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Sex pheromone of the processionary moths and biosystematic considerations within the genus *Thaumetopoea* (Thaumetopoeidae Thaumetopoeinae)

Abstract - Identification of sex pheromone within 5 species belonging to the Thaumetopoeidae family are reported in this paper. Chemistry of the pheromone components corroborated the split between two types of processionary moths on the insect biology: *ie* the winter and summer processionary moths. Pheromone chemistry tend also to support the fact that *T. pityocampa* and *T. wilkinsoni* should be considered as ecotypes. The same conclusion is proposed for *T. pinivora* and *T. bonjeani*.

Résumé - *Phéromones sexuelles des Processionnaires and considerations sistematiques sur le genre* Thaumetopoea *(Thaumetopoiedae Thaumetopoeinae)*.

L'identification des phéromones sexuelles de 5 espèces de lépidoptères appartenant à la famille des Thaumetopoeidae est rapportée dans cette publication. La séparation des processionnaires en deux groupes, proposée d'après les particularités de leurs cycles biologiques est validée par la chimie des composés phéromonaux. Une approche chimiotaxonomique étayée par des données de biologie démontre que l'on peut considérer *T. pityocampa* et. *T. wilkinsoni* comme des écotypes. La même conclusion est proposée pour *T. pinivora* et. *T. bonjeani*.

Riassunto - *Feromoni sessuali della Processionaria e considerazioni sistematiche sul genere* Thaumetopoea *(Thaumetopoeidae Thaumetopoeinae)*.

Viene riferita l'identificazione dei feromoni sessuali di 5 specie di Taumetopeidi. Le caratteristiche chimiche dei componenti feromonici confermano la separazione di questi insetti in due gruppi già proposta sulla base del loro ciclo biologico. La composizione chimica del feromone conferma che *T. pityocampa* e *T. wilkinsoni* possono essere considerati due ecotipi cosí come lo sono *T. pinivora* e *T. bonjeani*.

Key words: Lepidoptera, Thaumetopoeidae, chemotaxonomy, chemical identification, ecotypes.

INTRODUCTION

Moths belonging to the Thaumetopoeidae family are widespread throughout Eurasia, Africa and Australia from where numerous genera have been described (Kiriakoff, 1970). The processionary moths of the Mediterranean area belong to the genus *Thaumetopoea*. They share common morphological and biological characters and form an homogeneous group (Agenjo, 1941). All the species within this genus feed on rather primitive plants, except *Thaumetopoea processionea* (L.) which feed on oak trees. *Thaumetopoea pityocampa* (Den. & Schiff.), *Thaumetopoea wilkinsoni* (Tams.), *Thaumetopoea pinivora* (Tr.), *Thaumetopoea bonjeani* (Powell) and *Thaumetopoea libanotica* (Kiriakoff & Talhouk) feed on coniferous trees whereas *Thaumetopoea jordana* (Stgr.) and *Thaumetopoea solitaria* (Fr.) feed on Terebinthaceae plants. All of them, except *Thaumetopoea herculeana* (Rbr.) which feed on Geraniacae (Gomez-Aizpurua, 1986), are reported as serious pests on either deciduous trees or conifers.

Sex pheromone is the intraspecific sexual communication system used in virtually all moths. It is generally expressed as a female-produced chemical that activates the conspecific male sexual response. Identifications of sex pheromones were undertaken to provide a tool to detect the presence of these insects and to monitor their populations. On the other hand, the evidence of a relation between the identified molecules and the systematic position within the families studied and the ease with which sex pheromones can be used in determining specificity of taxa, has led to consider them as an important chemiotaxonomic tool (Roelofs & Comeau, 1969; Roelofs & Brown, 1982) and to take them into account for considerations in taxonomy (Foster & Dugdale, 1988; Frérot & Foster, 1991).

RESULTS AND DISCUSSION

Sex pheromone composition was studied in 6 of the quoted species to which we added for discussion the previously identified sex pheromone of *T. pityocampa* (Guerrero et al., 1980) (Fig. 1). Identifications were performed by gas chromatography analyses alone (GC) or linked with mass spectrometry (GCMS) and were achieved by comparisons of both retention times and mass spectra with those of synthetic samples.

