Development of a high-efficiency sex pheromone formula to control *Euproctis pseudoconspersa*[1]

LI Zhao-qun[1], YUAN Ting-ting[1], CUI Shao-wei[1], ZHAO Ying-jie[1], SHAO Yuan-hai[2], SHANG Jian-nong[3], LUO Zong-xiu[1], CAI Xiao-ming[1], BIAN Lei[1], CHEN Zong-mao[1]

1 Key Laboratory of Tea Biology and Resource Utilization, Ministry of Agriculture, Tea Research Institute, Chinese Academy of Agricultural Science, Hangzhou 310008, P.R.China
2 Tea Variety Research Institute of Wuxi, Wuxi 214122, P.R.China
3 Agricultural Technology Extension Service Center of Hangzhou Xihu District, Hangzhou 310063, P.R.China

Abstract

The tea tussock moth (*Euproctis pseudoconspersa)* is one of the most destructive chewing pests in tea plantations and causes a serious allergic reaction on the skin of tea plantation workers. The sex pheromone components of its Japanese population were first identified as 10,14-dimethylpentadecyl isobutyrate (10Me14Me-15:iBu) and 14-methylpentadecyl isobutyrate (14Me-15:iBu), with a stereogenic center. Only 10Me14Me-15:iBu has been identified in the Chinese *E. pseudoconspersa* population. However, field tests have shown that 10Me14Me-15:iBu cannot meet the demand of effective pest control in China. To develop a high-efficiency *E. pseudoconspersa* sex pheromone formula, electroantennogram (EAG) recordings of (*S*)- and (*R*)-enantiomers of 10Me14Me-15:iBu and 14Me-15:iBu were obtained in the present study. The results demonstrated that the EAG responses of male antennae to (*R*)-enantiomers were significantly higher than responses to the (*S*)-enantiomers, and 14Me-15:iBu c also elicited EAG activity. Field tests showed that the catch numbers of male moths by (*R*)-enantiomers were significantly higher than those of *(S)*-enantiomers. Addition of 14Me-15:iBu significantly increased the catch numbers of both the (*S*)- and (*R*)-enantiomers. The efficient pheromone formula containing 0.75 mg *R*-10Me14Me-15:iBu and 0.1 mg 14Me-15:iBu showed significantly higher attractiveness than commercial pheromone products. Our study demonstrated that *R*-10Me14Me-15:iBu was the major sex pheromone component of *E. pseudoconspersa,* and 14Me-15:iBu might

[1] Correspondence LI Zhao-qun, E-mail: zqli@tricaas.com

be the minor sex pheromone component. Furthermore, a high-efficiency sex pheromone formula for *E. pseudoconspersa* control was defined in this study.

Keywords: *Euproctis pseudoconspersa*, sex pheromone, chirality, electroantennogram

1. Introduction

Tea is one of the most popular beverages worldwide, and China is the largest tea-planting and producing country (Chen and Lin 2015). The tea tussock moth (*Euproctis pseudoconspersa*) is one of the most destructive chewing pests in the tea plantations of China. Its larvae damage tender and mature leaves of tea plants, causing severe yield losses. Moreover, it causes severe allergic reactions on human skin, making it difficult for workers to pick tea leaves and spray pesticides (Wang *et al.* 2021). In recent years, the occurrence of *E. pseudoconspersa* has expanded considerably.

Female moths secrete volatile compounds, the sex pheromones, which are utilized for interspecific communication (Witzgall *et al.* 2010). Synthetic sex pheromones have been widely used for monitoring, mating disruption, and mass trapping of pests (Knight *et al*. 2012; Liebhold and Tobin, 2008; Luo *et al.* 2020). Based on their chemical structure, pheromones are classified into four groups: Type I, unsaturated straight-chain hydrocarbons with an ester linkage, alcohols, or aldehydes as terminal functional groups (Ando *et al.* 2004); Type II, unsaturated straight-chain hydrocarbons and their epoxide derivatives; Type III, similar to Type I and Type II with one or more methyl branches (Lofstedt *et al.* 2016); and Type 0, a proposed group containing short-chain methylcarbinols and methylketones (Jurenka, 2021). One of the most important among them is methyl-branched sex pheromones because they include stereogenic centers (Ando and Yamakawa, 2015; Mori 2007).

Wakamura et al. first identified the sex pheromones of *E. pseudoconspersa* (Japanese biotype) as 10,14-dimethylpentadecyl isobutyrate (10Me14Me-15:iBu) and 14-methylpentadecyl isobutyrate (14Me-15:iBu) (Wakamura *et al.* 1994). According to their chemical structures, these components belong to the Type III pheromone group. The major component (10Me14Me-15:iBu) attracts wild male moths, and the attractant activity increases when it is blended with the minor component (14Me-15:iBu). 10Me14Me-15:iBu is

a chiral methyl-branched pheromone. The EAG tests have showed that the (*R*)-enantiomer induced a larger response than the (*S*)-enantiomer. However, field studies of the Japanese population have reported that the catch number difference between the (*R*)-enantiomer, (*S*)-enantiomer, and the racemic mixture was insignificant (Ichikawa *et al.* 1995; Wakamura *et al.* 1996).

