Chapter 8
Medical and Veterinary Impact of the Urticating Processionary Larvae

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1 Introduction

This chapter presents recent findings on the impact of processionary moths on human and animal health. The data obtained demonstrate that setae can be dispersed kilometers away from their origin, a fact that now explains why some sensitized subjects experience symptoms without a direct contact with larvae.

The use of molecular biology has made possible the study of some allergens present in the setae and Tha p 2, a major setae allergen from *T. pityocampa*, probably corresponds to thaumetopoein described many years ago. Therefore, setae must be considered as a source of allergens and not only as producers of irritant or toxic reactions.

The sensitizing capacity of moth allergens is clearly demonstrated with the help of epidemiological studies. Frequent contact seems to be the most relevant factor for sensitization and occupationally exposed workers should be carefully checked for sensitization in order to avoid further exposure to the allergens.

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Processionary moths constitute also a health hazard for animals. This chapter describes the results of epidemiological studies carried out to measure the relevance on different animal species, the main symptoms after contact and the degree of knowledge among veterinarians about this risk.

The relative risk of sensitization in France has been calculated and a program allows now to estimate the zones with highest danger. In this way medical and veterinary professionals as well as the whole population could be made aware about the danger of these moths and the preventive measures necessary to avoid contact with the setae.

Taking all these facts in consideration, the chapter updates our knowledge about the impact on health of *T. pityocampa*. Most of the findings will be useful for the remaining processionary moths, but the most relevant fact is that climate change will affect other territories where this problem was not previously known. It is therefore necessary that animal and human health professionals working in newly colonized areas are instructed about this emerging health hazard.

2 Comparative Structure of the Urticating Apparatus in Processionary Moths

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2.1 Introduction

Urticating setae are common in Lepidoptera, both larva and adult, and are generally considered to act as a defense against vertebrate predators (Gilmer 1925; Pesce and Delgado 1971; Kawamoto and Kumada 1984). Incidentally, these setae also pose a serious threat to human health when they get in contact with the skin or other parts of the body (Specht et al. 2008; Hossler 2009; Mullen 2009; Battisti et al. 2011).

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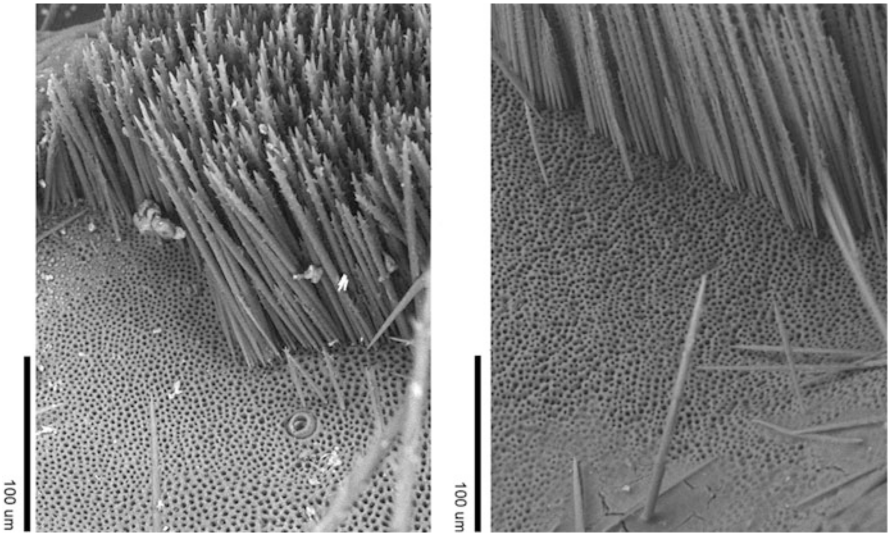

Fig. 8.1 Scanning electron photographs of the urticating setae in Thaumetopoea pinivora (*left*) and pityocampa (*right*). The setae are packed on special integument areas on the upper part of the abdomen. The holes indicate the sockets were their sharp proximal ends are implanted, while barbs indicate the distal ends of the setae. The penetration in the skin of the target organisms is made by the sharp proximal end, helped by the barbs of the distal end

Their nature is very different from other defensive hairs occurring in Lepidoptera, which are part of the integument and require contact with the larva to cause the reaction (e.g. the larvae of Saturniidae, Megalopygidae and Limacodidae), while they are similar to urticating setae released by some spiders from America (Theraphosidae) (Battisti et al. 2011). Setae are readily dehiscent from the integument and can disperse in the environment, often far away from the infested trees (Werno and Lamy 1990; Maier et al. 2003).

All studied species of the Thaumetopoeinae subfamily of Notodontidae are known to carry urticating setae, either as larva (genus *Thaumetopoea*) or adult (e.g. genus *Anaphe* from Africa and *Ochrogaster* from Australia) (Lamy et al. 1984; Floater 1998). The urticating setae of the larvae of a few *Thaumetopoea* species, namely the pine processionary moth *Thaumetopoea pityocampa* (Denis and Schiffermüller 1775), the northern pine processionary moth *Thaumetopoea pinivora* (Treitschke 1834), and the oak processionary moth *Thaumetopoea processionea* (Linnaeus 1758) (Lepidoptera, Notodontidae), have been well described (Fig. 8.1) and are reviewed in this chapter.

Setal production and morphology was first studied in the pioneering work of Fabre (1899), continued by Démolin (1963), Lamy et al. (1982) and Novak et al. (1987). The urticating setae are produced from the third larval instar or fourth larval instar, depending on the species, on integument areas of the abdominal tergites that are called "mirrors," owing to their property to reflect light (Figs. 8.2, and 8.3).

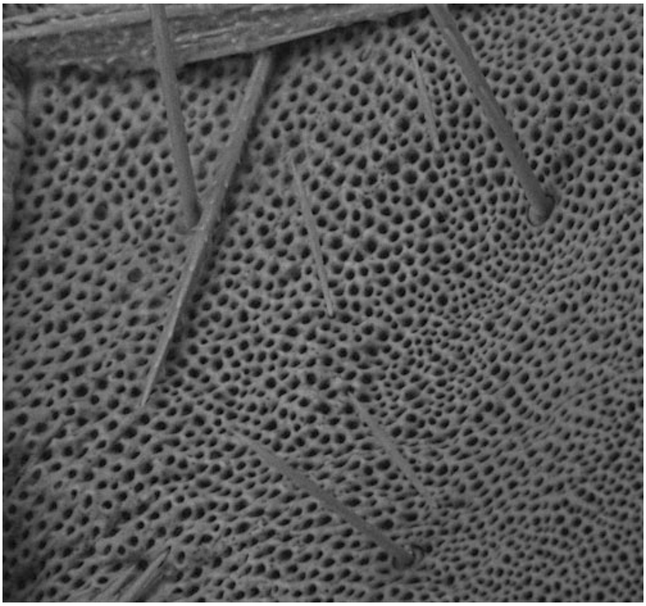

Fig. 8.2 Scanning electron photographs of the seta field of *Thaumetopoea pityocampa* with detached urticating setae laying on the floor and true hairs firmly implanted in the integument. The latter are identifiable by a collar at the base of the hair and are distributed all over the integumental field carrying the setae, having a role in the detachment of the urticating setae from the integument (see also Fig. 8.5)

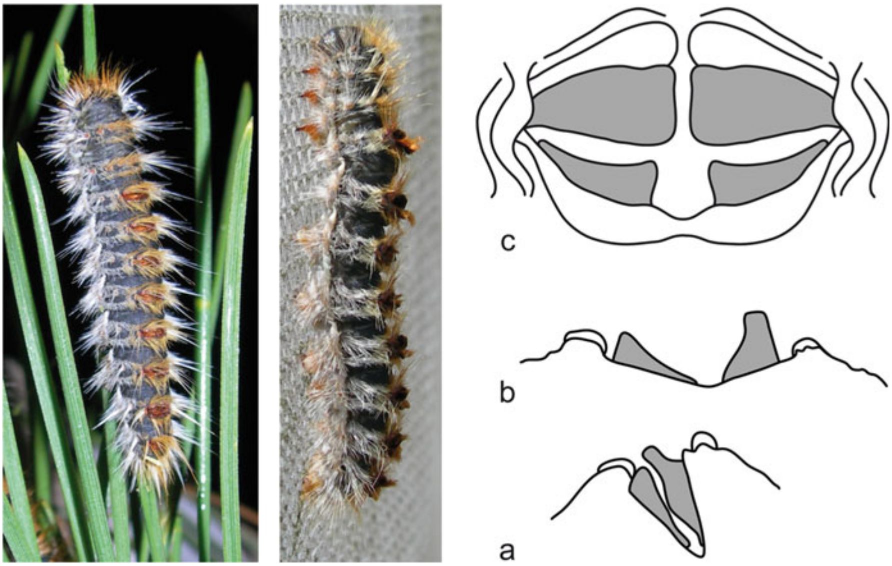

Fig. 8.3 Schematic drawing of the seta field of *Thaumetopoea pityocampa* when closed (*a*) and open (*b*) seen laterally, and open but viewed from *above* (*c*). On the left a fifth instar larva with the fields closed, with the longer setae protruding out of the fold like in (*a*). The setae are *bright orange* and are surrounded by *orange* and *white* hairs

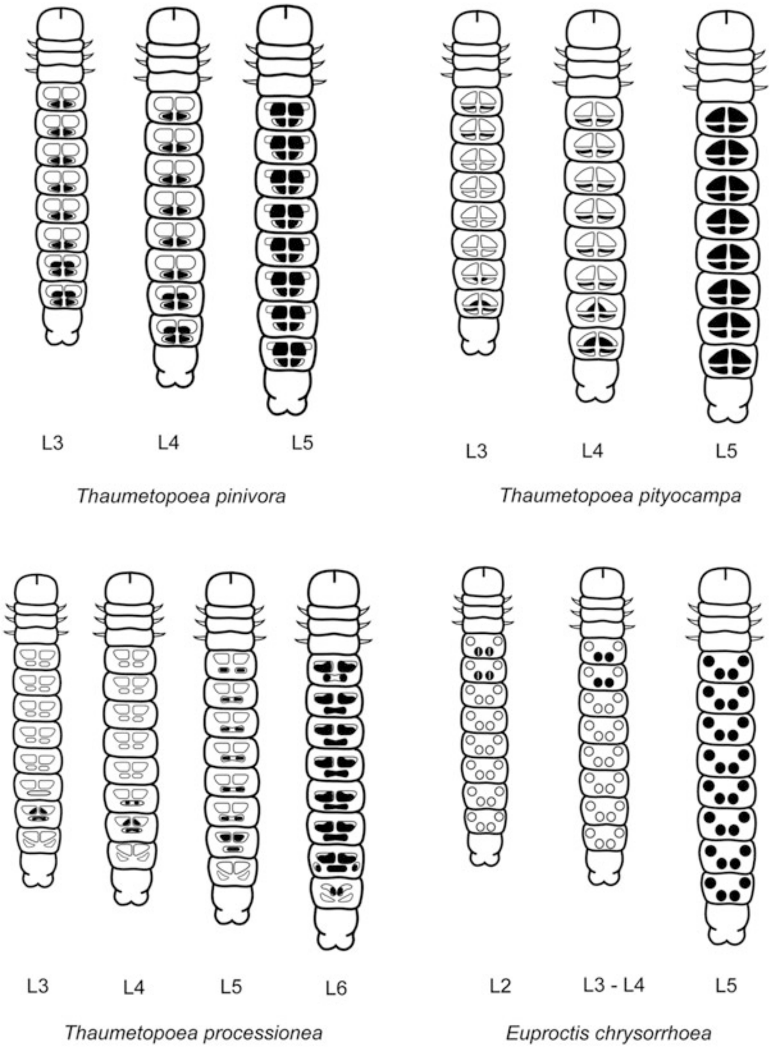

Fig. 8.4 Schematic drawing of the distribution of the seta fields in larvae of different instars of three *Thaumetopoea* species, compared with one lymantriid species

These mirrors increase in number with larval molts until the last larval instar, in which they occur on 8 abdominal segments (Fig. 8.4).

At this stage there are approximately 1,000,000 setae/larva in *T. pityocampa*, with a density of 60,000 setae/mm^2; setae are a few hundred micrometer long, with a sharp proximal end and pointed barbs directed distally (Lamy 1990). These setae are renewed at each molt and many are left with the old skin, so that they occur in a high number in tents used by larval colonies on trees as well as at pupation sites.

The release of setae by the larvae was explored by Démolin (1963), who showed that the larvae may actively open the integument mirrors when disturbed (Fig. 8.5).

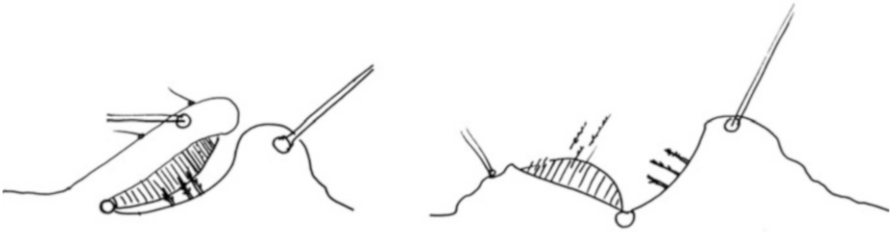

Fig. 8.5 Scheme of the opening of the setae field and release of the urticating setae by the action of normal hairs implanted in the field (*Thaumetopoea bonjeani*, modified from Démolin (1988))

Werno and Lamy (1990) found setae in pollen traps, with the abundance of setae progressively decreased moving away from the infested forest. Fenk et al. (2007) develop a setal dispersion model for the oak processionary moth, *T. processionea*, based on their physical properties in relation to prevailing wind. This is important because most reactions in humans are associated with airborne setae (Maier et al. 2003; Fuentes et al. 2006; Gottschling and Meyer 2006; Vega et al. 2011a, b). An aggravating factor is the long persistence of the setae in the environment even after exposure to rather extreme conditions: Hase (1939) demonstrates that dried insects retain their irritating power after 12 years of preservation and after treatment with temperature as high as 110 °C.

