

# Interference with the mate-finding communication system of the Chinese tortrix *Cydia trasi* (Lep., Olethreutidae) using synthetic sex pheromones

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**Abstract:** Interference with the mate-finding communication system of *Cydia trasi* (Meyrick) using sex pheromone dispensers (60% E8,E10-dodecadienyl acetate and 40% E8,E10-dodecadien-1-ol, 0.5 mg/dispenser) was investigated in three plots of the Chinese scholar-tree, *Sophora japonica* L., in Beijing, China, in 2000. Treatments were evaluated by pheromone-baited traps, caged virgin females, pupating and hibernating populations on tree trunks, and assessment of larval damage in petioles and seed pods. Six to eight natural rubber septum dispensers were placed in each tree at the pheromone-treated plots at three separate periods: at the beginning of overwintering generation flight, the first- and the second-generation flights. Application of pheromone dispensers reduced the incidence of mating of virgin females in treated plots relative to those in the control plots. Pheromone-baited trap catches were lowered on average by 96%, suggesting a high level of disruption. Average larval density on tree trunks was significantly lower in the pheromone-treated plots than in the untreated control plots. The mean percentage of Chinese scholar-tree petioles infested with first- and second-generation larvae, and seed pods infested with third-generation larvae was significantly greater in untreated control plots than in pheromone-treatment plots. These results suggest that disruption of pheromone-based communication in *C. trasi* is prospective, with synthetic sex pheromone on trees in urban environments.

**Key words:** *Cydia trasi*, Chinese tortrix, Lepidoptera, mating disruption, Olethreutidae, sex pheromone

## 1 Introduction

The Chinese tortrix, *Cydia trasi* (Meyrick), has been a significant pest of urban trees, including the Chinese scholar-tree, *Sophora japonica* L., in north China and the pagoda-tree, *Maackia amurensis* var. *Buergeri*, in Japan since the 1980s (ENDA and YAMAZAKI, 1987; CHEN and QI, 1996). In Beijing, the pest has two and a partial third generation (TG) each year, and the moths are active from early May to early or mid-October (CHEN and QI, 1992; ZHANG et al., 2001b). The larvae attack both the petioles and seed pods, and overwinter in seed pods, bark crevices and twigs (ENDA and YAMAZAKI, 1987; CHEN, 1992; CHEN and QI, 1992). Formerly, *C. trasi* was controlled mainly with insecticides. Applications of toxic chemicals have resulted in pest resistance to insecticides, disruption of biological control systems, accidental poisoning of gardeners, environmental contamination and health hazards. Reduction of insecticide usage is, therefore, of great importance, particularly in controlling urban tree pests. One promising alternative to insecticides for the control of tortricid pests is the use of synthetic sex pheromones to disrupt their mate-finding communication systems (RIDGWAY et al., 1990). Sex pheromones of

the major tortricid pests of fruit trees have been identified and used in mating disruption experiments. For instance, mating disruption of the codling moth, *Cydia pomonella* (L.), in apple and pear orchards produced encouraging results (MOFFITT and WESTIGARD, 1984; BARNES et al., 1992; PFEIFFER et al., 1993). Mating disruption of the Oriental fruit moth, *Cydia (Grapholitha) molesta* (Busck) was also reported (WEAKLEY et al., 1987; MENG et al., 1988; AUDEMARD et al., 1989; PREE et al., 1994). Results of these studies have indicated the potential of mating disruption as a control strategy.

*Cydia trasi* sex pheromone consists of a 2 : 3 mixture of E8,E10-dodecadien-1-ol (E8,E10-12:OH) and E8,E10-dodecadienyl acetate (E8,E10-12:Ac) (MENG et al., 1996), and pheromone dispensers containing this blend are available for mating disruption. Use of sex pheromone permeation to interfere with *C. trasi* mate-finding was first attempted by ZHANG et al. (2001a). Results indicated that this technique was able to reduce the ability of the male *C. trasi* to locate pheromone-baited and virgin female-baited traps and the infestations of petioles and seed pods. However, the mating frequency of females and the

population density in disrupted plots were not investigated. The results of studies with synthetic sex pheromone for suppression of *C. trassias* through interference with the mate-finding communication system as a low environmental impact control tactic in plots of Chinese scholar-trees in Beijing, China, have been reported.

