

## FIELD TRIALS WITH THE SYNTHETIC SEX PHEROMONE OF THE OAK PROCESSIONARY MOTH

### *Thaumetopoea processionea*

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**Abstract**—The biological activity of synthetic (Z,Z)-11,13-hexadecadienyl acetate, the major pheromone component found in female gland extracts of the oak processionary moth *Thaumetopoea processionea*, was evaluated in field trials. Traps baited with 10 mg of the chemical efficiently attracted a large number of males provided they were placed in the upper crown region of the oaks. Devices positioned 10–15 m high in the trees attracted significantly more males than those traps installed at 2 or 6–8 m above the ground. Pherocon traps were slightly more efficient than Delta traps, and lower or higher amounts of the attractant in the baits did not significantly influence the number of moths caught. The importance of the stereomeric purity of the lure and the easy isomerization of the (Z,Z)-acetate to other isomers, particularly to the *E,E* isomer, should be considered for the development of efficient formulations in the field.

**Key Words**—Sex pheromone, oak processionary moth, *Thaumetopoea processionea*, Lepidoptera, Thaumetopoeidae, field trials, pheromone traps.

## INTRODUCTION

The oak processionary moth *Thaumetopoea processionea* (L.) (Lepidoptera, Thaumetopoeidae) is a serious forest pest in many areas of western, southern,

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and central Europe. The larvae hatch during spring synchronously with sprouting oak leaves, develop within 2–3 months, and pupate in nests in early summer. The insect is univoltine, and the adults emerge in July–August, although the flight period is often restricted to a few weeks. In addition to the damage caused to oak trees, larvae disperse highly urticating microscopic hairs into the environment causing strong allergic reactions in animals and humans. Quite frequently, the skin and eyes are affected, and irritation of the respiratory mucous membranes can cause asthma symptoms and anaphylactic shocks (Lamy and Novak, 1987; Lamy, 1990). In public areas, therefore, control of the pest is mandatory, but thus far an efficient method to detect infested areas is lacking. In this regard, a promising way for monitoring the appearance and population of the pest might be through utilization of the sex pheromone.

In a previous paper, we reported the characterization of the sex pheromone of the pest as a mixture of (Z,Z)-11,13-hexadecadienyl acetate (**I**), (E,Z)-11,13-hexadecadienyl acetate (**III**), and (Z,Z)-11,13-hexadecadienol (**V**) (Figure 1) in a 88:7:5 ratio from female pheromone gland extracts (Quero et al., 2003). Acetate **I** and alcohol **V** had been previously reported as possible pheromone components (Frérot and Démolin, 1993), while Germinara et al. (2001) found compound **I** in gland extracts of females. However, no biological activity was disclosed. We found that in a wind tunnel the major compound **I** induced males to display all behavioral responses, and that (E,E)-11,13-hexadecadienyl acetate (**IV**) reduced the number of contacts with the source when mixed with the major compound in several ratios (Quero et al., 2003). In the field, although Tibery and Niccoli (1984) mentioned that traps baited with (Z)-13-hexadecen-11-ynyl acetate, the sex pheromone of the pine processionary moth *Thaumetopoea pityocampa*, also captured males of *T. processionea*, we (Breuer and De Loof, 1997) and others (Montoya and Hernández, personal communication, 1986) found that this compound was inactive in field trials carried out in Belgium and Spain, respectively. In this paper, we report a series of field tests conducted with the major compound **I**, as well as the effect of different parameters on the number of catches.

#### METHODS AND MATERIALS

*Chemicals and Dispensers.* The pheromone compounds were synthesized and encapsulated in polyethylene vials ( $3 \times 1.1$  cm i.d.) by Sociedad Española de Desarrollos Químicos (SEDQ, Barberá del Vallés, Barcelona, Spain). Most of the trials were done with baits containing the following composition: (Z,Z)-11,13-hexadecadien-1-yl acetate (**I**, 93.7%), (Z,E)-11,13-hexadecadien-1-yl acetate (**II**, 3.9%), (E,Z)-11,13-hexadecadien-1-yl acetate (**III**, 2.2%), and (E,E)-11,13-hexadecadien-1-yl acetate (**IV**, 0.1%) (Figure 1). To study the activity of the E,E isomer, a second formulation consisting of a mixture of (E,E)-acetate **IV**

