Organisation Européenne et Méditerranéenne pour la Protection des Plantes European and Mediterranean Plant Protection Organization

# Normes OEPP EPPO Standards

Diagnostic protocols for regulated pests Protocoles de diagnostic pour les organismes réglementés

PM 7/37



Organization Européenne et Méditerranéenne pour la Protection des Plantes 1, rue Le Nôtre, 75016 Paris, France

# Approval

EPPO Standards are approved by EPPO Council. The date of approval appears in each individual standard. In the terms of Article II of the IPPC, EPPO Standards are Regional Standards for the members of EPPO.

## Review

EPPO Standards are subject to periodic review and amendment. The next review date for this EPPO Standard is decided by the EPPO Working Party on Phytosanitary Regulations

# Amendment record

Amendments will be issued as necessary, numbered and dated. The dates of amendment appear in each individual standard (as appropriate).

# Distribution

EPPO Standards are distributed by the EPPO Secretariat to all EPPO member governments. Copies are available to any interested person under particular conditions upon request to the EPPO Secretariat.

# Scope

EPPO Diagnostic Protocols for Regulated Pests are intended to be used by National Plant Protection Organizations, in their capacity as bodies responsible for the application of phytosanitary measures to detect and identify the regulated pests of the EPPO and/or European Union lists.

In 1998, EPPO started a new programme to prepare diagnostic protocols for the regulated pests of the EPPO region (including the EU). The work is conducted by the EPPO Panel on Diagnostics and other specialist Panels. The objective of the programme is to develop an internationally agreed diagnostic protocol for each regulated pest. The protocols are based on the many years of experience of EPPO experts. The first drafts are prepared by an assigned expert author(s). They are written according to a 'common format and content of a diagnostic protocol' agreed by the Panel on Diagnostics, modified as necessary to fit individual pests. As a general rule, the protocol recommends a particular means of detection or identification which is considered to have advantages (of reliability, ease of use, etc.) over other methods. Other methods may also be mentioned, giving their advantages/disadvantages. If a method not mentioned in the protocol is used, it should be justified.

The following general provisions apply to all diagnostic protocols:

- laboratory tests may involve the use of chemicals or apparatus which present a certain hazard. In all cases, local safety procedures should be strictly followed
- use of names of chemicals or equipment in these EPPO Standards implies no approval of them to the exclusion of others that may also be suitable

• laboratory procedures presented in the protocols may be adjusted to the standards of individual laboratories, provided that they are adequately validated or that proper positive and negative controls are included.

# References

- EPPO/CABI (1996) *Quarantine Pests for Europe*, 2nd edn. CAB International, Wallingford (GB).
- EU (2000) Council Directive 2000/29/EC of 8 May 2000 on protective measures against the introduction into the Community of organisms harmful to plants or plant products and against their spread within the Community. *Official Journal of the European Communities* L169, 1–112.
- FAO (1997) International Plant Protection Convention (new revised text). FAO, Rome (IT).
- IPPC (1993) *Principles of plant quarantine as related to international trade.* ISPM no. 1. IPPC Secretariat, FAO, Rome (IT).
- IPPC (2002) *Glossary of phytosanitary terms*. ISPM no. 5. IPPC Secretariat, FAO, Rome (IT).
- OEPP/EPPO (2003) EPPO Standards PM 1/2 (12): EPPO A1 and A2 lists of quarantine pests. *EPPO Standards PM1 General phytosanitary measures*, 5–17. OEPP/EPPO, Paris.

# Definitions

*Regulated pest*: a quarantine pest or regulated non-quarantine pest. *Quarantine pest*: a pest of potential economic importance to the area endangered thereby and not yet present there, or present but not widely distributed and being officially controlled.

# **Outline of requirements**

EPPO Diagnostic Protocols for Regulated Pests provide all the information necessary for a named pest to be detected and positively identified by an expert (i.e. a specialist in entomologist, mycology, virology, bacteriology, etc.). Each protocol begins with some short general information on the pest (its appearance, relationship with other organisms, host range, effects on host, geographical distribution and its identity) and then gives details on the detection, identification, comparison with similar species, requirements for a positive diagnosis, list of institutes or individuals where further information on that organism can be obtained, references (on the diagnosis, detection/extraction method, test methods).