## 2 Materials and methods

### 2.1 Test sites

Plots along three roads, Road I, Road II, and Road III, located at Zhongguancun, Beijing, were selected for mating disruption test sites (fig. 1). At Road I, the pheromone-treated plot consisted of 110 trees planted in two rows of 55 trees each. Both rows were 280 m long, one on either side of the plot, separated by approx. 20 m. The control plot was located 100 m away from the treated plot and had 89 trees arranged similarly in two 220 m long rows separated by a distance of approx. 18 m. All trees were 7–10 years old, 7–9 m tall, and planted on a pedestrian path. Both ends of the disruption plot were at crossroads about 80 m wide, after which were the plots of *C. trassias* non-host plant trees, Chinese ash, *Fraxinus chinese* var. *acuminata* Lingelsh, and panicled goldrain tree, *Koelreuteria paniculata* Laxm., and ginkgo, *Ginkgo biloba* L., and Chinese white poplar, *Populus tomentosa* Carr. Both Roads II and III (the test sites I and II) had been described in the recent tests (ZHANG et al., 2001a). At Road II, all trees were 5–6 years old, 5–6 m tall, and planted on a pedestrian path. At Road III, all trees were 4–5 years old, 4–5 m tall, and planted on a piece of long lawn in a single row. Road I was separated from Road II by approx. 800 m, and from Road III by approx. 300 m, and Road II was separated from Road III by approx. 500 m.

