Journal of Chemical Ecology, Vol. 10, No. 4, 1984

VOLATILE COMPOUNDS FROM THE PREDATORY INSECT *Podisus maculiventris* (HEMIPTERA: PENTATOMIDAE):

Male and Female Metathoracic Scent Gland and Female Dorsal Abdominal Gland Secretions[1]

J.R. ALDRICH,[2] W.R. LUSBY,[2] J.P. KOCHANSKY,[2]
and C.B. ABRAMS[3]

[2]*USDA-ARS, Insect Physiology Laboratory,*
Bldg. 467, BARC-East, Beltsville, Maryland 20705
[3]*Summer Laboratory assistant, Washington University,*
St. Louis, Missouri 63130

(Received May 19, 1983; revised July 2, 1983)

Abstract—In the predatory spined soldier bug, *Podisus maculiventris*, the dorsal abdominal glands are much smaller in adult females than males. Females produce a mixture of (*E*)-2-hexenal, (*E*)-2-octenal, (*E*)-2-hexenoic acid, benzaldehyde, and nonanal in these glands. The female dorsal abdominal gland secretion may be a close-range pheromone since the dorsal abdominal gland secretion from males has been shown to be a long-range aggregation pheromone in this species. The metathoracic scent gland secretions of male and female spined soldier bugs are apparently identical, and similar to that of other pentatomids, except for the presence of the monoterpene alcohol, linalool.

Key Words—*Podisus maculiventris*, spined soldier bug, Hemiptera, Pentatomidae, allomone, pheromone, kairomone, linalool.

INTRODUCTION

Flying male and female spined soldier bugs, *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae), are attracted to a pheromone released from a pair of dorsal abdominal glands by calling males (Aldrich et al., 1984). Once sexually mature adults of this predaceous stink bug are in close proximity, the males readily recognize and court females. In addition, females of the egg

---

[1] Mention of a commercial product does not constitute an endorsement of this product by the USDA.

parasitoid, *Telenomus* n. sp. (Hymenoptera: Scelionidae), are phoretic on gravid female spined soldier bugs (Buschman and Whitcomb, 1980), but it is unknown precisely how this parasitoid locates mated female bugs.

In order to more fully understand the pheromonal system of *P. maculiventris* and the kairomonal cues of its parasitoids, we have identified the volatile components in the secretions from the small dorsal abdominal glands of adult females and the metathoracic scent gland of males and females.

METHODS AND MATERIALS

Insects and Gland Extracts. A culture of *P. maculiventris* was maintained in the laboratory on *Tenebrio molitor* L. larvae and pupae (Rainbow Mealworms, Compton, California) as previously described (Aldrich et al., 1978a). The culture was started with field-collected bugs from Geneva, New York, and augmented with field-collected bugs from Beltsville, Maryland. The insectary was kept at 28°C and 65% relative humidity, on a 16:8-hr light–dark cycle.

The dorsal abdominal glands opening between the third and fourth segments of mature adult females and the metathoracic scent glands of mature adult males and females were dissected from CO_2-anesthetized spined soldier bugs under tap water, dried with tissue paper, and immersed in 250 μl of triple-distilled CH_2Cl_2. After dissection of about 20 of each type of gland, they were macerated with a melting-point capillary tube and then removed from the solution. For one group of 15 ten-day-old female bugs, the lateral secretory tubules of the metathoracic scent gland (Everton et al., 1979) were removed and extracted separately from the median reservoir of the scent gland. Extracts of the lateral secretory tubules and the dorsal abdominal glands were concentrated under nitrogen to 25–50 μl for chemical analysis. Extracts of whole metathoracic scent glands and the sample of the median scent gland reservoir did not require concentration. Although the secretion from the dorsal abdominal glands of adult male *P. maculiventris* has been analyzed previously (Aldrich et al., 1978a), an extract of these glands was prepared as above and a gas chromatogram is included in Figure 1 for comparison to the dorsal abdominal gland secretion of adult females.

