{5,336,0.05}$ =21.807; P \leq 0.0001) for the series in which adults were in direct contact with the substrate. In this series, the average locomotor activity did not vary significantly with time (F_{1.336.0.05}=2.881; P=0.090) or sex $(F_{1,336.0.05}=0.241; P=0.624).$

Owing to the lack of significance of the time and sex variables, all the data were pooled and reanalyzed using one-way ANOVA. This analysis showed that the behavior of *C. sanguinea* adults differed significantly among treatments ($F_{5,354,0.05}$ =27.438; P≤0.0001). The lowest average locomotor activity was recorded in contact with infested pepper leaves (P≤0.05), the highest in the control treatment (filter paper). The average level of locomotor activity observed in contact with healthy leaves, with aphid molts and honeydew, or with healthy leaves sprayed with Actigard[®] did not differ significantly (Figure 1).

The locomotor activity of H. axvridis (Figure 2) exposed only to the volatiles differed significantly among treatments ($F_{5,312,0.05}$ =2.392; P=0.037). However, the results of the Tukev test did not identify the factor(s) responsible for the observed differences. In the presence of direct contact with the substrates, the locomotor activity of theis predator differed significantly among treatments ($F_{5,264,0.05}$ =2.974; P \leq 0.0001). The activity of this species did not differ according to sex (F_{5,312,0.05}=0.498; P=0.480) or time ($F_{5.312.0.05}$ =2.379; P=0.124) during the volatile only-exposure series of bioassays, nor did it differ during the series involving direct contact with the odor source (Fsexes_{1.312.0.05}=2.974; P=0.085; Ftime_{1.264.0.05}=0.654; P=0.419). No interaction among the factors was observed in the factorial ANOVA (P>0.05).

Owing to the lack of significance of the time and sex variables, all data were pooled and reanalyzed using one-way ANOVA. The reanalyzed data showed significant differences among treatments ($F_{5,282,0.05}$ =16.167; P≤0.0001). The highest locomotor activity was recorded



Figure 2. Locomotor activity of *Harmonia axyridis* when exposed to volatile semiochemicals or in direct contact with the evaluated substrates. Locomotor activity was estimated by counting the number of squares crossed by the insect during 60 seconds. References. Health-pl, healthy pepper leaves. High-pl, high-level infested pepper leaves. Low-inf, low-level infested pepper leaves. Hd&Mo, honeydew and molts on filter paper. AISAR, healthy pepper plants sprayed with Actigard[®] Ctrl, clean filter paper control. Mean locomotor activity and standard error are shown. Different letters indicate significant differences (α =0.05).

for the control treatment and for healthy leaves sprayed with Actigard[®]. *H. axyridis* locomotor activity did not differ significantly in contact with leaves at any prey density or with prey residues (Figure 2). The locomotor activity for contact with these substrates differed significantly from that observed in contact with healthy or with AISAR-sprayed leaves.

Electronic nose evaluation

The output produced by the QMB sensors from the MOSES E-nose produced a uniform nondiscriminated signal and had to be discarded because this result was equivalent to no response at all. Consequently, the PCA analysis had to be performed using only the original values reported by the SnO₂ sensors. The data had to be standardized as a prerequisite for PC analysis because the levels of the highest outputs (from S1 and S8) and the lowest output (from S4) differed by a factor of more than a thousand. The eigenvalues from the only two axes obtained from the PCA were 0.993 for CP1 and 0.07 for CP2. The second axis was considered negligible because it explained less than one percent (1%) of the variability and because the three treatments were clearly separated only on CP1 in the two-dimensional PCA plots. Based on these results, the PCA had to be discarded as valueless. The MANOVA analysis showed significant differences using the Pillai test (F=32.33; $P \le 0.0001$), and the Hotelling test showed significant rank differences among treatments (Table 1).

Table 1. Results from Hotelling test (α =0.05) from SnO₂ sensor output (100 points from release). Health-pl, healthy pepper plants. High-pl, high-level infested pepper plants. AISAR, healthy pepper plants sprayed with Actigard[®]. Error: covariance matrix, 3497 df. Different letters indicate significant differences (P≤0.05).

