

PHEROMONE SHARING: BLENDS BASED  
ON THE SAME COMPOUNDS FOR  
*Euschistus heros* AND *Piezodorus guildinii*

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**Abstract**—Pheromone compounds shared between two species of pentatomids from the soybean stink bug complex are reported. Two male-specific compounds ( $R_t$  27.47 and 29.62 min) were detected in the airborne extract of *Piezodorus guildinii* by gas chromatography and mass spectrometry analysis. The compounds were identified as methyl 2,6,10-trimethyldodecanoate and methyl 2,6,10-trimethyltridecanoate.

**Key Words**—Soybean, methyl 2,6,10-trimethyldodecanoate, methyl 2,6,10-trimethyltridecanoate, stinkbugs, pheromones, *Piezodorus guildinii*.

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\*\*In memoriam.

## INTRODUCTION

Soybean, *Glycine max* (L.) Merrill, is severely attacked by complexes of stink bugs around the world. In Brazil, the most important members of the complex are *Nezara viridula* (L.), *Piezodorus guildinii* (westwood), and *Euschistus heros* (F.) (Panizzi and Rossi, 1991). This attack has led to the use of more than four million liters of chemical insecticides to control these pests (Corrêa-Ferreira and Moscardi, 1996). The use of semiochemicals in biological control of these pests would be a more ecologically benign approach to their integrated pest management.

The first attractant pheromone identified for a member of the soybean stink bug complex was from the southern green stink bug, *N. viridula* (Baker et al., 1987; Aldrich et al., 1987). Field tests of synthetic blends have been conducted on a small scale, but very few bugs or tachinid parasites were attracted (Aldrich et al., 1993).

Recently, the chemical communication system of the Neotropical brown stink bug, *E. heros*, was elucidated during trapping studies of its Nearctic relative *E. obscurus* (Borges and Aldrich, 1994; Aldrich et al., 1994). The synthesis of a racemic mixture of methyl 2,6,10-trimethyltridecanoate and stereoisomeric mixture of methyl 2,6,10-trimethyldodecanoate, two components of the male-produced pheromone of *E. heros*, was conducted by Mori and Murata (1994) and Ferreira and Zarbin (1996), respectively. In spite of the identification and synthesis process, those compounds were never tested against *E. heros*. Recently, confirmation that methyl 2,6,10-trimethyltridecanoate is a male-produced pheromone of *E. heros* was presented by Borges et al. (1998a). In recent field tests we evaluated the response of stink bugs to natural (airborne extracts) and synthetic racemic mixtures of methyl 2,6,10-trimethyltridecanoate. The presence of stink bugs and their egg parasitoids was recorded in pheromone-baited traps. During these tests, *P. guildinii* was caught in the baited traps in significantly greater numbers than other insects, i.e., *P. guildinii* comprised 63% of the species present in the baited traps, followed by *E. heros* (10%) and *N. viridula* (10%). Other species, such as *Thyanta perditor*, *Acrosternum aseadum*, and *Edessa meditabunda*, comprised less than 5% of the catch (Borges et al., 1998b).

In this paper we report the identification of methyl 2,6,10-trimethyldodecanoate and methyl 2,6,10-trimethyltridecanoate in the airborne volatiles collected from *P. guildinii* males.

## METHODS AND MATERIALS

*Insects.* *P. guildinii* adults were obtained from a soybean field (1 ha) at Cenargen (National Research Center for Genetic Resources and Biotechnology),

and their offspring were reared on sunflower seeds, soybeans, and green beans at  $26 \pm 0.5^\circ\text{C}$  and  $65 \pm 10\%$  relative humidity with a 14L:10D photoperiod. To prevent olfactory interactions between the sexes, males were separated from females after the imaginal molt and cuticular hardening, but before sexual maturity.

*Collection of Volatiles.* Samples were collected from groups of 16 virgin adult male ( $N = 80$  or female ( $N = 48$ ) *P. guildinii* of known age by coaxing the bugs into a 500-ml glass column and trapping volatiles from the air drawn by vacuum (100 ml/min) over the bugs and through 30 mg of activated charcoal for 24 hr. Trapped volatiles were eluted from the filter with 150  $\mu\text{l}$  of *n*-hexane and stored at  $-20^\circ\text{C}$  (Borges and Aldrich, 1994).

*GC-MS Analysis.* GC-MS analyses were carried out on a Shimadzu QP-5000 GC-MS spectrometer linked to a TIC detector in splitless injector mode. The DB-5 capillary column (30 m  $\times$  0.53 mm  $\times$  0.25  $\mu\text{m}$ ) was operated at  $50^\circ\text{C}$  for 1 min, increased to  $150^\circ\text{C}$  at a rate of  $5^\circ\text{C}/\text{min}$ , held at this temperature for 5 min, increased to  $280^\circ\text{C}$  at a rate of  $10^\circ\text{C}/\text{min}$ , and finally held at this temperature for 10 min.

## RESULTS

Two male-specific compounds ( $R_t$  27.47 and 29.62 min) were detected in the airborne extract of *P. guildinii*, corresponding to methyl 2,6,10-trimethyldodecanoate (**1**) (Figure 1A and B) and methyl 2,6,10-trimethyltridecanoate (**2**) (Figure 1A and C). The same compounds were previously observed in the pheromonal blend of the stink bug *E. heros* (Aldrich et al., 1994).

Identity of the two compounds was checked by coinjection with a synthetic sample prepared in our laboratory (Ferreira and Zarbin, 1996) and with one prepared by Mori and Murata (1994). Gas chromatograms of volatiles collected from female *P. guildinii* (not shown) totally lacked compounds **1** and **2**.

