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# Influence of *Acarus siro* (Acari Acaridae) acetone extracts on conspecifics*

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**Abstract**

The influence of acetone extracts of male and female *Acarus siro* on the behavior of male and nymphal stages of this mite was tested. In the presence of a female acetone extracts, copulatory attempts between males occurred. The occurrence of a female sex pheromone in those acetone extracts is suggested. The same behavior was observed when males were in the presence of highly concentrated male acetone extracts. From this, we suggest the presence in males of the studied strain, of a semiochemical similar to the one emitted by females. Furthermore, male and female acetone extracts induced an aggregation effect as nymphal stages of the mite spent more time in an area treated with an acetone extract than in the control areas.

**Key words :** aggregation pheromone, sex pheromone, behavior.

## Introduction

*Acarus siro* L. is considered as an important pest of stored food products in most temperate regions (SOLOMON, 1962; VAN ASSELT *et al.*, 1996). Since cases of resistance to insecticides have been reported (WILKIN, 1973; WILKIN & STABLES, 1985), there is a need to search for alternative control strategies. Combined pheromones and acaricides have been used successfully on other mite species (BAKER & KRANTZ, 1984; NORVAL *et al.*, 1996).

For LEVINSON *et al.* (1989), there would be two different sex pheromones occurring in an acetone extract of *A. siro*. The sex pheromone produced by males attracted and stopped virgin females, while the sex pheromone produced by females attracted and stimulated males to mate.

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Our study investigated the effect of *A. siro* male and female acetone extracts on the behavior of males and immatures.

## Material and methods

Two bioassays were used : (1) the first tested the behavior of male individuals after the application of male and female acetone extracts; the total time spent by individuals in a treated areas was determined; (2) the second assay allowed us to investigate the effect of male and female acetone extracts on immatures; the number of immatures arrested for a given time on a treated area was studied.

The laboratory culture of *A. siro* was derived from a strain provided by Prof. A. FAIN (IRSNB-KBIN). It was maintained on a diet composed of two parts of wheat germ and one part of dried brewer's yeast. Test specimens were maintained at 23 ± 0.5°C, 75% relative humidity in darkness. Four hundred males and 400 females were harvested for testing. Since males and females mate several times during their adult life, no attention was paid to the age of the adult and to their physiological state.

Harvested individuals were immersed in 500µl acetone, at 20°C for 2 hours and then at 0°C for 16 hours. The acetone extract was concentrated until a quantity of 5µl was determined to contain the desired number of male or female equivalents, at least for concentrations corresponding to 5, 10, 20, 30 and 40 individuals. For higher concentrations (80 and 200 mite equivalents), a volume of 10µl was used.

A three-choice test was used to study the behavior of the mites. It consisted of three circular areas (diam. 6mm each) cut out of a filter paper and situated 1 cm apart at the vertices of an equilateral triangle. The filter paper was placed on a plaster plate colored with animal charcoal; the system was then humidified. In one of the three areas (the test area) 5 or 10µl of male (or female) acetone extract was added. The remaining areas received the same quantities of pure acetone (controls). Five males were then placed at the centre of the triangle. Each experiment was replicated 8 times. In each replicate, the behavior of the individuals was filmed during 30 minutes. The time spent in the areas was measured and the percentages of time spent were calculated for each area compared to the total time spent in the three areas.

Statistical analyses were done using a binomial test Bi (0.33, 8) (SOKAL & ROHLF, 1969). For this, we considered the test as positive when the percentage of time passed by mites in the test area was higher than the percentage of time lasted in either of the two control areas. In the other cases, the test was considered as negative.

## Results and discussion

Figure 1 shows the evolution of the average percentage of time spent by 5 males in the test area, when male acetone extract was tested. Compared to the control

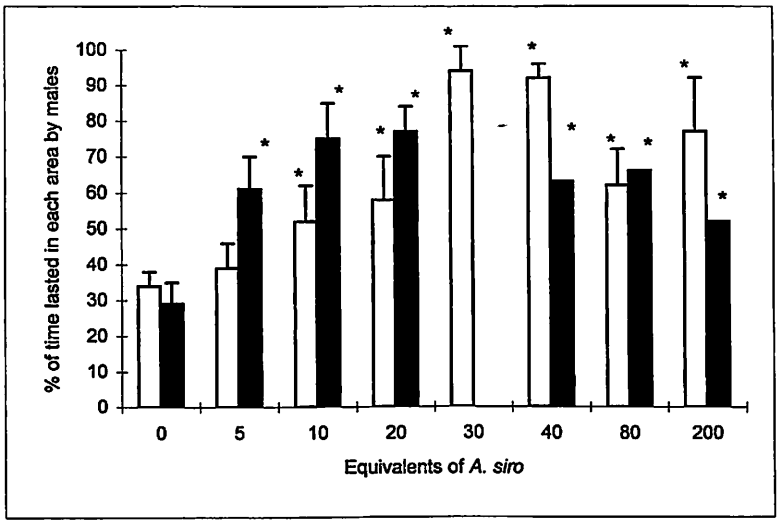


Fig. 1. Percentage of time spent by 5 male *Acarus siro* in the test area: effect induced by graded dosages of male acetone extracts (□) and female acetone extracts (■).