2.2 Comparative Analysis of the Urticating Apparatus of Thaumetopoea

To obtain samples of urticating setae of the 3 *Thaumetopoea* species, we collected larvae in the last instar from rearing in Padova University campus or from colonies in natural conditions. Samples were collected in 2008 from Tregnago (Italy) (45°30′N, 11°11′E, m 477) for *T. pityocampa*, from the island of Gotland (Sweden) (56°56′N, 18°16′E, m 7) in 2009 for *T. pinivora*, and from Caprino Veronese (Italy) (45°35′N, 10°47′E, m 245) in 2009 for *T. processionea*. To explore the length and diameter of setae, we randomly selected 10 individuals, each from a different colony, which were directly frozen after collection. Setae were randomly extracted with forceps from mirrors under stereomicroscope and were measured using a scanning electron microscope (SEM Hitachi TM-1000, Tokyo Japan) ($n = 108$ for *T. pityocampa*, $n = 120$ for *T. pinivora* and $n = 175$ for *T. processionea*). To setae were studied through their aerodynamic diameter, which is the hypothetic diameter that a water droplet would have in order to settle with the same velocity as the particle under consideration (Petrucco Toffolo et al. 2014). Last instar larvae of the 3 species were also used to observe the distribution of the setal length classes in the urticating apparatus. Larvae from the freezer were kept on ice under a stereomicroscope and dissected transversally to the fields carrying the setae. The specimens were observed using an SEM (Hitachi TM-1000) equipped with a cool stage unit (MK3 model, Deben United Kingdom), which maintained the specimens at temperature below zero to avoid water evaporation.

In *T. pityocampa*, the distribution of seta length showed a bimodal distribution (Fig. 8.6). The hypothetical horizontal distance traveled for a seta released at 20 m height in a day with a wind velocity of 2 m/s is 6.5 km for the short setae and 2.4 km for the long setae. The distribution of the length of *T. pinivora* is also bimodal (Fig. 8.6), and the corresponding dispersion distances are 21 and 7.4 km. In *T. processionea*, the distribution of length is unimodal, resulting in a dispersion of 8 km for a release at 20 m of height and a wind velocity of 2 m/s. It must be mentioned that the velocities and distances given above are for the mean aerodynamic diameter. Because the velocity is inversely proportional to the square aerodynamic diameter, the smaller setae will spread much further. Setal diameter and length were highly correlated in the three species according to a logarithmic function, with a higher slope for short setae. The picture of the setae in the mirrors showed differences among the three species. In *T. pityocampa* and *T. pinivora*, there were two well-defined levels of setae length that cohabit on the whole surface of the mirror, whereas this was not the case for *T. processionea* (Fig. 8.1).

2.3 General Considerations and Future Studies

Although the setal system of processionary moths has been the subject of a large number of studies, for both morphological and functional traits (Fabre 1899; Scheidter 1934; Démolin 1963; Lamy et al. 1982), the present analysis shows a markedly different pattern of setae length from that expected, with important implications for the dynamical properties. In particular, short setae like the ones that occur in two of the three species are able to spread farther away than long setae. The results showed a wide variation in setal length. In the case of *T. pityocampa*, the longest (680 μm) were approximately 14 times longer than the shortest (50 μm), whereas in *T. pinivora* (47–492 μm) and in *T. processionea* (56–351 μm) the same ratios were equal to 10 and 6 times, respectively. Previously published measures of the range were much lower for: *T. pityocampa*, 80–160 μm (Scheidter 1934), 93–415 μm (Hase 1939) and 150–250 μm (Lamy 1990); *T. pinivora*, 60–260 μm (Démolin 1963); and *T. processionea*, 150–250 μm (Lamy 1990) and 85–290 μm (Fenk et al. 2007). These differences may be ascribed to differences in setal extraction methods and sample sizes.

In *T. pityocampa* and *T. pinivora* the distribution of setal length can be considered bimodal, with a first peak in the class of 50–100 μm, and a second in the class of 200–250 μm, while it is unimodal in the oak processionary moth *T. processionea* (Fenk et al. 2007). Bimodality in *T. pityocampa* and *T. pinivora* can be explained by the overlapping of 2 normal distributions for each of the length classes. In fact, the short and long setae are intermixed throughout the mirror (Fig. 8.1). There are a few published examples of variation in setal size, for example, in the adult female of *Anaphe venata* Butler 1878 (Notodontidae) (Lamy et al. 1984), but no

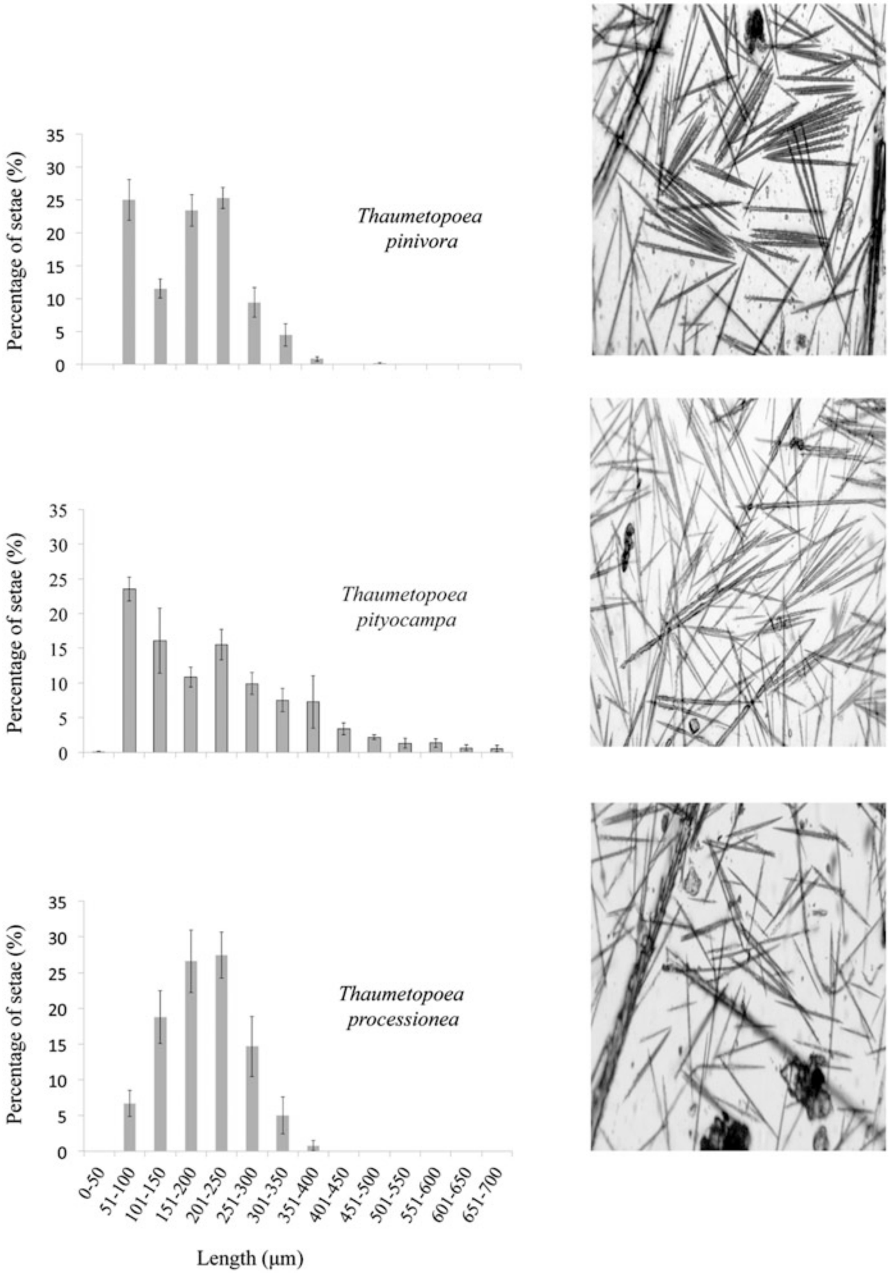

Fig. 8.6 Distribution of the length of setae in three species of *Thaumetopoea*, with photographs of the setae when they are removed from the integument with a forceps. Fragments of normal hairs are also visible

interpretation of the importance of setal size is given. As in the studied species of *Thaumetopoea*, the general shape of the seta is the same; the different size, however, may affect both dispersion and skin penetration. In other arthropods, however, such as the brown tail moth *Euproctis chrysorrhoea* (Linnaeus 1758) (Kemper 1955) and South American tarantula spiders (Cooke et al. 1972), the setae display a high variation of shape that could be potentially linked to a different function (Battisti et al. 2011). The absence of bimodality in *T. processionea*, despite a large interval of variation, remains unexplained and it is possibly related to a different evolutionary history of this species, the only species in the genus feeding on oaks (*Quercus* spp.) over all of Europe (Groenen and Meurisse 2012).

The role of seta size in protection from predation can be discussed in relation to what is known about the defense mechanism. Battisti et al. (2011) point out that the urticating setae provide an efficient defense system for the colony but not for the individual, as the symptoms appear with a delay of time, when the larva has already been killed. Although these conclusions have been made based on the reaction development in humans, it is unlikely that birds or mammalian predators would respond differently. As setae disperse as a cloud around the colony (Fenk et al. 2007), with high concentration of short setae up to 6 km (during the day) and 12 km (during the night), their function could be to keep away predators (Fig. 8.7). In this case, the larger and denser is the cloud, the stronger is the protection; the diversity of seta size may extend such a barrier much farther, with a direct benefit for the colony. Therefore, one could speculate that other prey of vertebrate predators could indirectly benefit from the protection, and, thus, competition among insect herbivores should increase. Although the mechanism needs to be elucidated with appropriate experiments, the large investment in urticating setae made by these species of processionary moths indicates that the benefits from extended protection are higher than the costs possibly imposed by competition.

In conclusion, the awareness of a great variation in size of urticating setae may open the way for better modeling of both the ecology of the systems and the risks to which humans and domestic mammalians are exposed. In the case of processionary moths, such a risk is very high due to the wide distribution of host plants in both urban and forest areas (EFSA 2009). This is emphasized by a continuous release of setae from the soil or from tents, because of their high persistence. The information provided in the present paper may offer an opportunity to explore the importance of setal size for protection from predation and for risk assessment toward non-target vertebrates and humans. In addition, it may be useful to pest managers and decision-makers in planning the control operations of these forest and urban tree pests.

3 Human Immune Responses to *Thaumetopoea pityocampa*

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Fig. 8.7 Relationships between insectivorous birds and larvae and pupae of the pine processionary moth *Thaumetopoea pityocampa*. A winter tent with a hole made likely by tits (Parus spp.) to extract the larvae, which are eaten by cutting off the head and taking the inside, thus leaving the skin with the urticating setae. A pupa inside the cocoon where the last larval skin carrying the urticating setae is present. When the hoopoe (Upupa epops) takes the cocoon (photo Z. Mendel), they open it and take only the pupa. For more information about bird predation of processionary moths (see Auger-Rozenberg et al. 2014, Chap. 7 in this volume)

3.1 Introduction

Stinging insects are known to cause allergic reactions, because they inject allergens that are recognised by the immune system, leading to a specific immune response with the synthesis of specific IgE against the injected allergens. After a new contact, these allergens cause the bridging of specific IgE located on the surface of basophils and mast cells. This induces the release of inflammatory mediators and initiates an allergic response. However, non-stinging insects, such as Lepidoptera, raise still today doubts about their mechanism of sensitization and many authors think that the lesions seen after contact with larvae are due to non-immune toxic mechanisms.

The pine processionary larva's ability to induce cutaneous lesions is well known since the first descriptions of Reamur in 1736 and Fabre in 1899 (described by Ducombs et al. 1981*). Thaumetopoea pityocampa* larvae induce skin lesions such as urticaria or dermatitis, conjunctivitis and rarely respiratory symptoms or even anaphylactic shock. These responses are caused by urticating hairs called setae disposed on cuticular plates and dispersed around the larvae as a defence mechanism. It is quite clear that the mechanical lesion induced by the penetration of the setae in the skin can be responsible for the pruritus that is always present, but setae store toxins and probably enzymes causing an additional injury that contributes to an increase of the inflammation observed in individuals who were in contact with larvae. Therefore, the dual additive mechanical-toxic effect was considered as a sophisticated defence mechanism (Ducombs et al. 1979).

3.2 Pine Processionary Moth Allergens

The first article that describes the antigens and proteins of the *T. pityocampa* setae was published in 1983 (Lamy et al. 1983). They described the protein content of the setae as a complex mixture of 16 proteins. Among them, they studied a 28 kDa protein, exclusive to the setae as it was present neither in the haemolymph nor in the cuticle. This protein was not associated to carbohydrates or lipids. It could be further separated in two subunits of 13 and 15 kDa. They called it thaumetopoein and proved its urticating properties in guinea pigs. Thaumetopoein induced mast cell degranulation by a non-immune mechanism. (Lamy et al. 1985)

Several years later, the same scientific group described a homologue of thaumetopoein in the setae of the oak processionary larva (Lamy et al. 1988). This protein exhibited the same urticating effect as thaumetopoein in the guinea pig's skin.

