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Evaluation of mass-trapping for control of tea tussock moth Euproctis pseudoconspersa (Strand) (Lepidoptera: Lymantriidae) with synthetic sex pheromone in south China

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Evaluation of mass-trapping for control of tea tussock moth *Euproctis pseudoconspersa* (Strand) (Lepidoptera: Lymantriidae) with synthetic sex pheromone in south China

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Abstract

Field experiments were conducted to evaluate the effect of dosage, trap placement, trap diameter and trap colour on the trap captures of tea tussock moth, *Euproctis pseudoconspersa*, in a tea plantation located in Guizhou province in southern China during the 2001 field season. The optimum dosage of synthetic sex pheromone was 1.5 mg/septum in a trap. Trap captures of males were maximized with traps paced 90 cm above the ground. Traps of 32 cm in diameter caught significantly more males than those of 20 cm, and white traps caught significantly more males than traps of other colors. The control effect by mass-trapping technique on tea tussock moth was investigated in the same tea plantation in 2002 and 2003. Twenty-five traps/ha were used in the 2-year, large-scale mass-trapping experiment, and a total of 146,767 males were captured. In the pheromone-treated field, mating rates were significantly reduced on nine of 12 sample dates. Larval and egg densities were reduced by 27.87-50.85 and 38.89-51.11%, respectively, compared with the untreated field. Our results suggest that mass-trapping is promising as a control agent for use against the tea tussock moth.

Keywords: Euproctis pseudoconspersa, tea tussock moth, mass-trapping, sex pheromone

1. Introduction

The tea tussock moth, *Euproctis pseudoconspersa* (Strand), is distributed throughout China, Japan, and Korea (Wakamura et al. 1994). It is a common and serious pest of members of the family Theaceae, including the tea, *Thea sinensis* L., an important crop in China. In addition, the larva of this insect interferes with hand-harvesting of tea, because its notorious urticating hairs can cause allergic reaction on human skin (Bleumink et al. 1982). There is, therefore, an urgent need to control tea tussock moth in commercial tea plantations.

Insecticides, the main measure for controlling tea tussock moth in China, can result in unacceptably high residues, which hinder tea exports due to issues of food safety (Jiang et al. 2002). Use of biologically based methods in controlling this pest insect would circumvent or minimize these problems. Sex pheromones are highly species-specific, non-toxic compounds, and have been proved effective in controlling some species of pest insects (Du 1986). The identification and synthesis of the sex pheromone of tea tussock moth provides opportunities for developing pheromone-based management for this pest. Wakamura et al. (1994) and Zhao et al. (1996) reported that the sex pheromone of *E. pseudoconspersa* is (R)-10,14-dimethylpentadecyl isobutyrate (R-10Me14Me-15:iBu), however (R)-, (S)-enatiomers and racemic mixtures of 10,14di-Me-15:iBu have equal attractiveness for males in fields (Wakamura et al. 1996; Zhao et al. 1998a). Mass-trapping is one of the general pheromone-based control methods. However, the efficacy of pheromone trapping may be influenced by many biological, environmental and operational factors. Understanding the effect of these factors will help to optimize and standardize the mass-trapping procedure for controlling the tea tussock moth.

The aim of our study was to determine optimum lure dosage, trap color, trap diameter and trap placement, and to evaluate how effective the masstrapping technique is in controlling this moth in field.

2. Materials and methods

2.1. Field

All experiments were conduced in a tea plantation, an area of c. 200 ha in area, near Duyun city, Guizhou province, Southern China, which is situated at $E107^{\circ}31'$, $N26^{\circ}16'$, c. 1000 m a.s.l. The variety of tea crop 'FuDing' was about 60-90 cm

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high and planted in rows about 1 m wide in 1994. Overwintering eggs result in adults emerging in early June, and first summer generation adults appear in early September in this area (Wang et al. 2003).