# **Existing EPPO Standards in this series**

Nineteen EPPO standards on diagnostic protocols have already been approved and published. Each standard is numbered in the style PM 7/4 (1), meaning an EPPO Standard on Phytosanitary Measures (PM), in series no. 7 (Diagnostic Protocols), in this case standard no. 4, first version. The existing standards are:

PM 7/1 (1) Ceratocystis fagacearum. Bulletin OEPP/EPPO Bulletin **31**, 41–44

PM 7/2 (1) Tobacco ringspot nepovirus. Bulletin OEPP/EPPO Bulletin **31**, 45–51

PM 7/3 (1) Thrips palmi. Bulletin OEPP/EPPO Bulletin **31**, 53–60

PM 7/4 (1) Bursaphelenchus xylophilus. Bulletin OEPP/ EPPO Bulletin **31**, 61–69

PM 7/5 (1) Nacobbus aberrans. Bulletin OEPP/EPPO Bulletin **31**, 71–77

PM 7/6 (1) Chrysanthemum stunt pospiviroid. Bulletin OEPP/ EPPO Bulletin **32**, 245–253

PM 7/7 (1) Aleurocanthus spiniferus. Bulletin OEPP/EPPO Bulletin 32, 255–259

PM 7/8 (1) Aleurocanthus woglumi. Bulletin OEPP/EPPO Bulletin 32, 261–265

PM 7/9 (1) Cacoecimorpha pronubana. Bulletin OEPP/EPPO Bulletin **32**, 267–275

PM 7/10 (1) Cacyreus marshalli. Bulletin OEPP/EPPO Bulletin **32**, 277–279

PM 7/11 (1) Frankliniella occidentalis. Bulletin OEPP/EPPO Bulletin **32**, 281–292

PM 7/12 (1) Parasaissetia nigra. Bulletin OEPP/EPPO Bulletin **32**, 293–298

PM 7/13 (1) Trogoderma granarium. Bulletin OEPP/EPPO Bulletin **32**, 299–310

PM 7/14 (1) Ceratocystis fimbriata f. sp. platani. Bulletin OEPP/EPPO Bulletin **33**, 249–256

PM 7/15 (1) Ciborinia camelliae. Bulletin OEPP/EPPO Bulletin **33**, 257–264 PM 7/16 (1) Fusarium oxysporum f. sp. albedinis. Bulletin OEPP/EPPO Bulletin **33**, 265–270

PM 7/17 (1) Guignardia citricarpa. Bulletin OEPP/EPPO Bulletin **33**, 271–280

PM 7/18 (1) Monilinia fructicola. Bulletin OEPP/EPPO Bulletin 33, 281–288

PM 7/19 (1) Helicoverpa armigera. Bulletin OEPP/EPPO Bulletin **33**, 289–296

Several of the Standards of the present set result from a different drafting and consultation procedure. They are the output of the DIAGPRO Project of the Commission of the European Union (no. SMT 4-CT98-2252). This project involved four 'contractor' diagnostic laboratories (in England, Netherlands, Scotland, Spain) and 50 'intercomparison' laboratories in many European countries (within and outside the European Union), which were involved in ring-testing the draft protocols. The DIAGPRO project was set up in full knowledge of the parallel activity of the EPPO Working Party on Phytosanitary Regulations in drafting diagnostic protocols, and covered regulated pests which were for that reason not included in the EPPO programme. The DIAGPRO protocols have been approved by the Council of EPPO as EPPO Standards in series PM7. They will in future be subject to review by EPPO procedures, on the same terms as other members of the series.

# Diagnostic protocols for regulated pests<sup>1</sup> Protocoles de diagnostic pour les organismes réglementés

# Thaumetopoea pityocampa

#### Specific scope

This standard describes a diagnostic protocol for *Thaume-topoea pityocampa*.

# Introduction

Thaumetopoea pityocampa is a pest of Pinus, Cedrus and Larix in the Mediterranean and North Africa. It is present throughout the littoral zone and warmer regions, and can be found in the interior of the continent at sites where the climatic and site conditions are suitable. T. pityocampa's dependence on relatively high temperatures has limited its northern spread, and it appears unable to survive lower winter temperatures. The caterpillars cause severe damage to pine plantations, especially in warm districts and low altitudes. Young pine plantations are the most susceptible, and may be completely destroyed if the attack is severe enough. Less severe larval feeding damage can pave the way for harmful secondary pests and pathogens. Mature trees may suffer reductions in growth but are rarely killed outright by the pest. For more information on geographical distribution and hosts, see EPPO/ CABI (1996).