An IgE mediated mechanism of sensitization to allergens present in *T. pityocampa* setae was demonstrated by ELISA and western blotting in 1993 (Werno et al. 1993). Two proteins bound IgE from the sera of exposed workers and one of them, a 28 kD allergen, was identified as thaumetopoein. Additionally a 45 kDa protein gave also a strong reaction with IgE. The authors, therefore, describe the presence of specific immune responses to allergens present in setae as potential relevant contributors to the lesions induced by these larvae, as well as the IgE-binding properties of thaumetopoein, previously described as a specific mast cell activator.

Some years later, our group described the presence of specific IgE antibodies in the serum from a patient suffering severe reactions after contact with *T. pityocampa* larvae. The allergen used for diagnosis was a whole body larvae's extract and this positive result suggested that the IgE-mediated mechanism found could be much more relevant than previously suspected.

3.3 Specific Antibodies to Crude Larval Extracts

A pilot study was designed by our group several years ago in order to evaluate the allergenicity of the crude whole larval extract and to characterize the principal allergens. Figure 8.8 shows how IgE from 15 patients with symptoms related to *T. pityocampa* exposure was able to bind proteins of a crude whole larval extract. The molecular weight of these proteins ranges from less than 6 kDa to more than 100 kDa, being the one around 15 kDa the most frequently recognised.

This protein could be the major allergen Tha p 1, described for the first time in 2003 (Moneo et al. 2003). In this paper, more than ten different proteins of the extract were able to bind patients' IgE, being the most frequently detected a protein of around 15 kDa. This protein was purified by ethanol fractionation by differential precipitation of a whole larval extract followed by separation by a reversed-phase high performance liquid chromatography (RP-HPLC). The amino terminal sequence GETYSDKYDTIDVNEVLQ for Tha p 1 was obtained, but, at that time, no similarities with other proteins were found using the web interface BLAST of the USA National Centre for Biotechnology Information (NCBI). Several years later, the complete sequencing of the silkworm *Bombyx mori* genome led to classify Tha p 1 as a chemosensory protein, similar to those found in this species (Picimbon et al. 2000).

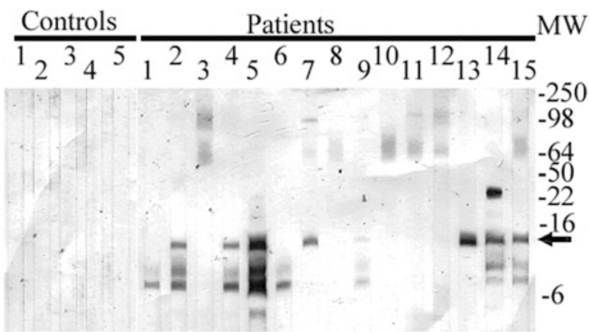

Fig. 8.8 Immunoblot of a crude whole body *Thaumetopoea pityocampa* extract revealed with sera from 5 healthy donors used as controls and 15 patients with allergic symptoms related to larva exposure. Estimated molecular weights are indicated. Tha p 1 is outlined by an *arrow*

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                                            cggggacagccagacg
aaggatcgtgcagttatcacgcggcgattgttaaaatgaaacttcttatcttagcgtta
                                     M  K  L  L  I  L  A  L
acgtgcgcggcggccgtctgggccagacccggtgaaacttactcagacaagtatgacacc
 T  C  A  A  A  V  W  A  R  P  G  E  T  Y  S  D  K  Y  D  T
atcgatgtgaatgaagtcctccagtccgagcgtttgttgaaaggctacgtggagtgcctg
 I  D  V  N  E  V  L  Q  S  E  R  L  L  K  G  Y  V  E  C  L
ttagataagggacggtgcacaccggacggcaaggagctgaaggacaccctccctgatgcc
 L  D  K  G  R  C  T  P  D  G  K  E  L  K  D  T  L  P  D  A
ctggaacacgaatgcagcaaatgtaccgagaaacagaagtccggcgcggacaccgtcatc
 L  E  H  E  C  S  K  C  T  E  K  Q  K  S  G  A  D  T  V  I
agacacctggtgaacaagcgcccggagctgtggaaagagttggcggttaagtacgacccc
 R  H  L  V  N  K  R  P  E  L  W  K  E  L  A  V  K  Y  D  P
gagaacatctaccaggagagatacaaggaccggctggaatcggtgaaggaacattaaacg
 E  N  I  Y  Q  E  R  Y  K  D  R  L  E  S  V  K  E  H  -
gtccaacagcaatccgattttaatgctgggtggaggagatggctcacgtaatactgatat
tataagaataaaagacgcaatatgtaaaaaaaaaaaaaaaaaaaaaaaa
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Fig. 8.9 Complete sequence of Tha p 1 mRNA and its translation to amino acids. Stop codon is identified as a dash. Signal peptide is underlined (European Nucleotide Archive accession number HE962022)

3.4 Tha p 1: Cloning and Amino Acid Sequence

Mature Tha p 1 mRNA (Fig. 8.9) could be sequenced isolating the larva RNA using the traditional method of TRIzol and performing a retrotranslation followed by a polymerase chain reaction (RT-PCR).

Tha p 1 was modelled upon the crystalline structure of the chemosensory protein 10 from *Bombyx mori*, using the web interface Fist approach mode of Swiss-model (available at http://swissmodel.expasy.org/). The predicted model (Fig. 8.10) showed that Tha p 1 was folded into 6 α-helices joined by random coils, with two possible disulfide bonds between Cys 56/Cys 58 and Cys 29/Cys 36.

Despite the high homology between the chemosensory proteins of *T. pityocampa* and *B. mori* (Fig. 8.11), patients sensitized to the pine processionary larva did not recognize any protein of a silkworm whole body crude extract (data not shown).

3.5 Allergens Present in Setae

To obtain a protein extract, setae were isolated from the larva body using forceps and a magnifying glass. We designed an especial extraction method which included several consecutive steps in order to measure differences in solubility of the potential allergens contained inside the setae or present on their surface.

Fig. 8.10 Predicted 3D model of Tha p 1, modelled upon the crystalline structure of *Bombyx mori* chemosensory protein 1. Cys residues are highlighted in *pink*

Fig. 8.11 Alignments of the chemosensory proteins' sequences of several moths and butterflies: *Bombyx mori* (NP_001037064 chemosensory protein 3 and ABH88203 chemosensory protein 10), *Papilio xuthus* (BAF91712 chemosensory protein), *Heliothis virescens* (AAM77040 chemosensory protein 2), *Heliconius melpomene* (CAJ01513 hypothetical protein) and *Manduca sexta* (AAF16721 sensory appendage protein 4). The alignment was performed using the web interface ClustalW available at http://www.ebi.ac.uk/Tools/clustalw2/index.html. *Red* indicates identical amino acids, *green* indicates semi-conserved substitutions and *blue* indicates conserved substitutions

1. As a first washing step, the setae were resuspended in PBS and after a short incubation at room temperature (5 min), setae were centrifuged and supernatant 1 (SP1) was reserved.
2. The pellet containing the setae was redissolved in PBS and sonicated. Sonication induced the fragmentation of the setae, exposing in this way its internal content. After centrifugation, supernatant 2 (SP2) was reserved.

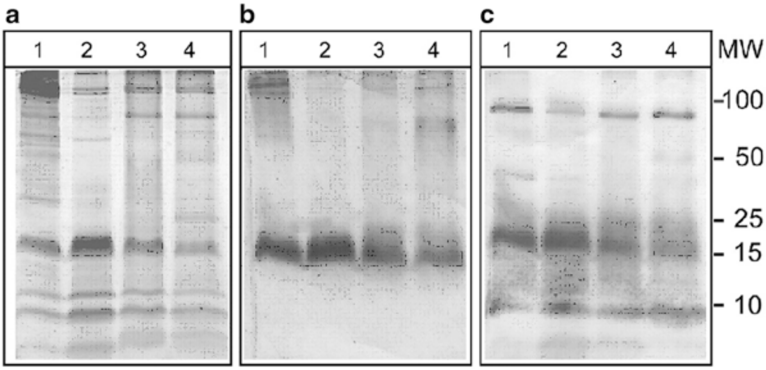


Fig. 8.12 Setae protein and allergen content studied by sequential extraction in different solutions. Coomassie stained SDS-PAGE (**a**), rabbit polyclonal immunoblot (**b**) and human IgE (**c**) immunoblot using a pool of positive sera. *Lane 1*: washing in PBS (SP1). *Lane 2*: SP2 supernatant obtained after sonication. *Lane 3*: SP3 extraction in SDS. *Lane 4*: SP 4 proteins obtained after extraction of the setae in hot SDS. Estimated molecular weights are indicated

3. The pellet was further redissolved in an ionic detergent solution (2 % SDS), incubated at RT and centrifuged to obtain supernatant 3 (SP3).
4. The pellet was redissolved again in the same detergent solution and then boiled and centrifuged to obtain supernatant 4 (SP4). This step tried to elute tightly non-covalently bound proteins attached to insoluble matrices, for instance to chitin.

The protein content of the different fractions was similar, ranging from 252 μg/ml (SP1) to 190 μg/ml (SP2).

After running the supernatants in a SDS-PAGE, proteins were visualized by Coomassie staining or transferred to nitrocellulose to perform Western blot (Fig. 8.12). The immunoblots were revealed with a rabbit anti-setae antiserum or a pool of *T. pityocampa* sensitized human sera. More than 25 different proteins were observed by Coommassie staining of the consecutive extracts (Fig. 8.12a). Some of these proteins (for example, the one of around 25 kDa, see Fig. 8.12a lane 4) were only extracted by boiling in detergent solution. A 15 kDa allergen was strongly detected when revealing the western blot with the rabbit antiserum (Fig. 8.12b). When revealing the Western blot with the pool of human sera (Fig. 8.12c), three proteins were mainly detected: a high molecular weight protein of around 90 kDa, a medium molecular weight protein of around 15 kDa and a low molecular weight protein of around 10 kDa.

The whole study demonstrated that allergens could be detected after an extraction that minimally broke setae and that sonication induced the release of an additional amount of protein. Harsher extraction conditions such as the use of high amounts of ionic detergents and the combined use of high temperature and detergents released further amounts of allergens from the setae, indicating that these allergens remained attached to the chitin core and were not solubilized by conventional extraction conditions. This fact led us to believe that after skin penetration, allergens could be delivered to the immune system in a dual way: a fast release of allergens present in the outside of the setae and a slow or very slow release that must occur after degradation of the setae by chitinases. Macrophage chitinases could directly deliver allergen to the macrophages and these cells could now present the allergens for optimal immune response, especially for IgE synthesis.

A second finding of the sequential extraction was that a protein of around 15 kDa was the most abundant component found in the different extracts, especially after mincing the setae by sonication (Fig. 8.12a). Furthermore, this protein bound specific antibodies from a rabbit antiserum obtained after immunization of rabbits with a whole setae extract (Fig. 8.12b) (Rodríguez-Mahillo et al. 2012). It was also by far the allergen most strongly detected by a pool of sensitized human sera (Fig. 8.12c). All these data suggested that this allergen could in fact correspond with thaumetopoein described by the group of Lamy several years ago (Lamy et al. 1985).

Figure 8.13 shows the IgE and IgG recognition patterns of 18 *T. pityocampa* sensitized patients. Again, several allergens present in the setae extract were detected, although the IgG recognition patterns of the whole body and the setae extracts were much weaker than the IgE recognition patterns. This fact implies that, for unknown reasons, sensitized subjects only produced IgE antibodies. In the IgE immunoblots, the 15 kDa protein was recognized by 72.2 % of the patients, while the low molecular weight allergen (10 kDa) was only detected by 27.7 %. Although the molecular weight of the 15 kDa setae allergen (Fig. 8.13, asterisk) was similar to that of the major body allergen Tha p 1 (Fig. 8.13, arrow), recognition of both allergens was not always paired (Fig. 8.13, patients 4 and 12), a fact that demonstrated that they are different proteins. The 15 kDa setae allergen, registered as Tha p 2, has been sequenced recently (Rodríguez-Mahillo et al. 2012). It has not similarity with Tha p 1 and it may correspond to the thaumetopein described in 1985, but unfortunately no information about the sequence of this allergen was provided. It is interesting to note that Tha p 2 showed similarity in the carboxy terminal region to a hypothetical protein of *Acyrthosiphon pisum*, the pea aphid (Fig. 8.14). This fact suggested that both proteins could be members of an unknown family of insect proteins and that more allergens of this family could be found in the future.

The proteins of the setae extract (SP1, Figure 8.12) were purified by reverse phase HPLC using a C18 column. More than 60 proteins were resolved (Fig. 8.15).

The amino end of the low molecular weight setae allergen (Tha p 3) has been sequenced (LAVETPEPISSN) and some other internal sequences have been obtained by the novo sequencing and MALDI-MS (EKDVHEWTGANWK ($m/z = 1{,}698.832$) y DVHEWTGANWK ($m/z = 1{,}441.685$) y VHVEWKGDN, where K can also be Q). None of these sequences had similarities with any other described protein.