2.2. Traps and synthetic pheromone

Since (R)-, (S)-enatiomers and racemic mixtures of 10Me14Me-15:iBu did not differ significantly in field tests (Wakamura et al. 1996; Zhao et al. 1998a), the racemate of 10Me14Me-15:iBu, synthesized in the Institute of Zoology, Chinese Academy of Science, was used as the sex pheromone of tea tussock moths in all experiments. The green rubber septum dispensers were impregnated with sex pheromone and antioxidant (butyl hydroxytoluene, BHT) and acted as lures. Each dispenser contained 1 mg of the synthetic sex pheromone except for those used in dosage trials. Plastic basin traps (purchased from market) baited with green rubber septum dispensers were used for this study. Unless otherwise indicated, the basin was 24 cm in diameter, white in color and contained a water/detergent mixture to capture moths attracted to the lure. Traps were set up in field with bamboo stick tripods at 60 cm height, except in the trap height experiments. The rubber septum dispenser was fixed about 1-2 cm above the water level in the basin. The release rate of sex pheromone is nearly constant and remains active for at least 40 days in the field (Zhao et al. 1998b). The rubber septa were replaced at about 30-day intervals.

2.3. Trap efficiency experiments

These experiences were carried in 2001. Traps were arranged in a randomized complete-block design. Traps within rows were separated by 25-m intervals. Captured moths were checked and removed every morning. At the same time, traps were replenished with water/detergent and replicated rows were rotated with each other to control variation due to inter-field density of tea tussock moth.

2.3.1. Pheromone dosage. Three kinds of dispensers, containing 0.5, 1.0 and 1.5 mg sex pheromone per septum, were tested from 20 September to 8 October, 2001. Each kind of dispenser was replicated 8 times.

2.3.2. Trap height. White traps were placed at 30, 60, 90 and 150 cm height above ground. The lowest traps were placed in midst of two rows of tea trees and below the tea canopy. Traps placed 60 and 90 cm in height were near or slightly above the top of the canopy. The same height trap was replicated 10 times.

2.3.3. Trap diameter. Traps of 20, 24, 27 and 32 cm in diameter were deployed to examine trap size effects on moth capture. Each kind of trap was replicated 8 times.

2.3.4. Trap color. Five kinds of traps, red, yellow, white, black and green, were used in the experiment from 10 September to 25 October, 2001. Each kind was replicated 10 times.

2.4. Control experiments by mass-trapping

The results obtained from preliminary experiments in 2001 were used to establish designs for large-scale control experiments by mass-trapping conducted from 2002 to 2003. White water basin traps of 24 cm in diameter baited with 1 mg sex pheromone were placed at a height of 90 cm in these experiments. A treated field and an untreated field, each 20 ha in area, were located 500 m apart from each other. Splitting each field into 10 plots created a total of 10 replicates. The experiment in 2003 was carried out in the same field as in 2002. Tea tussock moth larval density was investigated before mass-trapping began, to confirm that there was no significant difference between the two plots in early May 2002 $(24.82 \pm 1.95 \text{ vs. } 24.15 \pm 1.57 \text{ larvas/m}^2, \text{ d.f.} = 18,$ P = 0.794, by Student's two-tailed *t*-test). When overwintering and first generation moths began to emerge as adult in early June and early September, pheromone traps were set in the treated field. The inter-trap distance was 20 m (about 25 traps/ha) within the field and border traps along the edge were placed 15 m apart. The moths captured in each trap were counted and removed each morning. The trapping period and dispensers exchange dates are given in Table I. No pesticide was used during study periods in treated and untreated fields and other agricultural operations were the same in both fields.

2.4.1. Estimation of mating rate. Tethered virgin females have generally been used to determine the decrease in the mating rate of female moths (Meng et al. 1985). The procedure on a moth is delicate and it is difficult not to kill it, so we developed a new method. Virgin females (reared indoors, 2 days old) with a tip cut on one of their wings, were set free in round plastic boxes (15 cm depth, 20 cm diameter, white in color) with a smooth inner surface. They therefore they could not fly or crawl out of the boxes. The boxes were hung by 1-m long bamboo rods with hooks on the end. In the evenings, virgin moths (one virgin/box) were sent out in treated and untreated field, and they were collected the next

Table I. Summary of trapping period and dispenser replacing date (day/month).

Year	Generation [†]	Trapping period	Dispenser replacing date
2002	OG FG	6/6–30/6 9/9–21/10	29/9
2003	OG FG	8/6–30/6 7/9–20/10	28/9

[†]OG, overwintering generation; FG, first generation.

morning. These females were dissected under a stereomicroscope to determine if they contained a spermatophore in their vagina or bursa copulatrix. By this means, the mating rate in field was estimated. The female mating rate was estimated three times during middle mass-trapping period in each generation experiment.