The hosts vary in their susceptibility to attack. In order of pest preference, the hosts include the genus *Pinus (Pinus nigra var. austriaca, Pinus sylvestris, Pinus nigra var. laricio, Pinus pinea, Pinus halepensis, Pinus pinaster, Pinus canariensis)*, followed by *Cedrus atlantica* and finally *Larix decidua*.

#### Identity

Name: *Thaumetopoea pityocampa* (Denis & Schiffermüller, 1775)

**Synonyms:** *Traumatocampa pityocampa* (Denis & Schiffermüller, 1775) in de Freina & Witt (1987); *Cnethocampa pityocampa* Schiff.

**Taxonomic position:** Insecta: Lepidoptera: *Notodontidae: Thaumetopoeinae* 

**Specific approval and amendment** Approved in 2003-09.

Bayer computer code: THAUPI Phytosanitary categorization: EU Annex designation II/B

#### Detection

The branches of appropriate hosts may be completely or partially defoliated by the feeding larvae (Web Fig. 11), causing localized or more general dieback of the infested tree. The characteristic silken communal nests (Web Fig. 10) can be seen from a distance on branches with some lateral shoots. On younger trees, these nests are typically found in the upper part of the crown. The nests are 12–25 cm long, usually oblong in shape and white to light grey in colour.

Pupal cocoons (Web Fig. 9) can be found between February and July in the upper layers of the soil or in leaf litter. They are often found in the growing medium in which young plants are grown. Adults usually emerge between June and August but pupae can remain in diapause for 1–3 years.

Adult moths remain hidden in trees during the day and fly by night. Both sexes may be successfully caught using UV light traps. Male specimens can be captured using pheromone traps containing the female sex pheromone.

#### Identification

*Eggs*: eggs are spherical and white; laid in groups cylindrically around pine needles (Web Fig. 7), usually on the lower branches of the crown. The egg mass is 25-40 mm wide and about 5 mm high and can contain 70–300 eggs. It is covered with scales produced by the female from the tip of her abdomen and is of a grey-brown colour similar to branches so that it is well camouflaged.

*Larva*: the caterpillar has 5 larval stages. The body of the first instar caterpillar is a dull-green. After the second moult, the caterpillar assumes its definitive appearance. The full-grown caterpillar is 38–45 mm in length. The larva is covered in

<sup>&</sup>lt;sup>1</sup>The Figures in this Standard marked 'Web Fig.' are published on the EPPO website www.eppo.org.

urticating hairs that can cause serious skin irritations, conjunctivitis, respiratory congestion and asthma in man and animals. These urticating hairs are in tufts which appear to be arranged in pairs on each body segment. The coloration of the integument and hairs which clothe the body vary considerably depending on the race of moth. In general, the integument is darker in colder areas and varies from dull bluish-grey to black. The head capsule is black. The lateral and ventral hairs vary from white (mostly) to dark yellow. The dorsal hairs range from yellow to dull orange and are borne on red-brown verrucae. The lower part of the body is light brown to brown (Web Fig. 8).

*Pupa*: the pupa is in an oval silk cocoon of a brown-white colour. The obtect pupae are about 20 mm in length, oval, and of a pale brownish-yellow colour that later changes to dark reddish-brown. The cremaster is bluntly rounded, with two robust, curved spines.

*Adult*: the female moth has a wingspan of 35–50 mm, the male is smaller with a wing-span of 30–40 mm. They both have a very pronounced jagged crest on the front. The forewings are dull ashen-grey; the veins, margins and three transverse bands are darker. The pattern on the female wing is less pronounced. The cross lines in this species are not sharply jagged but rather obtuse. The hindwings are white, grey fringed, with a characteristic grey-brown spot in the anal region. The antennae appear filiform in females and pectinate in males (see Web Fig. 3), but they are bipectinate in both sexes. They are yellowish at the base and rather brown at the tip. Both sexes have a hairy thorax. The abdomen of the female is stout and its last segments are covered with a tuft of large scales; the abdomen of the male is brushy and sharp.

Rearing of eggs, caterpillars or pupae through to the adult stages is recommended for positive identification.

For illustrations, see Plate 7 in Reugeot & Viette (1978) and de Freina & Witt (1987).

