

Interspecific recognition through odours by aphids (Sternorrhyncha: Aphididae) feeding on wheat plants

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Abstract. In olfactometric experiments a wingless adult of *Sitobion avenae* was attracted by odours of a wingless adult of *Rhopalosiphum padi* feeding on a wheat plant. When the stimulus consisted of either a wingless *R. padi* without plant material or a wheat plant that had previously been infested by a wingless *R. padi*, the response by *S. avenae* was not significant, suggesting that recognition is due to volatiles produced by the combination of an aphid and a plant.

Introduction

The bird cherry-oat aphid *Rhopalosiphum padi* (L.) and the English grain aphid *Sitobion avenae* (F.) are two cosmopolitan aphids which usually coexist on many host plants (Dean, 1973, 1974; Carter et al., 1984). When *R. padi* infests cereal fields in early spring it feeds mainly on the lower parts of the young plants, in particular the base of the stem. Later in the season it moves to the upper parts such as flag leaves and heads. In contrast, *S. avenae* overwinters on wild perennial grasses and arrives in cereal fields later than *R. padi*, and feeds primarily on the upper leaves and ears at the reproductive stage (Dean, 1973, 1974; Carter et al., 1984). Consequently, both aphid species are often found together late in the development of cereal crops and may interact with each other (Carter et al., 1984).

Laboratory studies have shown that wingless adults of *S. avenae*, when feeding in mixed colonies with *R. padi*, made fewer probes, had shorter non-probing times, longer ingestion times, and increased fecundity on wheat seedlings, relative to pure colonies (Chongrattanmeteeikul et al., 1991a,b; Thirakhupt & Araya, 1992). This feeding behaviour was apparent in situations where low aphid densities prevailed. High aphid densities, on the other hand, had a detrimental effect on the performance of *S. avenae*.

The relative advantages obtained by *S. avenae* at low population density from living in mixed colonies with *R. padi* leads to the hypothesis that *S. avenae* is able to recognise the presence of *R. padi*. Since odours have been shown to be of importance in intraspecific communication in aphids (Pettersson, 1994; Pettersson et al., 1995), we studied odour communication between wingless adults of *S. avenae* and *R. padi*.

Material and methods

Plants

Wheat (*Triticum aestivum* L. cv. Paleta) seedlings at decimal growth stage 11, first leaf unfolded (Zadoks et al., 1974) were used. Plants were grown in a room kept between 18 and 22°C and 45 and 65% relative humidity, a light regime 12L : 12D, and light intensity of 200 $\mu\text{E m}^{-2}\text{s}^{-1}$.

Aphids

Aphids were collected in grass fields near the laboratory at Santiago. They were reared parthenogenetically on oat (*Avena sativa* L. cv. Nahuén) under the same conditions as for the wheat plants, for at least

five generations in order to ensure homogeneous material. Aphids used in all experiments were wingless adults.

Olfactometry

An olfactometer, as described by Pettersson (1970) was employed to test for interspecific recognition between aphids. One test aphid was enclosed in a square arena permeated by air coming from each of its four stretched-out corners and drawn through a hole in its centre with a vacuum pump at a flow of 250 ml/min. The exposure arena was thus divided into five different zones, a central one and four arms. Each arm had an inlet to which odours were applied. Two opposing arms were connected with plastic tubes to a glass belljar (approximate volume 800 ml) which contained the materials whose odours were to be tested ("treatment belljar"). The other two arms were connected to another belljar which contained control odour stimuli ("control belljar"). Air-tight seals and an air filter at the inlet of each belljar were used to eliminate external odours during the experiment. White paper was placed on the floor of the arena to facilitate walking by the test aphid and to make it easier to determine its position throughout the experiment.