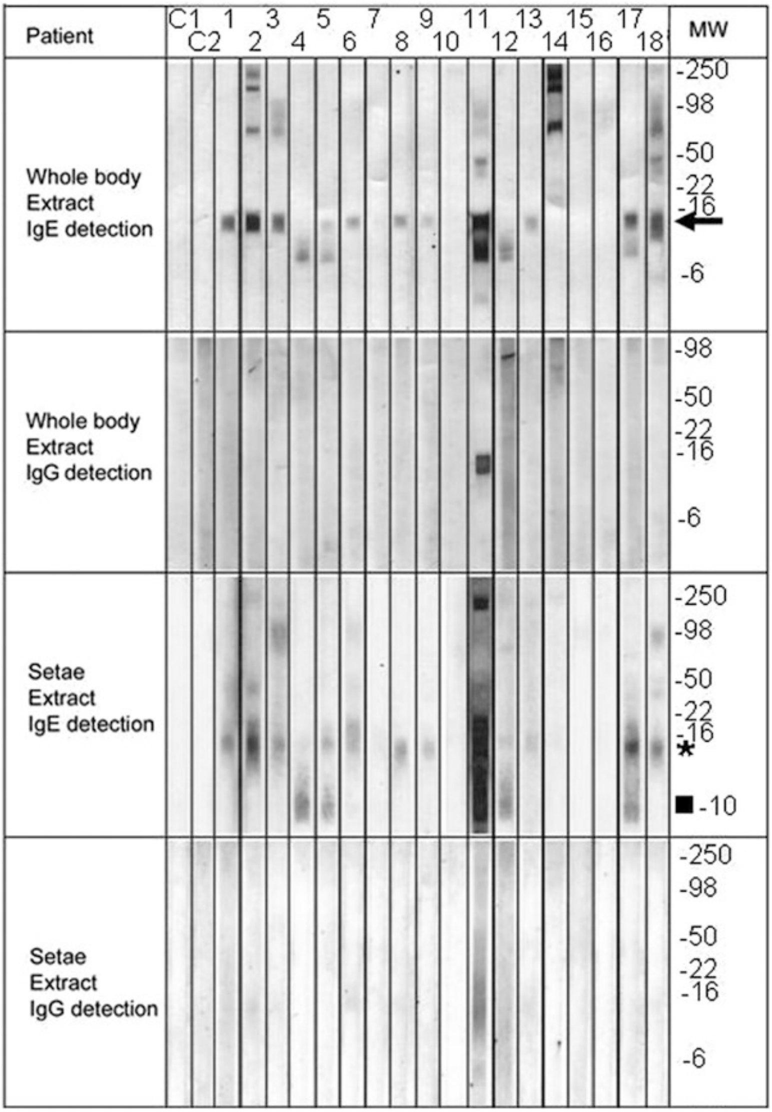


Fig. 8.13 IgE and IgG detection of proteins of *Thaumetopoea pityocampa* whole body and setae extracts. Individual sensitized patients (1–18) and healthy donors (C1 and C2) sera were tested. Tha p 1 is marked with an *arrow*. Tha p 2 is marked with an *asterisk*. Tha p 3 is marked with a *square*

3.6 Allergens Present in Larvae from Different Geographical Origins

When comparing the protein content of *T. pityocampa* from Spain (Valladolid and Madrid) and France (Charters, Orleans, Tours) by SDS-PAGE and immunoblot, we could find that they presented some differences. Figure 8.16 shows that although the

```
XP_001947225      MFAVVIFVLICAHAVHPSTVISGNTLTNRGSDKDNAAAGAAAAGDVIAATGGIIDEGSLA 60
Tha p 2           -----MKLLIFATLIALSSSVP--QLSEK-------------AEEAIDLT---------- 30

XP_001947225      AEHESDTDHALTKRGSGTDHAAQGAGTAGDVIAATGGIIAAGSLATGPLAPVVAGIGGVT 120
Tha p 2           ----YQEKNNLFDLGS----------VAGDILSRDG------------------------ 52

XP_001947225      AAVGGLVSVISKAADQSCREFGCHKNYCWSYC-------SLGNQWCYTTKTYSQSFEYVS 173
Tha p 2           CHVS---------------FGCHKGYCWAGCGNPTNPWSWGENWCYTTKSYSQSYSYVQ 96

XP_001947225      CTRDDECNGCWKCAGSCTL 192
Tha p 2           CTQDSECNGCWKCGGPCSA 115
```

Fig. 8.14 Alignment of the Tha p 2 sequence and a hypothetical protein from *Acyrthosiphon pisum*, a pea aphid (accession number XP_001947225). The alignment was performed using the web interface ClustalW available at http://www.ebi.ac.uk/Tools/clustalw2/index.html. *Red* indicates identical amino acids, *green* indicates semi-conserved substitutions and *blue* indicates conserved substitutions

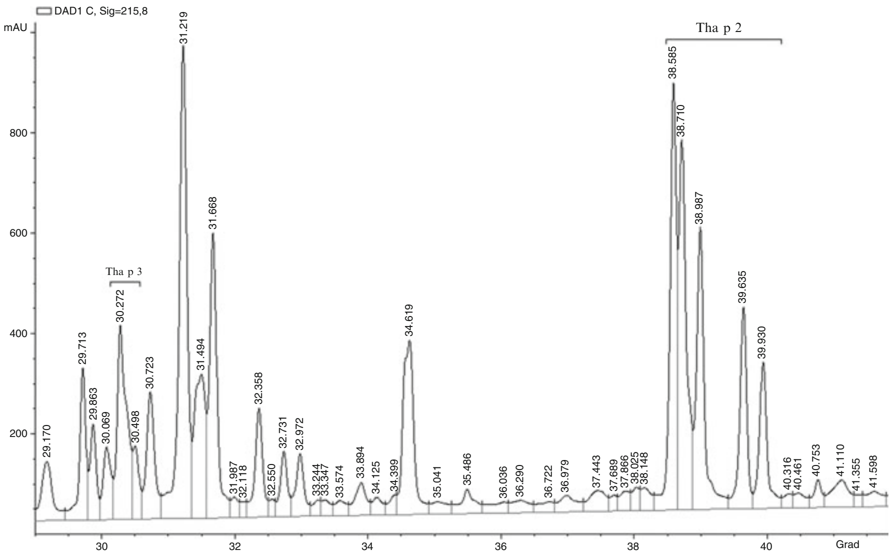


Fig. 8.15 RP-HPLC chromatogram ($A_{215\ nm}$) of the setae extract obtained in PBS after sonication. Detail from 20 to 67.5 % acetonitrile (ACN). Retention times of each peak are included. Tha p 2 corresponds to the peaks with retention times 36.586–39.930. Tha p 3 corresponds to the peaks with retention time 30.272 and 30.498

Coomassie stained SDS-PAGEs may be similar, patients with allergic symptoms related to exposure to larvae were able to detect different proteins from larvae collected in Spain and larvae collected in France. We do not know at present if this finding could have any clinical value or if it simply reflects normal variations in antigenic content of the different larvae.

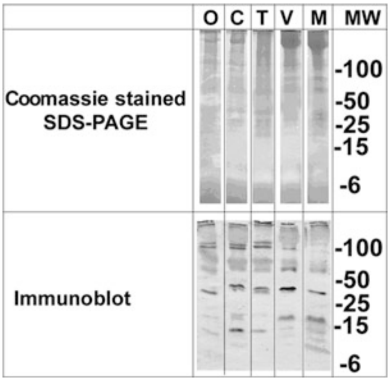


Fig. 8.16 Protein content of *T. pityocampa* from different origins in Spain and France. Coomassie stained SDS-PAGE and immunoblot revealed with a pool of *T. pityocampa*-allergic patients' sera. The whole body extracts were obtained using larvae from Orleans (*O*), Tours (*T*), Chartres (*C*), Valladolid (*V*) and Madrid (*M*). Estimated molecular weights (*MW*) are indicated

3.7 Differences Between Immune and Non-immune Reactions to Setae

As previously shown, around half of the people with suspected *T. pityocampa* larvae reaction showed positive skin prick test responses and clinical differences with respect to non- sensitized subjects indicating that an immune response was responsible for the symptoms (Vega et al. 1999). Therefore, the presence or not of immune reactions to larva allergens should be diagnosed early in order to avoid the progressive increase of the sensitization that occurs after new contacts with the allergen/s. The normal sensitization process implies that the sensitized subject responds to an allergen that he previously tolerated and that during the development of the immune response the symptoms appear after shorter exposure times or even after decreasing allergen exposure levels. As the immune response has a genetic background, not all the individuals show symptoms after exposure to setae, whereas toxic or irritant mechanisms normally affect all exposed subjects. However, the most practical way to demonstrate the presence of an immune response is the finding of positive skin tests or the presence of specific IgE antibodies on symptomatic subjects.

3.8 Factors Influencing a Specific IgE Response

One of the most intriguing findings of the immune response to this insect is the shift of the immune response to the IgE compartment. It is clear that other aspects of the immune response, such as non-antibody mediated cellular responses have not been yet explored, but the question remains unanswered: why are specific IgE antibodies synthesized in the absence of a specific IgG response? In other words, why are allergic responses preferred as a defensive response in spite of a classical IgG response as observed in a typical response to bacteria or viruses?.

Ducombs calculated that a single larva carries inside the mirrors around 1 million setae (Ducombs et al. 1979). This fact implies that a subject can be exposed to a high number of setae after direct contact to larvae, for instance after occupational exposure (pine cone collectors, etc). But the chronic exposure to low or very low number of setae can also occur. People living in the proximity of infested areas can be exposed to a low number of wind-carried setae for long time periods and this could lead to a high number of sensitized people such as found by us in epidemiological studies. The prevalence of positive skin prick tests reached 12 % in rural areas, 9.6 % in semi-urban areas with nearby pine forests and 4.4 % in urban areas (Vega et al. 2011a, b). Furthermore, this study demonstrated that 83.3 % of patients that fulfilled the criteria for a convincing reaction presented symptoms merely by walking or passing by pinewood areas without direct contact with larvae.

In addition to a long time exposure to low antigenic doses two other reasons can explain the shift towards an IgE response to setae antigens: the intradermal delivery and the presence of chitin or even the binding to chitin of the proteins present in the setae. Both conditions have been previously found to be especially relevant for IgE production (reviewed in Rodríguez-Mahillo et al. 2012) in animal models.

3.9 Relevance of the Immune Response

The presence or absence of an immune response to setae allergens is a relevant issue, because a sensitization to setae allergens changes the perspectives of the affected individuals. Toxic or irritant lesions produced by these larvae are usually more self-restricted to the contact areas. On the contrary, immune reactions can present systemic reactions that can require treatment at emergency rooms. Treatment of IgE-mediated reactions is similar to other allergic diseases and early diagnosis is required in order to avoid the development of the immune response.

3.10 Immune Reactions to Other Processionary Moths

Due to the close phylogenetic proximity between members of the Thaumetopoideae family, the presence of common allergens in other members of this family should be considered. A protein with similar molecular weight and showing cross-reactivity to the pine processionary was described to be present in the setae from the oak processionary larva (Lamy et al. 1988). Furthermore, workers exposed to oak processionary larvae presented severe anaphylactic reactions during work, a fact that suggests that IgE-mediated mechanisms were the cause of these reactions. It is therefore likely that other members of the same family could also have sensitizing properties (Bosma and Jans 1998; Licht and Jonker 1998).

3.11 Conclusions

The most relevant data about immune responses to larvae of pine processionary moth obtained so far can be summarized as follows:

- Both the whole larvae and the setae contain IgE-binding proteins (allergens).
- Some of them are restricted specifically to either the larva or the setae.
- In endemic areas, the IgE-immune response occurs in a high proportion of exposed subjects
- If present, the immune response is near always restricted to IgE, being the specific IgG response absent in most cases.
- Cell-mediated immune responses have not been tested so far and could be operative in some subjects.

4 Epidemiology and Clinical Symptoms of Human Reactions to Processionary Moths

Jose María Vega, Jesús Vega, José Carlos García-Ortiz, Ignacio Moneo, and Alain Roques

4.1 Introduction

In Europe and the Middle East, processionary moths (*Thaumetopoea* spp.) are among the main urticating Lepidoptera. From their third larval instars, they are armed with hair-like material called setae that can cause a variety of cutaneous reactions, ocular lesions, rhinitis and more rarely dyspnea and anaphylaxis. These reactions are attributable to a combination of non-allergic and allergic factors. IgE-mediated hypersensitivity reactions may be implicated when the process is rapid, recurrent and progressively more severe.

4.2 Epidemiology

4.2.1 *Thaumetopoea* Reactions are Underestimated: Possible Reasons

Although *Thaumetopoea*-related disease is common in the Mediterranean area, currently its importance is underestimated in both clinical practice and the scientific literature. This fact may be attributable to several factors including:

1. probably because most reactions are mild and transient;
2. in endemic areas, often the patient is aware of the cause and self-treats the condition;

3. there are not specific symptoms or signs of *Thaumetopoea*-related disease;
4. low knowledge of this pathology or inability of physicians to identify the suspected larvae;
5. diagnostic allergy tests are not commercialized.

However, we should also bear in mind that more serious reactions may occur and these are treated symptomatically without a diagnosis of the cause or without providing the patient with the necessary information to avoid future recurrences.

4.2.2 Epidemics of *Thaumetopoea* Reactions Can Occur

Another factor to take into account is that, although the reactions usually occur in isolated patients, at times, epidemics may occur. These can be related to exposure in areas of heavily infested tree forest or in communities located close to infested trees, the biological cycle of the insect or weather conditions, particularly strong winds.