areas, mites spent more time in treated area with doses higher than 10 male equivalents ($p<0.01$). When female extracts where testing on males, such a phenomenon was already found at doses as low as 5 female equivalents ($p<0.01$).

It should be noted that mounting attempts occurred between males in the presence of female extracts when high concentrations of the extract were used (40, 80 and 200 mite equivalents). The pairing attempts between males suggested, as LEVINSON *et al.* (1989) concluded, the existence of a female sex pheromone. In our experiments, the same behavior was observed when male individuals were in contact with male extract at 40, 80 and 200 mite equivalents (a test not performed by LEVINSON *et al.*, (1989)). Thus, we suggest the presence in the male extract of a substance inducing effects similar to those of the female sex pheromone. Yet, in natural conditions, this effect would be induced only by females.

KUWAHARA *et al.* (1992) after identification of a female sex pheromone (2-hydroxy-6-methylbenzaldehyde) in males, females and immatures *Aleuroglyphus ovatus* (Acari: Astigmata) concluded in a similar phenomenon. SATO *et al.* (1993) identified the presence in male, female and immature *Acarus immobilis* (Acari: Astigmata) of 2-hydroxy-6-methylbenzaldehyde, also considered as a female sex pheromone for this species. While MORI *et al.* (1998), observed the presence of rosefuran (a sex pheromone) in male, female and immature *Caloglyphus* sp. (Acari: Astigmata). Taking this into account, the production of a female sex phe-

romone by males and females should be considered as common in astigmatid mites. It is the first time that this phenomenon has been demonstrated in a strain of *A. siro*.

The effect of an acetone extract corresponding to 30 male or female equivalents was tested on immatures. The previously described system was used again but, instead of five *A. siro* males placed at the centre of the system, 10 immatures were placed in each of the three areas. The number of individuals present in each of the three areas after 30 minutes was counted. Fifteen replicates were conducted with larvae and 15 with a mixture of protonymphs and tritonymphs. A binomial test Bi (0.33, 15) was performed. For each repetition, we gave a positive score when the number of individuals in the test area was higher than the number of individuals in each of the two control areas. In the other cases, the test was considered as negative.

The presence of an aggregative effect could be suggested if, after a defined period of time, immature individuals were more numerous in the treated area than in the control areas.

This experiment showed the aggregation effect of the female and the male acetone extract on larvae and on protonymphs and tritonymphs ($p<0.001$ four times) (Table 1). As it may be assumed that there is no sexual activity in immatures, we conclude in the presence of an aggregative effect of the extracts used.

Table 1. Mean number ± standard error of larvae and proto- tritonymphs of *Acarus siro* present in each of the three test areas (15 replicates). E: treated area; $C_1$, $C_2$ : control. * $p < 0.001$

| | Larvae | | | Proto-tritonymphs | | |
|---|---|---|---|---|---|---|
| | E | $C_1$ | $C_2$ | E | $C_1$ | $C_2$ |
| Female extract | 6.8 ± 3.3* | 1.6 ± 1.2 | 1.6 ± 1.3 | 5 ± 1.6* | 0.8 ± 0.7 | 0.6 ± 0.6 |
| Male extract | 6.2 ± 1.4* | 1.1 ± 1.1 | 1.4 ± 1.1 | 4.1 ± 1.5* | 0.6 ± 0.7 | 0.9 ± 0.9 |

## Conclusions

Previously, the presence of two different sex pheromones in *A. siro* was suggested : a sex pheromone produced by males (attractant and arrestant) and a sex pheromone produced by females (attractant and copulatory stimulant) (LEVINSON *et al.*, 1989). According to our observations, we can hypothesize that, in our strain, *A. siro* males produced a semiochemical inducing a behavior similar to those induced by the copulatory stimulant female pheromone. The reason for its presence in males is still unknown.

The *A. siro* female and male acetone extracts seemed to contain an aggregation substance active on immature stages. This observation leads us to suggest a possible confusion in previous studies between the effects of both a sex and aggregation pheromone. Further chemical analyses should clarify these aspects.

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