Only a single electroantennogram (EAG)-active compound, 10Me14Me-15:iBu, has been identified from the sex pheromone gland of the Chinese biotype of *E. pseudoconspersa* (Zhao *et al.* 1996). Researchers believe that 10Me14Me-15:iBu is responsible for most of the biological activity associated with the sex-attractant pheromone of this species. The racemate of 10Me14Me-15:iBu has been used for commercial lure production. When used for mass trapping of *E. pseudoconspersa*, the synthetic racemate of 10Me14Me-15:iBu reduces larval and egg densities by only 27.87-50.85% and 38.89-51.11%, respectively (Wang *et al*. 2005). Therefore, the attractiveness of synthetic *E. pseudoconspersa* sex pheromones requires further improvement. In this study, we evaluated the EAG-active component and field attractiveness of the two enantiomeric forms of 10Me14Me-15:iBu and 14Me-15:iBu to develop a high-efficiency *E. pseudoconspersa* sex pheromone formula.

2. Materials and methods

2.1 Insects

Larvae of *E. pseudoconspersa* were collected from a tea plantation of the Tea Research Institute of Jiangsu Province in Wuxi City, Jiangsu Province, China (120.267°E, 31.46°N). The larvae were reared on fresh tea shoots in climate-controlled rooms with a 14-h light:10-h darkness photoperiod at (25±1)℃ and (70±5)% relative humidity. After pupation, male and female pupae were separated into different cages (50 cm×50 cm×50 cm) and kept in darkness until emergence. Adult moths were then provided with a 10% honey solution. Virgin male moths (2 days old) were used for EAG recordings.

2.2 Chemicals

The sex pheromone components *S*-10Me14Me-15:iBu, *R*-10Me14Me-15:iBu, and 14Me-15:iBu were synthesized by ZeQuan Bio-technology Co., Ltd. (Hangzhou, China).

S-10Me14Me-15:iBu and *R*-10Me14Me-15:iBu were synthesized from (*S*)- and (*R*)-citronellols, respectively, according to previously reported methods (Ichikawa *et al.* 1995). The NMR data of the components were: (1) *S*-10Me14Me-15:iBu ^{1}H NMR (400 MHz, Chloroform-d) δ 4.10 (t, *J*=6.7 Hz, 2H), 2.58 (p, *J*=7.0 Hz, 1H), 1.70–1.62 (m, 2H), 1.56 (m, 1H), 1.43–1.24 (m, 17H), 1.21 (d, *J*=7.0 Hz, 6H), 1.19–1.05 (m, 4H), 0.91 (d, *J*=6.6 Hz, 6H), 0.88 (d, *J*=6.5 Hz, 3H); (2) *R*-10Me14Me-15:iBu: ^{1}H NMR (400 MHz, Chloroform-d) δ 4.10 (t, *J*=6.7, 2H), 2.58 (p, *J*=7.0 Hz, 1H), 1.72–1.62 (m, 2H), 1.56 (m, 1H), 1.43–1.25 (m, 17H), 1.21 (d, *J*=7.0 Hz, 6H), 1.20–1.05 (m, 4H), 0.92 (d, *J*=6.6 Hz, 6H), 0.89 (d, *J*=6.5 Hz, 3H); (3) 14Me-15:iBu: ^{1}H NMR (400 MHz, Chloroform-d) δ 4.09 (t, *J*=6.7 Hz, 2H), 2.57 (p, *J*=7.0 Hz, 1H), 1.71–1.61 (m, 2H), 1.54 (m, 1H), 1.40–1.27 (m, 20H), 1.20 (d, *J*=7.0 Hz, 6H), 1.19–1.15 (m, 2H), 0.90 (d, *J*=6.6 Hz, 6H). Gas chromatography was used to ensure that the purity of each chemical was greater than 99%.