Chemical Analysis. Gas chromatography (GC) of the extracts was performed on two packed columns and one capillary column. A Packard 7400 GC equipped with a flame ionization detector was used with 1.8 m × 4 mm (ID) columns packed with either 3% SE-30 or 3% OV-275 on Chromosorb WAW (100/120). The temperature was held at 60°C for 2 min then programed to 250°C at 10°/min using nitrogen as the carrier gas. A 14-m

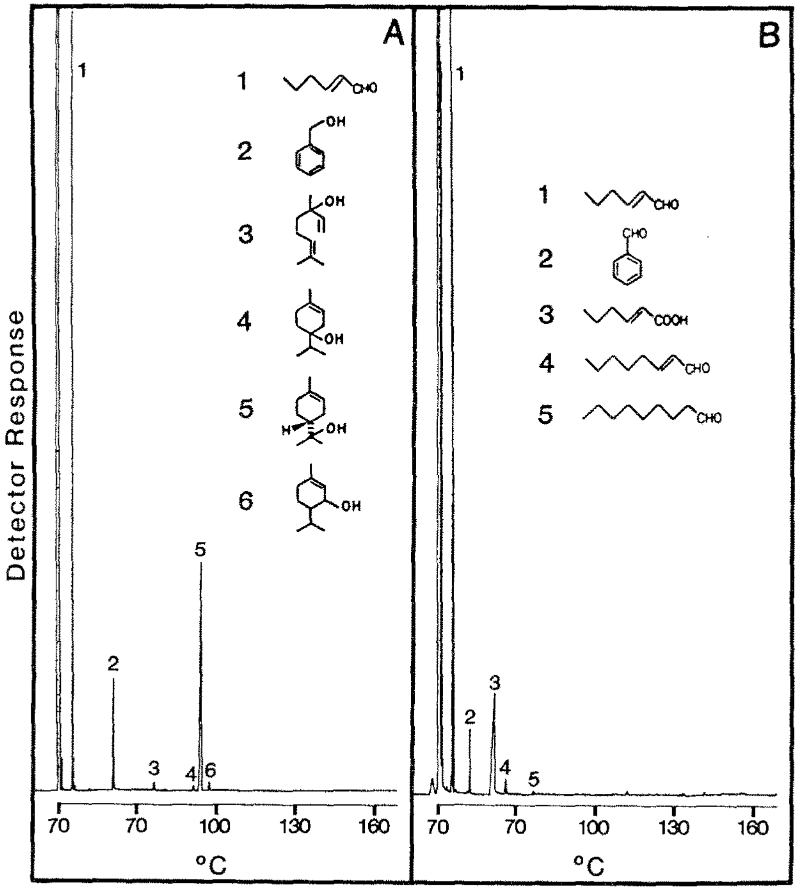

FIG. 1. Gas chromatograms of the dorsal abdominal gland secretions of *Podisus maculiventris* adult males (A) and females (B).

fused silica capillary column coated with a 0.25-μm film of DB-1^{TM} phase (J & W Scientific, Rancho Cordova, California) was used in a Varian 3700 GC equipped with a flame ionization detector. Helium, at a linear flow velocity of 40 cm/sec, was the carrier. The dorsal abdominal gland extracts and the extracts of whole metathoracic glands were run at 70° C for 2 min to 240° C at 15°/min, with a precolumn split ratio of 50:1. The lateral secretory tubule extract was run at 80° C for 2 min to 260° C at 10°/min, with a precolumn split ratio of 2.2:1. The metathoracic gland reservoir sample was run identically to the secretory tubule sample except a precolumn split ratio of 14:1 was used.

