

# Proceedings

## Population Dynamics, Impacts, and Integrated Management of Forest Defoliating Insects <sup>1</sup>

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**Edited by:**

**M.L. McManus**

*USDA Forest Service, Northeastern Research Station, 51 Mill Pond Rd.,  
Hamden, CT 06514-1703, USA*

**A.M. Liebhold**

*USDA Forest Service, Northeastern Research Station, 180 Canfield St.  
Morgantown, WV 26505, USA*

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# Biomass Production of *Pinus pinaster* after Defoliation by the Pine Processionary Moth (*Thaumetopoea pityocampa* Schiff.)

STEPHANOS MARKALAS

University of Thessaloniki, School of Forestry and Natural Environment, Laboratory of Forest Protection, P.O. Box 228, 540 06 Thessaloniki, Greece

**ABSTRACT** Biomass production rate was studied in young (5-year-old) *Pinus pinaster* trees at regular time intervals during the growing season following defoliation caused by the pine processionary moth. The change in shoot length, needle length and needle dry weight was measured for four different types of shoots in moderately defoliated, completely defoliated and undefoliated control trees. In defoliated trees, the shoots were significantly shorter and the needles were shorter and lighter than in the control trees. The percentage losses were also calculated during the total growing season. One year after defoliation, losses in total shoot biomass were 41-50% in moderately defoliated trees and 54-64% in completely defoliated trees.

THE PINE PROCESSIONARY moth (*Thaumetopoea pityocampa* Schiff.) is one of the most harmful insects that attack pine trees in many mediterranean countries (Androic 1957, Serafimovski 1959, Zankov 1960, Kailidis 1962, Rive 1966, Demolin 1969, Bellis and Cavalcaselle 1969, Souleres 1969, Buxton 1983, Geri 1983, Markalas 1985, 1986 and 1989a). Complete defoliation, even during consecutive years, seldom causes the death of attacked trees (Ruperez 1956, Kailidis 1962). More common is the dieback of the crown of young trees, especially in situations where the defoliation of artificially planted trees, growing on unsuitable sites, is followed by unfavorable growth conditions, such as severe drought during the spring months (Markalas 1985). In spite of the capability of defoliated trees to refoliate and survive in the following growing season, the effects of defoliation, especially on the so-called "commercial plantations", are very significant (Fratian 1973).

Most efforts to investigate the effects of defoliation caused by the pine processionary caterpillar measured the losses in growth of attacked pine trees (Joly 1969, Cadahia and Insua 1970, Bouchen and Toth 1971, Lemoine 1977, Markalas 1985, 1987 and 1989b, Avtzis and Chatziphilippidis 1993). Decreased biomass production also results in less vigorous and more stressed trees, which are susceptible to other biotic and abiotic factors. In the present study, we investigated the effects of defoliation on the development of biomass production of young *Pinus pinaster* trees attacked by the pine processionary moth.

## Methods and Materials

The study was conducted in an area forested with *Pinus pinaster* located near the village of Peristerona, about 50 km east of Thessaloniki. At the end of a severe infestation and defoliation (early April) of the 5-year-old trees caused by the pine processionary caterpillar, three groups of trees with different degrees of defoliation were distinguished:

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undefoliated (Control), moderately defoliated (45-55%) and completely defoliated (95-100%) trees. Each group consisted of 25 trees of approximately the same height, diameter and height increment during the last growing season.

The new foliage produced by defoliated and undefoliated trees was measured in four different types of shoots: terminal shoots of the top spondyl, side shoots of the top spondyl, terminal shoots of the second spondyl, and terminal shoots of the third spondyl. Data measurements were initiated in the middle of April and lasted until the end of September. At the beginning, measurements were repeated every 8-10 days, later every 10-11 days and after the middle of July every 13-15 days. In total, 15 measurements were made. Thirteen shoots in every tree was measured on: the terminal shoot and four shoots from each one of the other three shoot types. From the lower part of each shoot, a 2-needle cluster was cut off in order to measure the change in length and dry weight of the needles in the laboratory.

At the end of February of the next year (i.e., one year after the end of defoliation), one side shoot of the top spondyl and one terminal shoot of the second spondyl were cut off from each tree. Every shoot was divided into four equal parts, and from the middle of each part, 10 clusters of needles were cut off. In order to calculate needle biomass production, the average dry weight of the 40 needle clusters of each shoot was used. T-test was for all statistical analysis.

### Results and Discussion

The degree of defoliation of pines caused by *T. pityocampa* is a decisive factor in the development and final length of all four types of shoots measured in this study (Fig.1). The lack of photosynthetic biomass in defoliated trees during the beginning of the growing season did not result in the prolongation of the growing period. On the contrary, it was found that the shoot length increment in defoliated trees was completed earlier than in undefoliated control trees.

The development and final length of new needles were affected not only by the degree of defoliation, but also by the type of shoot as well. The differences in needle length between defoliated and undefoliated trees were always highly significant ( $P < 0.01$ ). However, the differences between moderately and completely defoliated trees were not significant in all shoot types. Specifically, the growth rate of needles of the terminal shoots of the top spondyl was almost the same in both moderately and heavily defoliated trees during the whole growing season (Fig. 2).



