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EFFECT OF EXOGENOUS JUVENILE HORMONE ON PHEROMONE TITER OF THE SMALLER TEA TORTRIX MOTH, *ADOXOPHYES* SP. (LEPIDOPTERA: TORTRICIDAE)[1]

RONG KOU[2], DING-SUI TANG and YING-SHING CHOW

Institute of Zoology, Academia Sinica,
Nankang, Taipei, Taiwan 11529,
Republic of China

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Rong Kou, Ding-Sui Tang and Ying-Shing Chow (1991) Effect of exogenous juvenile hormone on pheromone titer of the smaller tea tortrix moth, *Adoxophyes* sp. (Lepidoptera: Tortricidae). *Bull. Inst. Zool., Academia Sinica* **30**(4): 311-317. The juvenile hormone III (JH-III), when applied exogenously with 1 μg or 10 μg to day-1 virgin females of the smaller tea tortrix moth, *Adoxophyes* sp., significantly reduced pheromone titer at 44 h and 68 h after treatment. Exogenous treatment with 10 μg JH-III to day-3 virgin females also reduced pheromone titer at 20 h after treatment. However, a similar compound, juvenile hormone I (JH-I), had no effect on pheromone titer of the species at any of the concentrations tested.

Key words: Juvenile hormone I, Juvenile hormone III, Smaller tea tortrix moth, *Adoxophyes* sp., Pheromone titer.

Early studies of the neuroendocrine regulation of sex pheromone production indicated that the presence of corpora allata (CA), the source of juvenile hormone (JH), was essential for pheromone production and release in the long-living cockroach *Byrsotria fumigata* (Guerin), but that pheromone release was unaffected by CA removal in the short-living saturniid *Antheraea pernyi* Guerin and pyralid *Galleria mellonella* (L.) (Barth, 1961, 1965). Subsequent studies dealing with other species of moths (*Antheraea polyphemus* (Cramer), *Antheraea pernyi* and *Hyalophora cecropia* (L.), (Riddiford and Williams, 1971; Sasaki *et al.*, 1983), *Lymantria dispar* (L.) (Hollander and Yin, 1982, 1985; Tang *et al.*, 1987), *Manduca sexta* (L.) (Itagaki and Conner, 1986), *Utetheisa ornatrix* (L.) (Itagkaki and Conner, 1987) yielded similar results. Although recently a peptide hormone (pheromone-biosynthesis-activating neuropeptide, PBAN), produced by the brain-suboesophageal-ganglion (SOG) complex of several species of moths, induced pheromone biosynthesis when injected into neck-ligated *Heliothis zea* (Boddie) females (Raina *et al.*, 1986, 1987; Jaffe *et al.*, 1986), juvenile hormone is still shown to be essential to the initiation of both calling behavior and pheromone production in females of *Pseudaletia unipuncta* (Haworth) (Cusson and McNeil, 1989). In order to understand the role of JH, exogenous JH analog (ZR-512) had been

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2. To whom reprint request should be sent.

applied to the omnivorous leafroller moth, *Platynota stultana* (Walsingham), and the pheromone production in virgin females was blocked (Webster and Carde, 1984). On the other hand, incorporation of 1.0 ppm JH-III into the diet of adult cotton boll weevil, *Anthonomus grandis* Boheman, increased the biosynthesis of its 4 pheromone compounds by 3 times (Hedin *et al.*, 1982).

In this study, we investigate the effect of exogenous juvenile hormone on pheromone titer in the smaller tea tortrix moth, *Adoxophyes* sp., and propose a possible role for JH in its sex pheromone relationship in this moth.

MATERIALS AND METHODS

Insect

Female pupae of the smaller tea tortrix moth used in this study were sexed by and obtained from the Taiwan Tea Experiment Station, Hsinchu, Taiwan. They were maintained at 24-26°C under a 14:10 (L:D) photoperiod. Emerged moths were fed with 10% aqueous sucrose solution and maintained at the same conditions as the pupae. Only adults emerging just after the onset of a photophase were used in this test.

Operations

Juvenile hormone I and III (Sigma Chemical Company, St. Louis, Missouri, U.S.A.) were dissolved in acetone. A solution corresponding to the amount of of 0.01 μg, 0.1 μg, 1 μg or 10 μg was applied topically to the venter of abdominal segment 4-5 of each female with a microapplicator after the moths were CO_2 anesthetized. Topical application was done at the 2nd h of the photophase on the 1-day-old and 3-day-old virgin females which had exhibited calling behavior during their 1st scotophase.