Gas chromatographic–mass spectrometric (GC-MS) analyses were conducted using a Finnigan 4500 mass spectrometer. The GC-MS data for the metathoracic scent gland extracts were obtained using a 1.8 m × 0.315 mm

(ID) column packed with 3% SE-30 on Chromosorb WAW (100/120). The packed column temperature was held at 35°C for 1 min, then programed to 60°C at 25°/min, and finally increased at 5°/min to 250°C using helium as the carrier gas. Electron impact (EI) mass spectra were collected at 70 eV with the separator at 240°C and the source at 150°C. The GC-MS data for the dorsal abdominal gland extracts were obtained using a 15-m fused silica DB-1 capillary column held for 1 min at 40°C, then programed to 285°C at 90°/min with helium as the carrier gas. The EI spectra were collected as above and, in addition, methane chemical ionization (CI) spectra were collected for the components of the female dorsal abdominal gland secretion.

Each compound was identified by comparison of its mass spectrum with the published mass spectrum and/or the mass spectrum of the authentic standard (Heller and Milne, 1978; Stenhagen et al., 1969). Subsequently, all of the compounds identified by mass spectral data were cross-checked by comparison of the GC retention of the natural product to that of an authentic standard using the capillary column under isothermal conditions. Standards of (*E*)-2-hexenal, (*E*)-2-hexenoic acid, linalool, *n*-dodecane, *n*-tridecane, *n*-pentadecane, benzaldehyde, and 1-tridecanol were purchased from Aldrich Chemical Company (Milwaukee, Wisconsin). (*E*)-2-Decenal and (*E*)-2-decenyl acetate were purchased from Bedoukian Research Inc., (Danbury, Connecticut). Nonanal and (*E*)-2-octenal were synthesized by oxidation of the corresponding alcohols purchased from Aldrich Chemical Company. (*E*)-4-Keto-2-hexenal was synthesized by oxidation of (*E*)-2-hexenal with SeO_2 in dioxane/water.

RESULTS

Dorsal Abdominal Glands. (*E*)-2-Hexenal (1) is a major component in the secretion of both male and female dorsal abdominal glands (Figure 1). The other components in the male secretion (Figure 1A) are benzyl alcohol (2), linalool (3), terpinen-4-ol (4), (+)-α-terpineol (5), and *trans*-piperitol (6) (Aldrich et al., 1978a; Aldrich et al., 1984). Females (Figure 1B) produce benzaldehyde (2), (*E*)-2-hexenoic acid (3), (*E*)-2-octenal (4), and nonanal (5) in addition to (*E*)-2-hexenal.

The dorsal abdominal glands are much larger in males (>500 μg secretion/individual) than females (<10 μg secretion/individual) (Aldrich et al., 1978a). This sexual dimorphism is substantiated by the GC traces shown in Figure 1 in that the female dorsal abdominal gland extract was first concentrated and then run at 32 × greater detector sensitivity than the unconcentrated male gland extract in order to obtain equivalent peak sizes for the two extracts.

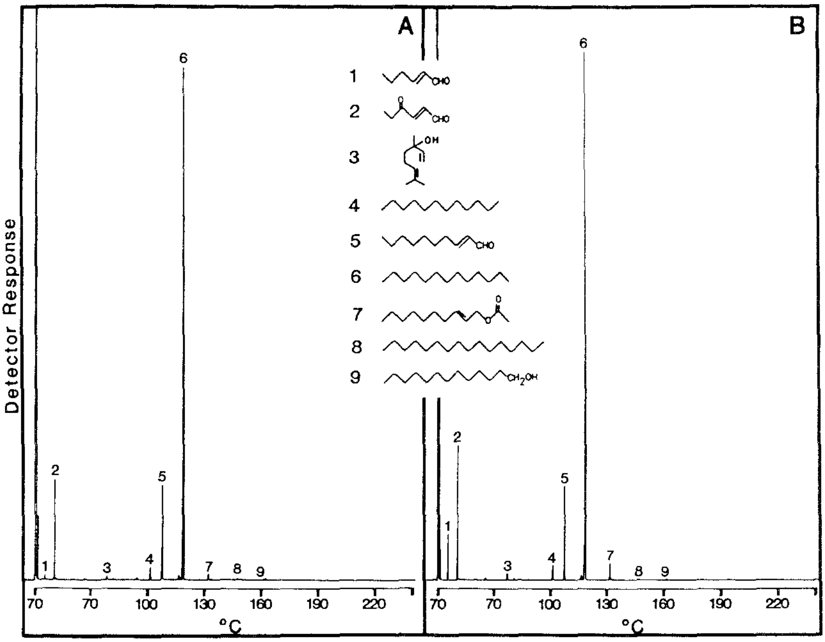

FIG. 2. Gas chromatograms of the metathoracic scent gland secretions of *Podisus maculiventris* males (A) and females (B).