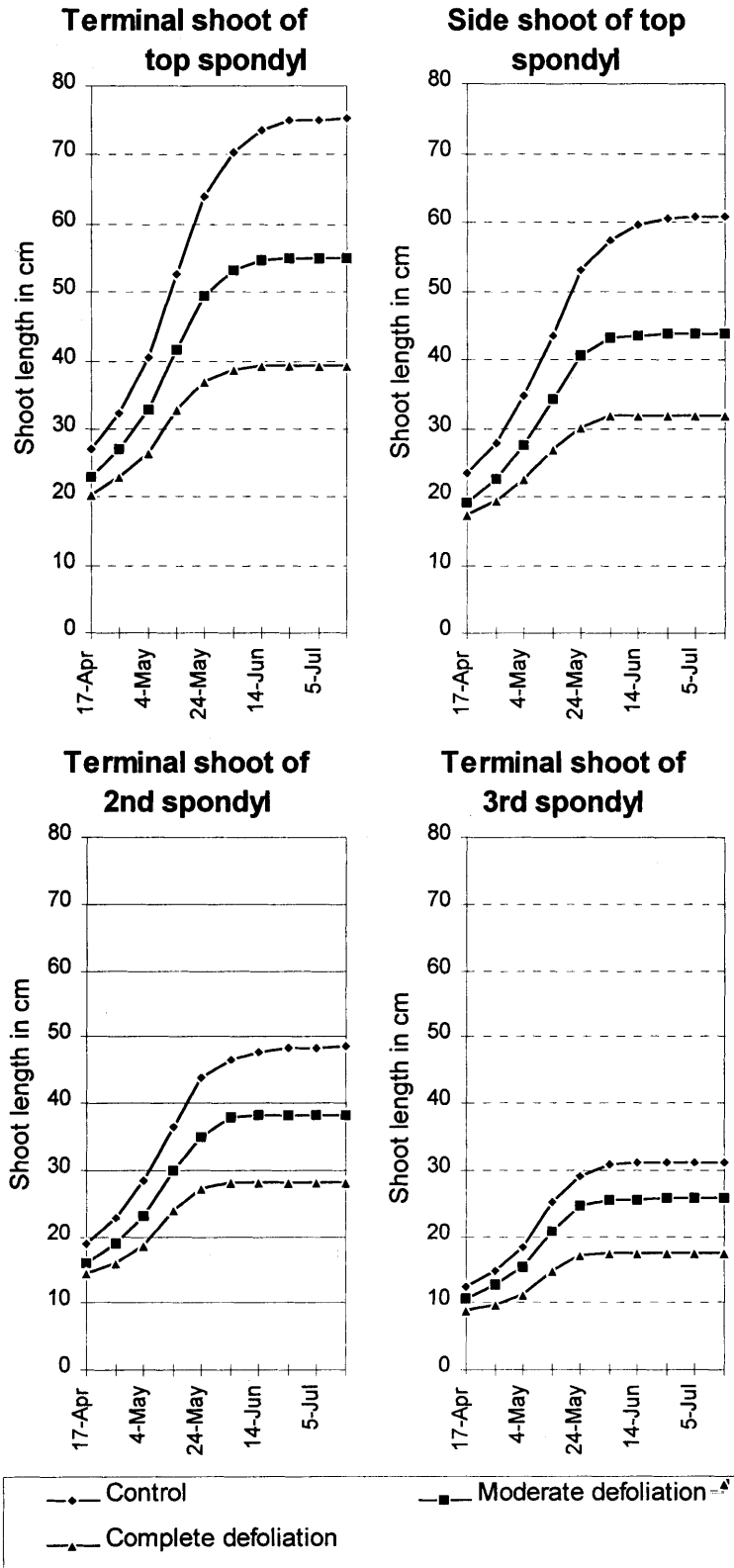


Figure 1. Change in shoot length of *P. pinaster* trees during the first growing season after defoliation by *T. pityocampa*.