4.2.3 Epidemiological Studies

Few epidemiological data have been published on reactions caused by species of the genus *Thaumetopoea*. Most studies describe isolated cases or series of patients attending in a hospital or incidence studies after outbreaks, such as one of *T. processionea* (6 % of 1,025 persons living in a radius of 500 m from infested trees in town) (Maier et al. 2003) and one of *T. pinivora* (18 % of 4,300 persons living in an infested area of about 3,500 ha) (Holm et al. 2009). However, two prevalence studies of reactions by *T. pityocampa* in general population have been reported and their results confirm that these reactions are common in endemic areas. In the first one, 9 % of 653 children living in a rural area had reactions with this Lepidoptera (Vega et al. 2003a, b). In the second, a cross-sectional study was carried out in 1,224 adults, the prevalence of skin reactions to *T. pityocampa* was 12 % in rural areas, 10 % in semi-urban areas with nearby pine forest and 4 % in urban areas (Vega et al. 2003a, b).

Children are particularly susceptible to Lepidoptera related conditions, this fact may be related to frequent outdoor activity; their natural curiosity leads them to touch the larvae or play with sand or vegetation that contains rests of them. As the article previously mentioned shows, these reactions are common in children from rural areas. However, peripheral urban areas with nearby pine/oak trees are also areas with a high incidence of *Thaumetopoea* reactions and where the treatment of the pest is particularly difficult (Maier et al. 2003; Gottschling et al. 2007; Artola-Bordás et al. 2008; Vega et al. 2011a, b).

4.2.4 Risk Factors

Exposure to the Larvae

The main risk of skin reactions to *T. pityocampa* is directly related to exposure to the larvae. This risk is significantly higher in people with daily exposure (Vega et al. 2011a, b).

Influence of Sex, Area of Residence, Age and Atopic Status

In adults, the reactions are more frequent in men from rural or semi-urban areas because they are more frequently exposed to larvae due to occupational exposure or hobbies. However, age influence or atopic status has not been found (Gottschling et al. 2007; Vega et al. 2011a, b).

Seasonal Influence

Thaumetopoea-related disease is usually seasonal. Most of the reactions occur in the months corresponding to urticating larval stages (*T. pityocampa*/*T. wilkinsoni*: January-April; *T. processionea*: May-July; *T. pinivora*: June-August). Within these months, the last larval stage has the highest risk because the number of setae and their allergenicity is maximal (Rebollo et al. 2002). In addition, the processionary larvae whose nymphosis occurs in the soil (all the species except *T. processionea*), in their last stage in which they descend from the trees, they cause a bigger exposure and a higher number of reactions (Vega et al. 2011a, b).

However, an out-of-season contact with setae can occur through dead larvae, tents, debris from an infested pine forest or with setae present in spit or hair from pets or livestock (Vega et al. 2004; Maronna et al. 2008). In addition, setae shed by the larvae can persist in the environment for many years. On the other hand, patients with hypersensitivity to the larvae can have symptoms with a minimal exposure to setae. These facts can explain why some people have recurrences of the reactions throughout the whole year.

Occupational Exposure

Exposed workers have a major risk of *Thaumetopoea*-related disease due to a major frequency and intensity of the exposure to setae. This risk is up to 5 times higher in forest workers exposed to *T. pityocampa* (Vega et al. 2011a, b). The jobs at greatest risk are pinecone collectors and lumberjacks (Vega et al. 2004; Vega et al. 2011a, b). Moreover, epidemic affectation of soldiers exposed to *T. processionea* has been reported (Hesler et al. 1999).

4.3 Clinical Manifestations

4.3.1 Dermatological Manifestations

Clinical Patterns

Cutaneous reactions are the most frequent clinical manifestation. The main symptom is an intolerable itching sometimes without visible lesions. Skin lesions by *Thaumetopoea* larvae are of two different types: immediate wheal and flare reaction (contact urticaria, Fig. 8.17) and delayed and persistent itchy eruption, usually reported as a papular eruption (Fig. 8.18). On the other hand, an overlapping of these skin lesions or a dual reaction can happen.

Most of the patients with contact urticaria by *Thaumetopoea* larvae are allergic (IgE-mediated) (Vega et al. 2000). The eruption appears within 30–60 min after exposure to larvae and disappears within 24 h and usually within a few hours. The whealing reactions can be strictly confined to the area of contact but they can also appear as generalized urticaria, facial oedema and sometimes can associate extracutaneous symptoms and anaphylactic reactions.

Delayed reactions do not appear until 2–24 h after exposure. The most frequent type, consist in infiltrated papules similar to insect bites that persists for several days. Sometimes, an eczematization of the lesions occurs due to the scratching or when the outbreaks are repeated. Less frequently, it has been described as pin-sized red papules, pustules and erythematous streaks which clear within 3–5 days (Maier et al. 2003); or pustular lesions on the palms of the hands of small children (Vega et al. 2003a, b); or vesicular dermatitis (Bogaerts-Rosbergen and van Wijk 2010); or blistering dermatitis, more frequently reported after *T. pinivora* exposure (Fagrell et al. 2008; Holm et al. 2009). In these delayed cutaneous reactions

Fig. 8.17 Intense contact urticaria with associated angioedema on the face and neck of a 5-year-old girl after playing in a sandpit close to pines infested with *Thaumetopoea pityocampa*. The allergological study showed a positive skin prick test with setae and whole larval extracts (Reprinted with permission of the authors and the editor (Vega et al. 2011a, b)

Fig. 8.18 Papular-erythematous rash on the anterior face of the wrist and the palm of the hand of a 7-year-old girl. Reprinted with permission from the authors and the editor (Vega et al. 2011a, b)

it is thought that an irritant mechanism is responsible, although a possible a delayed-type allergic reaction could also be present.

Distribution: *Thaumetopoea* larvae usually cause an airborne disease (Maier et al. 2003; Vega et al. 2011a, b). Skin lesions are more frequently located on exposed areas, typically on the neck and limbs, and in particular the wrists, forearms, flexure areas and ankles, although covered areas of the body may also be affected. Physical activity and scratching may increase the intensity of the dermatosis.

The palms of the hands and the interdigital spaces are more often affected in children, probably due to direct contact with larvae when playing on infested places (Vega et al. 2003a, b).

Ocular Manifestations

Ocular involvement due to processionary setae is also frequent and it may or may not be accompanied by dermatological involvement. It happens in about 11–30 % of individuals with *Thaumetopoeae* reactions (Maier et al. 2003; Holm et al. 2009; Vega et al. 2011a, b).

The setae can cause different ocular pathologies, which are aggravated by scratching. After setae exposure, immediate or delayed signs as conjunctivitis, keratitis and uveitis can appear (Fig. 8.19). Three cases of ophthalmia nodosa by *T. pityocampa* setae have been reported (Watson and Sevel 1966). Moreover, late signs (cataract, vitritis and retinitis) by an intraocular migration could happen (Trincao et al. 2012), although other authors with years of experience in ocular lesions induced by *T. pityocampa* have not found any case (Portero et al. 2013).

As in dermatological involvement, a combination of non-allergic and allergic factors can be responsible of the ocular manifestations. Patients with IgE-mediated hypersensitivity show often ocular itchiness with or without redness of the eye that disappears spontaneously or after treatment with antihistamines in minutes or few hours.

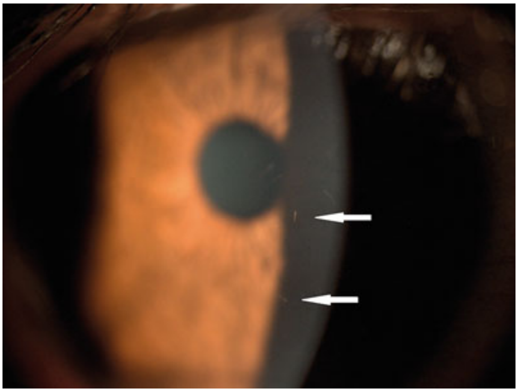


Fig. 8.19 Higher magnification, slit-lamp images of the cornea, showing the setae of *T. pityocampa* larvae (*white arrows*) (Image obtained by courtesy of A. Portero)

Other Manifestations

Respiratory involvement is less common and include rhinitis, dyspnea, cough and wheezing occasionally associated with an anaphylactic reaction with multiorgan involvement as described in several case reports (Licht and Jonker 1998; Bosma and Jans 1998; Vega et al. 1997, 1999; Shkalim et al. 2008).

Other systemic symptoms, such as fever (Gottschling et al. 2007; Maronna et al. 2008; Holm et al. 2009), vomiting, abdominal pain or hypertension (Kozer et al. 1999) have only rarely been reported. Moreover, a 15-month-old boy with severe orofacial edema after ingestion of a processionary larva has been reported (Inal et al. 2006). Up to date, fatal cases caused by *Thaumetopoea* species have not been informed, as it has happened in rare cases of lonomism by other Lepidoptera.

Hypersensitivity Reactions

In addition to mechanical irritation, Lepidoptera are capable of causing hypersensitivity reactions in susceptible individuals. The term hypersensitivity is used when a reaction causes objectively reproducible symptoms or signs, initiated by exposure to a defined stimulus at a dose tolerated by normal subjects. It can be classified in allergic (immunologic mechanism -IgE or not IgE mediated-) and nonallergic (Johansson et al. 2001).

With respect to *Thaumetopoea*-related disease, experimental application of setae cause a local inflammatory reaction in all subjects showing different intensity among them (Fagrell et al. 2008). Moreover, within the same population or workers group with similar exposure, stands out the great variability between subjects. There are cases practically free of symptoms, but others present severe reactions (Vega et al. 2011a, b; Holm et al. 2009). To date, there is evidence of a type I hypersensitivity (IgE-mediated) to *Thaumetopoea* setae, however, there has been no conclusive data of the participation of other types of hypersensitivity mechanisms, although cellular immunity is probably involved in some cases (Vega et al. 2011a, b). It also seems that chitin and its degradation products, which are powerful promoters and regulators of immune reactions, may play a role in the variable sensitivity to these insects (Battisti et al. 2011).

A difficulty to apply the term hypersensitivity to *Thaumetopoea* reactions is that in non-experimental conditions it is not possible to quantify the exposure (the number or amount of fixed setae) and therefore, there is a lack of definition in the magnitude of the reaction that could have a normal subject (not hypersensitive) after setae exposure. Moreover, an overlapping of pathogenic mechanisms (non-allergic and allergic) can happen, existing difficulties to value the clinic relevancy of each one.

IgE-Mediated Hypersensitivity

- Prevalence
  In endemic areas, approximately half of the adults with dermatological reactions due to *T. pityocampa* are allergic (IgE-mediated) (Ruiz 2011). In children from a rural area, a lower prevalence of allergic reactions has been found (7 % from 60 cases) (Vega et al. 2003a, b). However, this prevalence increases in children from communities located close to infested trees, as peripheral urban areas (data not shown).
- Risk factors
  The frequency and intensity of exposure to the larvae facilitates the sensitization to their allergens. Therefore, the main risk factor to IgE *T. pityocampa* hypersensitivity is the male sex because males are more frequently exposed to larvae by an occupational exposure or by some hobbies (Ruiz 2011).

  The atopic predisposition has not been found as a risk factor, as in the case of allergic reactions to other insects as Hymenoptera. The age and the presence of relatives with reactions to *T. pityocampa* were not found to be risk factors (Ruiz 2011).
- Clinical pattern
  The clinical pattern of patients with IgE-mediated hypersensitivity to *T. pityocampa* shows significant differences with respect to nonsensitized patients. Patients with IgE-mediated hypersensitivity experience immediate and progressively more severe reactions but of a lower duration. In addition, IgE-mediated reactions can appear even after a minimal exposure as walking through a pine forest. Moreover, the symptoms may occur throughout the year including the months in which the insect has not setae (Vega et al 1999; Ruiz 2011). In these patients, the characteristic skin manifestation is contact urticaria accompanied by angioedema -particularly on the eyelids- in half the cases. Rhinitis occurs more frequently and the most of cases of anaphylactic reactions have been reported among allergic individuals. Table 8.1 shows the differences between individuals with IgE-mediated and non allergic reactions to *T. pityocampa*.

Table 8.1 Differences between individuals with IgE-mediated and non allergic reactions to *T. pityocampa* larvae

	IgE-mediated	Non allergic
Affected people	Some	All
Prior contacts with the larvae (prior immunization phase)	Necessary	Not necessary
Intensity of the exposure	Minimal	High
Latency period	<1 h	>1 h
Number of reactions	Several	Few
Characteristic skin manifestation	Contact urticaria	Papular dermatitis
Distribution of cutaneous lesions	Exposed areas/ generalized	Exposed areas
Severity of the clinical manifestations	Greater	Lower, except for extensive exposure
Skin prick test	Positive	Negative
Specific IgE	Yes	No

Abbreviation: *IgE* immunoglobulin E

4.4 Diagnosis

There are no specific clinical signs of reactions to *Thaumetopoea* larvae. Diagnostic suspicion is based on several observations.