Treatment	S1	S2	S3	S4	S5	S6	S7	S8	n	
Health-pl	3004.15	2195.58	707.05	135.50	1335.78	2632.55	2204.85	2988.21	1200	а
High-inf	3376.52	2623.31	952.54	183.86	1607.77	3232.56	2589.27	3975.33	1000	b
AISAR	2820.79	2134.51	707.60	130.20	1325.13	2470.93	2155.84	2888.63	1300	с

MANOVA did not detect any interaction. This indication was confirmed by univariate graphic display and analysis (Figure 3 and Table 2). MANOVA distinguished the Actigard treatment as a third group (group (c) in Figure 3), probably because the outputs of S1 and S6 were the highest and had the most influence in terms of relative weight (Table 1 and Figure 3).

These results high-infestation treatment always showed the most significant response. The response for the high-infestation treatment always differed significantly from the response to the same compounds present in the Health-pl and AISAR treatments. Only a slight difference, involving only two of these same volatile compounds, was detected between these treatments.

Discussion

Several studies of tritrophic plant-herbivorecarnivore systems have indicated that the ability of the carnivores and parasitoids to discriminate among different odor blends depends strongly on their degree of dietary specialization (Dicke and Van Loon, 2000). Coccinellid responses to semiochemicals from aphids and to aphid honevdew, and related microorganisms, have been reported (Majerus, 1994; Ninkovic et al., 2001; Biesinger et al., 2005; Heit et al., 2008). Obata (1986, 1997) has claimed that Harmonia axyridis adults used olfactory cues to differentiate aphid-infested leaves from uninfested ones. However, he did not conclusively determine whether the observed locomotor response was related to substances associated with the aphids themselves or to plant semiochemicals whose production was induced in response to feeding by the aphids.

Our results suggest that a specific stimulus linked to aphid-host interaction would trigger a distinctive locomotor activity pattern in H. axvridis. This pattern would be displayed whether or not contact occurred with the aphids or the host plant. In contrast, C. sanguinea could not differentiate between the volatiles from infested or uninfected pepper leaves and could not distinguish these volatiles from the clean filter paper used as a control. This result may suggest that C. sanguinea adults are not receptive to the volatiles released from the aphids or aphidinfested plants or that they lack some additional cue not present in our tests. Similarly, Robertson et al. (1995) have argued that the value of an infochemical could also depend on contextual variation, including the simultaneous presence of other cues, which may modulate its intrinsic information value. For example, the presence of a particular substrate could modify the information value associated with one specific odor blend. In addition, a particular behavioral response may depend strongly on the level of plant induction (Holopainen, 2004). For this reason, treatment with Actigard could fail to evoke the same behavioral response produced by the aphid-infested treatment.

Once *C. sanguinea* was in contact with a prey patch, *i.e.*, aphid-infested pepper leaves (regardless of previous prey density), it moved slowly and scanned the area exhaustively. This response could be a consequence of the altered locomotor activity described by the Area-Concentrated Search (ACS) model. In this model, reduced durations of walking and increased durations of resting are particularly associ-

Table 2. Results from univariate analysis and comparisons among means (α =0.05, df=2). Health-pl, healthy pepper plants. High-pl, high-level infested pepper plants. AISAR, healthy pepper plants sprayed with Actigard[®]. Different letters within the same column indicate significant differences between the corresponding averages.

	S1	S2	S3	S4	85	S6	S7	S8
F	27.71	25.40	64.96	83.15	61.32	53.06	38.80	81.95
P- value	≤0.0001	≤0.0001	≤0.0001	≤0.0001	≤0.0001	≤0.0001	≤0.0001	≤0.0001
High-inf	а	а	А	а	а	а	а	а
Health-pl	b	b	В	b	b	b	b	b
AISAR	с	b	В	b	b	с	b	b



Figure 3. Odor profile of different substrates. Univariate averages and standard errors from SnO_2 outputs. Letters within the same sensor output indicate significant differences among the averages of different treatments for that SnO_2 sensor (α =0.05). Bars are standard errors.

ated with areas in which the probability of encountering prey is high. Similarly, *H. axyridis* showed significantly reduced locomotor activity when it was in contact with aphid-infested pepper leaves or aphid molts and honeydew, reliable signals of a food source.

