

References and Notes

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18. In our initial test of this hypothesis (presented at the Annual Meeting of the Canadian Psychological Association, Vancouver, B.C., 8 to 10 June 1977) latencies to discrete presentation of the letter and pattern pairs were collected under computer control. The results from a single subject who was tested in four 8-hour sessions were consistent with those reported here.
19. Subjects recorded any thoughts they had at the end of each 15-minute period.
20. All analyses were performed on a PDP-11 computer with software provided by DEC (Sparta) or written by R.K.
21. In one method (rank test) the power spectra values (root power) for each subject were ranked in order of increasing values from 1 to 16. For each period, a mean rank across the eight subjects was computed. Mean ranks above 12.3 are larger than one would expect by chance at the .01 level (one-tailed test). In the second method (randomization test) 500 random orderings of each subject's scores for each task were subjected to Fourier analysis. For each subject, task, and period, the mean root power and standard deviation of root power of the random sequences were used to determine the probability of obtaining, by chance, a root power as large or larger than that actually observed. The probabilities for each task and period were combined across subjects according to a method developed by R. A. Fisher [see W. Wallis, *Econometrika* **10**, 229 (1942) for a discussion] to determine for the group as a whole which periods had consistently larger peaks than would be expected by chance at the .01 level.
22. For each subject, the phase difference between the approximately 96-minute oscillations in spatial and verbal performance is represented on the circumference of the unit circle. Each of these points was treated as the end of a vector from the center, and their averaged vector sum (arrow in Fig. 1C) was computed. In this case, the length exceeds that required for significance at the .01 level (inner circle of Fig. 1C) [J. Greenwood and D. Durand, *Ann. Math. Stat.* **26**, 233 (1955)].
23. For each subject, the phase of the oscillations in the difference scores (verbal minus spatial performance) was used to determine the amount and direction of shift, which was then applied to both tasks. A three-point smooth, $0.25(i-1) + 0.5(i) + 0.25(i+1)$, was used to eliminate high-frequency oscillations and noise. Overhanging points (those contributed by fewer than eight subjects) have been dropped from Fig. 1D.
24. Complete verification of the proposal outlined by Broughton (7) requires the demonstration that these daytime oscillations in performance reflect changes in hemispheric functioning and that they bear an appropriate phase relation to the alternation between REM and NREM stages of sleep.
25. We thank B. Rusak for his comments and encouragement, R. Rodger for his advice, and the National Research Council of Canada for its financial support of this research.

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Trace Chemicals: The Essence of Sexual Communication Systems in *Heliothis* Species

Abstract. Analysis of heptane-soluble compounds from ovipositors of *Heliothis zea* and *Heliothis virescens* shows that both species produce relatively large amounts of (Z)-11-hexadecenal, with traces of (Z)-9-hexadecenal, (Z)-7-hexadecenal, and hexadecanal. *Heliothis virescens* females differ from *Heliothis zea* in that they also produce trace amounts of tetradecanal, (Z)-9-tetradecenal, and (Z)-11-hexadecenal-1-ol. In both species, trace compounds are essential to pheromonal activity and specificity of chemical signals.

The moths of *Heliothis zea* (Boddie) and *H. virescens* (F.) are among the most devastating pest insects of American agriculture. The species infest a wide range of crops, including cotton, corn, soybeans, sorghum, peanuts, tobacco, tomatoes, cabbage, and lettuce. We report the identification of trace chemicals that

heretofore have gone undetected in females of these pests and show that the newly discovered compounds are essential elements in the chemical signaling systems of these species.

The sex pheromones of *H. zea* and *H. virescens* have been studied by others (1-4). Females of *H. zea* have been

Table 1. Percentage composition of heptane washes from seven *Heliothis zea* and six *H. virescens* female ovipositors determined by capillary chromatographic analysis. Mean values and the range (in parentheses) of analytical values are shown.