Chemical structures of the identified compounds reveal the homogeneity of the group (Fig. 1). All the components possess an aliphatic chain of 16 carbons with insaturations situated in the 11^{th} and the 13^{th} positions, except for *T. solitaria*. Nevertheless, two different groups come into view: the first

characterized by a conjugated dienic structure represented by *T. processionea, T. bonjeani, T. pinivora*; the second with a conjugated enynic structure represented by *T. pityocampa, T. wilkinsoni* and *T. jordana*. Roelofs and Bjostad (1984) have demonstrated that within the Tortricidae elucidation of biosynthetic routes of pheromones is a useful criterion for determining evolutionary relationships. Biochemical studies have identified enzymes that regulated processes inside the sex pheromone gland of *T. pityocampa* (Arsequell et al., 1990) and have shown that (Z)-13-hexadecen-11-ynyl acetate, the main sex pheromone component, is biosynthesized from palmitic acid by combined action of delta - 11 and delta - 13 desaturases. For all the identified pheromones of processionary moths, except for *T. solitaria*, the trivial precursor should be the palmitic acid as described in *T. pityocampa*. This precursor is found in all the extracts of pheromone glands analysed. The delta - 13 desaturase is unusual in Lepidoptera and thus enhances the originality of this family within the Lepidopteran. The delta - 11 desaturase responsible for triple bond, present in only the second group, is also an unusual enzyme (Arsequell et al., 1990) among the Lepidoptera whereas the delta - 11 desaturase of the first group which is responsible for double bond should be identical to the most common desaturase involved in pheromone biosynthesis in Lepidoptera. Base methanolysis performed on *T. processionea* extract of pheromone glands evidence the presence of methyl (Z)-11-hexadecenoate and methyl (Z,Z)-11,13-hexadecadienoate, methyl (Z)-13-hexadecen-11-ynoate is not detected as described by Fabrias et al. (1989) for *T. pityocampa*. These results clearly show the difference between the two groups of processionary moths.

From biological characteristics such as diapause and caterpillar feeding periods, the different species of studied processionary moths can be also divided into two groups (Démolin, 1987): «the winter processionary moths» which develop with a pupal diapause during summer and with larvae feeding during winter i.e.: *T. pityocampa, T. wilkinsoni* and *T. jordana* and the «summer processionary moths» whose larvae feed during spring and summer i.e.: *T. processionea, T. bonjeani, T. pinivora* and *T. libanotica. T. solitaria* cannot be considered as a winter processionary moth because of the occurrence of a diapause at the egg stage and because of a biology particularly well adapted to desert conditions and to its host plants (*Pistacia* spp.) (Halperin, 1983).

Pheromone composition is correlated with this division into two groups based on the biology. The winter processionary moth corresponds with the enyne form of pheromone molecules and the summer processionary moth with the dienic form (Fig. 1).

Pheromone composition as a chemotaxonomic tool was of interest for Thaumetopoeidae, especially for *T. pityocampa* and *T. wilkinsoni*. The two