Pheromone titer measurements

The sex pheromone of the smaller tea tortrix moth, *A.* sp., in Taiwan consists of a 64:36 blend of (Z)-11- and (Z)-9-tetradecenyl acetate (Z11- and Z9-14:Ac) (Kou *et al.*, 1990). Therefore these two compounds were first quantified by the GC method as previously reported. The change in pheromone titer was then determined from moths at 20 h, 44 h and 68 h after topical hormonal application. That is, the ten ovipositors of the untreated, acetone-treated or JH-treated virgin females were excised at 8 h after the scotophase (The period which had the highest pheromone titer, (Kou *et al.*, 1991)). The ten ovipositors were then pooled in 10 μl hexane containing 0.5 μg (Z)-11-hexadecenyl acetate (Z11-16:Ac) as the internal standard. The ovipositors were soaked in the solvent for 5 min, then the extract was subsequently analyzed for Z11-14:Ac and Z9-14:Ac using the internal standard method of quantitative analysis (Kou *et al.*, 1990). Each treatment had three replicates. The results obtained were then analyzed by the Student-Newman-Keuls' method (Steel and Torrie, 1960).

RESULTS

Results obtained were presented in Fig. 1 to Fig. 3. The exogenous treatment with JH-I in day-1 virgin females had no significant effect on the change of pheromone titer (Figs. 1A and 1B). At 20 h after topical application, pheromone titers in untreated, acetone, 0.01 μg, 0.1 μg, 1 μg and 10 μg JH-I treated moths were 112.7, 133.7, 161.8, 141.0, 119.5 and 157.3 ng/♀, respectively (Fig. 1A). At 44 h after topical application, pheromone titers in untreated, acetone, 0.01 μg, 0.1 μg, 1 μg and 10 μg JH-I treated moths were 138.3, 157.6 165.7, 106.9, 101.2, and 130.0 ng/♀, respectively (Fig. 1B). No

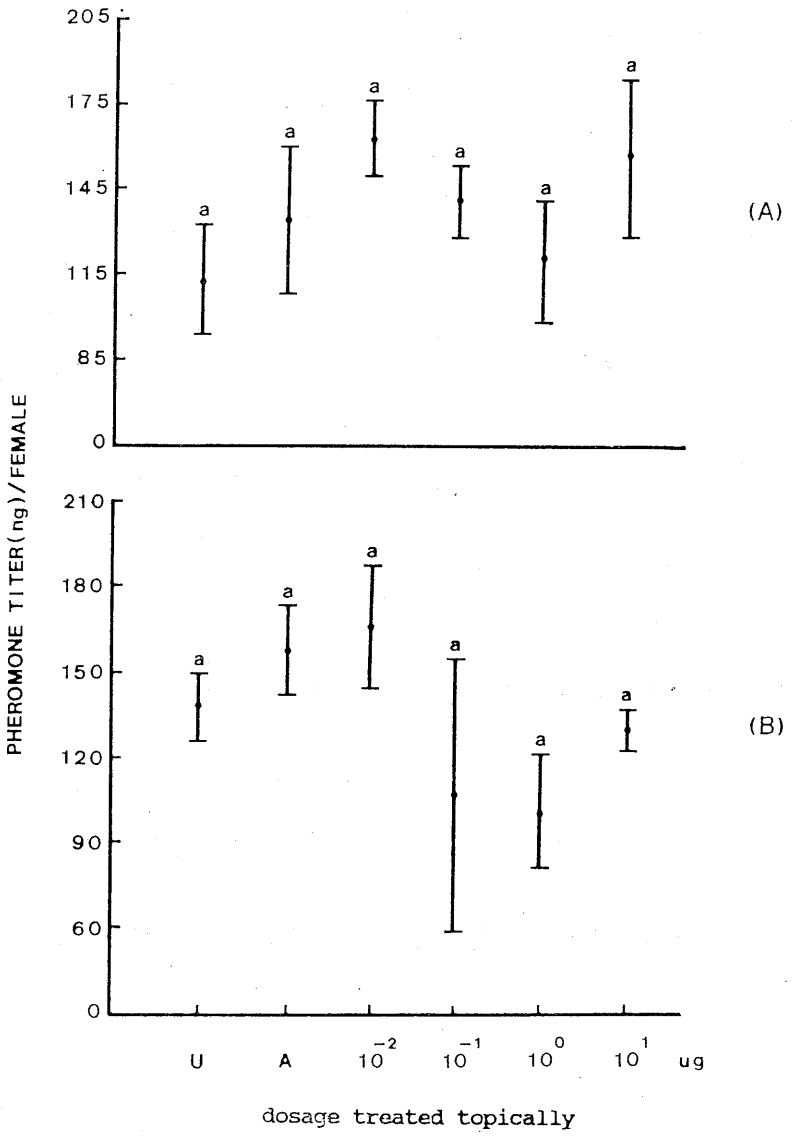

Fig. 1. Effect of different amounts of exogenous juvenile hormone I on pheromone titer of day-1 treated virgin females at (A) 20 h and (B) 44 h after treatment (U: untreated, A: acetone treated). The vertical bars represent ± standard error of each mean, bars topped by the same letter(s) are not significantly different at the 5% level according to the Student-Newman-Keuls' multiple range test.

substantial statistical differences were noted between each test.

