ODOUR RESPONSE PROFILES IN APHIDS DIFFERENTIATING FOR SPECIES, CLONE, FORM AND FOOD

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Summary

In order to evaluate the variation in responses of aphids to a range of odour compounds comprising plant volatiles, the alarm pheromone (*E*)-ß-farnesene and the sex pheromone components (+)-(4a*S*,7*S*,7a*R*)-nepetalactone and (-)-(1*R*,4a*S*,7*S*,7a*R*)-nepetalactol, electroantennogram (EAG) responses were measured and compared for (a) the cabbage aphid *Brevicoryne brassicae* (L.) and the peach-potato aphid *Myzus persicae* (Sulz.), both fed on Chinese cabbage, (b) two clones of *M. persicae*, (c) wingless and winged summer forms of *M. persicae*, and (d) *M. persicae* fed on Chinese cabbage or an artificial diet. The four comparisons differed significantly for 44 %, 20 %, 16 % and 12 %, respectively, of the compounds tested. These differences as well as the scatter plots of the comparisons (Figure 1A-D) support the conclusion that the rank order of factors for differentiating the EAG response profiles in aphids is: species, clone, form and food source.

INTRODUCTION

Semiochemicals such as the sex pheromones of lepidopterous species have been successfully applied in pest management programs as monitoring tools or mating disruptants. This approach is more ecologically sound than present-day insecticides and can also be useful in the control of other insect species, like aphids, which are major pests in temperate zones. The development of such strategies should involve a series of investigations directed to the identification of candidate chemicals, the evaluation of their effects on aphid behaviour and the elucidation of the impact of semiochemicals on crop colonization and aphid population dynamics.

The first step, the screening of potential semiochemicals, has been done for a number of aphid species, *e.g. Sitobion avenae* and *Metopolophium dirhodum* (Visser & Yan, 1995), *Megoura viciae* (Visser & Piron, 1995), *Aphis fabae* (Hardie *et al.*, 1994, 1995), *Brevicoryne brassicae* and *Myzus persicae* (Visser *et al.*, 1996), by the recording of electroantennogram (EAG) responses. From these datasets, a mean aphid response profile to plant volatiles has been constructed and species-specific traits have been indicated (Visser *et al.*, 1996). For the future application of semiochemicals, it is necessary to evaluate the biological variation in the responses to semiochemicals as controlled by genotype (the variation between aphid clones) and phenotype (the variation between aphid forms and the effects of food sources).

In the present study, we measured the variation in EAG responses by comparing (a) the cabbage aphid *Brevicoryne brassicae* (L.) with the peach-potato aphid *Myzus persicae* (Sulz.), both reared on Chinese cabbage, (b) two clones of *M. persicae*, (c) wingless and

winged summer forms of *M. persicae* and (d) *M. persicae* fed on Chinese cabbage or an artificial diet.

MATERIAL AND METHODS

Aphids

B. brassicae aphids were the Tjallingii clone from Spain. *M. persicae* clones M1 and M3 were obtained from Dieleman. The host-plant range of the M1 clone is more restricted than that of the M3 clone. In addition, the M1 clone cannot produce sexual forms, whereas the M3 clone will, under autumn conditions. Aphids were reared on Chinese cabbage, *Brassica chinensis* L. cv. Granaat, under long-day conditions (L16:D8) at 22 °C. In order to evaluate food effects on EAG responses, the *M. persicae* M1 clone was also reared on the MP 148 artificial diet as defined by Harrewijn (1983) at the same climatic conditions as the colonies on Chinese cabbage.

EAGs were recorded from alate virginoparae (winged summer forms) of *B. brassicae* and the *M. persicae* M1 and M3 clones, and from apterous virginoparae (wingless summer forms) of *M. persicae* M1 reared on Chinese cabbage or artificial diet. When reared on artificial diet, the 2nd generation born on the diet was used for EAG recordings.