#### Diagnosis of the genus Thaumetopoea

After Kiriakoff (1970), antennae of both sexes are bipectinate (although appearing filiform in females) to apex. Naked eyes. Chaetosemata absent. Frons sometimes with a jagged crest (the canthus). Proboscis vestigial. Palps short, porrect. Hind tibiae with a single pair of spurs. Females with woolly anal pilosity. Wings rather short and broad. Forewings, the margin more or less straight, apex forming a right angle, termen evenly convex, tornus not distinct, dorsum slightly convex, a bit longer than half of the margin of wing. Forewings have vein 2 arising from four-fifth of the cell, 3 and 4 separated, 5 arising from the middle of the discocellulars which are concave, areole absent, 6, 7, 8 + 9 and 10 stalked. Hindwings have the margin slightly convex, apex and tornus rounded. Hindwing venation: vein 2 arising from three-quarters of the cell, 3 and 4 generally separated, but sometimes stalked, 5 arising from the middle of the discocellulars, 6 and 7 with long stalks (sometimes on three-quarters of the length), 8 closest to the cell at threequarters of the cell. Male genitalia: short uncus, generally rounded, broad gnathos, bean-shaped or even semicircular. Broad tegumen, but constricting towards the apex. Valves more or less foliaceous, typically short process at the base of the costa, sometimes absent; sometimes a hooked costal or apical process. Aedeagus approximately as long as valve, slender, sickle-shaped in the distal part, lower fultura shield-shaped, proximal margin stretched to the angles. Saccus variable, typically rather long.

Female genitalia: Sterigma usually narrow, with distal angles often stretched and with distal margin concave, abdominal tergite IX narrow. Ductus bursae not sclerotized. Signum present.

For identification of larvae to family, see Patočka (1980).

# Diagnosis of species of the genus Thaumetopoea

See Table 1 (Agenjo, 1941). For identification of species, see also Web Figs 1–5.

#### Possible confusion with similar species

The related species *T. pinivora* has a more northerly range than *T. pityocampa*. The two species are very similar morphologically but differ as follows (see above and Web Figs 1–5):

- T. pityocampa cross lines on forewings are poorly or obtusely jagged, the postmedian line in the costal field is less divergent and reaches the inner margin of the wing at the second third. The fringes (scales on the edge of the wings) of the hindwings are white and brown-black only in the anal field. The thorax of the female is covered with light-grey scales.
- *T. pinivora* the cross lines on forewings are clearly and sharply jagged, the postmedian line in the costal field is very much divergent and reaches the inner margin of the wing on the third quarter. The fringes of the hindwings are grey. The thorax of the female is covered with dark-grey scales.

These two species can easily be differentiated because of the presence in both species of a jagged chitinous crest called the 'canthus', which allows the emerging adults to dig towards the soil surface; the posterior indentations of the canthus are higher in T. pityocampa than in T. pinivora (see Web Fig. 1 a-b). A study of female anal scales also helps distinguish between closely related species. The anal scales are arranged in a tuft at the tip of the female abdomen (see Web Fig. 2). They are deposited on the egg masses at oviposition. A study of male genitalia may show differences which are difficult to observe. The two species can be differentiated by studying the forms of the 'fulcrum', which is a jagged chitinous structure at the base of the aedeagus (Web Fig. 4b). The fulcrum allows the separation of T. pityocampa from T. pinivora, and of those from two other species T. processionea and T. herculeana (Gomez Bustillo, 1978a, 1978b). There is also a difference in shape of the valve (Web Fig. 4a).

Behavioral differences also allow differentiation between the two species. Both species are gregarious, but caterpillars of *T. pityocampa* aggregate in colonies and spin silken nests for shelter, which they enlarge as they develop and in which they

#### Table 1 Key to adult Thaumetopoea spp.

1	Canthus convex and smooth	2
	Canthus with protuberance	3
2	Hindwings without transverse band	T. solitaria
	Hindwings with transverse band	T. processionea
3	Underside of forewings grey	4
	Underside of forewings chestnut-brown or whitish	7
4	Males with the transverse lines on the underside of the forewings	
	black without yellow scales: females with anal scales longer than 2 mm	
	or with the proximal part not pointed	5
	Males with the transverse lines on the underside of the forewings	
	black without yellow scales: females with anal scales not longer than	
	2 mm or with the proximal part not pointed	6
5	First segment of the discoidal vein (bent-elbowed line) on the underside	
	of the forewing forming an angle of 35° with the costal margin	T. pityocampa
	First segment of the the discoidal vein (bent-elbowed line) on the	
	underside of the forewing forming an angle of 50° with the costal margin	T. wilkinsoni
6	Extrabasal lines on the underside of the forewings are closer to the inner	
	margin, than to the costal margin, where they converge together	T. pinivora
	Extrabasal and bent lines on the underside of forewings are equidistant	
	from the inner and costal margins	T. bonjeani
7	Underside of the forewing with discoidal area	T. herculeana
	Underside of the forewings without discoidal area	T. jordana

overwinter, whereas *T. pinivora* caterpillars aggregate freely near the pine needles where they were born. *T. pinivora* overwinters as eggs attached to the pine needle and protected by female anal scales. *T. pityocampa* feeds on *Pinus*, *Cedrus*, *Abies*, *Castanea*, *Larix*, *Quercus* and *Malus*, whereas *T. pinivora* feeds only on *Pinus* spp.