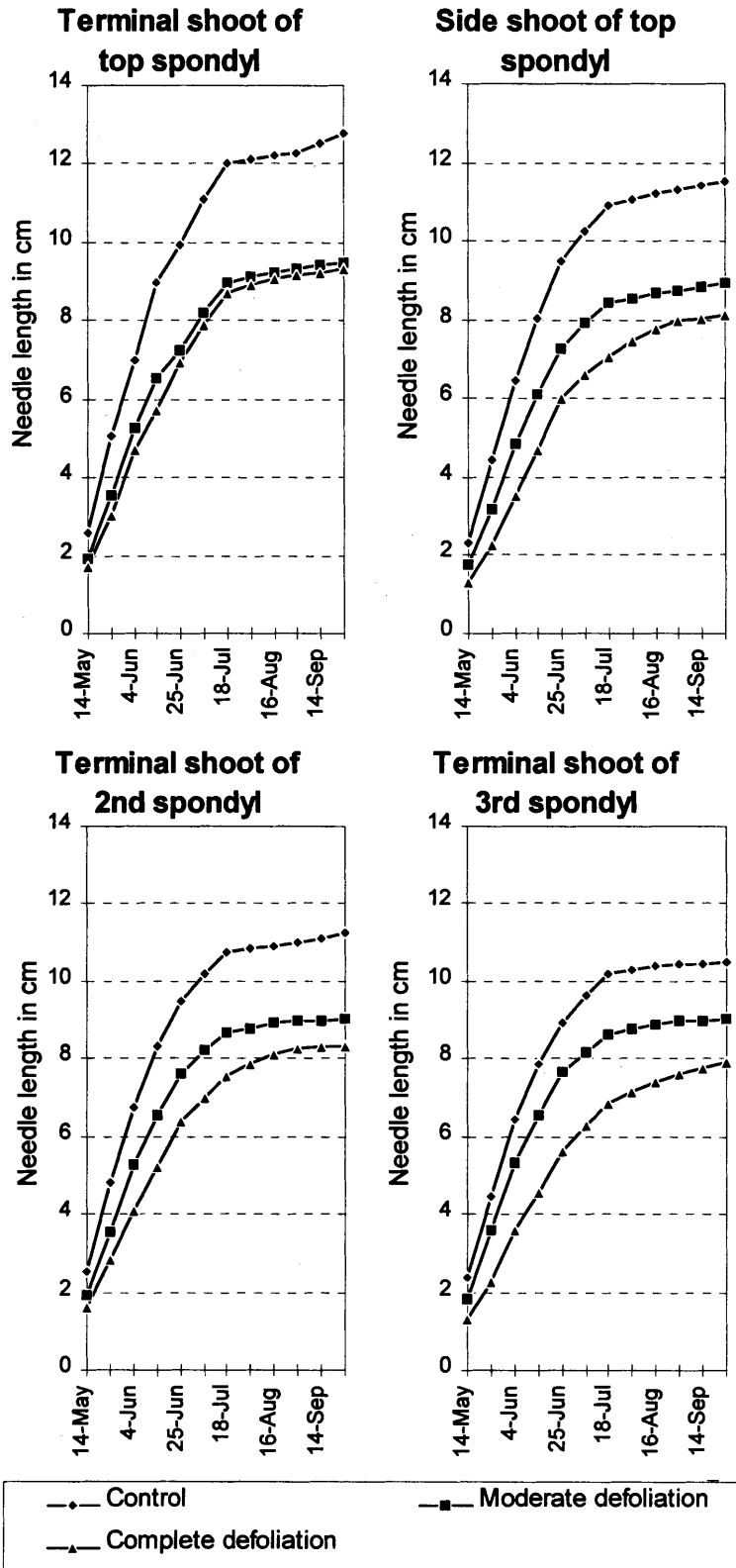


Figure 2. Change in needle length of *P. pinaster* trees during the first growing season after defoliation by *T. pityocampa*.

The increase in dry weight of new needles produced by undefoliated and defoliated trees demonstrated a similar pattern as that previously reported in needle length in almost all cases (Fig. 3). The only remarkable difference was that the increase in the dry weight of needles continued for about 1.5 months after the completion of the length increment.