2.3 EAG recordings

Solutions of *R*-10Me14Me-15:iBu, *S*-10Me14Me-15:iBu, and 14Me-15:iBu were prepared in hexane at different concentrations (0.001, 0.01, 0.1, 1.0, and 10 μg μL^{-1}). Both the basal and distal segments of the antenna of two-day-old virgin males were removed. The antenna was then connected to Ag-AgCl electrodes filled with saline solution. For each tested chemical compound, a filter paper strip (0.5 cm×5.0 cm) containing 10 μl of test solution was placed into a 13.5 cm long Pasteur pipette. The tip end of the Pasteur pipette was connected to a hole in a glass tube, which directed a charcoal-filtered and humidified airstream (800 mL min^{-1}) by an air flow controller (CS-55; Syntech Inc., Netherlands). The base of the Pasteur pipette was linked to a pulse airflow pipe. Each test compound was expelled into the airstream when the baseline of the EAG signal remained stable. Nonanal was selected as the reference compound. The recording trial was performed in the following sequence: n-hexane, reference compound, test compound (compounds were tested from low to high dosage), reference compound, and n-hexane. Each dosage was tested for at least five antennae, with a 30 s interval to allow the antenna to recover. The electrical signal was amplified and converted to a digital signal by IDAC (Syntech Inc., Netherlands). The digital signals were recorded with EAGPro software (version 2.0; Syntech Inc., Netherlands).

2.4 Field trapping tests

The field trapping tests were performed in the tea plantations of Wuxi City (Zhejiang, China; 120.267°E, 31.46°N) and Guilin (Guangxi, China; 110.17°E, 25.32°N) between June and September in 2019 and 2020. The synthetic sex pheromone compounds, *R*-10Me14Me-15:iBu, *S*-10Me14Me-15:iBu, and 14Me-15:iBu, were dissolved in distilled hexane (10 μg μL^{-1}). The sex pheromone solutions were then added to white rubber septa (8 mm O.D.; Sigma Aldrich Inc., St. Louis, MO) that were used as delivery materials. Control rubber septa were loaded with 100 μL of n-hexane. After the volatilization of n-hexane, each group of septa was packed and stored at -20℃ until their next use. The commercial *E. pseudoconspersa* sex pheromone lures used in this study were purchased from Ningbo Newcon Biotechnology Inc. (Ningbo, China), Pherobio Technology Co., Ltd. (Beijing, China), Hangzhou Zhongcha Technical Service Co., Ltd. (Hangzhou, China), and Zhangzhou Yingge'er Agricultural Technology Co., Ltd. (Zhangzhou, China). A combination of septa and commercial sex pheromone lures were placed in wing traps and hung separately 20 cm above tea plants at intervals of 12 m. Four replicates of each treatment were tested, and the captured males were counted every week.

2.5 Statistical analyses

SPSS (v.26.0 2019, IBM Corp., Armonk, NY) was used for the statistical analyses. The data was analyzed using a one-way analysis of variance and compared using Tukey's B with *P*-values adjusted for multiple comparisons. To homogenize the variance, means were transformed using the log(x+1) transformation.

3. Results

3.1 EAG responses of males

The sex pheromone components, *R*-10Me14Me-15:iBu and *S*-10Me14Me-15:iBu, were individually tested to verify whether male *E. pseudoconspersa* showed different EAG responses to both the (*S*)- and (*R*)-enantiomers. The results demonstrated that the EAG responses of male antennae to *S*-10Me14Me-15:iBu, *R*-10Me14Me-15:iBu, and 14Me-15:iBu were sigmoidal dose‑response curves (Fig. 1-A and B). However, EAG responses of (*R*)-enantiomers elicited significantly higher EAG activity in male *E.*

pseudoconspersa than those of (*S*)-enantiomers at 1 μg and 10 μg doses (Fig. 1-A).

3.2 Field trapping tests

To study the attractiveness of (*S*)- and (*R*)-enantiomers of *E. pseudoconspersa* major sex pheromone components, *R*-10Me14Me-15:iBu and *S*-10Me14Me-15:iBu were tested individually and in combination with 14Me-15:iBu in the tea garden of Wuxi (Fig. 2). The result showed that *E. pseudoconspersa* were captured with both the (*S*)- and (*R*)-enantiomers. However, the number of males captured with the (*R*)-enantiomer was significantly higher than those captured with the *(S)*-enantiomer. Moreover, almost no males were captured with only 14Me-15:iBu during the test period. Blending with 14Me-15:iBu significantly increased the number of males captured with both (*S*)- and (*R*)-enantiomers. Even after blending with 14Me-15:iBu, the catch numbers of (*R*)-enantiomers were significantly higher than those of (*S*)-enantiomers.

The dose effect of *R*-10Me14Me-15:iBu (blended with 14Me-15:iBu) on the sex pheromones of *E. pseudoconspersa* was then examined in the tea gardens of both Guilin and Wuxi (Fig. 3). The results showed that male catch numbers significantly increased when the dose of *R*-10Me14Me-15:iBu was raised from 0 mg per septum to 0.75 mg per septum. However, the catch numbers declined at the 1 mg dose level. Furthermore, the male catch number with *R*-10Me14Me-15:iBu was compared with the racemic mixture of 10Me14Me-15:iBu blended with 0.1 mg per septum of 14Me-15:iBu (Fig. 4). The comparative analysis showed that catch numbers with *R*-10Me14Me-15:iBu were significantly greater than those with the racemic mixture.

