Olfactory attractiveness of mixtures of some host plant and conspecific semiochemicals to the banana weevil, *Cosmopolites sordidus* (Germar, 1824)

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ABSTRACT

A simple "Y" shaped olfactometer was used in laboratory studies on the olfactory attractiveness of mixtures in various proportions of industrial analogues of some host plant and conspecific-based semiochemicals, or their combinations with banana rhizome, to the banana weevil. The aim was to identify factors that influence their attractiveness to the weevil, and consider the possibility for their use as lures for trapping the weevil in the field. *Cosmopolites sordidus* was attracted to the mixtures at specific concentrations and proportions of constituent chemicals. 6-methylhept-5-en-2-one was only attractive on its own at 1 μl/100 ml and in mixture with 4-mercaptophenol, but not at 10 μl, 0.01 μl, or in combination with banana rhizome. 4-mercaptohpenol and 2-n-butylfuran, which were compatible with most host plant-based chemicals and were attractive as a mixture, were perceived to be key elements in the composition of attractants to the weevil. It was concluded that in addition to the composition, other factors that may determine the attractiveness or otherwise of a mixture to *C. sordidus* are the proportions and concentrations of the constituent chemicals.

Original scientific paper. Received 20 Sep 02; revised 19 Feb 04.

RÉSUMÉ

BRAIMAH, H. & VAN EMDEN, H. F.: *Attraction olfactif de préparations de quelques plantes hôtes et des médiateurs chimiques conspécifiques aux charançon de banane* (Cosmopolites sordidus *(Germar 1824).* Une étude se déroulait pour évaluer l'attraction olfactif de préparations d'analogues industriels en proportions diverses des médiateurs chimiques basé sur quelques plantes hôtes et conspécifique ou leurs combinaisons avec les rhizomes de banane aux charançon de banane au laboratoire en utilisant un simple olfactomètre en forme de "Y". Le but était d'identifier les facteurs influençant leur attraction au charançon et de considérer la possibilité d'en utiliser pour attirer et prendre au piège les charançons aux champs. *Cosmopolites sordidus* était attiré aux préparations ayant des proportions et des concentrations spécifiques des produits chimiques constituants. 6-methylhept-5-en-2-one était seulement attirant qui lui est propre à 1μl/100 ml et en mélange avec 4-mercaptophenol mais pas à 10 μl, 0.01 μl, ou en combinaisons avec les rhizomes de banane. 4-mercaptophenol et 2-n-butylfuran qui était compatible avec la plupart de substance chimiques extrait de plante hôtes et étaient attirants comme une préparation étaient perçu d'être les éléments clés de la composition d'attractant aux charançons. La conclusion était tirée qu'en plus de la composition, d'autres facteurs qui pourraient déterminer l'attraction ou autrement d'une préparation à *C. sordidus* sont les proportions et les concentrations des substances chimiques constituants.

Introduction

Complex environmental and physiological factors influence the release, perception, and response of insects to semiochemicals (Haynes & Birch, 1985). Semiochemicals occur in blends and mixtures, and insects are equipped with sophisticated olfactory apparatus that enable them to filter and respond to specific ones. The complexity of the composition of semiochemicals that reach the insect depends on the heterogeneity of its habitat. It is, for example, known that the presence of other plants in a host plant patch of a specialised feeder constitutes 'chemical noise' that increases the difficulty of locating the host plant (Miller & Strickler, 1984). Plants, microorganisms and other fauna, particularly other insects within the same habitat, produce chemical signals that influence the composition of semiochemicals that reach a foraging insect. As a result of this, semiochemicals have been used to manipulate the behaviour of insect pests. Their use presents economically feasible and ecologically sustainable means of controlling insect pests in integrated pest management (IPM) programmes (Metcalf & Metcalf, 1992).

The traditional method of controlling the banana weevil, *C. sordidus,* is the use of split pseudostem and rhizome traps (Froggatt, 1924; Mitchell, 1980; Treverrow, 1993). The plant tissues are attractive only over a period of 7 to 14 days, depending on environmental conditions (Schmitt, Gowen & Hague, 1992). Some chemicals that originate from the banana plant and the banana weevil were individually attractive to the banana weevil (Braimah, 1997), and could act as cues for *C. sordidus* to locate its host plant. However, very little is known about the factors that may influence their attractiveness. Factors such as concentration, composition and proportions of chemicals in the blends, source, and isomeric configuration have all been found to affect the attractiveness of some semiochemicals to other insects (Dickens, 1978, 1986; Bedard *et al.*, 1980; Perez *et al.*, 1994).