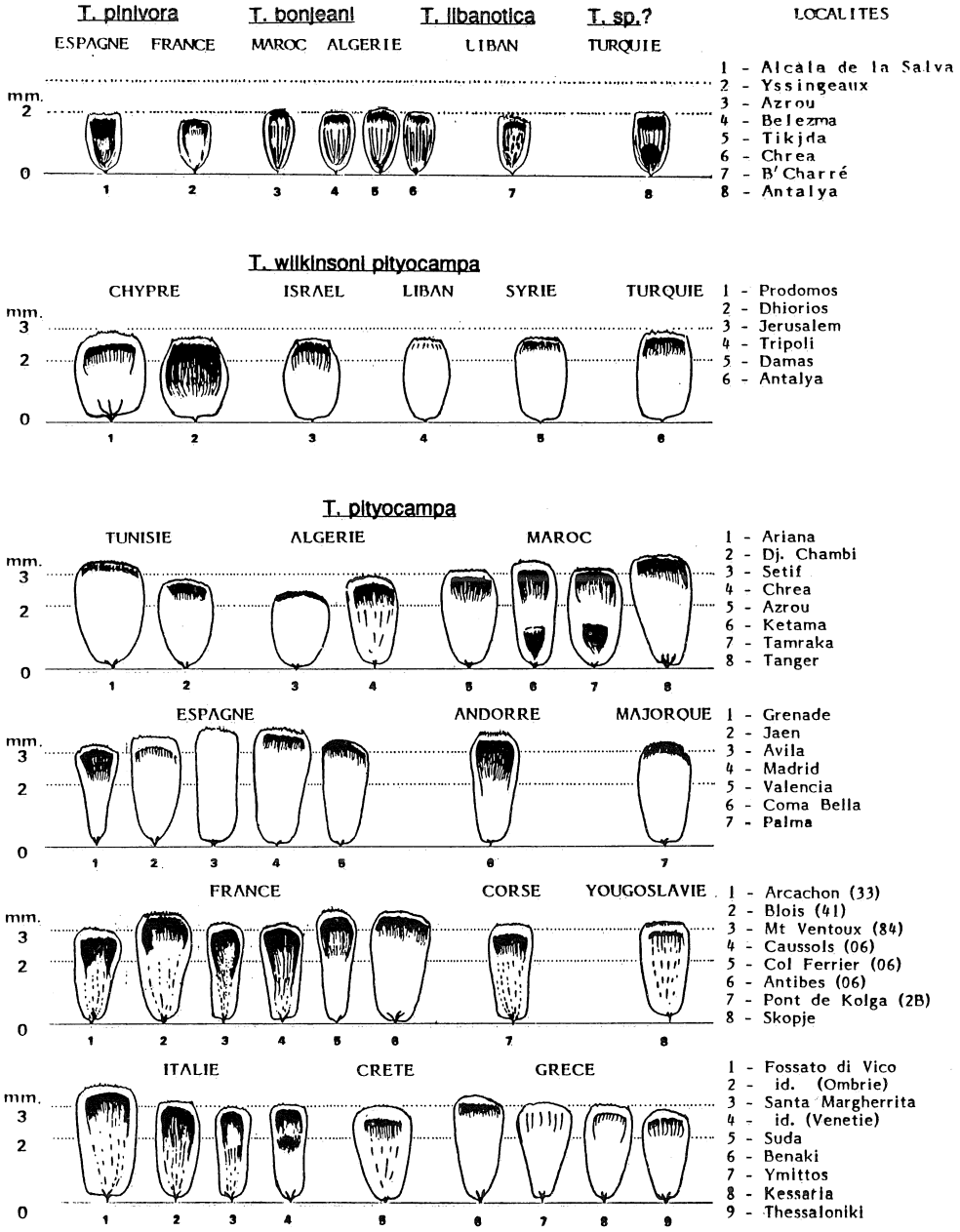

Fig. 1 - Variation of the shape of abdominal scales in 5 species of *Thaumetopoea*. The schema come from observation of about 1 hundred female samples per species and represent the average form.

species share the same biological and ecological singularities (Démolin, 1969; Huchon and Démolin, 1970; Wilkinson, 1926). The *T. wilkinsoni* species has been described from Cyprus samples. (Tams, 1924) and systematicians separated this species from *T. pityocampa* according to slight differences on male genitalia and on female abdominal scales. Valvae of *T. wilkinsoni* are more sclerified than those of *T. pityocampa* and the shape of the uncus is different but an extensive study on the variation of the female abdominal scales (Fig. 2) shows that this character should be considered cautiously.

Studies of the geographic distribution of the two species, based on the morphological differences (Démolin, 1965), reveal that *T. pityocampa* can be considered as the west winter processionary and *T. wilkinsoni* as the Middle East form. Females of both species exhibited the same mating behaviour. They