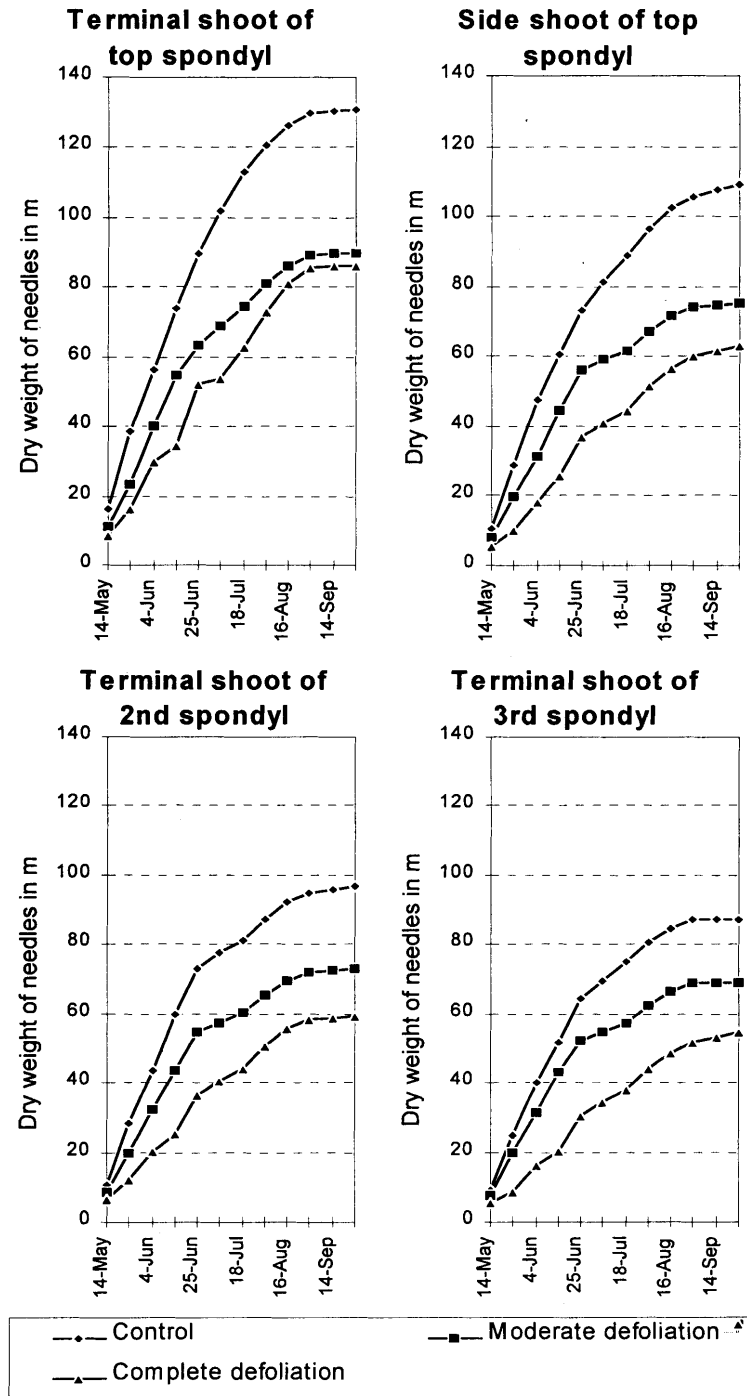
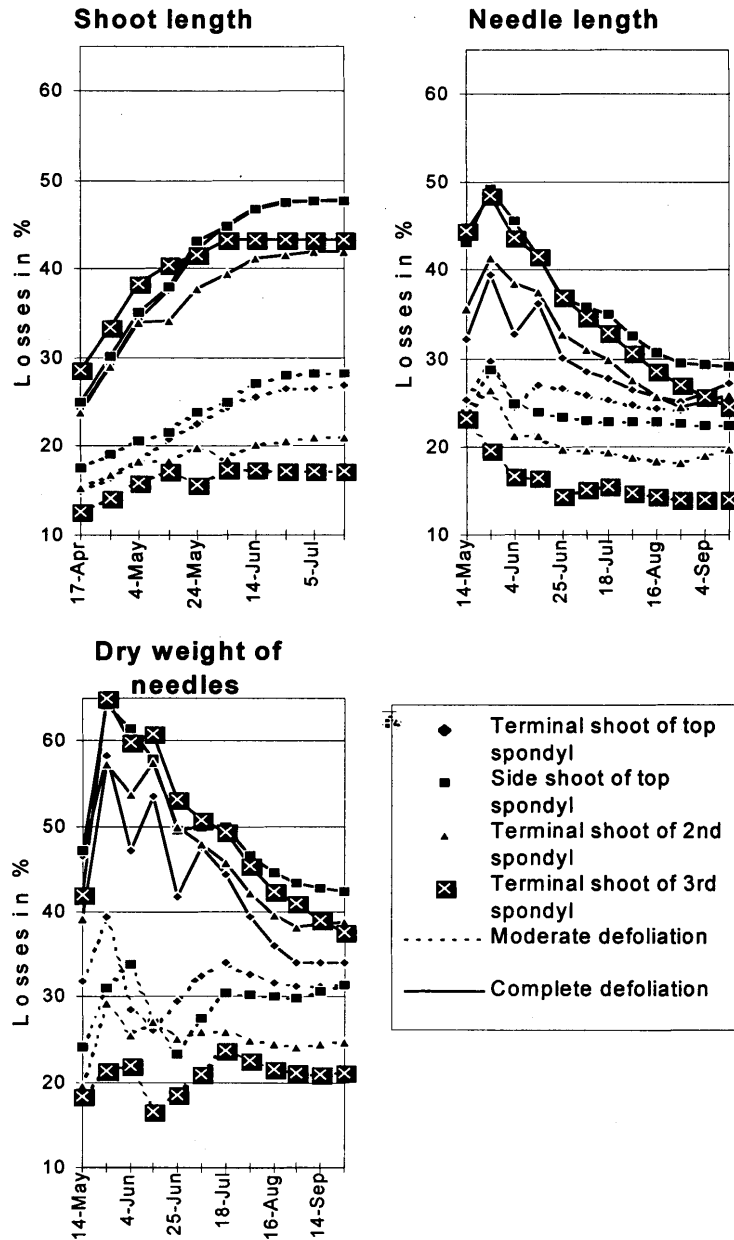


Figure 3. Change in dry weight of a 2-needle cluster in *P. pinaster* trees during the first growing season after defoliation by *T. pityocampa*.

In general, the increment in shoot length was completed in the first half of June, while the increment in needle length (originated always from the lower part of shoots) was completed within the first half of July. The increase in dry weight of needles was prolonged over a longer period of time, even after the end of August. Differences were also found among the three defoliation groups: in moderately defoliated trees, the growth in shoot length, needle length and needle weight was completed a few days earlier than in undefoliated control trees, and the growth of completely defoliated trees was completed earlier than that of moderately defoliated trees.



**Figure 4.** Change in losses in shoot length, needle length and needle weight of *P. pinaster* trees during the first growing season after defoliation by *T. pityocampa*.