4.4.1 Criteria of Clinical Suspicion (Vega et al. 2011a, b)

- A history of exposure in the previous 24 h in an area with trees infested with these insects is essential. Although *Thaumetopoea*-related disease has a seasonal importance, it can also cause symptoms throughout the whole year.
- Presence of urticaria, with or without angioedema, or papular eruption that are very pruritic and tend to appear on the neck and flexure and distal areas of the limbs. In children, the palms and interdigital spaces in particular should be checked for lesions. Keratoconjunctivitis is also frequent.
- Identification, whenever possible, of urticating hairs on the skin or clothes of the patient by applying an adhesive strip or directly through use of a dermatoscope.
- Reactions should not appear in other circumstances, and other differential diagnoses that may be associated with similar signs and symptoms should be excluded. These include reaction to bites of other insects, nodular or atopic prurigo, scabiosis, other contact eczemas and urticarias, photodermatoses, etc.
- Criteria for suspicion of an IgE-mediated allergic component include immediate and progressively more severe reactions, even with minimal exposure.

4.4.2 Pathologic Anatomy

Microscopic assessment of the different skin reactions to *Thaumetopoea* is also nonspecific and cannot be used for diagnosis. The microscopic pattern of the most characteristic delayed cutaneous manifestation, papular eruption, shows a nonspecific inflammatory reaction with epidermal oedema and a perivascular lymphohistocytic infiltrate with eosinophils, as seen in bites and reactions to other insects. There are not histopathological descriptions about vesiculopustular or blistering dermatitis.

4.4.3 Allergy Diagnostic Tests

Confirmation of an IgE-mediated mechanism would be made by a positive result in the skin prick tests with extracts to non-irritative allergen concentrations and/or specific IgE determination in serum using setae and whole body extracts from larvae in their last larval stage.

4.4.4 Skin Tests

Several studies have shown the safety and diagnosis usefulness of the skin prick tests using whole larval and more recently setae extracts. Biological standarization with both extracts has been performed and an extract concentration of 5 mg/ml was adequate for diagnosis (García-Ortiz et al. 2010; Ruiz 2011). When whole larvae and setae extracts have been compared by skin testing and immunoblotting, both extracts have demonstrated to be useful and complementary for diagnosis (Rodríguez-Mahillo et al. 2012; Ruiz 2011).

Patch testing with extracts at non-irritative concentrations can be a useful test in the investigation of *Thaumetopoea* delayed reactions such as has been demonstrated with other Lepidoptera.

4.4.5 Serological Determinations

IgE immunoblotting is a less sensitive technique than the skin prick test but it is very specific. Previous studies have shown a positive result in 72–74 % of the patients with positive skin prick tests (Vega et al. 1999; Ruiz 2011), thereby demonstrating the usefulness of this test. Specific IgE determination to *T. pityocampa* larvae by EAST method has been also reported (Fuentes et al. 2006).

Up to date, two major allergens from *T. pityocampa* have been described: Tha p1 from whole larvae extracts (Moneo et al. 2003) and Tha p2 from setae extracts (Rodríguez-Mahillo et al. 2012). These findings open the possibility of the molecular diagnosis for urticating Lepidoptera.

4.5 Treatment

Table 8.2 shows some of the preventive measures to avoid reactions to these insects. If a contact with these larvae has happened, washing with soap and water and removal of contaminated clothing is necessary. Scratching should be avoided as far as possible as this will exacerbate cutaneous symptoms or it will be able to cause ocular complications. Once symptoms have appeared, treatment is exclusively symptomatic: oral antihistamines to control pruritus and topical corticosteroids for eczematous lesions and papular eruption. A novel treatment with topical application of potassium dobesilate 5 % cream, a specific inhibitor of fibroblast growth factor, has been useful in a case of dermatitis due to contact with pine processionary larvae (Cuevas et al. 2011).

In patients with extensive or refractory lesions, oral corticosteroids can be used. In the event of anaphylactic reactions, early diagnosis is needed, along with immediate treatment with epinephrine as well as corticosteroids and antihistamine agents. Reactions caused by oral exposure can require urgent orotracheal intubation and they should be managed by a qualified specialist.

The acute ocular itchiness can be controlled by antihistamines. However, keep in mind the possibility of more serious consequences and if the symptoms persist or if the patient refers foreign body sensation, grittiness, photophobia, or visual worsening, the patient should be managed by an ophthalmologist. On the other hand, some authors report that the removal of intracorneal hairs significantly reduces the risk of intraocular penetration. However, other authors do not advise it because they have not found any case of setae-induced intraocular penetration and because efforts to physically remove them are likely to do more harm than good (Portero et al. 2013).

General doctors and specialists should become familiar with *Thaumetopoea*-related disease. Correct diagnosis and appropriate information that emphasizes preventive measures will reduce the incidence and severity of these reactions.

Table 8.2 Recommendations for avoiding reactions to *Thaumetopoea* larvae

Avoid heavily infested areas in the months in which the larvae are urticating, especially on windy days and when the larvae descend in processions from the trees. Pets (for example dogs) should also avoid these forests at this time
If larva processions are seen, keep children away and never disturb, touch, or sweep them
Do not collect objects (pine cones, wood, etc) from infested forests or touch the larva tents
In communities located close to infested trees, during the month in which the larvae are urticating, avoid to dry the clothes outdoors. Moisten the surrounding area to ensure that the setae of the larvae remain on the ground.
In edges and clears of infested pine/oak forests avoid removing the soil
In cases of occupational exposure, precautionary measures are essential when working in infested forests. It is important to leave as small an area of skin exposed as possible and to wear appropriate clothing and footwear. If the exposure is high use protective goggles or even a mask. Patients allergic to larvae should not work in infested forests

5 The Pine Processionary Moth: Assessment of Animal Health Issues

Julie Rivière, François Moutou, and Barbara Dufour

The pine processionary moth *Thaumetopoea pityocampa* causes significant forest damages (defoliation, weakening of trees, esthetic prejudice…) but can also induce important sanitary consequences in men and animals, both in pets (dogs, cats) and also in farm animals (horses, cattle, sheep, goats). Contact with irritating hairs of larvae can provoke oral, skin and eye damages which are most of the time benign but sometimes leading to allergic reactions up to anaphylactic shock.

5.1 The Pine Processionary Moth and Its Sanitary Consequences

The procession is the most dangerous period for men and animals, when larvae release irritant hairs when they feel assaulted or stressed. Damages may therefore result either from direct contact (with larvae or their tents, as they contain hairs remaining irritating even after the departure of the colony) or from indirect contact (with stinging hairs left on pasture during the procession or carried by the wind).

Urban, peri-urban and rural sanitary nuisances are more extensive and disturbing with the gradually spread of the pine processionary moth to northern France: populations of areas newly colonized are in fact usually not informed of these sanitary consequences.

5.1.1 Symptomatology

Envenomation by processionary larvae can be observed both in domestic pets (dogs, cats) and in farm animals (horses, cattle, sheep, goats). Clinical signs appear rapidly after exposure to stinging hairs, a few minutes to a few hours later on average. Symptoms depend on the terms of contamination but are globally similar whatever the species of larva involved (pine, oak, etc). Four main routes of exposure are classically described: skin contact, eye contact, inhalation and ingestion. The preferred involvement of the face and oral cavity can be explained by the behavior of animals. Thus, dogs are often contaminated by the nose and mouth by sniffing or even chewing and eating larvae in procession, whereas ruminants are contaminated when grazing grass on which larvae have left after their procession the stinging hairs (Charmot 1987).

Symptomatology in carnivores

Various studies report that the dog is by far the species most affected by envenomation (Charmot 1987; Darrasse 1991; Gleyze 1995; Pineau and Romanoff 1995; Pineau 1999; Scheiner 2003; Turpin 2006). This can be explained by their heightened curiosity during forest walks or even in gardens (procession is a curious phenomenon!), in contrast to the cat, which is more suspicious and so enters rarely in contact with larvae (oral exploration is less developed in the feline species).

- Local symptoms

  Oral damages following ingestion of larva or a contact with them are often predominant. Lingual impairment may also result from licking itching body parts of the animal. There is a strong inflammation and a swelling of the oral mucosa of the lips and tongue. Glossitis thus induced causes discomfort swallowing and results in an abundant salivation, anorexia and watering difficulties, which can lead to kidney failure (Blanchard 1994; Poisson et al. 1994). Necrosis of the tongue can also develop after blood coagulation of lingual vessels which prevent tissue oxygenation; it can go up to a loss of substance in the few days following envenomation, especially on dogs (Blanchard 1994) (Fig. 8.20).

  Skin lesions can also be observed: an acute onset of urticaria, erythematous and itching can be found in areas where the skin is thin, such as lips, chamfer, eyelids and ears (Poisson et al. 1994).

  Respiratory impairment can also be reported when dust loaded with irritating hairs reach the respiratory tract. This results in inflammation of the nasal mucosa, rhinitis and bronchitis, coughing violently and sometimes in dyspnea and shortness of breath (Bergia and Keck 1991). Some fatal cases have been reported after angioedema and asphyxia (Darrasse 1991).

Fig. 8.20 Edema of the tongue of a dog with blister on the ventro-rostral part of the tongue after envenomation by *T. wilkinsoni* (Bruchim et al., 2005)

Ocular impairment may also be observed but is less common. It takes the form of keratoconjunctivitis, glaucoma or sometimes corneal ulceration, causing pain, photophobia and lacrimation. Hairs can be embedded in ocular structures, in particular in cornea, causing relapse with chronic conjunctivitis.

In cats, the symptoms are similar to those observed in dogs. Acute pododermatitis can also be observed. Cases of envenomation in cats are nevertheless relatively rare (Darrasse 1991; Gleyze 1995; Pineau and Romanoff 1995; Poisson et al. 1994).

- General symptoms
  General symptoms are relatively rare, occurring primarily as marked exhaustion with hyperthermia which can go up to shock. A few animals sometimes develop an anaphylactic shock. Ingestion of larvae can also cause gastritis and vomiting (Chuzel 2004; Darrasse 1991).

Symptomatology in Ruminants

Ruminants, from the grip of fodder containing stinging hairs, develop vesicles on the apex of the tongue. They are very fragile, break easily and then lead to the formation of extensive superficial ulcers (Figs. 8.21, 8.22, and 8.23). Affected animals are often exhausted, anorexic and isolated themselves from the rest of the herd.

Fig. 8.21 Envenomation by a pine processionary larva (Photo J. M. Gourreau)

Fig. 8.22 Envenomation by an oak processionary larva with large superficial ulcer on the tongue of a cow (Braque 1999)

Fig. 8.23 Oral lesions (superficial ulcers) in small ruminants following envenomation by pine processionary larvae (Gourreau et al. 2002; photo M. Cornelis)

Symptomatology in Horses

In horses, congestion of the tongue, colics and extended urticaria were reported. According to the observations of Darrasse (1991), pruritus is sometimes intense and can lead to self-mutilation: for example, to relieve hives of the flanks, a mare gave herself frequently blows of hooves on the abdomen. Sweating may also be important (Charmot 1987). Many studies have recently been conducted to explore the envenomation in horses. Indeed, twice, massive waves of abortions were observed for which no known agent (microorganism, alkaloids, toxic plants . . .) have been identified, but epidemiological studies have revealed the presence of larvae in the environment; mares have certainly consumed them inadvertently (Cawdell-Smith et al. 2009; McDowell et al. 2010; Stewart 2009; Tobin et al. 2004). Thus, some species of stinging larvae can therefore now be considered as abortifacient potential agents in mare, as well as a few microorganisms, alkaloids and toxic plants (Stewart 2009).

5.1.2 Diagnosis and Differential Diagnosis

The clinical signs are generally nonspecific; the diagnosis is based on history: symptoms appearing suddenly, a few minutes to a few hours after a walk outdoor, in a forest or in a garden. Consultations are seasonal and take place mainly from February to May. Symptoms are dominated by a glosso-stomatitis which can be necrotic and ulcerative, and by an abundant salivation. Animals are often brought in emergency to the vet, because of the rapid onset of symptoms and their spectacular nature, which may suggest poisoning.

In carnivores, care must be taken not to confuse envenomation by the pine processionary moth with the ingestion of various caustics (irritating products), or with insect bites (wasps, bees, hornets) which also result in oral symptoms but rarely lead to necrosis. Ophidian envenomation may also be considered, although induced lesions are often more severe - the presence of fang can help to establish the diagnosis. Finally, the root of the tongue and the throat must systematically be inspected to eliminate any hairs which can cause inflammation and glossitis (Poisson et al. 1994).

In ruminants, oral symptoms may strongly suggest vesicular diseases such as foot-and-mouth disease (FMD) or bluetongue (BT). Epidemiological data must then be considered with interest in the differential diagnosis. Indeed, envenomations by processionary larvae are seasonal and occur mainly between January and May, as already mentioned. This affection can affect many animals grazing on the same area but the apparent contagion is less important than during outbreaks of FMD or BT. In addition, the udder and feet are not concerned in the case of envenomation by larvae (Gourreau 2002; Gourreau et al. 2002).

In the absence of specific test, diagnosis of envenomation by the pine processionary larva is primarily a diagnosis of exclusion based on clinical and epidemiological criteria, whatever the species affected. Specific tests depending on the location of symptoms (skin, eye, vascular . . .) can also be realized.

5.1.3 Treatment, Recommendations and Prognosis

The success of the treatment is related to three main factors (Demory 2004a, b):

- The precocity of the treatment: it allows limiting the consequences of envenomation (in particular lingual necrosis and loss of apical substance) and thus determines both the survival of the animal, but also the preservation of the tongue;
- The realization of a long-term treatment (during several weeks);
- The "non-breaking" of stinging hairs during treatment, so as not to facilitate the diffusion of the irritant substance.