Yet, in contrast, exogenous treatment with JH-III significantly reduced pheromone titer at 44 h and 68 h after treatment on day-1 moths (Figs. 2A, 2B and 2C). At 20 h after treatment, pheromone titers in the untreated, acetone, 0.01 μg, 0.1 μg, 1 μg, 10 μg JH-III treated moths were 142.6, 123.5, 121.0, 160.0, 119.4 and 88.8 ng/♀, respectively. Although both 0.1 μg and 10 μg JH-III treatments were not significantly different from the other treatments (Fig. 2A). The 0.1 μg JH-III

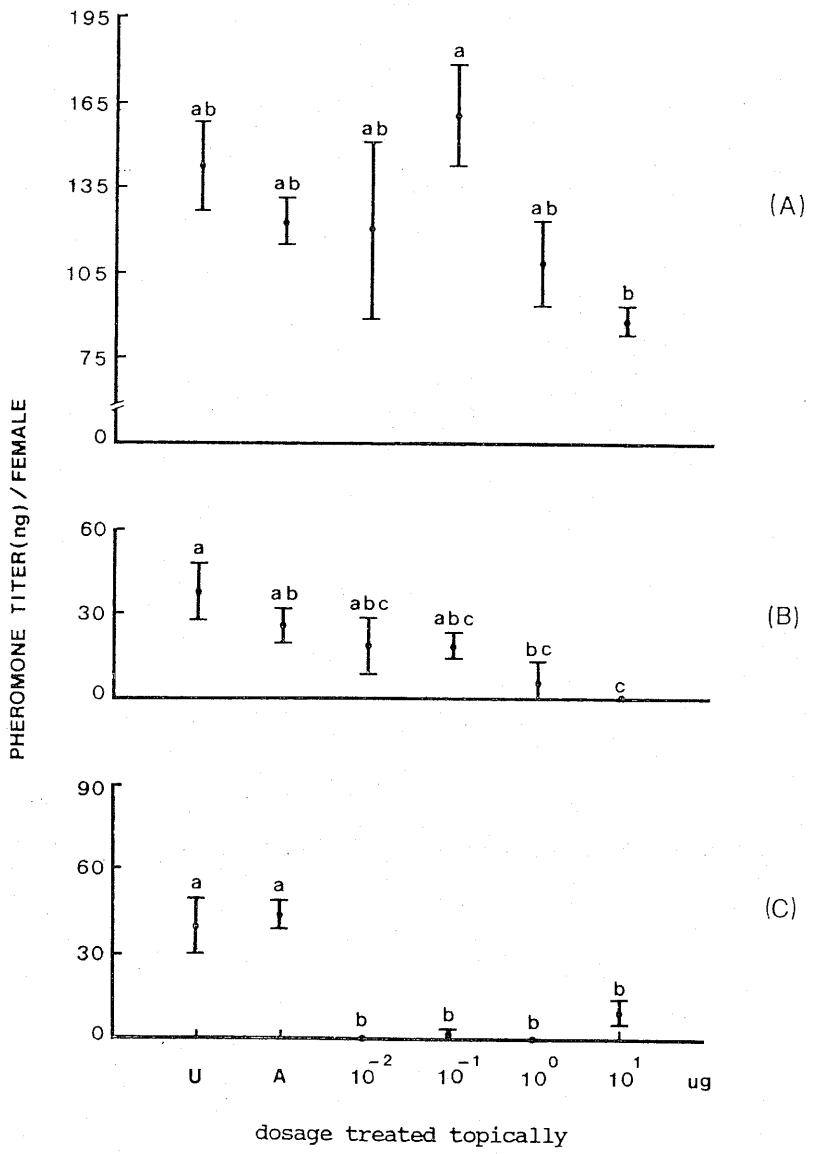

Fig. 2. Effect of different amounts of exogenous juvenile hormone III on pheromone titer of day-1 treated virgin females at (A) 20 h (B) 44 h and (C) 68 h after treatment (U: untreated, A: acetone treated). The vertical bars represent ± standard error of each mean. In each figure, bars topped by the same letter(s) are not significantly different at the 1% level according to the Student-Newman-Keuls' multiple range test.

treatment had slightly higher pheromone titer than the 10 μg JH-III treatment ($p<0.01$). At 44 h after JH-III treatment, pheromone titers in the untreated, acetone, 0.01 μg, 0.1 μg, 1 μg, and 10 μg JH-III treated moths were 38.3, 26.3, 18.8, 18.7, 6.5 and 0 ng/♀. The 1 μg and 10 μg JH-III treatment significantly reduced the sex pheromone titer ($p<0.005$) (Fig. 2B). At 68 h after JH-III treatment, although the pheromone titers in untreated and acetone treated moths were 39.6 and 44.1 ng/♀. The 0.01 μg, 0.1 μg, 1 μg and 10 μg JH-III treatments significantly reduced the pheromone titers to 0, 2.1, 0 and 9.9 ng/♀ ($p<0.05$) (Fig. 2C).