The percentage losses in shoot length, needle length and needle weight during the growing season are given in Figure 4. Shoot length losses increased with time for both degrees of defoliation. The losses in needle length and needle weight in the group of moderately defoliated trees were almost invariable, showing only small differences during the growing season, while in the group of completely defoliated trees they were decreasing. The greatest losses were found on the terminal and side shoots of the top spondyl, becoming progressively less as the spondyl's position was near to the ground.

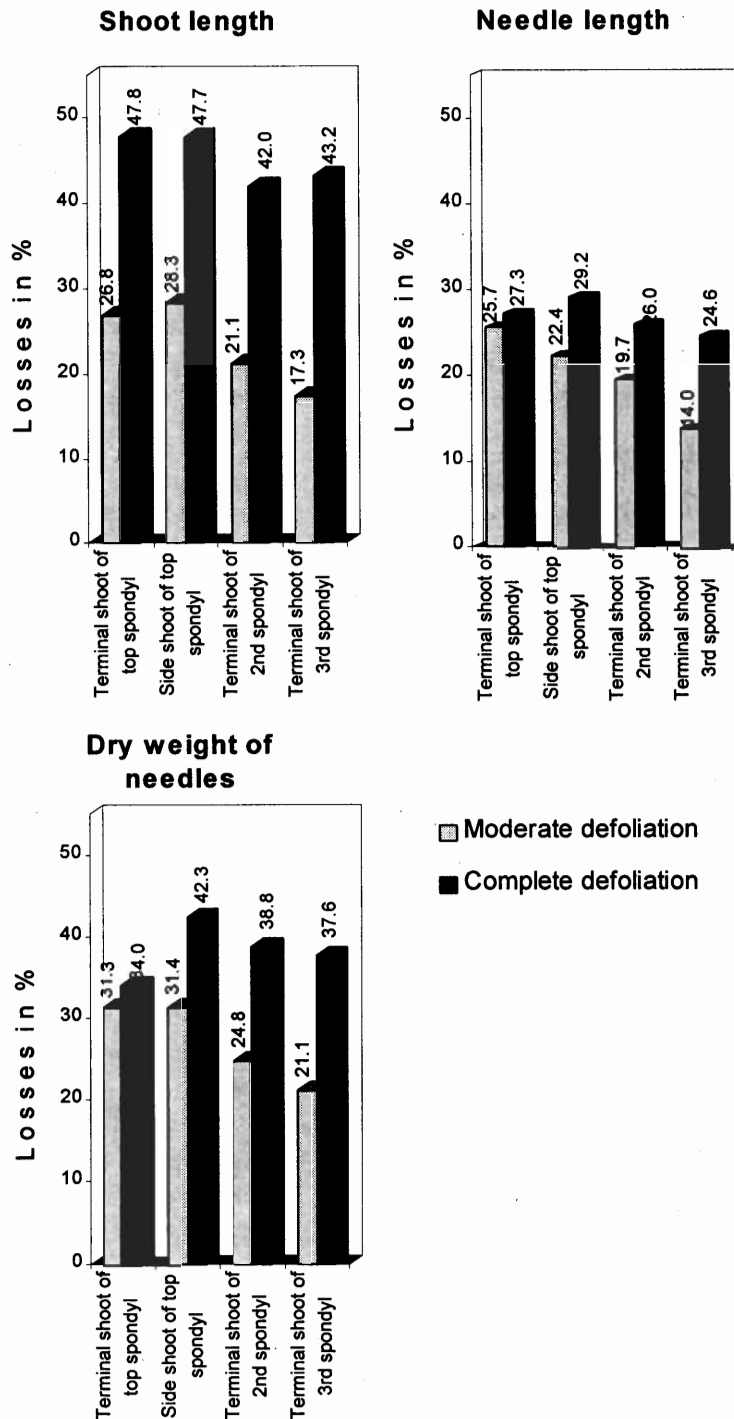
Final losses, measured by the increment of shoot growth, increased to 17.3-26.8% in moderately defoliated trees and to 42.0-47.8% in completely defoliated trees (Fig.5). The 47.8% loss observed after defoliation in the terminal shoot of the top spondyl at the end of the growing season is relatively low compared to the 67.5% and 67.2% losses measured in *P. nigra* and *P. radiata*, respectively (Markalas 1989b). This is because at the end of the defoliation (first days of April), the shoots of *P. pinaster* were already too long. More specifically, when the shoots of the severely defoliated trees were measured for the first time (17 April), they were longer than 50% of their final length.

Final losses in needle length were less than those in shoot length, increasing to 14.0-25.7% in moderately defoliated trees and 24.6-29.2% in heavily defoliated trees (Fig. 5). The losses in dry weight of needles reached 21.1-31.4% in the group of moderately defoliated trees and 34.0-42.3% in the group of completely defoliated trees. Analyzing the final losses in needle length and needle weight, we see that the differences between the two degrees of defoliation were relatively low in the terminal shoot of the top spondyl and greater in the other shoot types.