Metathoracic Scent Glands. The metathoracic scent gland secretions of male and female spined soldier bugs are qualitatively and quantitatively alike (Figure 2). (*E*)-2-Hexenal appears to be somewhat more abundant in females' secretion (Figure 2B) than males' (Figure 2A), but this difference may simply be due to individual or age variations. The outstanding feature of this metathoracic scent gland secretion (Figure 2) is the presence of the monoterpene alcohol, linalool (3), in a secretion otherwise dominated by straight-chain alkanes [*n*-dodecane (4), *n*-tridecane (6), and *n*-pentadecane (8)] and α,β-unsaturated carbonyl compounds [(*E*)-2-hexenal (1), (*E*)-4-keto-2-hexenal (2), (*E*)-2-decenal (5), (*E*)-2-decenyl acetate (7)]. 1-Tridecanol (9) is also a minor component in this secretion in males and females.

The lateral secretory tubules were dissected from the median reservoir of the metathoracic scent gland for one group of sexually mature female *P. maculiventris* and the two extracts were analyzed separately by GC (Figure 3). Linalool was undetectable in the lateral secretory tubules of *P.* maculiventris (Figure 3B). On the other hand, (*E*)-2-decenyl acetate was relatively much more concentrated in the lateral secretory tubules (Figure 3B) than in the median reservoir (Figure 3A).

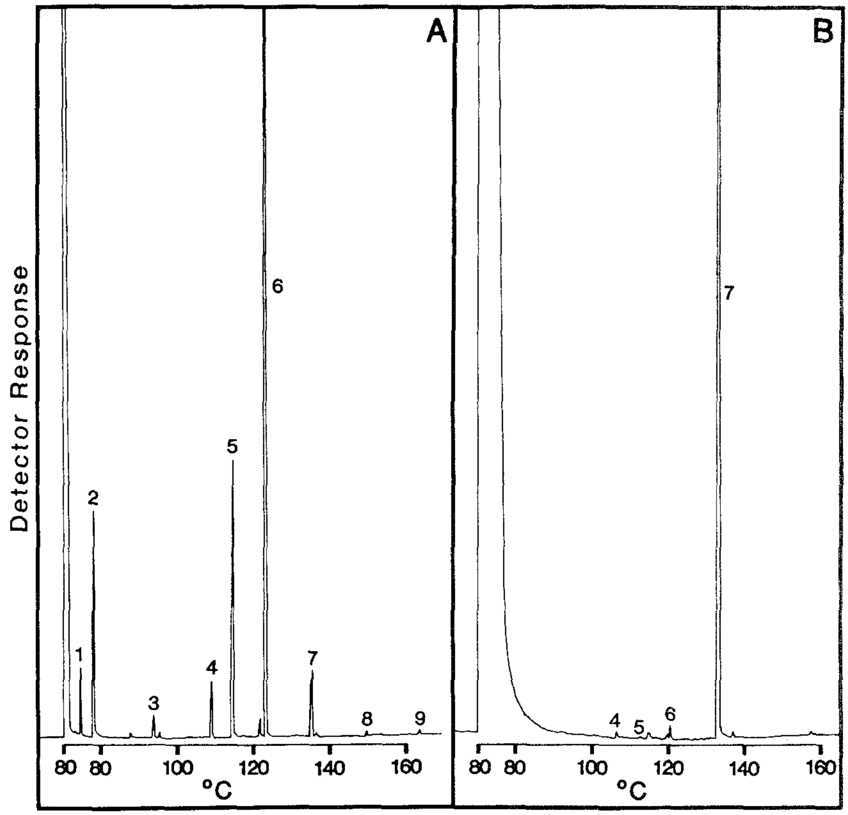

FIG. 3. Gas chromatograms of the secretions in the median reservoir of the metathoracic scent gland from female *Podisus maculiventris* (A) and the secretion in the lateral secretory tubules of these metathoracic scent glands (B). The compound numbers correspond to the structures listed in Figure 2.

