BIOLOGY

Unique Synergistic Effects produced by the Principal Sex Attractant Compounds of *Ips confusus* (LeConte) (Coleoptera: Scolytidae)

THERE is a powerful assembling scent or sex attractant in the frass produced by the male of *Ips confusus* boring in ponderosa pine (*Pinus ponderosa*)[1]. These attractants evoke the concentration flight or mass attack by individuals of both sexes[2], but the females are much more responsive than the males[3,4]. When the insects arrive at the source of the attractant, they take part in boring, feeding, mating and oviposition.

Recently, three terpene alcohols that are believed to be the principal components of the attractant were isolated from male frass[5] and identified[6,7] and synthesized[7]. These compounds are:

I: (−)-2-methyl-6-methylene-7-octen-4-ol

II: (+)-cis-verbenol

III: (+)-2-methyl-6-methylene-2,7-octadien-4-ol

In a laboratory bioassay, a mixture of compound I with either II or III evoked a response from female beetles[7], as did a mixture of all three compounds. The individual compounds were not attractive.

Field tests of isolated compounds I and III and synthetic compound II were begun on June 9, 1966, at the University of California Blodgett Research Forest, Eldorado County, California. The three compounds, individually and in various combinations, were presented together with a benzene extract of frass to flying populations on packets of filter paper placed on aluminium foil in the centre of 1 ft.^{2} masonite boards covered with 'Stickum Special'. Attractive male frass was also presented in the same way by placing it under screens of plastic mesh in Petri dishes. The packets and frass were renewed at hourly intervals to compensate for the loss of attractants by volatilization and for variations in the diurnal flight of the beetles. The boards were spaced about 25 ft. apart with no particular attention to exposure. A minimum of two replications was used for each test. All materials were presented in the amounts found in 1 g of male frass (I, 125 μg; II, 2·5 μg; III, 12·5 μg).

The combination of compounds I, II and III evoked the most potent response; the entire catch (fourteen males and eighteen females) occurred within about 20 min on the one day it was tested. Compounds I and III together attracted four females only. The beetles attracted to frass (fourteen males and forty-five females) and frass extract (twelve males and twenty-one females) were trapped over a period of 2 days. Compounds presented individually and in other combinations failed to attract a single specimen of *I. confusus*. Surprisingly, *Ips latidens* (LeConte) was attracted to compound I alone (five females) and to a combination of I and II (three males and thirty-four females). No bark beetles were trapped on the blank control boards.

After this unexpected development, *I. latidens* was exposed to all the synthetic compounds both individually and in combination, in the laboratory olfactometer. Compound I alone (20 μg), and in combination with II, definitely affected the behaviour of both sexes. The response to compound I alone is best described as an arrestant; the beetles circled very slowly in the air stream and some males eventually traversed to the receptacle containing the attractant compound. The characteristic klinotaxis[5] was not observed, however. Compounds I and II (2·0 : 0·1 μg) did evoke the typical klinotaxis, moderately for the female, weakly for the male. No response was elicited by the combination of I and III and by III alone. Compound II alone and II with III at 10 μg each evoked at most a weak response. But when 0·1 μg of compound III was added to 2·0 μg of I and 0·1 μg of II, the attraction for *I. latidens* was eliminated. This combination proved highly attractive to *I. confusus*, however, even at one-tenth this concentration.

The unusual synergistic system obtained by adding compound III to the combination of I and II, which blocked or masked the *I. latidens* attractant and created the *I. confusus* attractant, poses some challenging chemical and biological problems. We are at present comparing the chemical fractions of *I. latidens* frass with those of *I. confusus*.

These studies demonstrate the deviations between laboratory and field evaluations and emphasize the necessity of using both bioassays to identify pheromones.

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Volatility of Trail Marking Substance of the Town Ant

WE have found that the trail marking substance laid down by the town ant, *Atta texana* (Buckley), contains at least two components, one volatile and the other non-volatile; both are followed by workers. Both components partition into the organic phase of a methylene chloride water system.

Poison sacs from five medium-sized workers were crushed in 1 ml. of methylene chloride, and the solutions were dispensed with a pipette on circles 14 cm in diameter drawn on smooth cardboard. Polyethylene plastic sheets 0·13 mm thick were placed 0·1, 1, 2 or 4 cm over these artificial trails on solid circumferential spacers, thereby creating a closed air space between trail and plastic sheet. Holes were punched in the sheet directly above the trail with a No. 1 insect pin. For most tests, eight holes per cm were punched in each of two parallel rows 1·5 mm apart (about the distance between antennae of a minor worker).

In each test, five minor workers were released on the plastic sheet in the centre of the circle. After 10 min, actions of ants that responded were classed as (1) circling, when at least one worker travelled the entire circumference of the circle (44 cm); (2) partial circling, when a worker travelled only part of the circumference; and (3) detecting only, when workers briefly oriented toward a hole but did not follow the trail. The last two conditions were accompanied by zigzagging as the ants alternately found and lost the scent.

Height above the trail, distance between holes, and trail age affected insect behaviour. The ants circled as high as