Component	$\bar{X}$ percentage (range)	
	<i>H. zea</i>	<i>H. virescens</i>
(Z)-7-Hexadecenal	1.1 (0.4 to 1.9)	1.0 (0.1 to 1.56)
(Z)-9-Hexadecenal	1.7 (1.1 to 2.4)	1.3 (0.3 to 2.34)
(Z)-11-Hexadecenal	92.4 (89.8 to 95.7)	81.4 (76.8 to 91.1)
Hexadecanal	4.4 (1.8 to 7.1)	9.5 (3.0 to 19.0)
(Z)-9-Tetradecenal		2.0 (0.7 to 3.1)
Tetradecanal		1.6 (0.7 to 2.7)
(Z)-11-Hexadecenal-1-ol		3.2 (0.9 to 4.5)

Table 2. Responses of male *Heliothis zea* and *H. virescens* to synthetic compounds and to virgin females of the respective species (8). Means followed by the same letter are not statistically different from each other according to Duncan's multiple range test ($P = .05$).

Stimulus in trap	Males per trap per night	
	<i>H. zea</i>	<i>H. virescens</i>
<i>Test 1</i>		
115.5 μ g of (Z)-11-hexadecenal	0.3 b	0
133.6 μ g of mixture A*	11.7 a	0
<i>Heliothis zea</i> (four females)	12.0 a	0
<i>Test 2</i>		
115.5 μ g of (Z)-11-hexadecenal + 6.9 μ g of (Z)-9-tetradecenal	0	7.8 c
151.8 μ g of mixture B*	0	38.1 a
<i>Heliothis virescens</i> (four females)	0	21.7 b

*Mixture A consisted of 2.6 μ g of (Z)-7-hexadecenal, 4.5 μ g of (Z)-9-hexadecenal, 11.0 μ g of hexadecanal, and 115.5 μ g of (Z)-11-hexadecenal. Mixture B contained the same chemicals plus 6.9 μ g of (Z)-9-tetradecenal, 2.3 μ g of tetradecanal, and 9.0 μ g of (Z)-11-hexadecenal-1-ol.

shown to produce (Z)-11-hexadecenal (1, 2), but this compound lacks the biological activity characteristic of an insect pheromone and does not lure males to traps. Female *H. virescens* also produce (Z)-11-hexadecenal (2, 3), but in this species a small amount of (Z)-9-tetradecenal is also produced. The binary mixture [(Z)-11-hexadecenal and (Z)-9-tetradecenal, 16:1] was sufficiently attractive to be useful as a trap lure, but male responsiveness to the 16:1 synthetic mixture was not always equivalent to the responsiveness to crude solvent extracts of virgin females (3). Tumlinson *et al.* therefore speculated (3) that additional components might be produced by *H. virescens* females.

In *H. zea* and *H. virescens*, female sex pheromones are secreted by glandular cells situated in the intersegmental membrane (5) between the eighth and ninth abdominal segments that make up the ovipositor. The ovipositor is usually telescoped into the seventh abdominal segment. However, when the female emits pheromone, the ovipositor is extended, and male-attractive chemicals evaporate from its surface.

We analyzed the heptane washes of single ovipositors (6) for volatile components by glass open-tubular capillary chromatography (7). Repetitive analyses of washes from individual *H. zea* and *H. virescens* showed that the material from females of each species had reproducible but distinct chromatographic profiles (Fig. 1, A and B). Although quantitative variations between individual females were observed, four chromatographic components were consistently detected in washes from *H. zea* and seven components were found in those from *H. virescens*. The retention times of components 1 to 4 (Fig. 1), which were detected in both species, were coincident with the retention times of authentic (Z)-7-hexadecenal, (Z)-9-hexadecenal, (Z)-11-hexadecenal, and hexadecanal, respectively, on both polar and nonpolar capillary columns. The mass spectra (8) of the respective components were also identical to those of the authentic samples (9). Heptane washes of *H. virescens* differed qualitatively from *H. zea* washes in that they contained components 5 to 7 (Fig. 1B). These components proved to be (Z)-9-tetradecenal, tetradecanal, and (Z)-11-hexadecen-1-ol, respectively, according to chromatographic and mass spectral characterization (8). The data in Table 1 show that there was considerable intraspecific quantitative variation in the composition of the wash components from one female to another.