Our results also agree with those of Obrycky and Kring (1998), who have pointed out that as a general rule, area-restricted searching in coccinellids (i.e., extensive vs. intensive search) may be influenced by the substrate regardless of the presence of the prey. However, our results disagree with the findings of Ferran et al. (1994), who reported that Semiadalia undecimpunctata was able to detect its prey only after contacting it. Like other generalist predators, C. sanguinea and H. axyridis can consume different prey types, but they have a higher fitness when fed on aphids (Ninkovic et al, 2001; Isikber and Copland, 2002). Therefore, C. sanguinea and H. axyridis have probably developed the capacity to detect volatiles related to aphid-colonized patches. Therefore, our results suggest that these coccinellid species adjust their foraging behavior to short-term changes in environmental conditions, *i.e.*, the prey distribution when they arrive at a patch. The weak response of C.

sanguinea and *H. axyridis* to volatiles from uninfected plants may indicate that by themselves, these volatiles had a limited impact on prey searching behavior.

Our results showed that the E-nose is able to differentiate the blend of volatiles released from cut healthy and aphid-infested pepper leaves, although it is not able to identify the compounds perceived and then explored differentially by *H. axyridis* and *C. sanguinea*. Odor blends emitted by herbivore-infested plants are complex mixtures, often composed of more than 100 different compounds, many of which occur as minor constituents (Vet and Dicke, 1992). The chief concern in this context is to identify the key compound triggering the response, or alternatively to find the correct mixture of compounds that can be used to construct a blend capable of eliciting the response.

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Resumen

G. Heit, V., Messina, G. Mareggiani, R. Cohen y P. Carrizo. Utilización de actómetros y nariz electrónica, para evaluar la actividad locomotriz de dos Coccinellidae (Coleoptera). Cien. Inv. Ing. Agr. 38(2): 265-275. El efecto de los semioquímicos volátiles en la actividad locomotriz de Cycloneda sanguinea y Harmonia axyridis fue evaluado mediante el empleo de dos actómetros, a fin de estudiar cambios en las respuestas de los coccinélidos en contacto directo con los sustratos y los expuestos solamente a las sustancias volátiles. El perfil de volátiles implícitos en los bioensavos se analizaron mediante una nariz electrónica (Modular Sensor System Moisés II). Los tratamientos incluyeron el uso de los compuestos volátiles emitidos por hojas de pimiento sanas o infestadas con Myzus persicae, excretas de M. persicae, hojas sanas tratadas con Actigard (Acibenzolar-S-methyl). Los resultados del presente estudio sugieren que un estímulo específico vinculado a la interacción áfido-hospedero desencadenó un patrón de actividad locomotriz distintivo en H. axvridis, estando en contacto, o no, con la planta hospedera. En cambio, C. sanguinea no mostró cambios significativos en sus patrones de comportamiento ante la exposición a los volátiles. Una vez que C. sanguinea estuvo en contacto con hojas infestadas (independientemente de la densidad de áfidos), se movió lentamente, examinando exhaustivamente la zona. H. axvridis mostró una reducción significativa de la actividad locomotriz cuando estuvo en contacto con las hojas de pimiento infestadas o excretas de áfidos, actuando éstas como señales de una fuente confiable de alimento. El tratamiento con Actigard no evocó, en ambas especies de Coccinellidae, la misma respuesta comportamental que las plantas de pimiento infestadas con áfidos. Además, se demuestra que la nariz electrónica es capaz de diferenciar entre las mezclas de volátiles liberados de hojas de pimiento sanas o infestadas con áfidos.

Palabras clave: Acibenzolar-S-methyl, actividad locomotriz, Coccinellidae, nariz electrónica, semioquímicos.

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