The results of the last measurement are presented in Table 1, which took place at the end of February of the next year. As expected, the number of needle clusters per shoot was significantly greater in undefoliated control trees than in defoliated trees, since this depends mainly on the shoot length. Interestingly enough, the number of needle clusters per unit length of shoot (10 cm) demonstrated an opposite trend, increasing with the degree of defoliation. Thus, the side shoot of the top spondyl contained 55.1 clusters in the control trees and 97.4 in the completely defoliated trees, while in the terminal shoot of the second spondyl the corresponding numbers were 58.3 and 103, respectively (Table 1).

Taking into account the data in Table 1, we see that in the group of moderately defoliated trees, the needles were 24.7-26.1% shorter and 31.8-33.2% lighter, while in the group of completely defoliated trees, they were 27.4-27.8% shorter and 43.5-45.8% lighter.

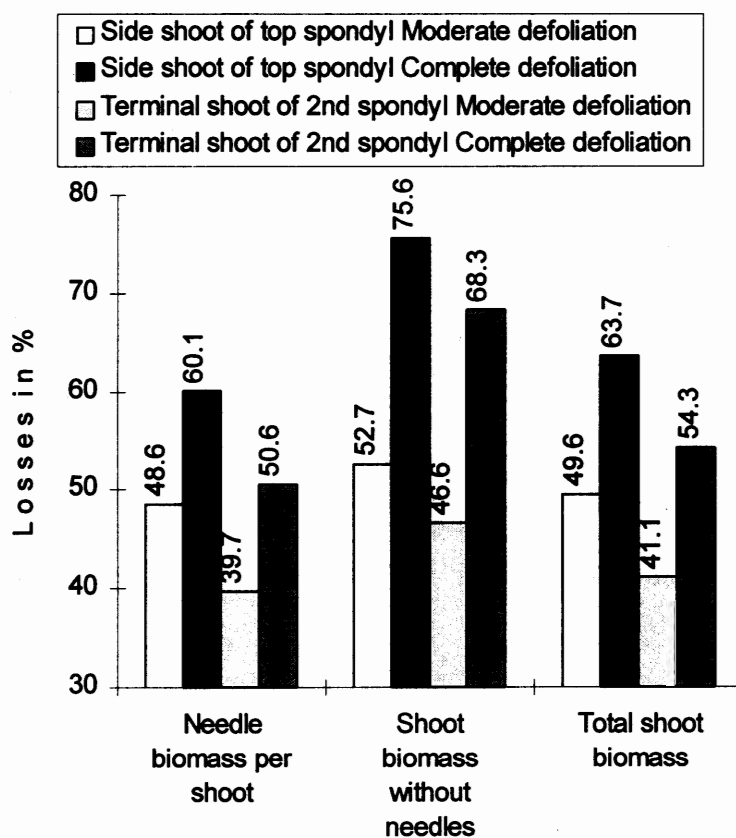
The biomass of new shoots produced by pine trees during a growing season is comprised of needle biomass and the biomass of the woody stem of the shoots. The differences found between defoliated and control trees were greater in biomass of the woody stem of the shoot (shoot without needles) than in the biomass of needles (Table 1). Thus, the percentage losses in needle biomass were 39.7-48.6% in moderately defoliated trees and 50.6-60.1% in heavily defoliated trees, but for the woody stem of the shoot the losses were 46.6-52.7% and 68.3-75.6%, respectively (Fig. 6). The losses in total shoot biomass were found to be 41.1-49.6% in moderately defoliated trees and 54.3-63.7% in the completely defoliated trees. In all cases, losses were greater in the terminal shoots of the second spondyl than in the side shoots of the top spondyl.



**Figure 5. Final losses in shoot length, needle length and needle weight of *P. pinaster* trees at the end of the first growing season after defoliation by *T. pityocampa*.**

**Table 1. Refoliation and biomass production of *P. pinaster* trees measured one year after defoliation by *T. pityocampa***

Parameter	Side shoot of top spondyl			Terminal shoot of 2nd spondyl		
	Control	Moderate defoliation	Complete defoliation	Control	Moderate defoliation	Complete defoliation
Total shoot length (cm)	64.2	45.5	31.4	51.9	37.4	27.4
Shoot length carrying needles (cm)	60.6	39.4	26.5	50.2	35.3	25.8
Average needle length (cm)	11.7	8.8	8.5	11.5	8.5	8.3
Number of needle clusters per shoot	334.5	269.0	246.3	291.6	256.7	255.7
Needle clusters per 10 cm shoot length	55.1	67.9	97.4	58.3	73.0	103.0
Average needle cluster weight (mg)	147.1	98.3	79.8	134.0	91.4	75.7
Needle biomass per shoot (gr)	50.26	25.84	20.07	39.64	23.92	19.60
Shoot biomass without needles (gr)	15.42	7.29	3.76	10.69	5.71	3.39
Total biomass per shoot (gr)	65.68	33.13	23.83	50.33	29.63	22.99



**Figure 6. Percentage losses in biomass production of *P. pinaster* trees measured one year after their defoliation by *T. pityocampa*.**

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# Crown Fauna of Birch Stand Caterpillars in a Pollution Area and their Population Dynamics

EMANUEL KULA

Faculty of Forestry and Wood Technology, Zemedelská 3, CZ 613 00 Brno, The Czech Republic

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**ABSTRACT** In a forested area affected by air pollution in FD Snezník (north Bohemia), the fauna of butterfly caterpillars in birch crowns was sampled by shaking foliage. Among 6,068 individuals collected, 119 Lepidoptera species were identified; the most dominant species were those with high outbreak potential: *Cabera pusaria*, *Operophtera fagata* and *Coleophora serratella*. Changes in population dynamics significantly influenced which species was dominant. An apparent 3-to 4-year population density cycle was observed for *C. pusaria*. Two periods of increased caterpillar occurrence in birch crowns were observed each growing season: a spring peak with culmination at the end of May and June, and a summer peak with culmination in mid August.