The most abundant component of heptane washes of both *H. zea* and *H. virescens* ovipositors was (Z)-11-hexadecenal (about 10 to 20 ng per female). Field bioassays (10) subsequently showed that the trace compounds we detected in the

respective insects are essential to the chemical signaling systems of the two species (Table 2). (Z)-11-Hexadecenal by itself was ineffective as a lure for males of either insect, and a mixture of (Z)-9-tetradecenal and (Z)-11-hexadecenal was only weakly attractive to *H. virescens* males.

In contrast, when the full chemical complements identified from the *H. zea* and *H. virescens* females were deployed in traps, male responses equaled or exceeded those elicited by virgin females of the respective species. The males were specifically attracted only to the synthetic mixture that was representative of their own species, even though both species were present in the fields. Inasmuch as washes from the *H. virescens* ovipositor differ from those of *H. zea* by the presence of (Z)-9-tetradecenal, tetradecanal, and (Z)-11-hexadecen-1-ol (Fig. 1, components 5 to 7), the observed specificity of pheromonal signals must result from the presence of one or more of these three compounds. We do not fully understand the ethological roles of all compounds produced by *H. zea* and *H. virescens* females; however, indications are that (Z)-11-hexadecenal, (Z)-9-hexadecenal, (Z)-7-hexadecenal, and hexadecanal (Fig. 1, components 1 to 4) represent a congeneric biochemical theme. Whether other species of *Heliothis* also reflect this theme and which compounds in the set might have pheromonal function in one species but not in another remain to be determined.

No effective synthetic male lure has been available for monitoring *H. zea* populations. However, appropriate combinations of the trace compounds that we identified from *H. zea* and *H. virescens* with the principal pheromone component, (Z)-11-hexadecenal, make for effective male attractants for both species. In case of *H. virescens*, the new lure is at least five times more effective than the one previously available.