EAG recordings and odour stimuli

The methods of EAG recording from aphid antennae, the instruments and data processing involved have been described in detail by Visser & Piron (1995). In the present study, a wide range of odour components were used as stimuli, including (a) plant volatiles as described by Visser & Piron (1995), (b) the alarm pheromone (*E*)-ß-farnesene (from KRI-TNO, 98% pure) and (c) the sex pheromone components (+)-(4a*S*,7*S*,7a*R*)-nepetalactone and (-)-(1*R*,4a*S*,7*S*,7a*R*)-nepetalactol (both from IACR-Rothamsted, 98% pure).

All odour components were diluted to 1 % (v/v) in paraffin oil (Merck, Uvasol). Stimulation cartridges were prepared by applying 25 µl of each solution to a piece of filter paper which was subsequently placed in a Pasteur pipette. The aphid antenna was stimulated for 2 s by pushing air through the Pasteur pipette into an air flow directed over the preparation. EAG waveforms were recorded and the largest deflection of the baseline was measured as the absolute EAG response.

Absolute EAG responses were normalised and expressed as percentage responses relative to the responses of adjacent standards consisting of 1% (*E*)-2-hexenal at the source (Visser & Piron, 1995). All odour components were tested on at least 10 different antennal preparations and means ± 95% confidence intervals (2-tailed) were calculated. EAG responses were considered significantly different when their means ± 95% confidence intervals did not overlap.

RESULTS AND DISCUSSION

In order to evaluate the effects of aphid species, clone, form or food source on the EAG responses to a range of volatiles, four comparisons were made (Figure 1A-D).

Comparing *B. brassicae* with *M. persicae*

The absolute EAG responses to the standard, 1% (*E*)-2-hexenal, were not significantly different between alate virginoparae of *B. brassicae* (mean ± 95% c.i.: 269±82 µV) and the *M. persicae* M3 clone (457±125 µV), both being reared on Chinese cabbage. The mean relative EAG responses (Figure 1A) were significantly different for 35 out of 80 components, namely: (*E*)-2-hexenol-1, (*Z*)-3-hexenol-1, hexanol-1, hexyl acetate, (*E*)-2-hexenyl propionate, (*Z*)-3-hexenyl acetate, (*Z*)-3-hexenyl propionate, heptanol-1,

3-heptanone,3-octanone, 3-methoxybenzaldehyde, 4-methoxybenzaldehyde, 2-hydroxybenzaldehyde, 1,2-dimethoxybenzene, (+)-(*S*)-carvone, (-)-(*R*)-carvone, α-ionone, ß-ionone, linalool, citral, (+)-limonene, (-)-(*E*)-caryophyllene, (*E*,*E*)-farnesyl acetate, (*E*,*E*)-α-farnesene, hexanonitrile, 1-hexanethiol, 1,6-hexanedithiol, butyl isothiocyanate, *tert*-butyl isothiocyanate, allyl isothiocyanate, 3-butenyl isothiocyanate, 2,5-dimethylpyrazine, methylsalicylate, (+)-(4a*S*,7*S*,7a*R*)-nepetalactone and (-)-(1*R*,4a*S*,7*S*,7a*R*)-nepetalactol.

Comparing clones of *M. persicae*

The absolute EAG responses to the standard were not significantly different between alate virginoparae of the *M. persicae* clones M1 (407±109 μV) and M3 (457±125 μV), both being reared on Chinese cabbage. The mean relative EAG responses (Figure 1B) were significantly different for 11 out of 54 components, namely: 3-octanone, butyl isothiocyanate, allyl isothiocyanate, 3-butenyl isothiocyanate, 2-methoxybenzaldehyde, 3-methoxybenzaldehyde, 4-methoxybenzaldehyde, 2-hydroxybenzaldehyde, 1,2-dimethoxybenzene, *m*-cresol and (-)-(*R*)-carvone.