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FROM THE EXISTING spectrum of more than 300 phytophagous insect species developing on birch, the majority of representatives are in the order *Lepidoptera*. Free living or mining caterpillars first damage leaves, then buds and catkins. Caterpillar damage is minimized due to a high birch regeneration capacity, weather conditions and a natural enemy complex.

The crown fauna of birch was studied by Gninenko (1974), Kozlov (1984), Kolomic and Artamonov (1985) and Kutenkova (1986). *Epirrita autumnata* (Eidmann 1964), *Erannis defoliaria* (Badalík 1988), *Eriocrania* sp. (Koricheva 1994) and *Coleophora serratella* (Kula and Vaca 1995) are considered serious birch pests.

Kula (1995a) used light traps to sample in the same area as we report upon here and collected a total of 861 butterfly species of which 123 feed upon birch. The goal of this paper is to evaluate the caterpillar fauna of birch crown: their species and numerical representation, phenology, and seasonal and population dynamics.

## Materials and Methods

From 1986 to 1995, we collected 6,068 birch caterpillars by shaking crowns. Butterfly fauna in the caterpillar stage were studied in six birch stands of Forest District (FD) Snezník by shaking caterpillars onto 2 x 2 m linen squares. Caterpillar control in birch crowns was carried out in every locality on 5 trees at 14-day intervals throughout the entire vegetative season (mid-April until the end of October). Different sample trees were treated between successive control applications so that the stand, not only sample trees, could be monitored. Collected caterpillars were preserved in 75% ethyl alcohol.

### Description of the Study Area

The experiment was carried out in Forest District (FD) Snezník (north Bohemia) between 450 and 600 m in altitude. The area is characterized by rough topography, mountainous climate with an average annual temperature of 6° C, a total annual precipitation of 800 mm, a vegetative season of 110 to 120 days and a long-term influence of enhanced SO<sub>2</sub> concentrations. Forest stands are situated in the area of maximum (A) and heavy (B) air-pollution endangerment, in complex of forest types (6K) acidic spruce beechwood and is characteristic of substantial weed infestation with dominant representation of *Calamagrostis villosa* (Chaix.) Gmel., *Avenella flexuosa* (L.) Pirl. (Table 1).

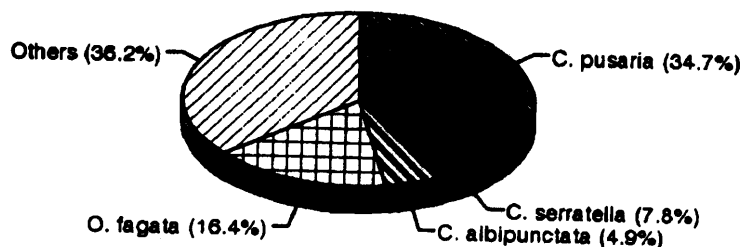
**Table 1. Characteristics of stands under study**

Locality	Birch representation (%)	Year of Forestation	Aspect	Altitude (m)	Degree of pollutant load	Forest types composition	Weed infestation (%)
Vlcák	100	1980	NW	450	B	6K4	100
K. Hrádek	100	1980	N	500	A	6K4	100
Snezník	60	1979	S	560	B	6K1	90
Tisá	100	1980	plain	600	A	6K4	100
Ostrov	100	1979	N	550	A	6K8	50
Letadlo	70	1983	SE	450	B	6K4	75

### Results and Discussion

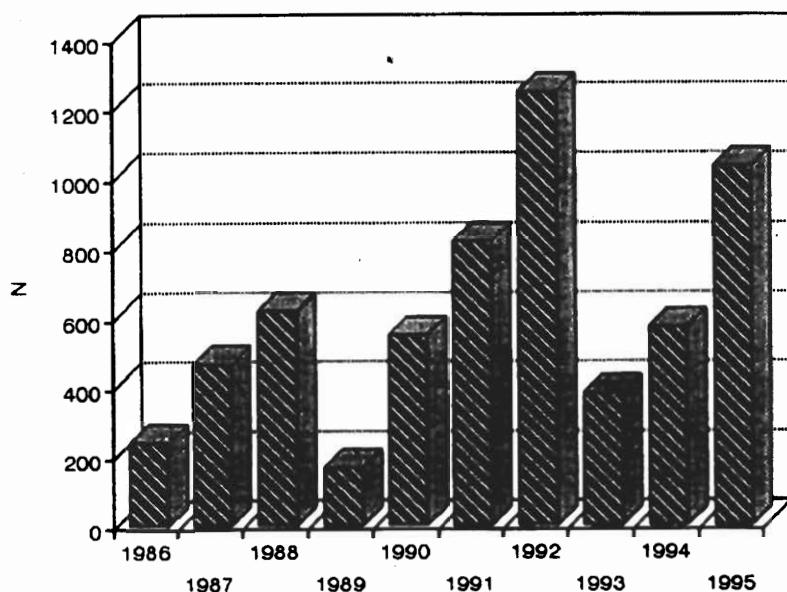
We collected 6,068 caterpillars belonging to 119 species of *Lepidoptera*. The majority of individuals were collected in birch stands within the K. Hrádek and Vlcák localities over 10 years using the shake down method.