This study aimed to ascertain some of the factors that affect the attractiveness to the banana weevil of the industrial analogues of its host plant and conspecific semiochemicals. The ultimate aim was to generate information that would facilitate their use, either singly or as blends, in lures for trapping and managing *C. sordidus* in the field.

Materials and methods

Chemicals

The chemicals studied were those identified from analysis of headspace gases of dead banana leaves and fresh pieces of banana rhizome with male *C. sordidus* feeding on them. The chemicals were selected from among several that were identified through standard Gas Chromatography and Mass Spectrophotometry (GC-MS) and Gas Chromatography-Mass Spectrophotometry and coupled Electro-antennograhpy (GC-MS-EAG) techniques. Those selected for this study were found to be individually attractive to the banana weevil at either one or both of two concentrations (Braimah, 1997).

Chemicals identified from GC-MS of dead leaves were isobutyraldehyde, 2-methylbutyraldehyde, 2-n-butylfuran, and 3-methylbutyraldehyde. 4-mercaptophenol was identified through GC-MS of chopped banana rhizome on which male *C. sordidus* fed. 6-methylhept-5-en-2-one was identified as a minor active peak of a GC-MS-EAG trace which showed strong electro-physiological effects on an excised *C. sordidus* antenna (Braimah, 1997). Isobutyraldehyde, 2-methylbutyraldehyde, and 3-methylbutyraldehyde had been found to be attractive to the banana weevil on their own, while 4-mercaptophenol and 2-n-butylfuran were only weakly attractive on their own, but seemed to be synergised by the presence of host plant material (Braimah, 1997). All the chemicals were obtained as industrial formulations and were at least 90 per cent pure. They were not re-assessed for purity.

Simple mixtures of these chemicals were assayed in an olfactometer (Braimah & van Emden, 1999) for their attractiveness to the banana weevil. In another test, 6-methylhept-5-en-2-one was assayed on its own at three concentrations, in mixture with only 4-mercaptophenol and together with fresh banana rhizome material.

Dilution of chemicals

The dilution of the chemicals was originally guided by the results of Dickens (1986) and Leal *et al.* (1994). The chemical solutions were prepared by dissolving required quantities of chemicals in 1 ml of absolute ethanol. The solution in ethanol was then dissolved in 99 ml of distilled water. For a mixture of more than one chemical, all the constituent chemicals of the mixture were dissolved in 1 ml of ethanol, and then in 99 ml of distilled water.

6-methylhept-5-en-2-one was tested at three concentrations; 0.1, 1 and 10 ul each in 100 ml of distilled water. The 1 μl concentration was prepared by dissolving 1 μl in 1 ml ethanol, and then in 99 ml distilled water. For the 0.1 μl, 10 ml of the 1 μl concentration was dissolved in 90 ml of distilled water. For 10 μl, this amount of chemical was dissolved in the ethanol, and was then mixed with 99 ml of distilled water. Where 6-methylhept-5-en-2-one was assayed in combination with 4-mercaptophenol, both chemicals were diluted in the same vial at the rate of 1 μl/100 ml of water. For the combination of 6-methylhept-5-en-2-one and banana rhizome, 1 ml of the chemical was used. Fresh banana rhizome and 4-mercaptophenol were used as the control and test materials, respectively (Fig. 2).

Olfactometer bioassay

The assays were conducted in a 'Y' olfactometer (Braimah & van Emden, 1999). Twenty millilitres of the diluted chemical were soaked into a 2-g piece of cotton wool in a small plastic pill box. This was then compared with a similar box of cotton wool that was soaked with distilled water mixed with only ethanol. Twenty weevils were then placed in the response chamber and allowed 30 to 45 min to respond to the test materials. At the end of the test period, the number of weevils in each chamber was counted and recorded. Weevils that remained in the response chamber were recorded as neutral. The tests were replicated six times each, except for tests involving 6-methylhept-5-en-2-one where eight replicates were run. The first half of replications of each assay were run with the test materials in the same arms of the olfactometer. After this, the olfactometer was wiped clean with cotton swabs soaked with ethanol and then washed in distilled water. It was then allowed to dry in a glassware oven at 45° C for 30 min before it was re-used. In the second half of the assay, the positions of the test materials in the arms of the olfactometer were interchanged to eliminate the effects of position on the responses of the weevils.