Comparing forms of *M. persicae*

The absolute EAG responses to the standard were significantly different between alate (407±109 μV) and apterous (256±35 μV) virginoparae of *M. persicae* , both being the M1 clone reared on Chinese cabbage. As apterous *M. persicae* do not possess secondary rhinaria on their antennae, whereas alate aphids do, the difference in absolute EAGs to (*E*)-2-hexenal is attributed to the secondary rhinaria responding to this component.

The mean relative EAG responses (Figure 1C) were significantly different for 8 out of 50 components, namely: butyl isothiocyanate, *tert*-butyl isothiocyanate, allyl isothiocyanate, 3-butenyl isothiocyanate, geraniol, (+)-citronellol, 2-methoxybenzaldehyde and *m*-cresol.

Most compounds show the same relative EAG levels in alate and apterous *M. persicae*. However, the absolute response to the standard is larger in alate than apterous aphids, which implies that absolute levels for most components are raised as well. In other words, the secondary rhinaria will be responsive to most components tested.

Comparing food sources for *M. persicae*

The absolute EAG responses to the standard were not significantly different between *M. persicae* reared on an artificial diet (314±62 μV) or on Chinese cabbage (256±35 μV), both being apterae of the M1 clone. The mean relative EAG responses (Figure 1D) were significantly different for 6 out of 50 components, namely: (*E*)-2-hexenyl propionate, hexanonitrile, 3-methoxybenzaldehyde, sabinene, (-)-(*E*)-caryophyllene and (-)-(1*S*)-ß-pinene.

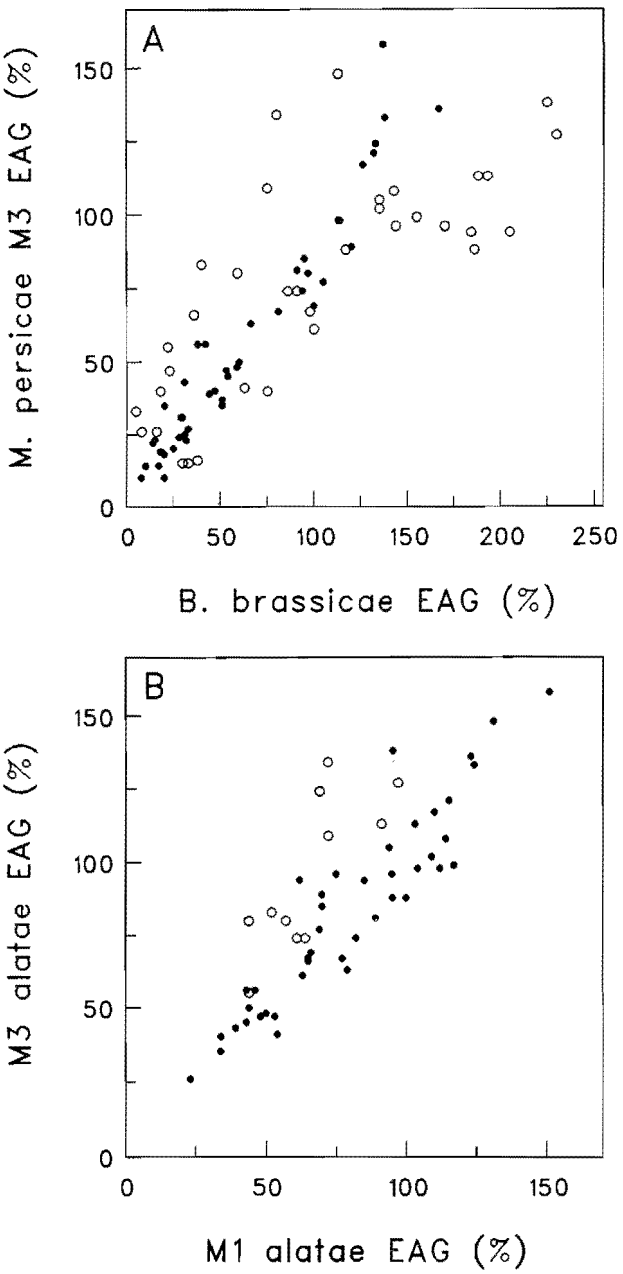

Figure 1. *Comparison of mean relative EAG responses to a range of odour compounds for (A)* Brevicoryne brassicae *with* Myzus persicae *M3 clone, and (B)* M. persicae *M1 clone with M3 clone. Open circles indicate that, between aphid groups, compounds differ significantly in their relative EAG responses.*