The sex pheromone formula combining different doses of 14Me-15:iBu (0, 0.1, 0.5, 1.0 mg per septum) and 1.0 mg per septum of *R*-10Me14Me-15:iBu were evaluated for their attractiveness to *E. pseudoconspersa* male moths in Wuxi (Figure 5). The trap test catch numbers demonstrated that 0.1 mg per septum of 14Me-15:iBu blended with *R*-10Me14Me-15:iBu showed the highest attractiveness, and that the male catch number declined at higher doses of 14Me-15:iBu.

According to the above results, an efficient sex pheromone formula should contain 0.75 mg per septum of *R*-10Me14Me-15:iBu and 0.1 mg per septum of 14Me-15:iBu. To evaluate its attractiveness, the proposed formula was compared with the commercial *E.*

pseudoconspersa sex pheromone lures from four companies in the tea gardens of Wuxi and Guilin (Fig. 6). The field trapping test showed that the *E. pseudoconspersa* male catch numbers of the proposed sex pheromone formula were significantly (more than two times) higher than those of the four commercial sex pheromone lures. The catch numbers among the four commercial sex pheromone lures were similar.

4. Discussion

Synthetic sex pheromones have been successfully used in pest control as an alternative to insecticides (Cui and Zhu 2016; Witzgall *et al.* 2010). However, mass trapping experiments have shown that the synthetic sex pheromones of *E. pseudoconspersa* cannot meet the demands of effective pest control. To improve the attractiveness of synthetic *E. pseudoconspersa* sex pheromones, EAG and field trapping tests were performed to verify the following: (1) whether different chiral isomers have different activities; (2) how the chiral isomers work; and (3) the function of the minor sex pheromone component in field trapping.

The major sex pheromone component of *E. pseudoconspersa*, 10Me14Me-15:iBu, is a chiral compound (Ichikawa *et al.* 1995). Enantiomers can influence the activity of the chiral pheromone if: (1) both enantiomers are active; (2) one enantiomer is active and the other is inactive; (3) one enantiomer is active but the other is inhibitory; and (4) enantiomers are active in combination but individually inactive. Our study demonstrated that both the (*S*)- and (*R*)-enantiomers of 10Me14Me-15:iBu showed EAG responses and field attractiveness, and the male catch number of the (*R*)-enantiomers was significantly higher than those of the (*S*)-enantiomers and racemic mixture. Therefore, the results indicated that *R*-10Me14Me-15:iBu is the major sex pheromone component of *E. pseudoconspersa*. In contrast, previous studies on the Japanese biotype of *E. pseudoconspersa* reported that the catch numbers of the (*R*)-enantiomers and the racemic mixture were similar (Wakamura *et al.* 1996). This difference might be related to the pheromone polymorphism between geographical populations.

As a minor sex pheromone component, 14Me-15:iBu could increase the male catch numbers of the Japanese biotype of *E. pseudoconspersa* when blended with

10Me14Me-15:iBu (Wakamura *et al.* 1994). However, it has not been identified from the Chinese biotype of this species (Zhao *et al*. 1996). Our study showed that 14Me-15:iBu elicited EAG activity of male antennae with a typical sigmoidal dose - response curve. Furthermore, field tests demonstrated that this compound significantly increased the catch numbers with *R*-10Me14Me-15:iBu and *S*-10Me14Me-15:iBu. We speculated that 14Me-15:iBu might also be the minor sex pheromone component of *E. pseudoconspersa* in China. It may likely not have been identified because of its low abundance in the sex pheromone gland.

The ratio and dose of components directly affect the attractiveness of synthetic sex pheromones (Ma *et al.* 2016; Yan *et al.* 2019). Our field tests showed that *R*-10Me14Me-15:iBu and 14Me-15:iBu have the highest catch numbers at doses of 0.75 mg per septum and 0.1 mg per septum, respectively. Our speculation that an efficient formula containing 0.75 mg *R*-10Me14Me-15:iBu and 0.1 mg 14Me-15:iBu has a higher attractiveness was successfully proved by the field tests. The efficient sex pheromone formula captured more than twice the number of male moths captured by commercial sex pheromone products. This formula will improve the control efficiency of sex pheromones in *E. pseudoconspersa*. Further research on the application of the efficient formula in mass trapping and mating disruption is required to achieve pheromone-based *E. pseudoconspersa* control.