2.4.2. Investigation of larval and egg density. Larval densities in treated and untreated fields were investigated on 28 July, 8 August, 17 August, 2002, and 25 July, 7 August, 17 August, 2003, to determine actual control effectiveness of mass-trapping, following the trapping period. The larvae were counted in every sampling unit, a randomly selected 5-m long tea row, and 10 sampling units were checked in every replicate. Eggs laid by first summer generation moths would not hatch until the following spring, so control effectiveness on the first generation was evaluated by determining egg density. The sample method was the same as that of the larval investigation.

2.4.3. Statistical analysis

Trap catches, transformed to $(x+0.5)^{1/2}$, were analyzed by one-way ANOVA, and Tukey's multiple range test was used to separate means. Larva and egg data investigated from treated and untreated fields were compared by independent-samples *t*-test. The incidences of mating occasion of the two fields were compared by Fisher's exact test for a 2 × 2 table. All the statistical analyses were carried out using SPSS statistical software (SPSS, Inc. Version 10.0). Arithmetic rather than transformed means, are given in the results.

3. Results

3.1. Pheromone dosage response

Significantly more males were captured in traps with highest dosage of lure (F=8.710; d.f. = 2, 21; P=0.002) (Table II). The 1.5-mg/septum dispenser captured significantly more males than the

Table II. Trap catches of male *E. pseudoconspersa* with different dosage of lure (2001).

Dosage of lure (mg/septum)	No. of males caught/trap/night (mean \pm S.E.) [†] Sept. 20–Oct. 8
1.5 1.0	$11.88 \pm 1.58a \\ 6.44 \pm 0.93b$
0.5	$4.74\pm1.19\mathrm{b}$

[†]Values indicated are means and standard errors with eight replicates for 19 nights. Data were transformed to $(x + 0.5)^{1/2}$ and then subjected to ANOVA. The means followed by the same letter are not significantly different (P > 0.05) by Tukey's multiple range test for mean separation. lower dosages (P < 0.05). However, no significant difference was observed between dispensers of 1.0 and 0.5 mg/septums (P > 0.05).

3.2. Trap height

Height of trap significantly influenced trapping efficiency of the pheromone trap (F = 3.776; d.f. = 2, 21; P = 0.022) (Table III). Traps placed 60 cm height captured significantly more males than those at 30 cm (P < 0.05). However, no significant difference was observed between traps of 90 and 60 cm height (P > 0.05). Captures at 150 cm were intermediate in terms of numbers, between those caught at 30 cm and 60 or 90 cm.

3.3. Trap diameter

Trap diameter also significantly affected trapping efficiency (F = 3.297; d.f. = 3, 36; P = 0.031) (Table IV). Traps of 32 cm in diameter caught significantly more males than those of 20 cm (P < 0.05), whilst traps of 27 and 24 cm were not significantly different in trapping efficiency.

3.4. Trap color

Significantly different trapping efficiencies were observed among the five kinds of color traps

Table III. Trap catches of male *E. pseudoconspersa* with pheromone traps placed at different height (2001).

Height of trap (cm)	No. of males caught/trap/night (mean \pm S.E.) [†] Sept. 29–Oct. 11			
30	3.21 ± 0.50b			
60	$8.33 \pm 1.65a$			
90	7.18 ± 1.31 a			
150	4.78 ± 1.00 ab			

[†]Values indicated are means and standard errors with eight replicates for 13 nights. Data were transformed to $(x + 0.5)^{1/2}$ and then subjected to ANOVA. The means followed by the same letter are not significantly different (P > 0.05) by Tukey's multiple range test for mean separation.

Table IV. Trap catches of male *E. pseudoconspersa* with pheromone traps of different diameter (2001).

Diameter of trap (cm)	No. of males caught/trap/night (mean \pm S.E.) [†] Sept. 20–Oct. 1			
32	$3.62 \pm 0.40a$			
27	$3.17 \pm 0.40ab$			
24	$2.76 \pm 0.27ab$			
20	$2.12 \pm 0.31b$			

[†]Values indicated are means and standard errors with 10 replicates for 12 nights. Data were transformed to $(x + 0.5)^{1/2}$ and then subjected to ANOVA. The means followed by the same letter are not significantly different (P > 0.05) by Tukey's multiple range test for mean separation. (F = 2.723; d.f. = 4, 45; P = 0.041) (Table V). White traps captured significantly more males than green and blue traps did (P < 0.05). Red and yellow

Table V. Trap catches of male *E. pseudoconspersa* with pheromone traps of different colour (2001).