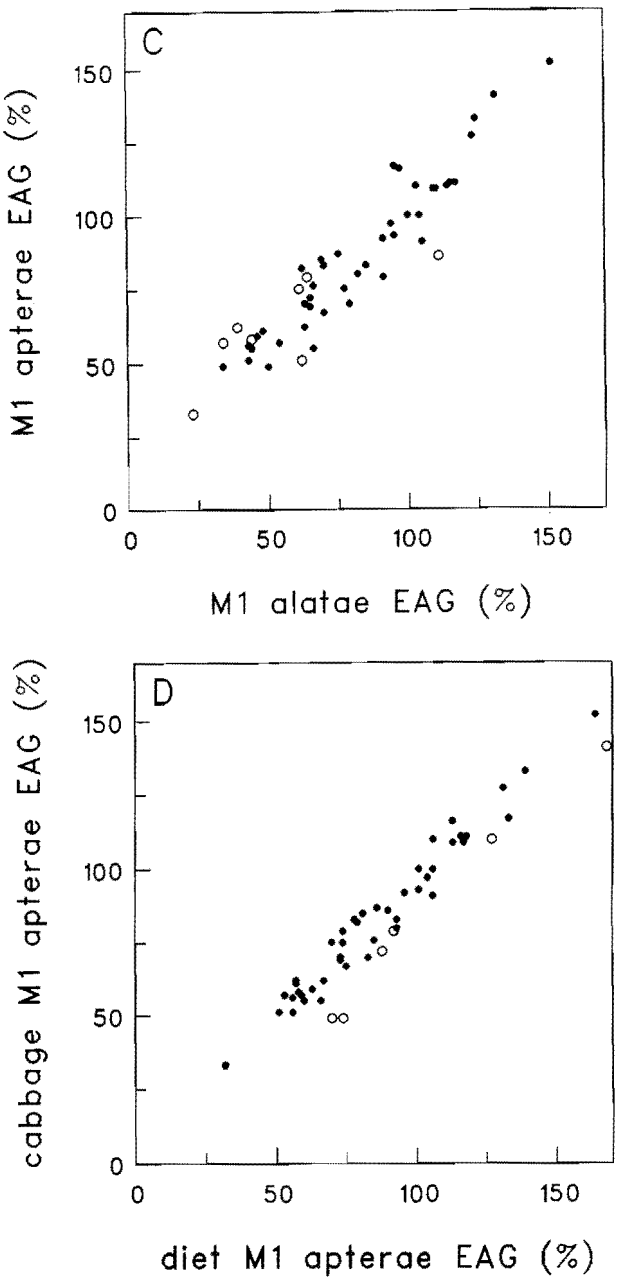

Figure 1 (continued). *Comparison for (C)* Myzus persicae *M1 clone alate with apterous virginoparae, and (D)* M. persicae *M1 clone apterae reared on artificial diet with apterae reared on Chinese cabbage. Open circles indicate that, between aphid groups, compounds differ significantly in their relative EAG responses.*

Factors differentiating response profiles

The comparison of *B. brassicae* with *M. persicae* differed significantly for 35 out of 80 compounds (44 %), *M. persicae* M1 clone with M3 clone, 11 out of 54 (20 %), *M. persicae* alatae with apterae, 8 out of 50 (16 %), and *M. persicae* diet-reared with cabbage-reared, 6 out of 50 chemicals (12 %). These differences as well as the scatter plots of the comparisons (Figure 1A-D) support the conclusion that the rank order of factors for differentiating the response profiles in aphids is: species, clone, form and food source.

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