5. Conclusion

Only 10Me14Me-15:iBu has been identified from the Chinese biotype of *E. pseudoconspersa*. Accordingly, the racemate of 10Me14Me-15:iBu has been used for commercial lure production. However, it cannot meet the demands of effective pest control. The present study focused on EAG activities and field attractiveness of chiral enantiomers of 10Me14Me-15:iBu and the minor sex pheromone component of the Japanese population, 14Me-15:iBu. It demonstrated that *R*-10Me14Me-15:iBu is the major sex pheromone component of *E. pseudoconspersa,* and 14Me-15:iBu can elicit the EAG activity of male antennae and significantly increase male moth catch numbers. Therefore, 14Me-15:iBu

might be the minor sex pheromone component. Furthermore, an efficient sex pheromone formula containing 0.75 mg *R*-10Me14Me-15:iBu and 0.1 mg 14Me-15:iBu has significantly higher attractiveness than commercial pheromone products. These findings will improve the control efficiency of sex pheromones in *E. pseudoconspersa*.

Acknowledgements

This study was funded by the National Key Research & Development (R&D) Plan (2021YFD1601100), the Key Research and Development Program of Zhejiang Province, China (2019C02033), the National Natural Science Foundation of China (31701795), and the Modern Agricultural Industry Technology System (CARS-19).

Declaration of competing interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

References

Ando T, Inomata S, Yamamoto M. 2004. Lepidopteran sex pheromones. In: Schulz S, ed, *The Chemistry of Pheromones and Other Semiochemicals I*. Vol. 239. Springer: Berlin Heidelberg, Germany, pp. 51-96.

Ando T, Yamakawa R. 2015. Chiral methyl-branched pheromones. *Natural Product Reports*, **32**, 1007-1041.

Chen ZM, Lin Z. 2015. Tea and human health: biomedical functions of tea active components and current issues. *Journal of Zhejiang University Science B*, **16**, 87-102.

Cui GZ, Zhu JJ. 2016. Pheromone-based pest management in China: past, present, and future prospects. *Journal of Chemical Ecology*, **42**, 557-570.

Jurenka RA. 2021. 2-Lepidoptera: Female sex pheromone biosynthesis and its hormonal regulation. In: Gary JB, Richard GV. Insect Pheromone Biochemistry and Molecular Biology (Second Edition). Elsevier: London, United Kingdom, pp. 13-88.

Ichikawa A, Yasuda T, Wakamura S. 1995. Absolute-configuration of sex-pheromone for tea tussock moth, *Euproctis-pseudoconspersa* (strand) via synthesis of (r)-10,14-dimethyl-1-pentadecyl and (s)-10,14-dimethyl-1-pentadecyl isobutyrates. *Journal of Chemical Ecology*, **21**, 627-634.

Knight AL, Stelinski LL, Hebert V, Gut L, Light D, Brunner J. 2012. Evaluation of novel semiochemical dispensers simultaneously releasing pear ester and sex pheromone for mating disruption of codling moth (Lepidoptera: Tortricidae). *Journal of Applied Entomology*, **136**, 79-86.

Liebhold AM, Tobin PC. 2008. Population ecology of insect invasions and their management. *Annual Review of*

Entomology, **53**, 387-408.

Lofstedt C, Wahlberg N, Millar JG. 2016. Evolutionary patterns of pheromone diversity in Lepidoptera. In: Jeremy DA, Ring TC. *Pheromone Communication in Moths: Evolution, Behavior, and Application*. Elsevier: London, United Kingdom, pp. 43-78.

Luo ZX, Magsi DH, Li ZQ, Cai XM, Bian L, Liu Y, Xin ZJ, Xiu CL, Chen ZM. 2020. Development and evaluation of sex pheromone mass trapping technology for *Ectropis grisescens*: a potential integrated pest management strategy. *Insects*, **11**, 15.

Ma T, Xiao Q, Yu YG, Wang C, Zhu CQ, Sun ZH, Chen XY, Wen XJ. 2016. Analysis of Tea Geometrid (*Ectropis grisescens*) Pheromone Gland Extracts Using GC-EAD and GCxGC/TOFMS. *Journal of Agricultural And Food Chemistry*, **64**, 3161-3166.

Mori K. 2007. Significance of chirality in pheromone science. *Bioorganic & Medicinal Chemistry*, **15**, 7505-7523.

Sun ZF, Zhou LN, Meng YF, Zhang T, Du ZT, Zheng HJ. 2017. Concise asymmetric synthesis of the sex pheromone of the tea tussock moth. *Tetrahedron-Asymmetr*, **28**, 1562-1567.

Tsai RS, Yang EN, Wu CY, Tseng HK, Chow YS. 1999. A potent sex attractant of the male tea tussock moth, *Euproctis pseudoconspersa* (Strand) (Lepidoptera : Lymantriidae) in Taiwan: Field and EAG responses. *Zoological Studies*, **38**, 301-306.

