The Significance of Blends of Two Sex Pheromone Components on the Behavior of Male *Spodoptera litura* F. (Lepidoptera: Noctuidae)

Kenjiro KAWASAKI

*National Institute of Agro-Environmental Sciences*

Tsukuba, Ibaraki, 305 Japan

Abstract — Behavioral responses of male *Spodoptera litura* to the female sex pheromone components, (Z, E)-9, 11-tetradecadienyl acetate (compound A) and (Z, E)-9, 12-tetradecadienyl acetate (compound B), and mixtures of compounds A and B were analyzed in the laboratory. Male orientation flight in a wind tunnel was induced by a lure dispenser onto which 1.1x10^-3 ng of a mixture (A:B=10:1) was absorbed, while 1 ng of compound A was required to induce the same level of behavior. The blend ratio was important for trap catch and orientation flight of males and a 10:1 mixture of compounds A and B was most effective. A mixture (10:1) was more effective than compound A alone in induction of movement, walking, and flying in resting males in a glass tube. These results indicated that 2 sex pheromone components act as a set from the initial to final steps of male attraction to female.

The common army worm moth, *Spodoptera litura* F. (Lepidoptera: Noctuidae) is known as a pest of vegetables in Japan. Its sex pheromone was identified as a mixture of (Z,E)-9, 11-tetradecadienyl acetate (Compound A) and (Z,E)-9, 12-tetradecadienyl acetate (Compound B) and the blend ratio of compounds A and B in extracts of females is 9:1 (Tamaki et al. 1973). Mixtures of these chemicals ranging from 8:2 to 39:1 showed equally high attractiveness in a field trapping test (Yushima et al. 1974).

Analyzing the attraction process of males to females, Nakamura (1976) proposed the hypothesis of “double active space”, which is created by different functions of compounds A and B, in which compound A determines a long range active space (Nakamura & Kawasaki 1977) and compound B determines a close range active space when blended with compound A (Nakamura 1979, 1981). Kawasaki (1981) observed flight tracks of males to various blends of the sex pheromone sources in a field cage and concluded that a blend of compounds A and B is an essential factor in inducing upwind and searching flight near the sex pheromone source. However, male orientation behavior at long distances from the sex pheromone source was not observed and the “double active space” hypothesis was not confirmed by direct observation. As it is difficult to observe moth behavior far from the sex pheromone source in the field, the responses of males to each compound and their mixture in low concentration was observed using a wind tunnel and a glass tube.

Materials and Methods

Insects were reared on an artificial diet (Fujjie & Miyashita 1973) at 25°C under a 16L8D condition. After pupation, male pupae were kept at 20°C under continuous light and emerged moths were kept under the same conditions till the experiment. Chemicals were purified by column chromatography or high performance liquid chromatography. Purities of compounds A and B were checked by capillary GLC and were 95.3 and 96.3%, respectively. Most of the impurities were geometrical isomers of each compound. Compounds A and B were serially diluted by n-hexane and adsorbed in a polyethylene capsule separately or as a mixture. After aging for 3 days in a draft chamber, the capsules were kept in a freezer till used for experiments.

Male responses to various amounts of a 10:1 mixture of compounds A and B, individual compound A or B, and various blend ratios of both compounds were examined in a wind tunnel. The method of wind tunnel study was the same as reported in Kawasaki (1985). The size of the wind tunnel was 1 x 1 x 3 m and light intensity and wind speed were controlled at 0.1 lux and 0.5 m/s, respectively. Temperature ranged from 20 to 23°C. Each group of 15 male was kept in a glass container and moved into the dark. Experiments were conducted from 2 to 8 h after the moths were moved into the dark. A lure source was set above a water pan trap 12 cm in diameter at 50 cm from the air intake end. Moths which showed orientation flight within 50 cm of the...
lure source was counted as "orientated". In this experiment, orientation flight means slowdown of flight speed accompanied by casting or hovering flight in a sex pheromone plume downwind of a sex pheromone source. Fifteen males kept in a glass container were released as a group 50 cm from the exhaust end and this was repeated at least 3 times. If a male was not caught in a trap during the first orientation flight, it was possible that the male was counted 2 or more times as orientated. Observation was continued for 10 min at which time the number of trapped males was counted. A male orientation flight was sometimes interfered with by other males in the case of a lure source which showed high attractiveness, however, the male resumed orientation flight immediately after interference in most cases.

The effect of low amounts of compounds A or A+B on a resting male was observed in a glass tube 7.5 cm in diameter and 70 cm long. A fan fixed to a box which was connected to one end of the glass tube generated a constant air flow of 0.5 m/s in the glass tube. Before each experiment, a male was put in a plastic petri dish (3.5 cm x 1 cm) and kept in a dark condition. The moth was used for experiment 2 to 6 h after the transfer. A capsule of sex pheromone was put into the middle of the tube and then a male in a plastic petri dish was put into the tube about 5 cm from the exhaust end. Generally, a resting male shows the following behaviors before the start of flight behavior: some movements such as slightly stretching the legs or slightly and slowly swinging the body (movement [MO]), then beginning wing vibration (WV), walking (WA) and then flying (FL). The time required to induce movement, wing vibration, walking, and flying was scored. Observation was continued for 10 min unless the male did not start FL. The air containing sex pheromone was exhausted from the draft chamber. Illumination was by a photographic safe light bulb and light intensity was controlled at 0.3 lux by a transformer. Temperature ranged from 20°C to 23°C.

Results

I. Wind Tunnel Test

Effect of the Amount of a 10:1 Mixture

Response of males increased with an increase in the amount of a 10:1 mixture of compounds A and B (Fig. 1). Trap catch clearly occurred with $1 \times 10^{-4}$ ng and increased with the amount of chemical till 10 ng. Trap efficiency (no. of males caught in the trap / no. of males orientated x 100 [%]) ranged 45.8% - 51.9% within the concentration range from $1 \times 10^{-5}$ to $1 \times 10^{-1}$ ng, and 60.5% - 68.8% in the range from 1 to $1 \times 10^{-4}$ ng. This result suggests that trap efficiency was lower in the low concentration range than in the higher concentration range.

![Fig. 1. Effect of the amount of chemicals on trap catch and orientation flight of males in a wind tunnel. Blend ratio of compounds A and B was fixed at 10:1 and the amount is expressed by that of compound A. Circles and triangles indicate the total number of males captured and orientated, respectively. The standard number of males released was 45. If the total number released was not 45, the numbers were adjusted. Circles or triangles with same letters do not differ significantly by Duncan's New Multiple Range Test (P $\leq$ 0.05).](image-url)