DISCUSSION

Adult male and female spined soldier bugs both have synthetically active dorsal abdominal glands whose secretions are sex specific, but in males these glands contain enormously more secretion than they do in females. (*E*)-2-Hexenal occurs in the dorsal abdominal gland secretion of each sex, while all other components are unique to one or the other sex—benzaldehyde, (*E*)-2-hexenoic acid, (*E*)-2-octenal, and nonanal in the female secretion; benzyl alcohol, linalool, terpinen-4-ol, (+)-α-terpineol, and *trans*-piperitol in the male secretion (Aldrich et al., 1978a; Aldrich et al., 1984). Nonanal has not been previously found in true bugs (Staddon, 1979).

In contrast, the metathoracic scent gland secretions of male and female *P.*

maculiventis appear to be identical. The composition of this glandular secretion is similar to that of other pentatomids (e.g., Gilby and Waterhouse, 1965), except for the presence of the monoterpene alcohol, linalool. Cotton stainer bugs in the genus *Dysdercus* produce and store linalool in the lateral secretory tubules of their metathoracic scent glands (Everton et al., 1979; Daroogheh and Olagbemiro, 1982). Apparently linalool enhances the attractiveness of *Dysdercus* females to courting males (Osmani and Naidu, 1966; Hebbalkar and Sharma, 1982). In *P. maculiventris*, (*E*)-2-decenyl acetate is relatively more concentrated in the lateral secretory tubules than in the median reservoir of the metathoracic scent gland, confirming that the tubules are the site of biosynthesis for the esters in the secretion (Aldrich et al., 1978b; Everton and Staddon, 1979), but linalool does not accumulate in the lateral secretory tubules. It seems likely that linalool plays some communicative role in *P. maculiventris*. This compound is known to be a pheromonal component of certain beetles (Young et al., 1973; Hedin et al., 1974), bees (Bergstrom and Tengo, 1978; Hefetz et al., 1979), and butterflies (Hayashi et al., 1978; Honda, 1980).

Spined soldier bug males only sporadically release their aggregation pheromone and, when silent, they are essentially invisible to their parasitoids (Aldrich et al., 1984). The females produces a much fainter odor than male bugs. We suggest that this mating system may have evolved because *P. maculiventris* adults are long-lived and vulnerable to parasitism (Warren and Wallis, 1971). Evolution has favored males that endure the risk of discovery by parasitoids for the potential bonanza of mating with more and healthier females.

Acknowledgements—We wish to thank Mary Holyst and Clarence Greer for technical assistance, and Martin Weinrich and Sherry Cohen for typing the manuscript. We also thank Drs. James A. Svoboda and Mark F. Feldlaufer (Insect Physiology Laboratory, USDA), and Dr. Ralph E. Webb (Florist and Nursery Crops Laboratory, USDA) for reviewing the manuscript.

REFERENCES

Aldrich, J.R., Kochansky, J.P., and Abrams, C.B. 1984. Attractant for a beneficial insect and its parasitoids: Aggregation pheromone of the predatory spined soldier bug, *Podisus maculiventris* (Hemiptera: Pentatomidae). *Environ. Entomol.* (submitted).

Aldrich, J.R., Blum, M.S., Lloyd, H.A., and Fales, H.M. 1978a. Pentatomid natural products: Chemistry and morphology of the III–IV dorsal abdominal glands of adults. *J. Chem. Ecol.* 4:161–172.