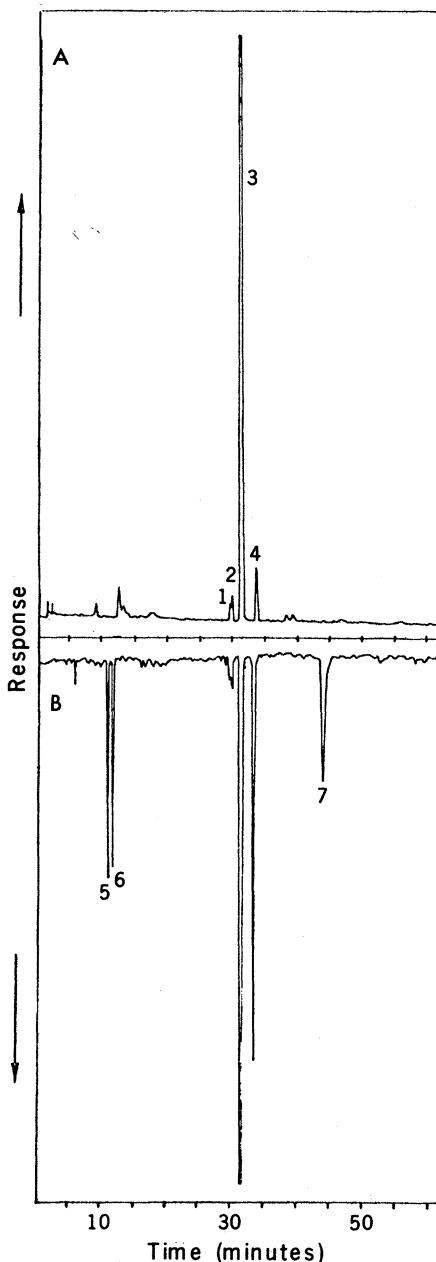


Fig. 1. Gas chromatograms of heptane washes of ovipositors of individual female *Heliothis zea* (A) and *H. virescens* (B). Chromatograms [60 m by 0.25 mm (inside diameter) SP 2100 glass open-tubular capillary column] of the washes from respective species are displayed as mirror images to accentuate the qualitative differences. 1, (Z)-7-hexadecenal; 2, (Z)-9-hexadecenal; 3, (Z)-11-hexadecenal; 4, hexadecanal; 5, (Z)-9-tetradecenal; 6, tetradecanal; and 7, (Z)-11-hexadecen-1-ol. The major chromatographic component 3 was always well off printer-plotter scale. Other unnumbered trace chromatographic peaks in (A) and (B) are unknowns that did not appear with regularity in washes from all females.

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7. The gas chromatograph (Hewlett-Packard model 5840A) was equipped with a splitless injector system and a flame ionization detector. Apolar (SP2100) and polar (SP1000) 60 m by 0.25 mm (inside diameter) glass open tubular capillary columns were used. Conditions of the chromatography were: helium flow 2 cm³/min at 120°C; injector, 225°C; injector purge, 1.1 minute after injection; and temperature programmed 120°C at injection and held for 2 minutes followed by heating at 30°C per minute to 180°C (SP 1000 column) or 200°C (SP2100 column).
8. Combined glass open tubular capillary chromatography-mass spectrometry (GOTC-MS) studies of washings of the ovipositor from *H. zea* and *H. virescens* were conducted with a Finnigan model 4000 mass spectrometer equipped with Finnigan model 6110 data system. Heptane-soluble components were also epoxidized and studied by GOTC-MS to establish the position of unsaturation in the carbon chain of the olefinic compounds. The geometry of the olefins was determined by capillary chromatography retention data (J. A. Klun *et al.*, in preparation; J. A. Klun *et al.*, *J. Chem. Ecol.*, in press).
9. All Z olefinic compounds, except (Z)-9-hexadecenal, were prepared by the reaction of lithium acetylides with tetrahydropyranyl ethers of haloalcohols in tetrahydrofuran-hexamethylphosphorotriamide [M. Schwarz and R. M. Waters, *Synthesis* **10**, 567 (1972)]; hydrogenation to the Z olefin followed the method of C. A. Brown and V. K. Ahuja [*J. Org. Chem.* **38**, 2226 (1973)], and oxidation of the resulting olefinic alcohols to aldehydes was by the method of R. W. Ratcliffe [*Org. Synth.* **55**, 84 (1976)]. (Z)-9-Hexadecenal was prepared by reduction of methyl palmitoleate (Tridom Chemicals Inc., Hauppauge, N.Y. 11787) with LiAlH₄ and subsequent oxidation to the aldehyde. Hexadecenal was prepared by oxidation of hexadecan-1-ol (Aldrich Chemical Co., Inc., Milwaukee, Wis. 53233) and tetradecanal was purchased from Aldrich Chemical Co., Inc. The compounds were purified first by high-pressure liquid chromatography on AgNO₃-impregnated silica or silica, with toluene as eluent, and subsequent gas-liquid chromatography showed purity > 99 percent.
10. Field tests were conducted in cotton fields near Tifton, Ga. Insect traps were baited nightly with cotton dental rolls (test 1) or cigarette filters (test 2) that were freshly treated with heptane solution containing test chemicals plus 5 μ g 2,6-bis(1,1-dimethylethyl)-4-methylphenol, as antioxidant. Virgin females (5) used in the tests were 24 to 72 hours old. In test 1, three wind-vane-type insect traps [U.S. Dep. Agri. ARS-S-173 (1978)] were used. The test was conducted in a complete randomized-block design. Treatment locations in the field were randomized nightly, and the test was replicated over six consecutive nights. In test 2, three electric grid traps [U.S. Dep. Agri. ARS-S-42-3-1, (1963)] were used. The test was conducted over 25 nights, and treatment locations in the field were randomized every 5 days.
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Development of the Capacity for Tactile