## DISCUSSION

Our finding of methyl 2,6,10-trimethyldodecanoate and methyl 2,6,10-trimethyltridecanoate in volatiles collected from *P. guildinii* explains by *P. guildinii* was attracted to field traps (Borges et al., 1998b) containing synthetic methyl 2,6,10-trimethyltridecanoate during pheromones studies of *E. heros*. Both compounds are present in the pheromone of *E. heros*, and both are part of the *P. guildinii* communication system. Males of *P. guildinii* release them in a 1:3 ratio of methyl 2,6,10-trimethyldodecanoate–methyl 2,6,10-trimethyltridecanoate. We have no information on the stereochemical importance of the stereogenic centers in the molecules to attraction of *P. guildinii*.

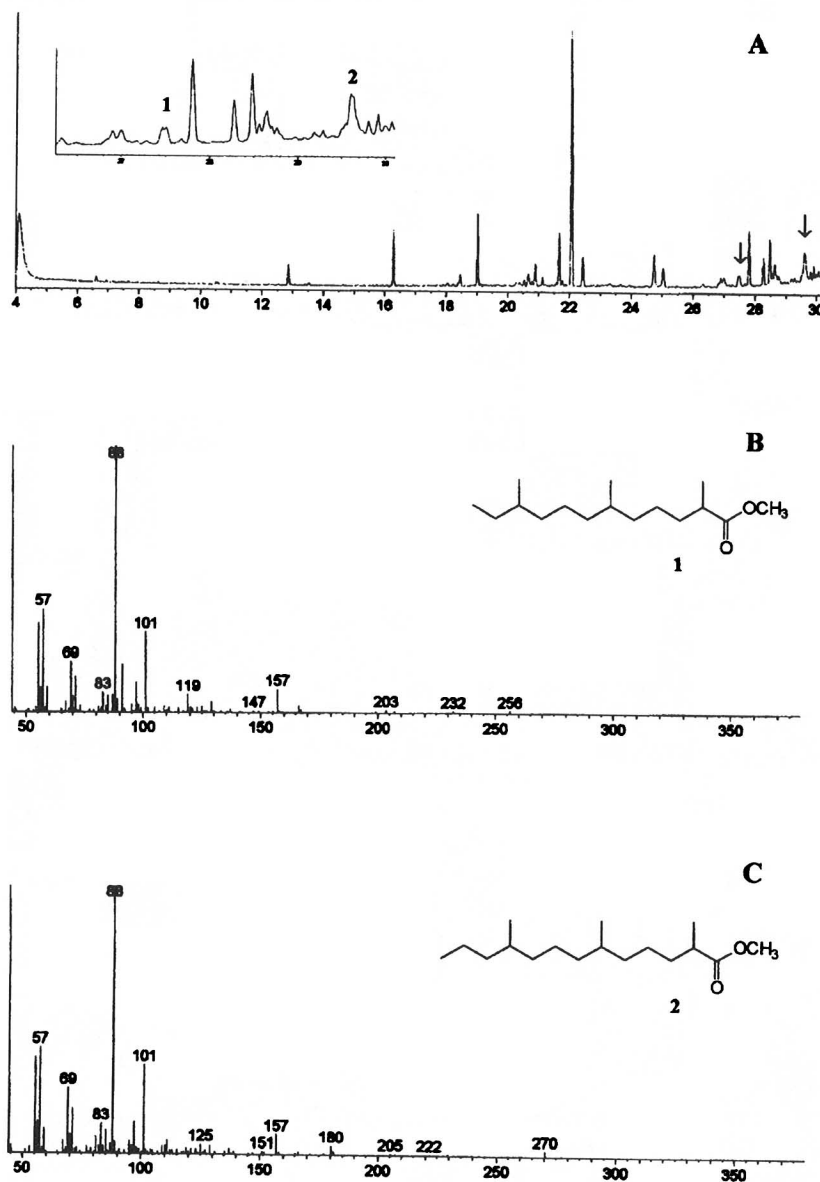


FIG. 1. GC-MS analysis of the airborne pheromone extract of *Piezodorus guildinii*. (A) chromatogram chart (arrows indicate the active compounds in the full chart. 1: methyl 2,6,10-trimethyldodecanoate, and 2: methyl 2,6,10-trimethyltridecanoate). (B) Mass spectrum of compound 1. (C) mass spectrum of compound 2.

Species-specific blends based on the same compounds among *Euschistus* species were reported by Aldrich et al. (1991). This phenomenon of pheromone sharing was also reported for *N. viridula* and the sister genus *Acrosternum* (Aldrich et al., 1993). Although the different species liberate a similar pheromone blend, unique ratios of these compounds might be involved in species isolation.

Synthetic methyl 2,6,10-trimethyltridecanoate is a very stable molecule (Prof. Kenji Mori, personal communication), and its stability may enable an integrated pest management program (IMP) to adopt another tool to control and monitor *E. heros* and *P. guildinii*. In addition, it may be possible to manipulate a stereoisomeric mixture containing methyl 2,6,10-trimethyl-tridecanoate to make a blend attractive for other species from the soybean stink bug complex.

Additional components from the *P. guildinii* pheromone blend remain to be identified. We believe, however, that these components have little or no influence in the long-distance communication behavior of this species because efficient attraction in the field is possible with the synthetic racemic mixture of methyl 2,6,10-trimethyltridecanoate (Borges et al., 1998b).

Pheromone blends based on the same compounds for two pentatomid species, *N. viridula* and species in the sister genus *Acrosternum*, were reported by Aldrich et al. (1993). In this work, we report the sharing of pheromone compounds between two species of the soybean stink bug complex in Brazil, *P. guildinii* and *E. heros*. Furthermore, we have now identified two male-released compounds from the sex pheromone of the stink bug, *P. guildinii*.

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