Aldrich, J.R., Blum, M.S., Hefetz, A., Fales, H.M., Lloyd, H.A., and Roller, P. 1978b. Proteins in a nonvenomous defensive secretion: Biosynthetic significance. *Science* 201:452–454.

Bergstrom, G., and Tengo, J. 1978. Linalool in mandibular gland secretion of *Colletes* bees (Hymenoptera: Apoidea). *J. Chem. Ecol.* 4:437–449.

Buschman, L.L., and Whitcomb, W.H. 1980. Parasites of *Nezara viridula* (Hemiptera: Pentatomidae) and other Hemiptera in Florida. *Fla. Entomol.* 63:154–162.

DAROOGHEH, H., and OLAGBEMIRO, T.O. 1982. Linalool from the cotton stainer *Dysdercus superstitious* (F.) (Heteroptera: Pyrrhocoridae). *Experientia* 38:421-422.

EVERTON, I.J., and STADDON, B.W. 1979. The accessory gland and metathoracic scent gland function in *Oncopeltus fasciatus*. *J. Insect Physiol.* 25:133-141.

EVERTON, I.J., KNIGHT, D.W., and STADDON, B.W. 1979. Linalool from the metathoracic scent gland of the cotton stainer *Dysdercus intermedius* Distant (Heteroptera: Pyrrhocoridae). *Comp. Biochem. Physiol.* 63B:157-161.

GILBY, A.R., and WATERHOUSE, D.F. 1965. The composition of the scent of the green vegetable bug, *Nezara viridula*. *Proc. R. Soc.* 162B:105-120.

HAYASHI, N., KUWAHARA, Y., and KOMAE, H. 1978. The scent scale substances of male *Pieris* butterflies (*Pieria melete* and *Pieris mapi*). *Experientia* 34:684-685.

HEBBALKAR, D.S., and SHARMA, R.N. 1982. An experimental analysis of mating behavior of the bug *Dysdercus koenigii* F. (Hemiptera: Pyrrhocoridae). *Indian J. Exp. Biol.* 20:399-405.

HEDIN, P.A., GUELDNER, R.C., HENSON, R.D., and THOMPSON, A.C. 1974. Volatile constituents of male and female boll weevils and their frass. *J. Insect Physiol.* 20:2135-2142.

HEFETZ, A., BATRA, S.W.T., and BLUM, M.S. 1979. Linalool, neral and geranial in the mandibular glands of *Colletes* bees—and aggregation pheromone. *Experientia* 35:319-320.

HELLER, S.R., and MILNE, G.W.A. 1978. EPA/NIH Mass Spectral Data Base. U.S. Government Printing Office, Washington, D.C.

HONDA, K. 1980. Odor of a papilionid butterfly: Odoriferous substances emitted by *Atrophaneura alcinous alcinous* (Lepidoptera: Papilionidae). *J. Chem. Ecol.* 6:867-873.

OSMANI, Z., and NAIDU, M.B. 1966. Evidence of sex attractant in female *Dysdercus cingulatus* Fabr. *Indian J. Exp. Biol.* 5:51.

STADDON, B.W. 1979. The scent glands of Heteroptera. *Adv. Insect Physiol.* 14:351-418.

STENHAGEN, W.S., ABRAHAMSSON, S., and MCLAFFERTY, F.W. 1969. Atlas of Mass Spectral Data. Wiley, New York.

WARREN, L.O., and WALLIS, G. 1971. Biology of the spined soldier bug, *Podisus maculiventris* (Hemiptera: Pentatomidae). *J. G. Entomol. Soc.* 6:105-109.

YOUNG, J.C., BROWNLEE, R.G., RODIN, J.O., HILDEBRAND, D.N., SILVERSTEIN, R.M., WOOD, D.L., BIRCH, M.C., and BROWNE, L.E. 1973. Identification of linalool produced by two species of bark beetles of the genus *Ips*. *J. Insect Physiol.* 19:1615-1622.