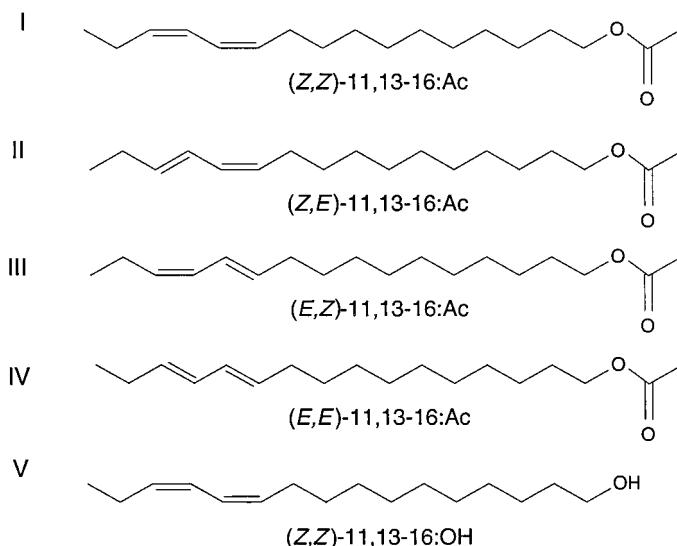


FIG. 1. Structures of pheromone compounds and analogues considered.

(72.8%), (*E,Z*)-acetate **III** (11.1%), (*Z,E*)-acetate **II** (3.1%), and (*Z,Z*)-acetate **I** (2.9%) was also tested. In both formulations, 10 mg of the major component, either the *Z,Z* or *E,E* isomer, and 1% of BHT (*t*-butylhydroxytoluene) as stabilizer, were encapsulated in polyethylene vials, packed in aluminum foil, and stored at  $-20^{\circ}\text{C}$  until use. Stereochemical analyses were performed by GC either on a cross-linked HP-FFAP capillary column (25 m  $\times$  0.2 mm i.d., 0.33  $\mu\text{m}$ ) (Agilent Technologies, Barcelona, Spain) or on an HP-1 column (12 m  $\times$  0.2 mm i.d., 0.33  $\mu\text{m}$ ) (Agilent Technologies, Barcelona, Spain).

**Pheromone Traps.** All trials were conducted using Delta traps (12 cm high) (Servei de Protecció dels Vegetals, Generalitat de Catalunya, Spain) containing a nondrying adhesive base (10  $\times$  10 cm) (Tangle-Trap Paste, The Tanglefoot Company, USA). In addition and to study the effect of other types of traps, Pherocon traps (10 cm high) (Trécé Inc., California, USA) with a sticky bottom (10  $\times$  10 cm) were also used. All traps were provided by SEDQ (Barcelona, Spain).

**Field Trials.** The tests were carried out in three different oak forests near Genthin, Germany (NE of Magdeburg) from July 23 to September 12, 2001, in a region that had been highly infested in previous years by the oak processionary moth. The main purpose of the trials was the evaluation of the effects on catches of the following parameters: (a) trap height, (b) charge of dispenser, (c) trap design, (d) infestation level, and (e) pheromone components.

Most of the traps were suspended on oak trees along arbitrary lines through the forest to facilitate compilation of data. The distance between two adjacent traps

was 30–50 m, according to the forest structure, a distance considered sufficient to prevent one trap from “poaching” moths from the plume of the closest trap. Poaching is suppression of the trap catches occurring from effects of overlapping active spaces of other traps relative to the catches by a single isolated trap. For the first series of experiments, traps were installed at 2, 6–8, and 10–15 m above the ground. For the two lower heights, the traps were provided with hooks, which were hung on the tree branches either directly by hand or with the help of a telescopic bar. To reach the upper crown region (10–15 m high), a thread was shot over a branch. The trap was fixed at one end of the thread and lifted into the crown of the tree by pulling the other end. In this way, further control of these traps could be easily done. For the other experiments, traps were all installed in the upper crown region, since more individuals were caught at this maximum height (see below).

During the flight period, traps were inspected at intervals of 3–7 days, the moths were counted, and the adhesive bottom was renewed if necessary. Except for the experiment on the trap height, traps were rotated on each inspection to minimize the possible effects of their location in the forest.

To estimate the degree of infestation in the different study areas, the percentage of leaf mass consumed by *T. processionea* in at least 10 trees randomly selected in each area was determined and averaged relative to healthy unattacked trees.

## RESULTS AND DISCUSSION

In the three study areas, the flight periods of the moths were similar. Most males were caught in the period July 24 – August 29, with a peak of catches during the 1st week of August. It is worth noting that in the more open stands, the maximum number of catches occurred earlier than in the more closed forest areas (August 2–5 vs. August 8–9, respectively). The temperature in all areas ranged from 10 to 36°C through the season.