Colour of trap (cm)	No. of males caught/trap/night (mean \pm S.E.) [†] Sept. 10–Sept. 25			
White	$1.69\pm0.33a$			
Red	$1.44\pm0.27\mathrm{ab}$			
Yellow	$1.37\pm0.26\mathrm{ab}$			
Green	$0.83\pm0.17\mathrm{b}$			
Blue	$0.74\pm0.15\mathrm{b}$			

[†]Values indicated are means and standard errors with 10 replicates for 16 nights. Data were transformed to $(x+0.5)^{1/2}$ and then subjected to ANOVA. The means followed by the same letter are not significantly different (P > 0.05) by Tukey's multiple range test for mean separation. traps did not appear to be significantly more efficient than green and blue traps (P > 0.05), nor did they catch significantly less males than white traps did (P > 0.05).

3.5. Capture amount and its occurrence during 2002-2003

Changes in capture amount from Figures 1 and 2 clearly show that tea tussock moth completed two separate generations a year in Duyun city, China. The overwintering generation of adults emerged in early June and the flight period lasted for about a month; the first generation emerged in early September and that flight period lasted for about 2 months.

Similar peak-trapping dates occurred in 2002 and 2003. Most males were trapped near to peak-trapping dates. 13,769 males in 460 traps were captured during the overwintering generation period, and 74,246 males in 497 traps were trapped during the

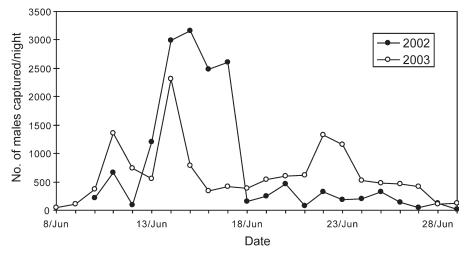


Figure 1. Captures of *E. pseudoconspersa* males during overwintering generation in pheromone traps on each night with 460 traps in 2002 and 488 traps in 2003.

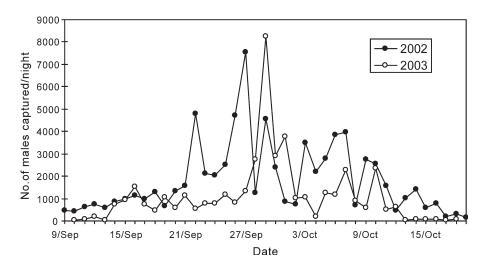


Figure 2. Captures of *E. pseudoconspersa* males during first generation in pheromone traps in each night with 497 traps in 2002 and 470 traps in 2003.

first generation period in 2002. In 2003, traps set in overwintering and first generations were 480 and 470 in number, and captured 15,668 and 43,084 males, respectively. In the 2-year large-scale mass-trapping, a total of 146,767 males were captured. The males captured were reduced from 88,015 in 2002 to 58,752 in 2003 in almost the same number of traps, indicating that the population of *E. pseudoconspersa* was suppressed by mass-trapping in a sense.

3.6. Mating rate after mass-trapping

Female mating rate in the treated field was significantly lower (Fisher's exact test, P < 0.05) than that in the untreated field on nine of 12 sampling dates in the four generation experiments (Table VI). The mating rates were reduced from 61-77% in the untreated field to 29-50% in the treated field.

3.7. Population abundance after mass-trapping

Larval abundance of tea tussock moth in the treated field was significantly lower than that in the untreated field in every investigation during 2002 and 2003 (*t*-test, d.f. = 18, P < 0.05, Table VII).

It was reduced by 27.87-50.85% in the treated field compared with that of the untreated field for 2 years. The egg density of the first generation in the treated field was also significantly lower than that in the untreated field in every sampling (*t*-test, d.f. = 18, P < 0.05, Table VIII), which was reduced by 38.89-51.11% when comparing treated with untreated fields for the 2 years.