Treatment is mainly symptomatic, with its main aims at fighting against the effects of histamine-releasing irritant substance. A corticosteroid, anti-histaminic and diuretic association is often recommended to support or restore the general condition of the animal. Antibiotics may also be prescribed if the lesions are already at an advanced stage (oral administration of amoxicillin or spiramycin). It is also important to ensure the good nutrition and good watering of the animal.

Prescription of local care is required; it should be established as soon as possible after the envenomation to limit the risk of lingual necrosis and to improve prognosis. Gentle washing should be done without rubbing the affected areas, to avoid breaking hairs and thus to prevent the release of the irritating substance. Injections of heparin in the wake of necrosis may be recommended. Lingual plasty can sometimes be considered after necrosis to restore the tongue to a form compatible with normal eating and watering (Darrasse 1991).

The prognosis of envenomation by pine processionary larvae can be severe, although many cases are mild. It depends largely on the intensity of contact with stinging hairs, the precocity and the duration of the treatment (Blanchard 1994). When the treatment is implemented early, in the first hours after envenomation, the prognosis is generally good and the animal's life is not in danger. Evolution is generally favorable and benign lesions heal in 8 to 10 days. When treatment is delayed (more than 24 h after envenomation) or if the contact with stinging hairs was intense (many bristles or long-term contact), the glosso-stomatitis evolves rapidly to necrotizing ulcerative on a large area (Blanchard 1994).

Thus, the short-term prognosis depends mostly on the severity of the systemic implications (acute renal failure, laryngeal edema . . .), while the longer-term prognosis depends on the severity of the lingual lesions, loss of apical part being relatively common during envenomation (Demory 2004a). Fatal cases are fortunately rare, but sometimes euthanasia is the only possible outcome when the lingual necrosis extends to the throat (Gleyze 1995).

5.2 Importance of Clinical Cases of Envenomation in Animals in France

As part of the URTICLIM project, two retrospective epidemiological surveys (Rivière 2011; Rivière et al. 2011) were conducted among veterinary practitioners, in order to:

- Identify veterinarians confronted with cases of envenomation in livestock and pets: this allowed identifying areas that have been most affected and highlighting geographical areas at risk (detection of cases of envenomation in animals may allow alerting public health actors and population of the newly colonized areas),
- Identify the main clinical signs and lesions of envenomation,
- Highlight the main diagnostic difficulties that may be encountered by veterinarians, especially the difficulties in diagnosing an envenomation in ruminants because of potential confusion with vesicular diseases,
- Find a serological test able to distinguish envenomation by the pine processionary moth from other vesicular diseases like FMD or BT, because these diseases could have severe sanitary, social and economic consequences. Unfortunately, this part of the project has not been successful, because laboratories contacted for the development of the diagnostic tool did not respond to the request -there is in fact actually not commercial reason to develop this tool.

5.2.1 Material and Methods

The two retrospective surveys were conducted 2 years apart, the first in 2008, covering the period 2005–2008, the second in 2010, covering the period 2008–2010. An electronic questionnaire including some sixty items all closed or mixed was sent to French veterinary practices all over the country through the SNGTV,[1] an association (NGO) including more than 600 contacts (a combination of single or multi practices, meaning one or more than one practitioner, in both rural and urban areas). We also contacted some respondents by phone to gather expended information. The second survey was conducted for the purpose of comparing the results with the first survey and to assess the temporal and geographical evolution of envenomation cases, in order to follow the expanding migration of the pine processionary moth via the location of animals affected. Both surveys were administered and stripped using "Sphinx" survey software online.

Moreover, a study of a data base located in the National Center for Veterinary Toxicological Information of Lyon (CNITV), which provides assistance for advices, diagnosis, treatment or prognosis about requests concerning poisoning of wild and domestic animals, was performed from 2008 to 2010.

5.2.2 Results

Epidemiological Surveys Among Veterinary's Clinic in France

A total of 122 answers were obtained in the first survey and 49 in the second (which equals respectively a response rate of 20 and 8 %). Between 2005 and 2008, 58.2 % of respondents said that they have seen cases of envenomation (Table 8.3). By the time of the second survey, that number had climbed to 63 %: the rate of practitioners who have been confronted with cases of envenomation by pine processionary moth was however equivalent in the two surveys and concerns approximately 60 % of practitioners in each study.

Table 8.3 Number of veterinarians who have been confronted with cases of envenomation by the pine processionary moth between 2005 and 2008 and between 2008 and 2010

	No. 2005–2008	% 2005–2008	No. 2008–2010	% 2008–2010
No	51	41.8	19	38.8
Yes	71	58.2	30	61.2
Total	122		49	

[1] Société Nationale des Groupements Techniques Vétérinaires (National Society of Veterinary Technical Groups)

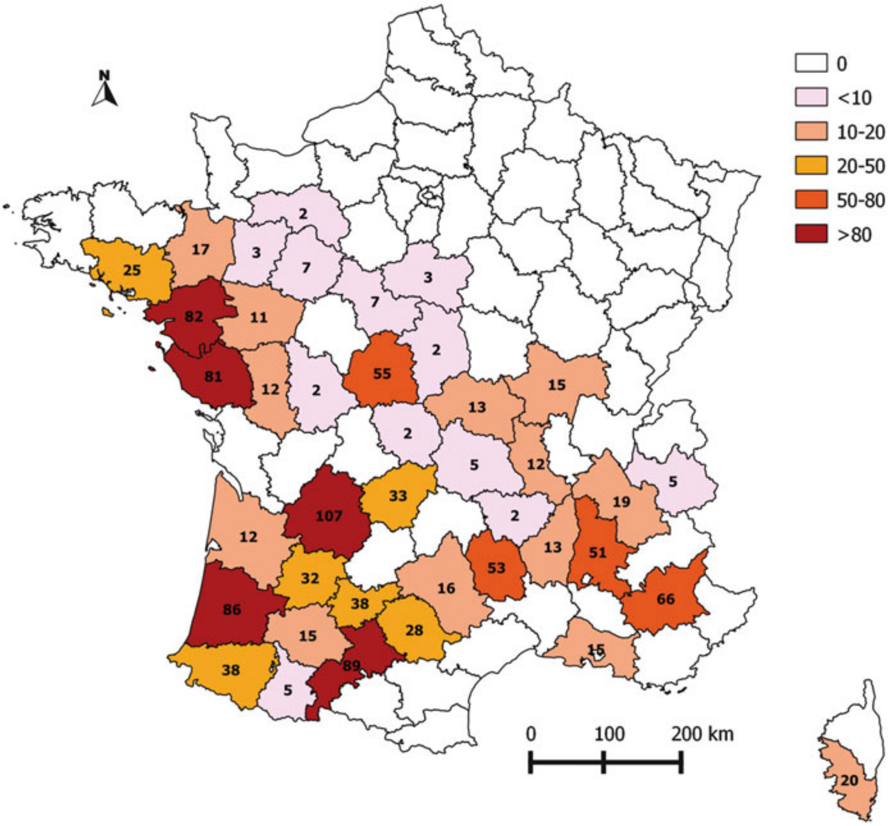


Fig. 8.24 Departmental location and number of envenomation cases by the pine processionary moth in animals

Geographical Location of Concerned Practitioners

For the geographical localization, the practitioners who have seen cases were mainly located in south and west regions of France, which corresponded to other scientific data (and especially consistent with data from the French Department of Health Forestry (DSF)) (Fig. 8.24). The number of cases of envenomation mentioned by veterinarians was higher in the south-west and north-west of France, which corresponds to endemic areas where the processionary moth has been present for several years.

Affected Species

In total, 94 veterinarians have seen at least one case of envenomation between 2005 and 2010 and 1,112 cases have been recorded over the 6 years period, but some respondents admitted that they have not recorded all the cases that they have seen.

Table 8.4 Number of veterinary practitioners who have observed cases of envenomation between 2005 and 2010 and number of affected animals by species

	Cattle	Cats	Horses	Dogs	Small ruminants
Animals	39	106	96	826	45
% Animal group	3.5	9.5	8.6	74.3	4.1
Practitioners	7	15	7	94	4

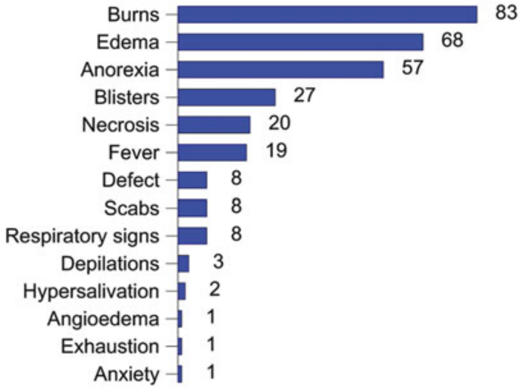


Fig. 8.25 Symptoms of envenomation by processionary larvae in dogs (number of veterinarians who observed the symptoms mentioned in dogs between 2005 and 2010)

Table 8.4 shows the number of cases mentioned in each species and the number of practitioners who have seen cases in each species. All respondents have seen at least one case in dogs, and the cases in this species represent 74 % of all cases. Only few cases have been reported in farm animals (cattle, sheep, and goats): they represent 7.5 % of the total number of cases observed between 2005 and 2010.

When comparing the number of animals affected by report number of concerned practitioners, we noticed that most cases were sporadic in dogs and cattle (about 5–10 cases by practitioner and by year), although some massive envenomation can be reported (for example a practitioner who reported 50 cases of envenomation in horses, or another who faced 30 cases in small ruminants).

Symptoms

- Dogs
  The main symptom of envenomation by the processionary larva in dogs is burning (with vesicles) in 87 % of the cases mentioned (Fig. 8.25). The swelling (edema) of the tongue, lips and sometimes of the face is also a common sign, which is accompanied in some cases, and logically, by lesions on the tongue and anorexia. Lesions could evolve into necrosis, with a possible loss of a part of the tongue. The appearance of "blisters" (term used in the questionnaire) and fever is more random, such as respiratory problems, crusts and depilation. The chamfer was also sometimes reached, with blisters and scabs. Local signs, oral signs, are predominant. No nervous disorder was observed.

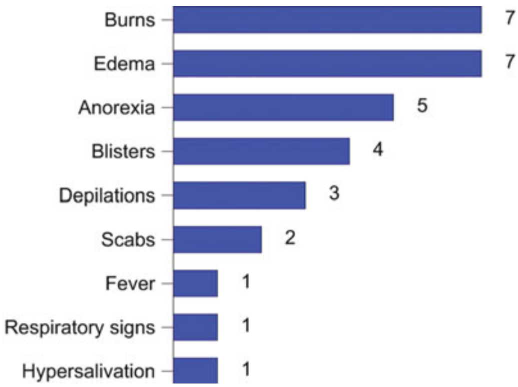


Fig. 8.26 Symptoms of envenomation by the processionary larvae in ruminants (number of veterinarians who observed symptoms mentioned in ruminants between 2005 and 2010)

Treatment must be implemented quickly. Anti-inflammatory, antihistaminic and antibiotics have been used to manage the affection and prevent the potential complications, associated with local care on oral cavity. Some veterinarians have made a fluid therapy and have fixed a nasogastric esophageal catheter to provide needed fluids and nutrition to animals.

Three practitioners have said that envenomation could be fatal for dogs: two veterinarians have notified that euthanasia was necessary and one had observed a death consecutive to the necrosis of the lips and the nose with a loss of substance, despite hospitalization and treatment implemented quickly.

- Ruminants

  The main clinical sign of an envenomation by processionary larvae in cattle is also burning and edema. Then occur other signs, more inconstant: "blisters", edema, depilation, crusts and anorexia. Finally, non- specific signs have been mentioned: fever, respiratory disorders, salivation. As in dogs, nervous symptoms were not observed (Fig. 8.26).

  Affected animals stop eating and separate themselves from the herd. Treatment is also globally similar, and some animals could die.

Differential Diagnosis

Regarding the differential diagnosis, this survey focused only on the possible diagnostic difficulties among farm animals. Indeed, the potential confusion of envenomation with certain regulated diseases is to be considered, because confusion could have serious economic consequences (including commercial constraints, restriction on movement. . .).

Of the 10 practitioners who have seen cases in ruminants from 2005 to 2010 (39 reported cases in cattle and 45 in small-ruminants), five have actually encountered difficulties to establish the diagnosis; the main disease which can be confused with an envenomation by the processionary larvae has been BT. Then were mentioned FMD and malignant catarrhal fever (MCF). Confusion with an irritant

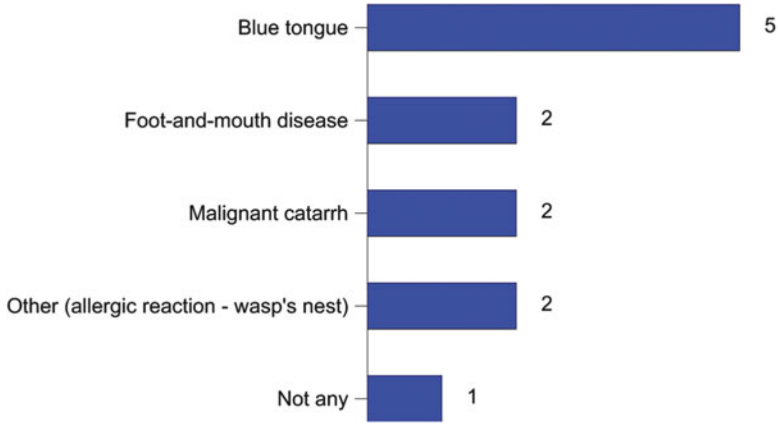


Fig. 8.27 Main diseases in the differential diagnosis of envenomation by the pine processionary moth among farm animals identified in both epidemiological surveys

or allergic reaction from an insect bite (wasp tent for example) had also been cited (named "other" in Fig. 8.27). One practitioner had no difficulty to establish the diagnosis (Fig. 8.27).