The position of the traps on the trees had a noteworthy effect on the number of males caught. Devices positioned at the upper crown of the tree (10–15 m) attracted more males than did those installed at 6–8 or 2 m. The difference between the lowest and highest elevation was significant at  $P < 0.05$  (Mann–Whitney *U* test) (Figure 2). These are important findings for practical application. At least two possibilities may explain these results. First, males may preferentially fly at the highest altitude of the trees. Leafy twigs may play a role in orientation. It has been observed that males fly to and around such structures, and traps attach to foliated branches attract more moths than those on brittle branches (H.-G. Kontzog and M. Breuer, unpublished). Second, in a forest, wind intensity usually increases with height and, therefore, more pheromone molecules may be perceived by the male antennal receptors orienting males to the traps. In a few other pest insects, an improvement of catches by placing traps in or even over the top of trees has

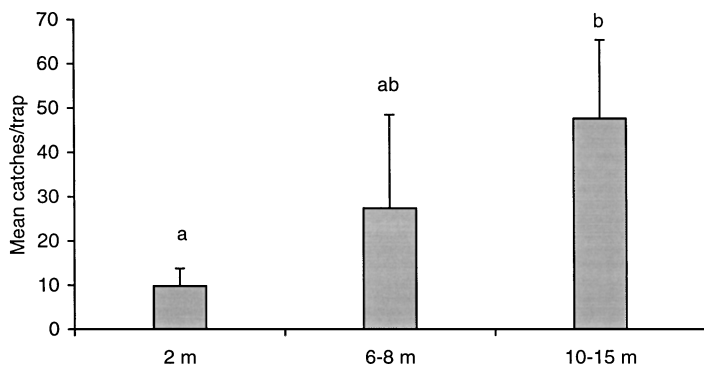


FIG. 2. Mean number of catches of *Thaumetopoea processionea* ♂♂ ( $\pm$ SD) in Delta traps placed at different heights on the tree. The dispensers were loaded with 10 mg of (Z,Z)-11,13-hexadecadienyl acetate (**I**) (stereochemical purity: Z,Z 93.7%; E,E 3.9%; E,Z 2.2%; E,E 0.1%). Similar letters on top of the bars indicate no significant differences at  $P < 0.05$  (Mann–Whitney *U* test).

been reported. Pasqualini et al. (1992, 1999) found that the level of capture of the leopard moth *Zeuzera pyrina* was improved by setting the traps at least 1 m above the apple or pear orchard. The authors speculated that in this position males may find it easier to locate traps, and at the same time competition with females may also be lower than when traps are placed inside the orchard. Similarly, Riedl et al. (1979) demonstrated that male codling moth catches are highest when traps were positioned near the top of the apple tree canopy.

Increasing or decreasing the load (5, 10, and 15 mg) of the major component **I** had no effect on the number of male catches (data are not shown). On the other hand, Pherocon traps worked more efficiently (twofold) than Delta traps, catching 59 males/trap vs. 28 males/trap, respectively. The difference was significant at  $P < 0.05$  (Mann–Whitney *U* test) (Figure 3). It is well known that trap size and shape is one of the critical parameters for a given active formulation to achieve an efficient level of male captures and, consequently, a more successful competition with virgin females (Cardé and Elkinton, 1984; Ridgway et al., 1990).

There was a clear relationship between the level of infestation (measured as defoliation caused by the larvae) and the number of moths caught (Figure 4). Traps in the center of an infested forest area attracted more males than those hanging in areas slightly damaged by the pest. The level of catches diminished when traps were placed 50–100 m away from the infested area, suggesting that the attractive range of the pheromone preparation is apparently limited. This is in contrast with the close species, *T. pityocampa*, the pine processionary moth, which was attracted to pheromone even at 1400 m away from the pine grove (A. Shani, unpublished). It is possible that, under summer conditions, the easy isomerization

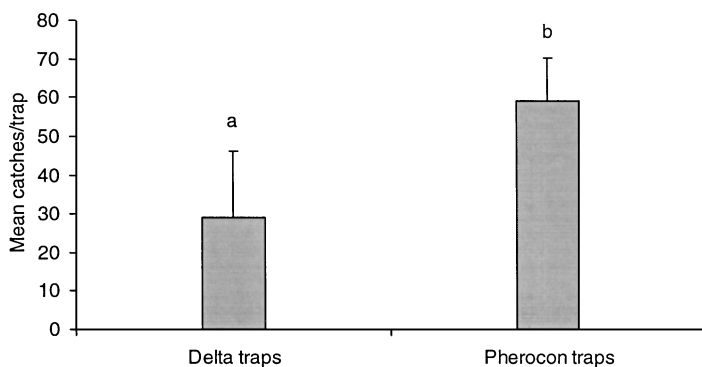


FIG. 3. Mean number of catches of *Thaumetopoea processionea* ♂♂ (±SD) in Delta and Pherocon traps baited with 10 mg of (Z,Z)-11,13-hexadecadienyl acetate (**I**) (stereochemical purity: Z,Z 93.7%; Z,E 3.9%; E,Z 2.2%; E,E 0.1%). Different letters on top of the bars indicate significant differences at  $P < 0.05$  (Mann-Whitney  $U$  test).