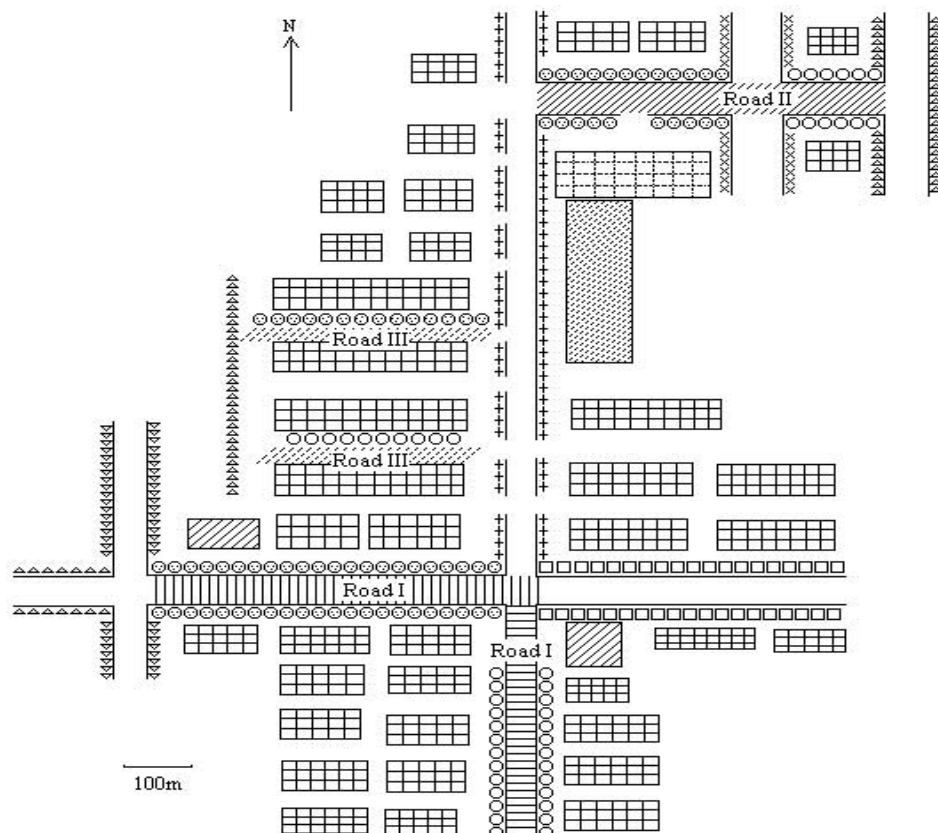
### 2.2 Pheromone-disruption dispensers

Natural rubber septum (Shunyi Rubber Com., Beijing, China) dispensers (produced by our laboratory), made of two parts, the first part solid (6 × 5 mm in diameter) and the second part hollow (8 × 6 mm i.d. and 10 mm o.d.), were used throughout these studies (ZHANG et al., 2001a). The dispensers contained 60% E8,E10-dodecadienyl acetate (E8,E10-12:Ac) and 40% E8,E10-dodecadien-1-ol (E8,E10-12:OH) (0.5 mg/dispenser). Pheromone dispensers were tethered to twigs in the upper third of the tree canopy approx. 0.5–1.0 m from the top of the central leader, or on the first lateral branch on 15 May prior to the first capture of moths [overwintering generation flight (OwG)] in pheromone-baited traps located in control plots. Dispensers were located approx. 1 m apart. Each tree held six to eight dispensers. A total of 850, 450 and 200 dispensers were installed at test Roads I, II and III, respectively. Second [first generation (FG) flight] and third [second generation (SG) flight] installations of equal numbers of dispensers were on 8 July and 16 August, respectively.

### 2.3 Pheromone traps

Four (exception: three in the control plot of Road III) sticky traps, triangular in cross-section (21 cm base, 19 cm × 19 cm), 25 cm long, with a movable sticky base liner, were placed in both pheromone treated and control plots of each test road. The traps were baited for *C. trassias* with 0.25 mg sex pheromone (60% E8,E10-12:Ac and 40% E8,E10-12:OH) on rubber septa (MENG et al., 1996; ZHANG et al., 2001b) and operated for 1 week before the first installation of the pheromone dispensers (7 May, 2000). Traps were hung approx. 3 m above ground at 30 m intervals in all plots. Catches of male moths were recorded every other

**Fig. 1.** A map of mating disruption test Roads I–III in Zhongguancun, Beijing, China. Round crown, Chinese scholar tree; dots, pheromone treated and blank, untreated control; triangle crown, Chinese white poplar; del crown, ginkgo; cross (+) crown, paniculated goldrain tree; ork (X) crown, silk tree albizzia; square crown, Chinese ash; checked building (solid lines), residential building; checked building (broken lines), primary school; oblique line building (solid lines), supermarket; oblique line building (broken lines), kindergarten



day. Pheromone lures were replaced every 3 weeks throughout the season, and sticky liners every 2–4 weeks, or more often if needed.

## 2.4 Caged female assessment

Caged virgin female *C. trassias* moths were used to assess mating in both disruption and control plots. At Road I, during the peak periods of the OwG and SG flights, 50 OwG and 40 SG mating stations (non-sticky-liner triangular traps) were deployed in each disruption and control plot, respectively (the period of the FG flight is usually rainy and unfavourable for the test). A modified cylindrical polyvinylchloride (PVC) mating cage (55 × 35 mm diameter) (FITZPATRICK and TROUBRIDGE, 1993) containing a 1-day-old virgin female moth was pinned in each station (two pieces of tape were stuck around the cage to allow attachment to the station with thumb pins). Each PVC cage had a wire-mesh cone with an opening (5 mm diameter) on the each end of the cage to allow moth entry. The test was performed three times for each generation. A total of 40–50 (OwG) or 30–40 (SG) virgin female moths were used in each test. Females were obtained from isolated seed pods (containing overwintering larvae collected during spring) held in individual glass tubes (kept under natural photoperiod and temperature conditions (21.0–32.5°C, 14 : 10 to 16 : 8 hours L : D) and then at 14°C for about 10 h before they were placed in the test plots. The caged virgin females were placed in pheromone-treated and control plots at 15:00 hours daily, and 24 h later, retrieved and dissected under a stereoscope to determine if they had spermatophores in their spermatheca. Accordingly, the mating rates of the females in the field were estimated.

## 2.5 Estimation of population density on tree trunks

At each of the three roads during the end of the FG and SG moth eclosion, as well as after the TG larvae entered hibernacula, 40 treatment (T) and 40 control (CK) trees (Road I), 30 T and 30 CK trees (Road II), and 28 T trees and 16 CK trees (Road III) were sampled randomly by inspecting trunks for larvae which had pupated in the FG, SG, or TG generation on the tree trunks in each disruption and control plot. In the process of sampling, one tree was subsampled from one to three trees according of the size of the plot, in order to make the sampling effort representative (Li et al., 2000). That is, one tree was subsampled from two to three trees at Road I, one tree from one to two trees at Road II, and one tree from one to three trees at Road III.