Wakamura S, Ichikawa A, Yasuda T, Arakaki N, Fukumoto T. 1996. EAG and field responses of the male tea tussock moth, *Euproctis pseudoconspersa* (Strand) (Lepidoptera: Lymantriidae) to (R)- and (S)-enantiomers and racemic mixture of 10,14-dimethylpentadecyl isobutyrate. *Applied Entomology and Zoology*, **31**,623-625.

Wakamura S, Yasuda T, Ichikawa A, Fukumoto T, Mochizuki F. 1994. Sex attractant pheromone of the tea tussock moth, *Euproctis pseudoconspersa* (Strand) (Lepidoptera: Lymantriidae): identification and field attraction. *Applied Entomology and Zoology*, **29**, 403-411.

Wang X, Gu Q, Zhang W, Jiang H, Chen S, Smagghe G, Niu J, Wang JJ. 2021. Prevalence of a novel bunyavirus in tea tussock moth *Euproctis pseudoconspersa* (Lepidoptera: Lymantriidae). *Journal of Insect Science*, **21**(4), 5.

Wang YM, Ge F, Liu XH, Feng F, Wang LJ. 2005. Evaluation of mass-trapping for control of tea tussock moth *Euproctis pseudoconspersa* (Strand) (Lepidoptera : Lymantriidae) with synthetic sex pheromone in south China. *International Journal of Pest Management*, **51**, 289-295.

Witzgall P, Kirsch P, Cork A. 2010. Sex pheromones and their impact on pest management. *Journal of Chemical Ecology*, **36**, 80-100.

Yan Q, Liu X-L, Wang Y-L, Tang X-Q, Shen Z-J, Dong S-L, Deng J-Y. 2019. Two Sympatric Spodoptera Species Could Mutually Recognize Sex Pheromone Components for Behavioral Isolation. *Frontiers in Physiology*, **10**, 1256.

Zhang HL, Sun ZF, Zhou LN, Liu L, Zhang T, Du ZT. 2018. Synthesis of the sex pheromone of the tea tussock moth based on a resource chemistry strategy. Molecules. **23**, 1347.

Zhao C, Millar JG, Wen Z, Wang S, Wang X, Zhu Y. 1996. Isolation, identification and synthesis of the female sex pheromone of the tea tussock moth, *Euproctis pseudoconspersa* (Lepidoptera: Lymantriidae). *Entomologia Sinica*, **3**, 58-69 (in Chiense) .

Figure legends

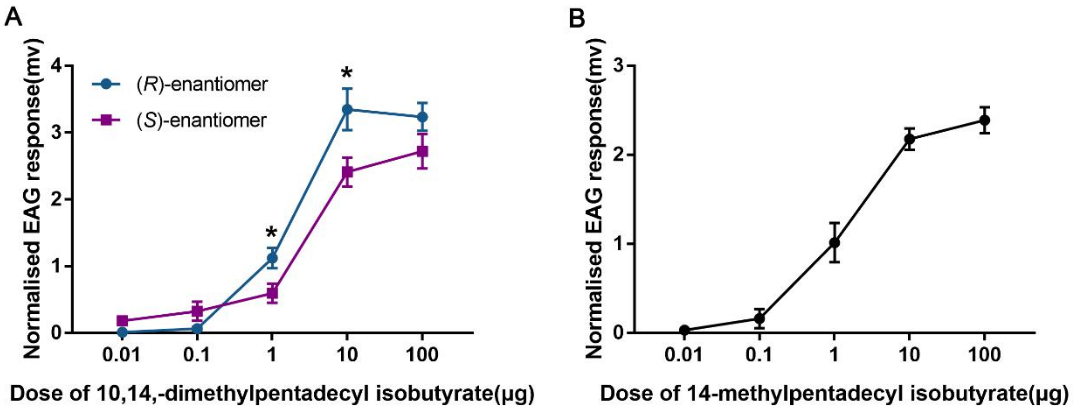

Fig. 1 Electroantennographic (EAG) responses (mean mV±SE, *n*=6) of male *Euproctis pseudoconspersa* antennae to synthetic pheromone components. A, EAG responses to the (*S*)- and (*R*)-enantiomers of 10Me14Me-15:iBu. B, EAG responses to 14Me-15:iBu. Asterisks above the lines indicate significant differences assessed via analysis of variance (ANOVA) followed by Tukey's B (*P*<0.05).