Statistical analysis

The data recorded were analysed, using a χ^2 test that incorporates a test of heterogeneity (van Emden, 1993). Significant χ^2 tests that were followed by significant heterogeneity tests were considered to show inconsistency of replicates, and were viewed with caution. This was necessary to check the wrong interpretation of the data. Weevils that were neutral were left out of the χ^2 analysis.

Results and discussion

The results showed that the weevil responded to the mixtures at specific concentrations and in specific proportions. It was, for example, found that a mixture of 4-mercaptophenol, isobutyraldehyde, 3-methylbutyraldehyde, and 2-methylbutyraldehyde at ratios of 1:1:10:10 was very attractive ($P<0.001$), while their blend at the ratio of 1:1:1:1 was not ($P>0.05$) (Table 1). A mixture of 4-mercaptophenol with 3-methylbutyraldehyde at a ratio of 1:1 was also not attractive, while their mixture at 10:1 was highly attractive ($P<0.001$) (Table 1). Other workers have reported similar findings. Lin, Phelan & Bartelt (1992) reported that a three-component blend of acetaldehyde, ethyl acetate, and 2-methylpropanol was significantly less effective in trapping the dried fruit beetle, *Carpophilus lugubris* Erichson, in the field than a seven-component blend. It is, therefore, possible that when mixed at the right proportions and concentrations, the chemicals identified from banana and banana weevil by-products can be attractive lures. This is borne out by the attractiveness of some simple mixtures tested (Table 1).

TABLE 1

Olfactory Attractiveness to the Banana Weevil, C. sordidus, *in an Olfactometer of Simple Mixtures of Some Identified Host Plant and Conspecific Based Semiochemicals*

	Mean number of weevils attracted to the test material			
Chemical blend tested	*Chemical mixture*	*Control*	χ^2 *1df*	χ^2 *n-1df*
4-mercaptophenol plus others[1]	13.50	3.30	36.88***	5.58ns
4-mercaptophenol plus 2-n-butylfuran	17.00	1.83	72.28***	2.85ns
4-mercaptophenol plus 3-methylbutryaldehyde[2]	5.17	13.33	21.63***	3.66ns
4-mercaptophenol plus 3-methylbutyraldehyde[3]	14.33	3.50	39.49***	6.96ns
2-n-butylfuran plus 3-methylbutyraldehyde	9.50	4.33	11.59***	16.08*
3-methylbutyraldehyde plus others[4]	5.50	11.33	12.12***	27.95**

ns = not significant (*P*>0.05) * = (*P*<0.05) ** = (*P*<0.01) *** = (*P*<0.001)
[1] = 3-methylbutyraldehyde + 2-methylbutyraldehyde + isobutyraldehyde + 4-mercaptophenol @ 10:10:1:1 µl
[2] = 3-methylbutyraldehyde + 4-mercaptophenol @ a ratio of 1 : 1 µl
[3] = 3-methylbutyraldehyde + 4-mercaptophenol @ a ratio of 10 : 1 µl
[4] = 3-methylbutyraldehyde + 2-methylbutyraldehyde + isobutyraldehyde + 2-n-butylfuran @ a ratio of 1 : 1 : 1 : 1 µl

The bioassay of 6-methylhept-5-en-2-one indicated that it was only attractive (P<0.01) to the weevil on its own at the low concentration of 1 µl/100 ml (Fig. 1). At the high concentration of 10 µl and the lowest concentration of 0.01, the differences between the attractiveness of the chemical and distilled water control were not statistical (Fig. 1). This confirms the finding above that the weevil responds to specific concentrations of chemicals. The combination of the chemical with banana rhizome resulted in a significant repellence (*P*<0.05) of the banana rhizome to the weevil (Fig. 1), though significant heterogeneity of replicates was recorded (Fig. 1). However, the assay of 6-methylhept-5-en-2-one in a mixture with 4-mercaptophenol resulted in significant attractiveness to the weevil (*P*<0.001) (Fig. 2). This further supports the idea that the proportions of a blend contributed by the constituent chemicals are important for its attractiveness or otherwise to the banana weevil.