Experimental procedure

Aphids used as stimulus and as test individuals were removed from the colony and starved for 1 h. After this period the test aphid was allowed to move in the arena for 10 min before the belljars were connected to the olfactometer and the experiment started. During the 15 min which the experiment lasted, the time the test aphid spent in each olfactometer arm was recorded. In order to avoid intraspecific interactions between aphids during the experiments (Pettersson et al., 1995), only one individual was present at any time in the olfactometer arena or the treatment belljar. Eighteen repeats of each treatment-control combination were performed. In order to avoid pseudoreplication the setup and the biological materials were changed for every repetition. Wilcoxon's two-tailed rank sum test was used to compare the total time spent by each individual aphid in the two treated vs. the two control arms.

Experiments were performed to test intraspecific interactions between *R. padi* and *S. avenae* and to determine the source of the effect, i.e. whether it was the aphid or the host plant.

Results

Aphid responses to a wheat seedling with an aphid of a different species

In this experiment, the treatment belljar contained a pot with a single wheat plant growing in soil and supporting an aphid individual of a different species than the aphid in the arena. The control belljar contained a single wheat plant growing in soil with no history of aphid feeding. The "stimulus" aphid was exposed to the plant in the treatment belljar for 15 min before the "test" aphid was placed in the olfactometer and the airflow towards the olfactometer began. *S. avenae* was significantly attracted to *R. padi* but the inverse combination was not significant (Table 1).

TABLE 1. Olfactometric response of a wingless aphid adult to a wheat seedling infested with a heterospecific wingless aphid individual. Number of repetitions is given as "n". One standard deviation is shown in parenthesis. Comparisons were performed with the Wilcoxon two-tailed rank sum test. The level of significant differences was set at $p < 0.05$.

Stimulus aphid	Test aphid	n	Average time in treated arm (min)	Average time in control arm (min)	Significance level (p)
<i>R. padi</i>	<i>S. avenae</i>	18	3.7 (± 1.1)	2.4 (± 0.9)	0.017
<i>S. avenae</i>	<i>R. padi</i>	18	3.5 (± 0.9)	3.6 (± 0.9)	0.925

To test whether the "attraction" of *S. avenae* towards *R. padi* was caused by the aphid, the plant or the aphid/plant combination, the following experiments were performed.

Aphid response to an aphid exposed to wheat

An individual of *R. padi* was allowed to remain on a wheat seedling (outside the belljar) for 40 min and was then transferred to an empty treatment belljar. The control belljar was either empty or it contained a wheat seedling. The test aphid was *S. avenae*. The responses were not statistically significant in either of the two control setups (Table 2).

TABLE 2. Olfactometric response of a wingless adult of *S. avenae* to different treatments. Number of repetitions is given as "n". One standard deviation is shown in parenthesis. Comparisons were performed with the Wilcoxon two-tailed rank sum test. The level of significant differences was set at $p < 0.05$.

Treatment belljar	Control belljar	n	Average time in treated arm (min)	Average time in control arm (min)	Significance level (p)
<i>R. padi</i>	wheat	18	3.4 ($\pm$ 1.0)	2.6 ($\pm$ 1.1)	0.19
<i>R. padi</i>	empty	18	2.5 ($\pm$ 1.3)	3.0 ($\pm$ 1.4)	0.35
Preinfested wheat	wheat	18	2.5 ($\pm$ 1.5)	3.2 ($\pm$ 1.8)	0.81

Aphid response to a wheat seedling exposed to aphid

A wheat seedling that had supported the stimulus aphid (one individual of *R. padi*) for 40 min was introduced into the treatment belljar following aphid removal. A wheat seedling with no history of aphid feeding was used as control. The test aphid was *S. avenae*. No significant differences were observed in this experiment (Table 2).

Discussion

The results show that wingless adults of *S. avenae* were attracted by wingless adults of *R. padi*. No significant response was obtained when the stimulus consisted of an aphid without plant material, or when the stimulus was a wheat plant that had previously been infested by an individual of *R. padi*. An interpretation of these results is that the recognition response to *R. padi* by *S. avenae* is due to volatile compounds produced by the interaction of *R. padi* with the plant. Work is underway to isolate and determine the identity and origin of the volatile compounds responsible for such recognition behaviour.

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