# Requirements for a positive diagnosis

The procedures for detection and identification described in this protocol should have been followed. The specimens should have the diagnostic characteristics of the genus *Thaumetopoea* as described in this protocol.

# Report on the diagnosis

A report on the execution on the protocol should include:

- information and documentation on the origin of the infested material
- · how many specimens were available for examination
- drawings or photographs of the morphological features of the genus and species required for a positive diagnosis
- comments as appropriate on the certainty or uncertainty of the identification.

Preserved specimens of the pest should be retained.

## **Further information**

Further information on this organism can be obtained from: S. Gomboc, Biotechnical Faculty, Agronomy Department, Institute of Phytomedicine, Jamnikarjeva 101, 1000 Ljubljana, Slovenia; H. Evans, Forest Research, Alice Holt Lodge, Wrecclesham, Farnham GU10 4LH, UK; J-C. Martin. INRA Unité Expérimentale Forestière Méditerranéenne – Avenue A. Vivaldi F-84 000 Avignon.

#### Acknowledgements

This protocol was originally drafted by: S. Gomboc, Institute for Phytomedicine, Ljubljana (SI); J-F. Germain, LNPV, Montpellier (FR).

#### References

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Fig. 1 a Canthus shape in different species in the genus *Thaumetopoea* (from De Freina and Witt, 1987)

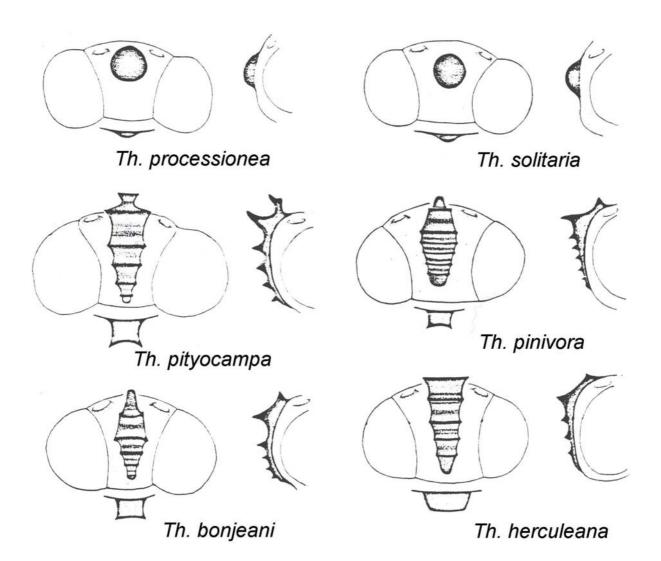


Fig. 1 b Canthus in *Thaumetopoea pityocampa* and *T. pinivora* (photos J.F. Germain, LNPV-Unité d'Entomologie)



T. pinivora



T. pityocampa

Fig. 2 Variation in shape of female anal scales for four *Thaumetopoea* species on conifers: *T. bonjeani*, *T. pinivora*, *T. wilkinsoni*, *T. pityocampa* (from Demolin *et al.*, 1994)

The scales shown were chosen from amongst the largest and are typically representative of the average shape of the regions.