4. Discussion

4.1. Trap efficiency experiments

Various blends of (R)- and (S)-10Me14Me-15:iBu, the sex pheromone of the tea tussock moth, differed in attractiveness to males, but the differences were not significant (Wakamura et al. 1994). The synthetic (R)- and (S)- enantiomers of 10Me14Me-15:iBu and racemic mixtures did not demonstrate significant differences in field tests (Wakamura et al. 1996; Zhao et al. 1998a). Thus, the racemic mixture was used in this experiment.

Lure, trap design and trap placement significantly influenced moth catches in pheromone traps (Trematerra 1993; Cork et al. 2003). Traps baited

	Sampling date (DAM) [‡]	Treated field		Untreated field		
Generation [†]		Test virgins	% mated	Test virgins	% mated	P^{\S}
2002 OG	10	24	42	33	64	0.084
	12	31	42	32	75	0.008
	13	26	38	28	61	0.086
2002 FG	19	30	50	30	63	0.217
	20	32	44	31	74	0.014
	23	35	29	28	68	0.002
2003 OG	9	32	44	32	66	0.066
	10	38	31	30	70	0.001
	13	30	37	30	67	0.019
2003 FG	20	34	35	33	73	0.002
	21	27	41	26	77	0.008
	24	34	38	34	68	0.014

[†]OG, overwintering generation; FG, first generation. [‡]DAM, days after mass-trapping. [§]Data were subjected to the Fisher's exact test (one-sided) for 2×2 table.

Table VII. Larva densities of E. pseudoconspersa during first generation after mass-trapping.

		No. of larvas/m ² (mean \pm S.E.) [†]				
Year	Sampling date	Treated field	Untreated field	t	Р	Reduce (%)
2002	28 July	$25.73 \pm 3.93b$	$35.67 \pm 3.59a$	-1.866	<0.05	27.87
	8 Aug.	$16.68 \pm 2.35b$	$28.94 \pm 5.73a$	-1.981	<0.05	42.36
	17 Aug.	$11.86 \pm 2.25b$	$20.74 \pm 2.55a$	-2.614	<0.001	42.82
2003	25 July	$21.52 \pm 4.78b$	$37.08 \pm 4.17a$	-2.455	<0.05	41.96
	7 Aug.	$16.12 \pm 4.67b$	$32.80 \pm 3.89a$	-2.759	<0.001	50.85
	17 Aug.	$9.45 \pm 2.17b$	$18.39 \pm 4.59a$	-1.765	<0.05	48.61

[†]Values indicated are means and standard errors of 10 sample units. Means within rows followed by the same letter are not significantly different (P > 0.05) (one-tailed *t*-test, d.f. = 18).

		No. of egg clumps/m ² (mean \pm S.E.) [†]				
Year	Sampling date	Treated field	Untreated field	t	P	Reduce (%)
2002	7 Nov. 16 Nov.	$0.22 \pm 0.04b$ $0.30 \pm 0.05b$	$0.45 \pm 0.07a$ $0.58 \pm 0.08a$	-2.725 -2.977	<0.01 <0.01	51.11 48.28
2003	10 Nov. 15 Nov.	$0.19 \pm 0.04b$ $0.22 \pm 0.05b$	$0.34 \pm 0.07 a$ $0.36 \pm 0.06 a$	$-1.848 \\ -1.878$	<0.05 <0.05	44.12 38.89

Table VIII. Egg densities of E. pseudoconspersa during overwintering generation after mass-trapping.

[†]Values indicated are means and standard errors of 10 sample units. Means within rows followed by the same letter are not significantly different (P > 0.05) (one-tailed *t*-test, d.f. = 18).

with a higher dose of synthetic pheromone captured significantly more male tea tussock moths than those with lower doses (Wakamura et al. 1994; Zhao et al. 1996; Tsai et al. 1999). Wakamura et al. (1994) found that 24 μ g of the pheromone had a similar attractiveness to 80 and 240 μ g when treated on a filter paper pad, showing that lure dosage an upper limit to obtain maximal attractiveness should exist. Our experiment indicated that lures of 1.5 mg/ stepum were significantly more attractive than those of 1.0 or 0.5 mg/stepum in the field. Higher dosage than 1.5 mg/stepum lures should be tested to determine whether there is a more optimal dosage.