## 2.6 Damage assessment

At the end period of each generation, the twigs bearing compound leaves (injured by the FG and SG larvae tunnelling in the petiolar base) or clusters of seed pods (injured by the TG larvae) were inspected for larval injury in each disruption and control plot. Seven twigs or clusters were inspected in each of five cardinal directions (east, west, south, north and top) on each of randomly selected trees (ZHANG et al., 2001a). At Road I, 34 and 28 trees were sampled at each generation in the disruption and control plots, respectively. At Road II, 20 and 20 trees were sampled during the FG and SG, and 19 and 15 trees during the TG. At Road III, 20 and 14 trees were sampled during each of the FG and SG, and 12 and 10 trees during the TG. Petioles and seed pods were recorded as either damaged or undamaged. Data, summarized for each plot, were expressed as the percentage of petioles or seed pods

infested. A paired-sample *t*-test or an analysis of variance LSD-test (SPSS; Microsoft®) was performed to test the equality of means.

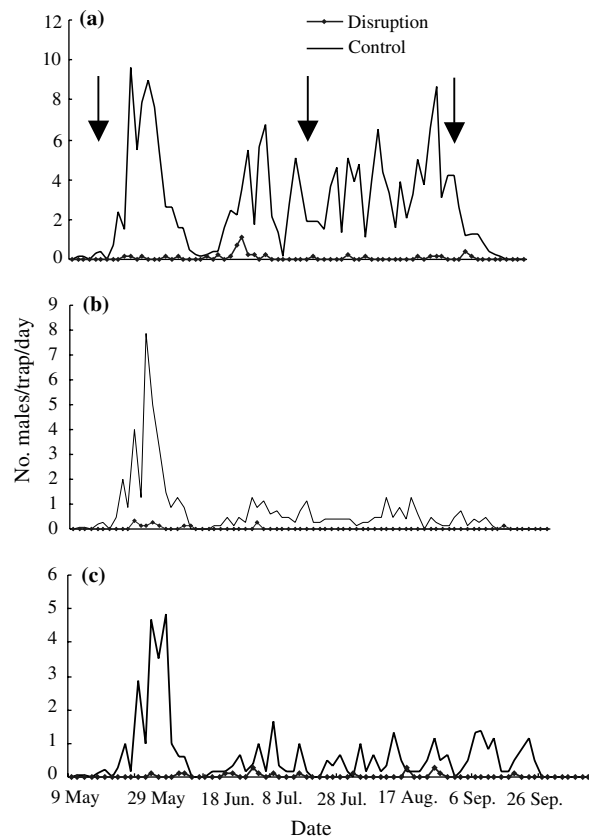
## 3 Results

### 3.1 Disruption of orientation ability

The numbers of male moths captured in traps operated over the same period in three similar but untreated plots indicated the phenology of the pest, and the contrasting inhibition of orientation ability in the pheromone-treated plots. As shown in fig. 2, the presence of the pheromone dispensers almost completely disrupted male orientation towards the pheromone-baited traps for the season following the three pheromone applications in the three treated plots. Trap captures in pheromone-baited traps in the three pheromone-treated plots were 94.0–98.9% lower than those in the untreated plots (table 1).

### 3.2 Mating of caged female moth

At Road I, in the presence of the pheromone dispensers, the numbers of mated *C. trassias* females after 1-day exposure in the pheromone-treated plots were suppressed 76.9–94.6% during the OwG and SG flights when compared with controls (table 2).



**Fig. 2.** Catches of *Cydia trassias* males in pheromone-baited traps located in three test roads in Zhongguancun, Beijing, China, 2000. Arrows indicate dates of pheromone applications in the pheromone treated plots. (a) Road I; (b) Road II; (c) Road III

**Table 1.** Trap catches in four (three in control plot of test Road III) pheromone-baited traps in pheromone-treated and control plots at three test roads, in mating disruption trials against the Chinese tortrix *Cydia trasi* in Zhongguancun, Beijing, in 2000

Road code	Generation	Captures		
		Pheromone treated	Control (untreated)	Percentage reduction
Road I	Overwintering	7	468	98.50
	First	25	414	93.96
	Second	11	800	98.63
Road II	Overwintering	9	231	96.10
	First	2	84	97.62
	Second	1	90	98.89
Road III	Overwintering	4	127	97.64
	First	3	48	95.31
	Second	6	98	95.41

**Table 2.** Mating status of caged *Cydia trasi* females in 24 h at South Zhongguancun road. Dates of overwintering and second generations: 27 May to 6 June and 29 August to 4 September 2000