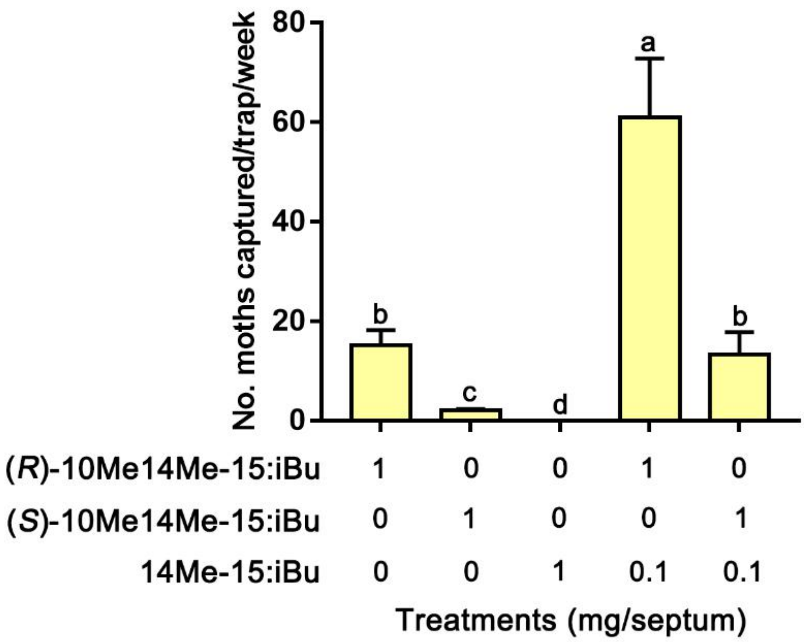

Fig. 2 Catch numbers of *Euproctis pseudoconspersa* males with different synthetic pheromone components. Data are mean±SE (*n*=4). Letters above the bars indicate significant differences assessed *via* ANOVA followed by Tukey's B (*P*<0.05).

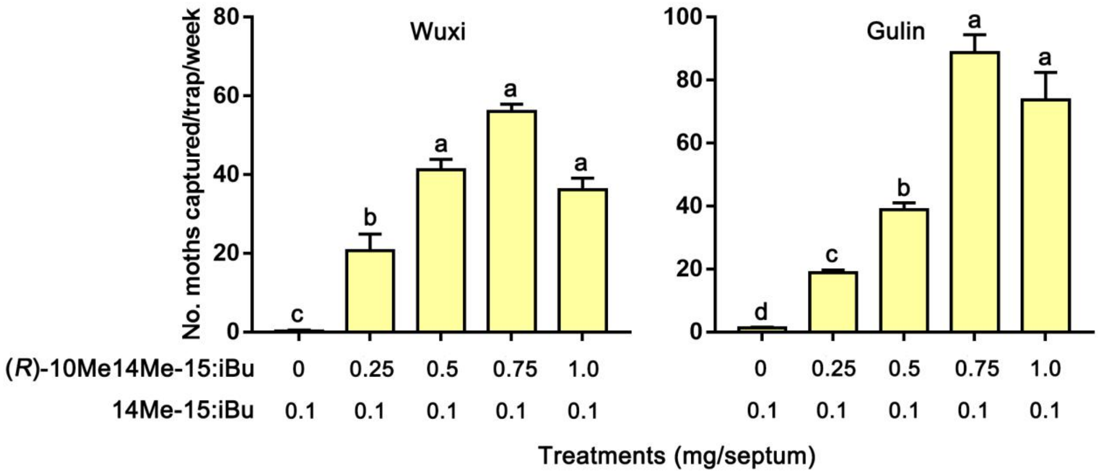

Fig. 3 Catch numbers of *Euproctis pseudoconspersa* males with different dosages of *R*-10Me14Me-15:iBu. Data are mean±SE (*n*=4). Letters above the bars indicate significant differences assessed *via* ANOVA followed by Tukey's B (*P*<0.05).

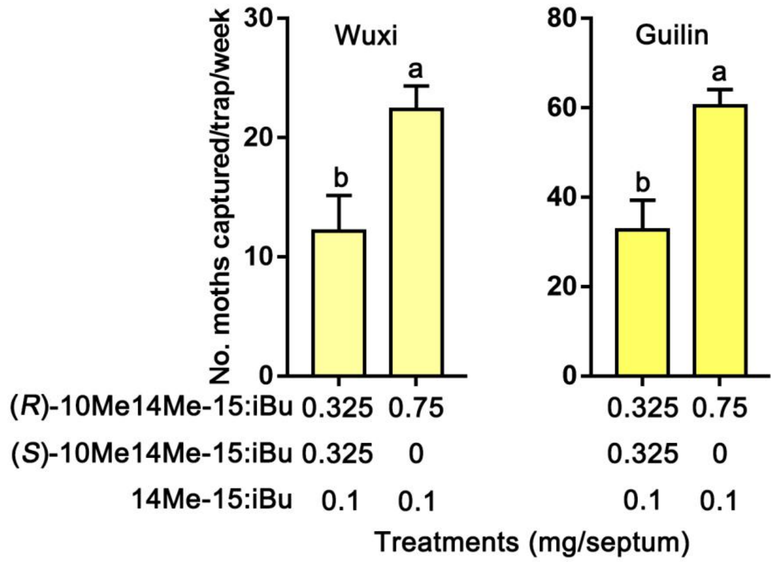

Fig. 4 Catch numbers of *Euproctis pseudoconspersa* males with *R*-10Me14Me-15:iBu and a racemic mixture of 10Me14Me-15:iBu. Data are mean±SE (*n*=4). Letters above the bars indicate significant differences assessed *via* ANOVA followed by Tukey's B (*P*<0.05).