For many species, traps within a crop canopy catch more moths than traps above the canopy. For example, traps located 10 cm above the top of the canopy caught significantly more European corn borer, Ostrinia nubilalis (Lepidoptera: Pyralidae), than those located 50 cm above (Mason et al. 1997). Similarly, insect pests associated with tree crops are most frequently trapped in highest numbers within a canopy (Bartlet et al. 1994). However, David and Horsburgh (1989) obtained opposite results in trapping leafroller, Platynota flavedana Clemens (Lepidoptera: Pyralidae). Unlike other crops, tea tree is a shrubby plant and has a dense canopy. According to our observation, most males flew around the tea crop canopy, so that traps located at crop height caught significantly more males than those above or below the canopy. Our results presumably reflected the fact that the pheromone plume extended further above the crop canopy than below, which enabled more males to respond to the pheromone and successfully locate the traps.

More males were caught as trap size increased from 27 to 32 cm, but the captures did not increase significantly (P > 0.05). Thus, traps of 27 cm in diameter were optimal in practice. In addition, larger traps were more difficult to manage. In other studies, sticky traps were used to capture tea tussock moth (Tsai et al. 1999; Wakamura et al. 1996). In general, the main drawback in the use of adhesive traps is that the sticky surface is often 'overloaded' with moths, dust or wing scales, which reduces trapping efficiency (Beasley and Adams, 1994). Furthermore sticky traps were relatively costly in Guizhou province where labor is very cheap and water is easy to obtain. The water basin trap is most practical at this location.

Although long-distance attraction was primarily an olfactory response, some evidence showed that visual cues were important in eliciting short-range orientation in some lepidopteran pests. Childers et al. (1979) obtained increases in captures of lesser peach tree borer and peach tree borer males with colored traps; however, McLaughlin et al. (1976) reported that white traps performed as well as or better than colored traps. As for tea tussock moth, initiation of copulation was observed even after sunrise, which suggested that light conditions might not inhibit mating behavior (Wakamura et al. 1996). Our results also indicated that white traps captured significantly more moths than other color traps.

4.2. Mass-trapping experiments

In tea plantations in China, it is necessary to reduce insecticide usage by the use of biologically based methods, such as synthetic sex pheromone. Compared with the mating disruption technique, the concept of mass-trapping appears to be simpler. The use of mass-trapping for control of insect pests has been suggested for a number of lepidopteran pests. However, unlike coleopteran and dipteran pests, there are only a few examples of the successful application of mass-trapping in Lepidoptera in the literature (Cork et al. 2003). This is probably because it is generally assumed that trapping 80-95% of males is needed to suppress population growth (Knipling and McGuire 1966). Mass-trapping methods were more effective if the target species can only mate one time or can only mate one per night (Roelofs et al. 1976). According to our observations, most males of tea tussock moth mate only once (Wang et al. 2003). Males of tea tussock moth emerge 1.5 days earlier than females on average (Wang et al. 2003), so that males can be trapped when they are waiting for their mates. Also, the flight period of tea tussock moth is only about 3 months in a year (Figures 1 and 2). Thus, it is advantageous to control tea tussock moth with synthetic sex pheromone. In our large-scale mass-trapping experiments, the field egg density and field larval density was reduced by 38.89-51.11 and 27.87-50.85% in the treated field compared with that of the untreated field, showing that mass trapping males exerted an effect on suppressing tea tussock moth population.

Different trap density was selected on mass-trapping researches of other lepidopteran pests. Trematerra (1993) considered three densities, six, 12 and 24 traps/ ha, in mass-trapping Synathedon myoapaeformis, and concluded that 12 traps/ha was optimal effectively and economically. Faccioli et al. (1993) reported that a density of over 10 traps/ha was unnecessary in severely infested orchards for the control of Cossus cossus. Ranga Rao et al. (1991) found there was no significant improvement in captures when four or more traps per ha were installed. Optimal trap density is likely to depend on crop coverage and population density of the target insect. To achieve the best control by mass-trapping on tea tussock moth at lowest cost, a series of trials need to be carried out to determine optimal trap density.

In this experiment, the egg density of 2003 was lower than that of 2002 in the treated field (Table VIII). The same tendency was observed in larval density (Table VII), but we hesitate to conclude that masstrapping is more effective in controlling tea tussock moth when it is applied generation by generation because the pest insect population generally changes from year to year. However our experiment proved that mass-trapping with synthetic sex pheromone demonstrated significant control effect on the population of tea tussock moth. It may act as a potential IPM component in managing tea tussock moth.

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