Generation	Percentage mated (mean $\pm$ SEM)		Percentage reduction
	Pheromone treated	Control (untreated)	
Overwintering	1.33 $\pm$ 1.33	24.83 $\pm$ 5.13	94.64
Second	4.45 $\pm$ 2.22	19.28 $\pm$ 2.06	76.92

### 3.3 Pupating or hibernating populations on tree trunks

The results in table 3 demonstrate that interference with the mate-finding communication system by synthetic sex pheromone during the whole season significantly reduced the density of *C. trasi* on the trees. During the whole season, the pupating (FG and SG) and hibernating (TG) status of the FG, SG and TG larvae differed between treatments ( $t = 5.984$ , d.f. = 8,  $P < 0.01$ ; SPSS: paired-sample  $t$ -test) with significantly fewer larvae pupating and hibernating on tree trunks in the treated plots than in untreated controls (table 3) after three applications of pheromones.

### 3.4 Petiolar and seed pod infestation

As shown in table 4, percentage petiolar and seed pod damage in each of the three generations were clearly less in the three pheromone-treated plots than in the three controls. The percentage petiolar damage in the three pheromone-treated plots during the end of the FG and SG were 0.9 and 1.1%, well below the 4.4% ( $t = 10.377$ , d.f. = 2,  $P = 0.009$ ; SPSS: paired-sample

**Table 4.** Infested petioles and seed pods during the end of the first and second (tunnelling petioles), and third (bored seed pods) generation larvae, in mating disruption trials against the Chinese tortrix, *Cydia trasi* in Zhongguancun Beijing, in 2000

Generation	Road code	Infested [% (mean $\pm$ SEM)]		Percentage reduction
		Pheromone treated	Control (untreated)	
First (tunnelling of petioles)	Road I	0.98 $\pm$ 0.11	4.50 $\pm$ 0.54	78.22
	Road II	1.35 $\pm$ 0.22	5.32 $\pm$ 0.79	74.62
	Road III	0.46 $\pm$ 0.12	3.29 $\pm$ 0.64	86.02
Second (tunnelling of petioles)	Road I	1.28 $\pm$ 0.14	6.52 $\pm$ 0.47	80.37
	Road II	1.61 $\pm$ 0.25	7.71 $\pm$ 0.76	79.12
	Road III	0.54 $\pm$ 0.14	2.84 $\pm$ 0.26	80.99
Third (boring of seed pods)	Road I	0.93 $\pm$ 0.12	5.60 $\pm$ 0.65	83.39
	Road II	1.73 $\pm$ 0.20	7.15 $\pm$ 1.01	75.80
	Road III	0.27 $\pm$ 0.07	5.29 $\pm$ 0.98	94.90

**Table 3.** Average numbers of pupating (first- and second-generation) and hibernating (third generation) larvae on tree trunks observed during mating disruption trials against the Chinese tortrix *Cydia trasi* in Zhongguancun, Beijing, in 2000

Generation	Road code	No. larvae/tree (mean $\pm$ SEM)	
		Pheromone treated	Control (untreated)
First (pupating)	Road I	1.43 $\pm$ 0.26	2.35 $\pm$ 0.73
	Road II	0.33 $\pm$ 0.11	1.07 $\pm$ 0.28
	Road III	0.04 $\pm$ 0.04	0.31 $\pm$ 0.15
Second (pupating)	Road I	2.80 $\pm$ 0.55	3.88 $\pm$ 0.61
	Road II	0.10 $\pm$ 0.06	0.50 $\pm$ 0.41
	Road III	0.04 $\pm$ 0.04	1.06 $\pm$ 0.47
Third (hibernating)	Road I	0.08 $\pm$ 0.04	0.80 $\pm$ 0.20
	Road II	0	0.33 $\pm$ 0.10
	Road III	0	0.31 $\pm$ 0.12

$t$ -test) and 5.7% ( $t = 3.952$ , d.f. = 2,  $P = 0.058$ ; SPSS: paired-sample  $t$ -test), respectively, observed in the three controls. During the end of the TG, the percentage seed pod damage in the three pheromone-treated plots was 1.0%, well below 6.0% ( $t = 23.246$ , d.f. = 2,  $P = 0.002$ ; SPSS: paired-sample  $t$ -test), in the three controls. On average, the control efficiency in the three pheromone-treated plots was 80.8%.