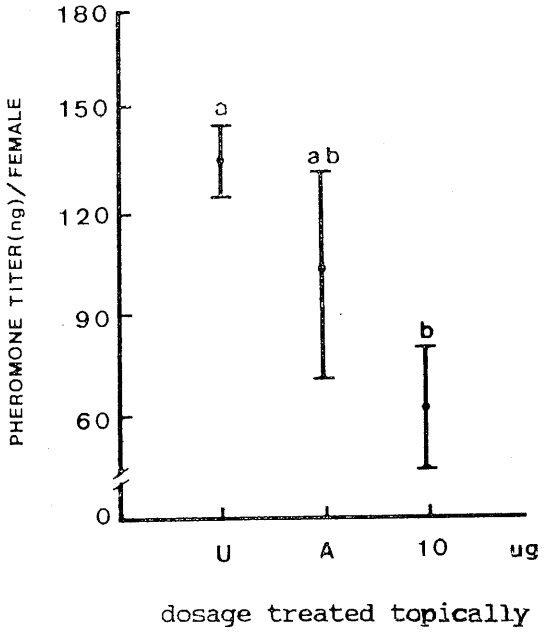

Fig. 3. Effect of 10 μg exogenous juvenile hormone III on pheromone titer of day-3 treated virgin females at 20 h after treatment (U: untreated, A: acetone treated). The vertical bars represent ± standard error of each mean. Bars topped by the same letter(s) are not significantly different at the 5% level according to the Student-Newman-Keuls' multiple range test.

Exogenous treatment with 10 μg JH-III also reduced pheromone titer at 20 h after treatment on day-3 moths (Fig. 3). Pheromone titers of the untreated and acetone-treated months were 136.1 and 103.6 ng/♀, but that of the 10 μg JH-III treated moths was 62.7 ng/♀, being significantly different from the untreated one ($p<0.05$) but not different from the acetone-treated control.

DISCUSSION

In this study, the exogenously applied JH-I had no significant effect on the pheromone titer of the smaller tea tortrix virgin females, but the exogenously applied JH-III significantly reduced the pheromone titer at 44 h and 68 h after treatment. In another tortricid moth, *Platynota stultana* (Walsingham), a JH analogue (ZR-512), JH-I, II and III applied exogenously to virgin females elicited many of those changes normally associated with the switch from virgin to mated behavior observed in mated moths. The JH analogue also appeared to block pheromone production in virgin females (Webster and Carde, 1984). But in the cotton boll weevil, *Anthonomus grandis* Boheman, incorporation of 1.0 ppm JH-III into the diet of an adult male can increase the biosynthesis of its 4 pheromone compounds by 3 times. The biosynthesis at lower and higher levels of JH-III was less, and JH-I was not active at any of the concentrations tested (Hedin *et al.*, 1982).

Recently, in females of the true armyworm moth, *Pseudaletia unipuncta* (Haworth), JH is shown to be essential to the initiation of both calling behavior and pheromone production. Females without corpora allata, the source of JH, do not call and do not produce pheromone, but injection of JH into allatectomized females restored these activities (Cusson and McNeil, 1989). If the smaller tea tortrix moth also has the same physiological regulation as *Pseudaletia unipuncta* (Haworth), then the insect may already have a relatively precise low level of JH. The failure of exogenously applied JH to elicit a greater effect on pheromone titer may then be attributed to a cellular sensitivity which limits the capacity of the cell to respond to a given stimulus as suggested by Staal (1975). The reduction of the pheromone titer by the JH-III might suggest that the exogenous hormone can switch the virgin female to a mated condition. But this assumption on the smaller tea tortrix moth still needs further confirmation.

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青春激素對茶姬捲葉蛾性費洛蒙含量之影響

寇　融　唐丁水　周延鑫

以 1 μg 或 10 μg 青春激素Ⅲ處理一日齡之茶姬捲葉蛾雌蟲，於 44 小時或 68 小時後會顯著地降低雌蟲性費洛蒙含量。以 10 μg 青春激素Ⅲ處理三日齡雌蟲亦於 20 小時後降低其性費洛蒙含量。青春激素Ⅰ對雌蟲性費洛蒙含量無顯著影響。