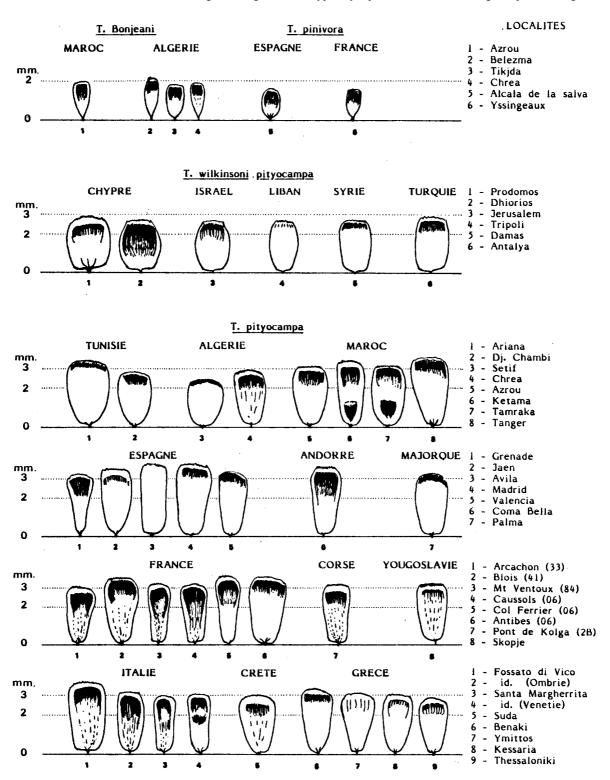
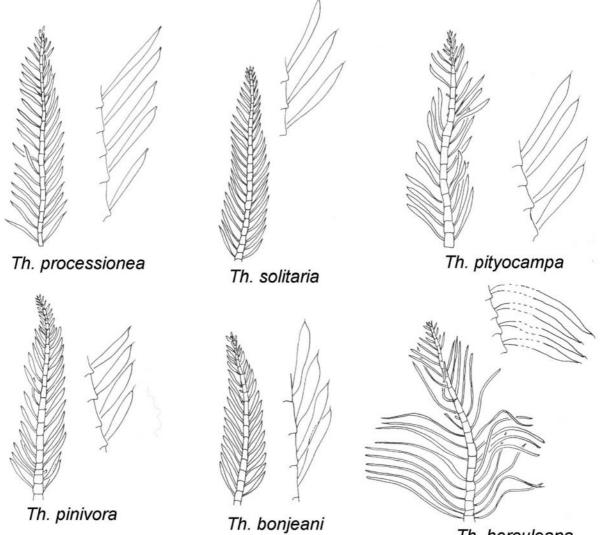
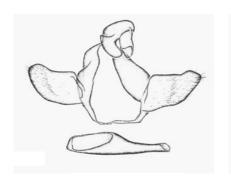


Fig. 3 Male antennae pectination in West Palearctic Thaumetopoea species (from De Freina and Witt, 1987).

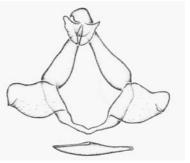


Th. herculeana

Fig. 4 a Male genitalia in three closely related *Thaumetopoea* species (from De Freina and Witt, 1987).



Th. pityocampa

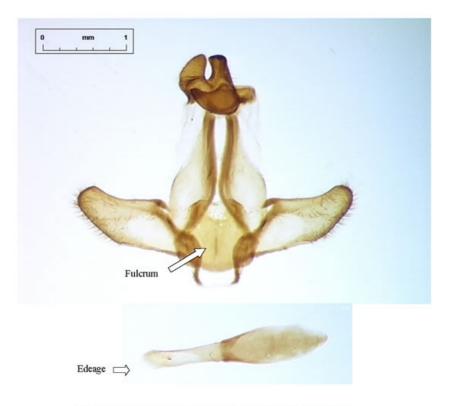


Th. pinivora

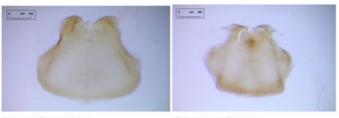


Th. bonjeani

Fig. 4 b Male genitalia.



Génitalia male Thaumetopoea pityocampa (collection INRA/Chambon)



Fulcrum T. pityocampa Fulcrum T. Pinivora (photos J.F. Germain, LNPV-Unité d'entomologie)

Fig. 5 Habitus Thaumetopoea pityocampa, male and female, originating from Krk island in Croatia (Photo Gomboc S.).



Fig. 6 Habitus Thaumetopoea pityocampa and T. pinivora (photo J-F Germain)



Habitus *Thaumetopoea pityocampa* (Denis & Schiffermüller)

Habitus *Thaumetopoea pinivora* (Treitschke)

Fig 7 Egg mass of *Thaumetopoea pityocampa* (photo B. Hrasovec)



Fig. 8 Caterpillar of *Thaumetopoea pityocampa* (photo B. Hrasovec)



Fig. 9 Pupa of Thaumetopoea pityocampa (photo B. Hrasovec)



Fig 10 Owerwintering nest of Thaumetopoea pityocampa on Krk Island in Croatia (Photo Gomboc S.)



Fig. 11 Heavily damaged young tree of *Pinus nigra* from larvae of *Thaumetopoea pityocampa* on Krk Island in Croatia (Photo Gomboc S.).