## 4 Discussion

Three applications of the pheromone containing 60% E8,E10-12:Ac and 40% E8,E10-12:OH with six to eight dispensers per tree per generation appear to have

a strong effect in disrupting communication between male and female *C. trasi*s moths. During the year of investigation, male catches in pheromone-baited traps were significantly lower in disruption plots than in the controls. Mating status, using caged virgin female stations, provided a more meaningful assessment of mating disruption than reduction of captures at monitoring traps (ROTHSCHILD, 1981), and was lower in pheromone-treated plots at Road I. On average, the numbers of larvae pupating and hibernating on tree trunks were lowered by 63% in pheromone-treated plots when compared with that in the controls. More important, the percentage damage of petiole and seed pod were significantly reduced in the three sex pheromone-treated plots when compared with controls.

These results are in line with those obtained by MENG et al. (1988), who stated that natural rubber septum dispensers containing 0.2 mg mixture of the sex pheromone Z8-dodecenyl acetate (Z8-12:Ac, 90%), E8-dodecenyl acetate (E8-12:Ac, 10%), and Z8-dodecen-1-ol (Z8-12:OH, 10%) were effective in suppressing of the Oriental fruit moth when placed at a density of five dispensers per tree in pear orchards in Suizhong, Liaoning, China.

Successful mating disruption of *C. trasi*s was achieved in 1999 with synthetic sex pheromone (a 2 : 3 mixture of E8,E10-12:OH and E8,E10-12:Ac) as indicated by male catches in pheromone-baited and virgin female-baited traps, and percentage damage of petiole and seed pod in two pheromone treated plots (ZHANG et al., 2001a). In the tests conducted in 2000, effective mating disruption of *C. trasi*s was evident, based on observed male catches in pheromone-baited traps, mating status of caged virgin females, population density on tree trunks and percentage damage of petioles and seed pods in three pheromone-treated plots. Among the three test areas of the present study, two of them (both Roads II and III) were the test sites of the recent 1999 tests (test sites I and II). At Road II, the control efficiency in the present study was 13.4% more than the 1999 tests ( $t = 2.480$ , d.f. = 2,  $P = 0.131$ ). At Road III, the control efficiency of the seed pods in the present study was 5.7% more than the 1999 tests.

Using synthetic sex pheromone resulted in excellent reduction in mate-finding communication between the *C. trasi*s sexes, as evidenced by the reduction in response of males to pheromone-baited traps (on average, 96.9% trap disruption). However, the control efficiency (on average, 80.8%) was not as successful as trap disruption ( $t = 11.624$ , d.f. = 2,  $P = 0.007$ ). The inconsistency between the trap disruption and the control efficiency may be due to the incidence of mating in pheromone-treated plots. CARDÉ and MINKS (1995) stated that point-source release may tend to leave pheromone-free gaps where mating could occur. Moreover, the movement of mated females may undoubtedly play a significant role in the degree of suppression obtained with mating disruption using sex pheromone (ROTHSCHILD, 1981, 1982; MOFFITT and WESTIGARD, 1984; PREE et al., 1994; CARDÉ and MINKS, 1995; KEHAT et al., 1995; TRIMBLE, 1995). For example, at Road II (located

between a primary school and two residential buildings), the control efficiency measured by petiolar and seed pod damage was not as good as that at Road III ( $t = 2.169$ , d.f. = 2,  $P = 0.162$ ) according to the 2-year pheromone treatment. Besides the age and height of the trees (ZHANG et al., 2001a), the immigration of mated females is the key factor which influences the control efficacy. The air mobility was much greater at Road II than at Road III, in part, because of greater building spacing in the pheromone treatment plot at Road II. However, at Road III, the air mobility was much less than at Road II as it was located between two residential buildings, with less building spacing.

These results suggest that pheromone-mediated mating disruption can effectively shut down pheromone-baited trap catches, reduce caged virgin female mating, reduce damage from *C. trasi*s larvae and lower pupating and hibernating populations on tree trunks of urban trees (in this case, Chinese scholar-tree). Although, it would be impractical to deploy dispensers widely as used in the present study, it is a possible management strategy under urban conditions. Further research is required to determine the effect of other formulations (such as a sprayable microencapsulated formulation) on the efficacy of mating disruption.

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