According to some practitioners contacted by telephone following investigations, cases of envenomation in ruminants are certainly under-diagnosed, and essentially based on a diagnosis of exclusion. In addition, it seems that farmers are unaware of this affection, especially because the loss of production is minimal and mortality very rare.

In endemic areas, practitioners are very aware of this affection and have no particular difficulty in establishing the diagnosis, thanks to the seasonality of envenomation to avoid confusion with other diseases.

5.3 Data of the National Center of Veterinary Toxicological Information

For the data from the CNITV, they were 116 calls concerning envenomation by larva (whatever the species of larva involved) between 2008 and 2010, including 69 cases considered as certain or probable. 65/69 calls related to dogs, 4/69 cats and 1/69 horses, no appeal having been identified for ruminants. Altogether, 72 cases of envenomation have been reported in dogs, 12 in cats and 1 in horses.

Digestive disorders are the majority, accounting for nearly 57 % of the symptoms mentioned (Fig. 8.28).

Signs could be most extensive, for example with cutaneous disorders when dogs roll in a procession. They are also respiratory difficulties or cardiovascular difficulties like hemorrhage, or anaphylactic shock. Eyes could also be affected, with burns.

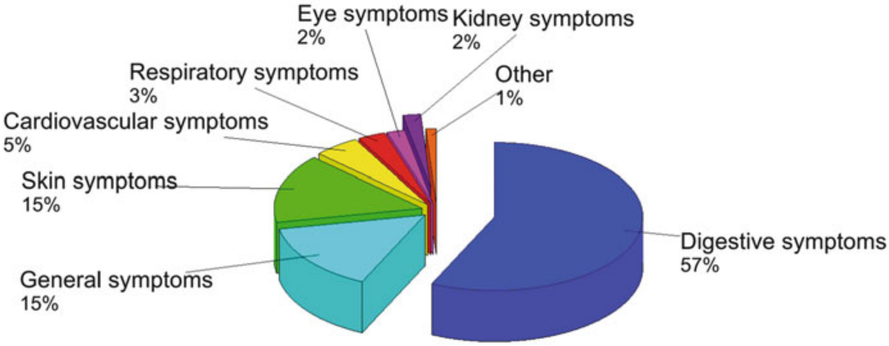


Fig. 8.28 Symptoms of envenomation by pine processionary moth notified in the CNITV between 2008 and 2010

5.4 Discussion

These exploratory surveys show various biases (including practitioners' selection bias because practitioners were contacted via the SNGTV, an NGO composed mainly of rural practitioners) do not allow any generalization of the results obtained at the national level. However, the results and the distribution of the cases match other results and scientific data.

Only 20 % of veterinarians contacted responded to the first survey, and 8 % to the second. If this rate of answer for an anonymous questionnaire by computer is usual (it is even rather pretty good for the first survey), it is however important to note that it is impossible to know why some veterinarians did not respond to these investigations. The assumption that they were not concerned by the problem is obviously not excluded.

Geographical maps with veterinarians who have been confronted with cases of envenomation have been established to evaluate the expansion of pine processionary moth. However, the geographical representativeness can be discussed.

We received in fact some false negative answers, because some practitioners who have observed no cases of envenomation have nevertheless seen cocoons of pine processionary moth and processions around their workplace. Furthermore, some veterinarians admitted that they are unaware of the symptoms although they have observed clinical signs like burns, vesicle, edema of the tongue... and veterinarians are the main applicants of information to the national center for toxicologic information. The main reason of calls to this center is a need of advices for the treatment and for the diagnosis, which highlights the need of information about, among others, pine processionary moth for veterinarians and owners (Fig. 8.29).

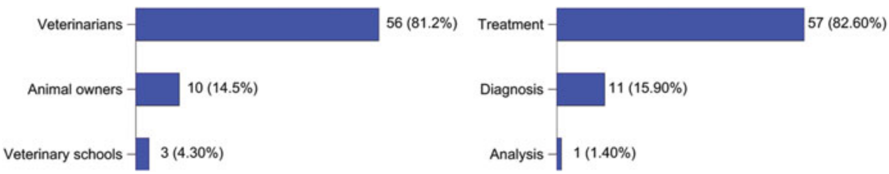
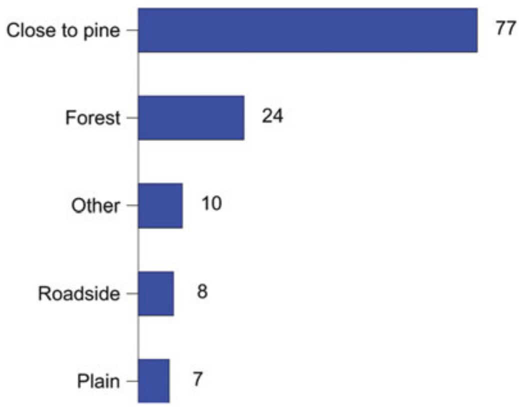


Fig. 8.29 Applicants for information regarding the processionary larvae from the CNITV and reasons calls

Fig. 8.30 Environment of affected animals



Similarly, we have also received some false positive answers: it cannot be excluded that some of the observed envenomation have been caused by the oak processionary larva (*T. processionea*), which also owns the stinging properties from May to July (however, the procession and pupation of the oak processionary are taking place on the tree, and not on the ground; so pets are significantly less frequently in contact with this insect). In fact, the majority of cases had been observed within proximity of pine trees, but some veterinarians did not specify the environment of the animal affected and the specific identification of trees, giving answer like in plain, forest and garden – and one practitioner had evoked a pear tree (Fig. 8.30).

Therefore, the geographical distribution of affected animals does not reflect accurately the geographical evolution of populations of processionary larvae.

Otherwise, the comparison between the two surveys did not highlight a significant evolution in the number of cases of envenomation, or even a particular geographic evolution. Among the 171 practitioners who participated in this study, 12 responded to both questionnaires. Only two veterinarians have reported an occurrence of cases of envenomation in animals of their clinic.

5.5 Conclusions

The pine processionary moth, which continues its geographical expansion inexorably from the south to the North of France in conjunction to climate change, caused a number of envenomation on animals; the intensity and frequency vary according to regions. A thousand cases of envenomation were recorded over the 6 years under study and this affection could also be actually considered as a real problem in animal health, especially for dogs. Envenomation is mainly benign, but the severity and outcome of infected animals may depend on the precocity and effectiveness of the treatment started by the practitioner. However, according to the investigations, some practitioners of still uncolonized areas are unaware of envenomation symptoms, which could prove problematic in the coming years, if the geographical expansion of the pine processionary moth continues at the current rate. Indeed, some mathematical models consider the presence of this insect in Paris in 2025 (Robinet et al. 2007).

We highlighted also the difficulty of diagnosing envenomation in livestock and the importance of the seasonality of the cases as a help for the diagnosis. However, some respondents have observed that this seasonality has been modified by the global warming in recent years, and this could make the diagnosis even more difficult in the future. Due to the geographical extension of the pine processionary moth, populations must be informed of health risks, especially in newly colonized areas.

The realization of two informative leaflets, one for veterinary practitioners and the second for animal owners, would be an interesting way to better promote the dissemination of information. We thus realized, in project, two informative brochures in French to explain the biology and the danger of the pine processionary moth, the clinical signs and the main recommendations to prevent and manage an envenomation for humans and animals. It could be also interesting to make more local epidemiological surveys around clinics really concerned or localized along the front of the geographic areas of the pine processionary moth distribution.

6 URTIRISK, a Software for Assessing the Allergy Risk

Lionel Roques

6.1 Objectives

The software URTIRISK allows the user to observe the evolution of the allergic risk associated with the presence of pine processionary moth, over a year and throughout the French territory. This software is mainly based on the observations of the population range carried out by INRA URZF (UR633).

6.2 Data

The software is based on the following data:

1. Position of the colonization front (observed by INRA URZF): Northern front (2010–2011) and altitudinal front in the Massif Central (2005–2006).
2. IGN map showing the location of pine trees.
3. Chart describing the life cycle of the pine processionary moth and its variations between southern France and the colonization front, as a function of latitude. This chart was obtained by adapting the observations of Huchon and Démolin (1970) to the current geographical distribution of the pine processionary moth.

6.2.1 Computing the Allergy Risk

The allergy risk depends on the time of year, and, of course, on the presence of larvae. The production of urticating setae begins during the larval stage "L3" (Huchon and Démolin 1970). We begin by defining a potential allergy risk R_0 which depends on the latitude and on the time of year, but which is independent on the presence of pine processionary moth. The computation of this potential allergy risk is based on the following assumptions:

(i) There is a time t_0, which corresponds to the beginning of the larval stage L3 and a time t_1 which corresponds to the beginning of the procession. These times depend on the latitude only. They are computed on the basis of the chart in Huchon and Démolin (1970), adapted to the current geographical distribution of the pine processionary moth.
(ii) The duration of the procession is fixed to 30 days.
(iii) Urticating setae are released with a constant rate between t_0 and t_1, and with another (higher) rate during the procession.
(iv) The half-life of the setae is fixed to 10 days in the current version of the software.

Then, we define a function R_1, which depends on the position (latitude, longitude), and roughly describes the distribution of the tents of pine processionary larvae:

(i) We build a map of the potential area of distribution of the pine processionary moth, using only the northern front and the altitudinal front in the Massif Central together with the altitudinal limit of 1,400 m.
(ii) Using the IGN map showing the location of pine trees, we restrict the area of distribution of the pine processionary moth to the locations where pine trees are present.

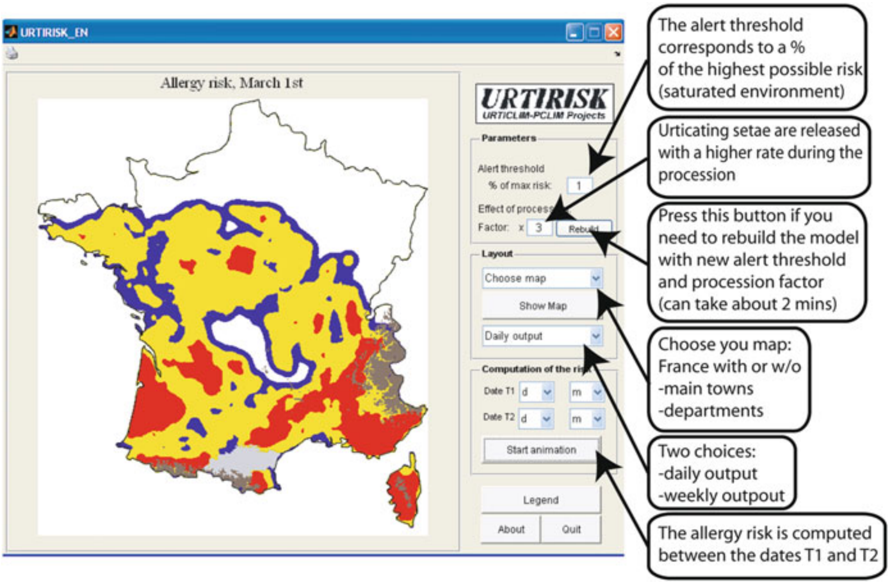


Fig. 8.31 A screenshot from the URTIRISK software

(iii) We use a smoothing kernel to take into account the uncertainty related with the presence of unlisted isolated host trees and pine processionary tents beyond the colonization front.

By overlaying maps R_0 and R_1, we obtain a model for the risk R as a function of position (latitude, longitude) and date.

6.2.2 Outputs

The risk R is normalized (in time and space), so that the maximum risk value is $R = 1$. We define a warning threshold S in terms of the maximum risk. The URTIRISK software produces a map with four risk levels: no risk for $R < S/10$ (in white), low risk to $S/10 \leq R < S$ (blue), medium risk for $S \leq R < 10S$ (in yellow) and very high risk for $R \geq 10S$ (red) (Fig. 8.31).

6.3 Downloading the Software

The Matlab® source code (for Matlab R2011a or later) and executable versions for Windows x32 and x64 of the URTIRISK software are available on the website: http://urtirisk.biosp.org.

7 General Conclusions

Processionary moths constitute an economic problem in the affected countries, but the impact on animal and human health is becoming progressively clear. The fact that an immune response can appear after direct or indirect contact with setae and the high number of proteins present in setae makes research on new antigens/ allergens mandatory. Besides IgE, other type of immune responses should be explored in sensitized people, especially cellular-mediated reactions that could explain the papular lesions frequently observed on affected patients.

Future research on animals and humans will need the development of reliable diagnostic methods. Serological approaches, such as ELISA tests could be very helpful for clinical diagnosis as well as for epidemiological studies. These epidemiological studies will show the expansion of the sensitization to moth allergens and the impact on different animal and human groups, for instance on occupationally exposed workers due to climate change. However, dealing with moth allergens has also an occupational risk for researchers, therefore the production of recombinant allergens for diagnosis should be done in the near future in order to obtain reliable and safe diagnostic tests.

These future new diagnostic methods would finally help in the follow-up of the different biocontrol methods, because a decrease of the moth population would be followed in a short time by a decrease in the number of sensitized humans and animals. Let us hope that this could be reached in a near future!

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