Jaffé *et al.* (1993) showed that for the palm weevil, *Rhynchophorus palmarum* (F.), mixtures of compounds, in proportions comparable to their occurrence in attractive host plant tissues, were as attractive as natural coconut palm tissue. They also reported that rhynchophorol, 2(E) 6-methyl-2-hepten-4-ol, the active component of the aggregation pheromone attracted *R. palmarum* in the field only when plant tissue, ethyl acetate or other host plant chemicals were present. The findings of Budenberg *et al.* (1993), on the other hand, that reconstituted host plant odours did not attract the banana weevil, was probably due to inappropriate proportions and concentrations of the constituent compounds in their test mixtures.

The compatibility of 4-mercaptophenol and 2-n-butylfuran with the other host plant chemicals (Table 1) and the attractiveness of their mixtures to *C. sordidus* indicate that purely synthetic variants of these chemicals could be used for trapping the weevil. The use of parakairomone analogues (synthetic analogues of the natural attractants) in slow-release formulations would overcome problems of maintaining their concentration threshold over longer periods in the field. Perez *et al.* (1994), for example, had already concluded from their work that mixtures of four stereoisomers of the pheromones of the

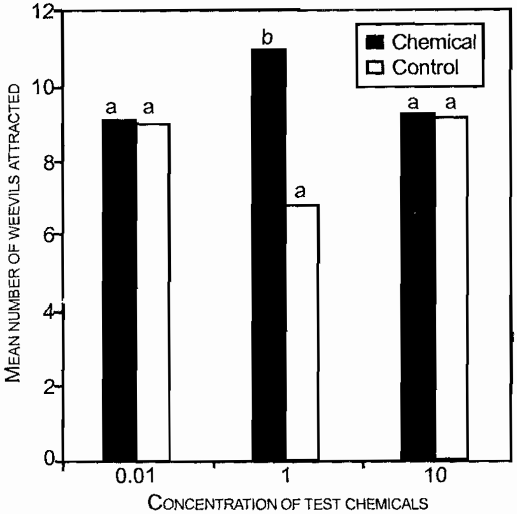

Fig. 1. The olfactory attractiveness of 6-methylhept-5-en-2-one to the banana weevil, *C. sordidus*, at three concentrations.

Comparisons can be made only for bars in each test concentration. Bars are mean numbers of weevils attracted to any test chemical for eight replications. Bars for a test concentration superscripted with the same letter are not significantly different ($\chi^2_{1\,df}$, $P<0.05$). $\chi^2_{1\,df}$ = 0.07 ns, $\chi^2_{n-1\,df}$ = 11.69 ns; $\chi^2_{1\,df}$ = 7.72**, $\chi^2_{n-1\,df}$ = 13.28 ns; and $\chi^2_{1\,df}$ = 0.007 ns, $\chi^2_{n-1\,df}$ = 15.70* for the tests using 0.01, 1 and 10 µl, respectively.

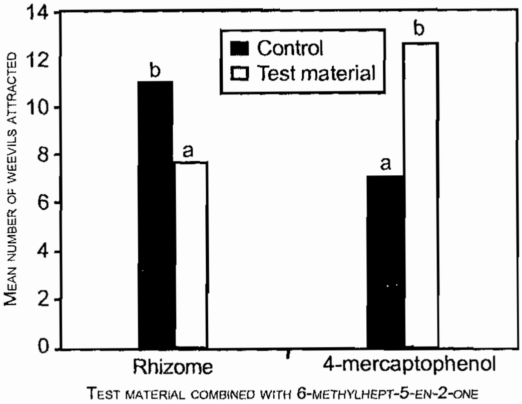

Fig. 2. The effect of 6-methylhept-5-en-2-one on the attractiveness of fresh banana rhizome and 4-mercaptophenol to the banana weevil.

Comparisons can be made only for bars in each test material. Bars are mean numbers of weevils attracted to each test chemical in eight replications. Bars for each test material superscripted with different letters are significantly different ($\chi^2_{1\,df}$, $P<0.05$). $\chi^2_{1\,df}$ = 4.25*, $\chi^2_{n-1\,df}$ = 19.37* and $\chi^2_{1\,df}$ = 13.24***, $\chi^2_{n-1\,df}$ = 4.71 ns for the combination of rhizome and 4-mercaptophenol with 6-methylhept-5-en-2-one, respectively.

palm weevils, *Ryhnchophorus pheonicis* (F.) and *R. cruentatus*, could be used in combination with host plant materials to monitor or mass-trap them.

From the foregoing, it is clear that the chemical composition of a blend or mixture and the proportions and concentrations of its constituent chemicals determine the final response that a material or semiochemical elicits in the banana weevil.

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