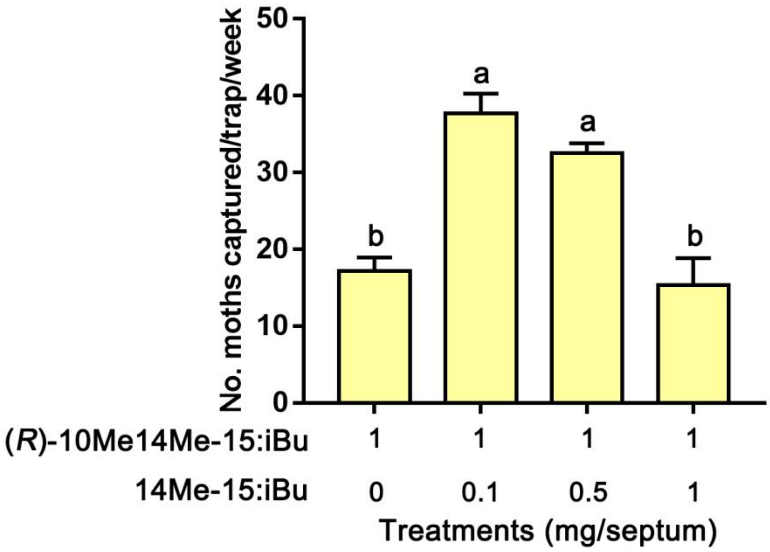

Fig. 5 Catch numbers of *Euproctis pseudoconspersa* males with different dosages of 14Me-15:iBu. Data are mean±SE (*n*=4). Letters above the bars indicate significant differences assessed *via* ANOVA followed by Tukey's B (*P*<0.05).

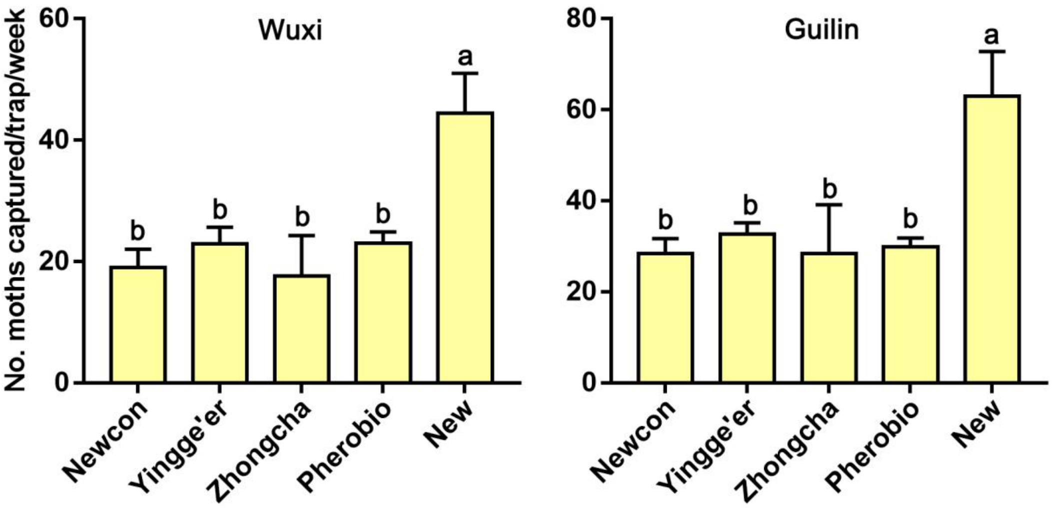

Fig. 6 Comparison of *Euproctis pseudoconspersa* male catch numbers with the efficiency pheromone formula and commercial pheromone products. NK, Newcon commercial pheromone products from Ningbo Newcon Biotechnology Inc.; YG, Yingge'er commercial pheromone products from Zhangzhou Yingge'er Agricultural Technology Co., Ltd.; ZC, Zhongcha commercial pheromone products from Hangzhou Zhongcha Technical Service Co., Ltd.; ZJ, Pherobio commercial pheromone products from Pherobio Technology Co., Ltd.; New, efficiency pheromone formula developed by the present study. Data are mean±SE (*n*=4). Letters above the bars indicate significant differences assessed *via* ANOVA followed by Tukey's B (*P*<0.05).