and relatively low thermal stability of the Z,Z-conjugated system in isomer **I** of the oak processionary moth pheromone, in contrast to the much more stable enyne group present in the pine processionary pheromone, may account for the difference in attractiveness and potency of both attractants. In this regard, in wind tunnel experiments, the E,E isomer (**IV**), the most stable isomer into which the active Z,Z isomer can degrade, has been found to be an inhibitor of contacts with the source when males are subjected to mixtures of compounds **I** and **IV** in 1:1 and 1:10 ratios in comparison to the activity of compound **I** alone (Quero et al., 2003).

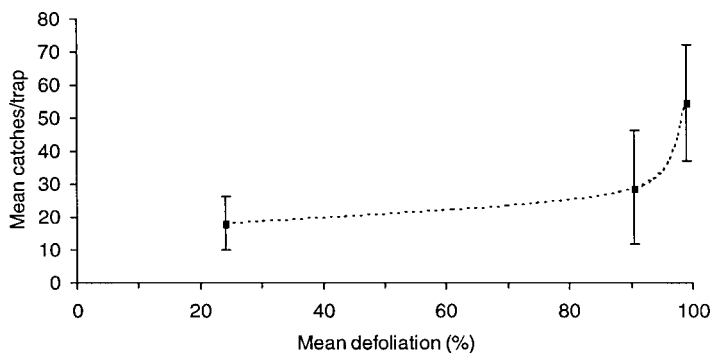


FIG. 4. Relationship between the percent of defoliation in the study areas caused by *Thaumetopoea processionea* larvae and the number of catches per trap (±SD). Delta traps baited with (Z,Z)-11,13-hexadecadienyl acetate (**I**) (stereochemical purity: Z,Z 93.7%; Z,E 3.9%; E,Z 2.2%; E,E 0.1%) were used.

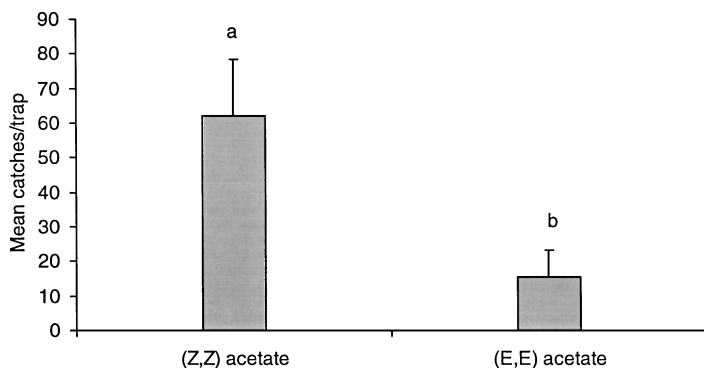


FIG. 5. Mean number of catches of *Thaumetopoea processionea* ♂♂ (±SD) in Delta traps loaded with (Z,Z)-11,13-hexadecadienyl acetate (I) (stereochemical purity: Z,Z 93.7%; Z,E 3.9%; E,Z 2.2%; E,E 0.1%) and (E,E)-11,13-hexadecadienyl acetate (IV) (stereochemical purity: E,E 72.8%, E,Z 11.1%, Z,E 3.1%, Z,Z 2.9%). Different letters on top of the bars indicate significant differences at  $P < 0.05$  (Mann Whitney  $U$  test).

To confirm the importance of the stereomeric purity of the lure, the activity of traps baited with a formulation enriched with the *E,E* isomer (stereochemical purity: *E,E* 72.8%; *E,Z* 11.1%; *Z,E* 3.1%; *Z,Z* 2.9%) was compared with that of the standard formulation. While the former attracted on average only 16.0 males/trap, the standard lure caught 62.2 males/trap (Figure 5). These data confirm the importance of the stereomeric purity of the pheromone, and the very low activity, if any, of the *E,E* isomer (the low content (2.9%) of the *Z,Z* isomer present in the second formulation may be responsible for the number of males caught). Because of the chemical and thermally easy interconversion of the isomers, the possible inhibitory effect of the *E,E* isomer in the field has not been tested, but, nevertheless, the results from the wind tunnel suggest that stereomerically pure *Z,Z* isomer is advisable for an efficient formulation, and also that measures (e.g., stabilizers, UV-protecting agents) should be taken to prevent isomerization to the *E,E* isomer under field conditions.

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