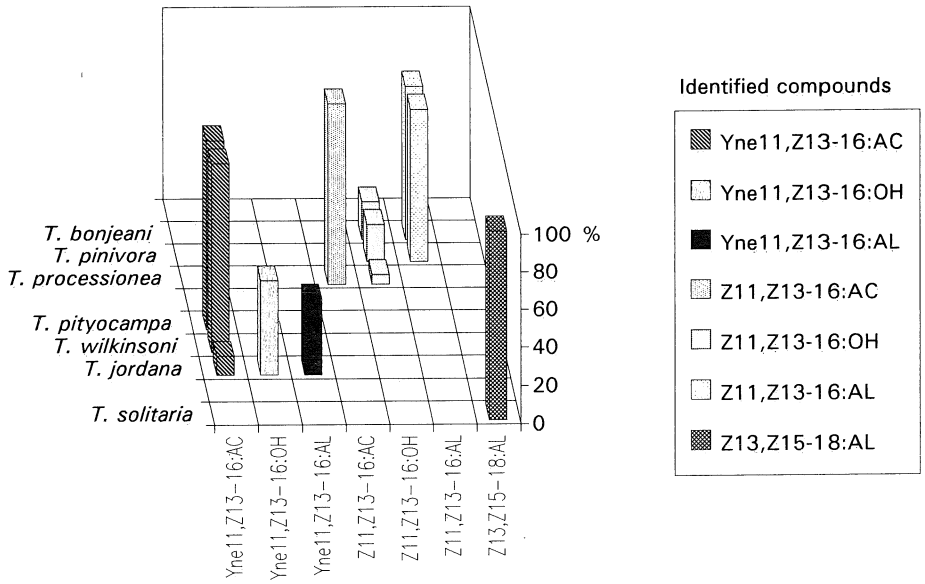

Fig. 2 - Compounds identified by GC and GCMS from gland extracts of different species within *Thaumetopoea* genus.

emerge at the sunset and start to emit the pheromone four hours after emergence. Males are attracted towards calling females and copulate without exibiting any particular courtship behaviour. In the laboratory cross-breeding can be obtained and produced larvae and adults. These two processionary moths could be thereby considered as ecotypes. Sex pheromone identifications enhanced this hypothesis owing to the fact that they both share the same active

compound as sex pheromone (Frérot, unpublished results; Halperin et al., 1984).

The same phenomenon occurred with *T. pinivora* (Montoya & Robredo, 1972) and *T. bonjeani* (El Yousfi, 1989; Démolin, 1987) which have been described as very closely-related species by systematicians and biologists. Larvae hatch when the new shoots of cedar or pine trees appear, at the beginning of spring. No common nest is spun but they have a gregarious behaviour. Pupation takes place in the soil and adults emerge in August. The diapause occurred at the first instar larvae. Morphological differences are weak and male genitalia did not differ consequently and female abdominal scales showed slight differences. For both, females produced the pheromone, only the second night following the emergence and they both produced the same pheromone blend (Frérot et al., 1990); results confirmed by field trials on *T. pinivora* (Montoya & Démolin, pers. comm.)

Thaumetopoea processionea (L.) which feed on oak trees and which could be considered either as an autochthonus species or as a more recently evolved member of the genus is the last summer processionary moth studied. Based on the pheromone identification, it did just differ slightly from the others. The aldehyde function is replaced by an acetate. It presupposed just one more step in the pheromone biosynthesis, involving a fatty alcohol acetyltransferase that catalyses the conversion of fatty alcohol to acetate ester (Jurenka & Roelofs, 1989). Nevertheless, pheromones of the summer processionary moth should have a common precursor, a fatty hexadecadienol converted further into either an aldehyde or an acetate (Teal & Tumlinson, 1987).

T. solitaria appeared to differ from the other processionary moth according to the biology and the identified pheromone with 18 carbons and 2 double bonds in 13 th and 15 th positions on the chain. This compound must be confirmed but it could promote the intervention of a delta 11 and delta 13 desaturase on palmitic fatty acid followed by an elongation of the chain as proposed for some noctuids, arctids and geometrids (Roelofs & Bjostad, 1984).

Pheromone identifications corroborated the biological differences between the species of Thaumetopoidae and also reinforced the hypothesis that *T. pityocampa* and *T. wilkinsoni* could be the same species and that *T. pinivora* and *T. bonjeani* are closely related species if not ecotypes. However pheromone of these species can already be used to monitor their populations (Démolin, unpublished results; Halperin, 1984; 1985; Tiberi & Niccoli, 1984). From a biochemical point of view, all these processionary moths form an homogeneous groups that can be divided into two sub-groups. We hope eventually to be better able to characterize the *T. solitaria* sex pheromone and to be able to undertake pheromone identification on *T. herculeana*.

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