Information Transfer Between Hemispheres in Normal Children

Abstract. *The hypothesis of less direct interaction between hemispheres in young children was supported by a behavioral test. Fabric samples were compared with either the same hand (same hemisphere) or with opposite hands (between hemispheres). Crossed errors were a significantly larger proportion of total errors in 3-year-olds than in 5-year-olds.*

Interaction between the two halves of the brain is mediated in part by the neocortical commissures. Developmental studies may offer a key to understanding hemispheric cooperation and conflict because the corpus callosum and other commissures are not completely formed at birth and mature very slowly. In very young children, therefore, there may be little communication between the hemispheres; they may each function relatively independently as in adult "split-brain" patients who have had the connections surgically severed (1-3).

The sequence of myelination in a given pathway has been taken as an index of the sequence of functional maturity. The corpus callosum is one of the last systems to begin and to complete myelination. In humans, myelination of the corpus callosum does not begin until the end of the first year; it is reported to be substantially advanced by age 4 and continues to increase to age 10 and beyond (4, 5). Thus, the principal anatomical substrate for interhemispheric integration develops only slowly over the first decade of life. Recent electrophysiological evidence also supports this view (6).

Different functional components in the neocortical commissures may myelinate at different times, some callosal functions being fully established while others are still completely undeveloped. It is therefore important to test the behavior of young children and establish which aspects of hemispheric integration are functionally mature at different ages.

Since each hemisphere receives sensory input primarily from one side of the body or one visual half-field, an important commissural function is the simple transfer of sensory information between hemispheres, which makes possible a world picture integrated across the midline. For example, an inability to perform crossed tactile matching has been demonstrated as an enduring effect of surgical disconnection of the commissures (1, 7). We have investigated the development of commissural transfer in young children with a simple texture discrimination test. Tactile matching with the same hand (same hemisphere) and between hands (between hemispheres)

was compared in 3- and 5-year-olds. We predicted that the 3-year-olds would show more frequent errors in the crossed condition than in the uncrossed, and that 5-year-olds would show less if any difference between crossed and uncrossed errors.

Fifteen 3-year-old and 15 5-year-old right-handed girls (8) were tested. Approximately half of each age group was black and half white. The children were drawn from several San Francisco nursery schools including a working-class day-care center and an upper-middle-class private school. Handedness was assessed by asking the child to demonstrate the use of a pencil, spoon, comb, hammer, toothbrush; to pick up a coin; and to throw a ball. Right-handed responses were required on at least five of the seven items, and had to include right-handed use of the pencil. Screening information was gathered from parents and teachers; children who did not speak English at home or who had neurological, cognitive, or emotional problems were disqualified.

In testing very young children it is important to ensure that they understand and can comply with the demands of the task. We therefore developed screening and training procedures for the 3-year-olds to be certain that they had developed the concept of "same" and "different" and that they could and would use these verbal responses reliably (9).

Cotton-filled pillows approximately 2 inches square were covered with fabrics of various textures—linen, denim, rayon, rough wool, and so forth—and divided into sets of four different pillows each. In one set, each fabric was quite different from the others ("easy"), in one set they were quite similar ("difficult"), and the other two sets were "medium easy" and "medium difficult."

In order to eliminate ceiling effects, we tested each child to find a set of fabrics which, for her, elicited 15 to 30 percent errors in the uncrossed condition. This procedure left room to observe an excess of crossed errors (maximum = 50 percent). Thus, different materials were used for different children.

At the beginning of each tactile testing