Dominant species included *Cabera pusaria* (34.7%), *Coleophora serratella* (7.8%) and *Operophtera fagata* (16.4%); subdominant species included *Cyclophora albipunctata* (4.9%) and *Teleoides proximellus* (2.6%). *Biston betularius* (1.5%), *Operophtera brumata* (1.3%) and *Orthotaenia undulana* (1.3%) were classified as receding, and the 111 remaining species were classified as subreceding (< 1%) (Fig. 1).



**Figure 1. Dominant crown caterpillar species of birch stands (FD Snezník, 1986-1995).**

Over the 10 years of the study, three peaks of caterpillar abundance were recorded (1988, 1992, 1995), followed by a significant decrease. It is not yet known if there is a 3- or 4- year gradation phase between peaks of population density (Fig. 2).



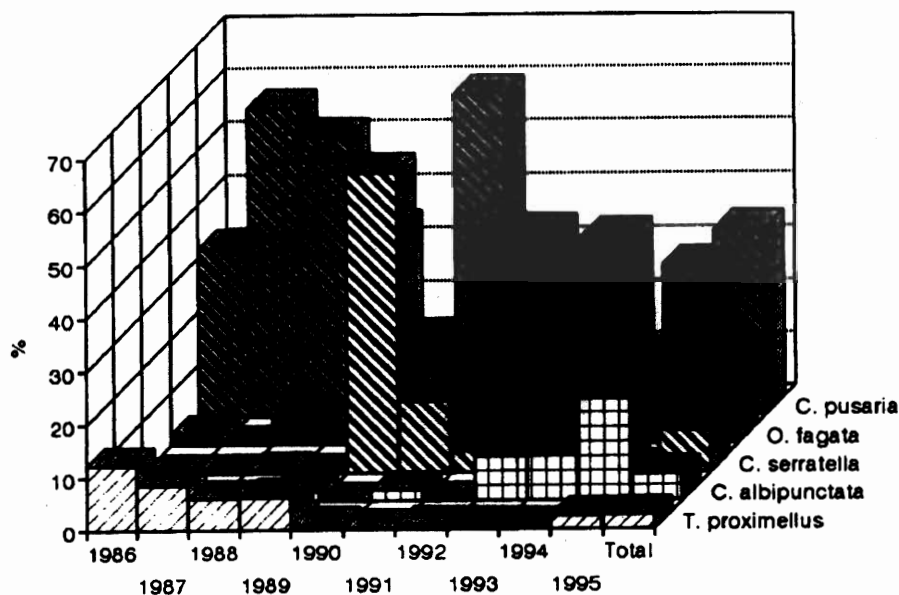
**Figure 2. Dominance of caterpillars in the crown fauna of birch stands (FD Snezník, 1986-1995).**

The seasonal and population dynamics of the crown fauna varied substantially among eudominant and dominant caterpillar species. The decline of *C. pusaria* in 1990 was substituted by local outbreaks of *C. serratella*, and its decreased dominance in 1991-1994 was offset by increases in *O. fagata* and partially by *C. albipunctata*. Subdominant species usually had one population peak in the course of 10 years (5-8%): *O. brumata* (1988), *Paradiarsia similaria* (1989), *Semiothisa notata* (1992) and *O. undulana* (1993).

Eudominant species *C. pusaria* changed dominance in the following pattern: a peak occurred in 1987 (57.9%); after three successive years of decline to 14.6% in 1990, a second peak occurred (60.6%) in 1991, followed by a significant and rapid decrease of dominance (34.4% and 8.9%) in 1992 and 1993, respectively. In 1995, this species had a partial increase of dominance (28.2%). When comparing absolute values of caterpillars shaken down from the same number of trees, a significant population trend appeared, characterized by 2-year peaks in an almost regular, 3-year cycle in 1987-1988, 1991-1992 and 1995 (268 x 329, 494 x 428 pieces, 292 caterpillars).

*C. pusaria* was the most significant species of the study area and reached a eudominant position in all localities. Looper *O. fagata* was a eudominant species from 1986 to 1995 with relatively low dominance from 1986 to 1991 (0 - 7.8%). In 1992, we recorded a peak in this species (31.2%) with a mild decrease the following year. The next peak during the 10-year period occurred in 1994 (39.1%). Evaluation of population dynamics from the absolute number of caterpillars caught showed that since 1989, abundance of this caterpillar species in birch crowns was gradually increasing with a peak in 1992 (389 caterpillars)

followed by a decrease. The density of this species remained 2-4 times higher than the density recorded one year before residual (Fig. 3).

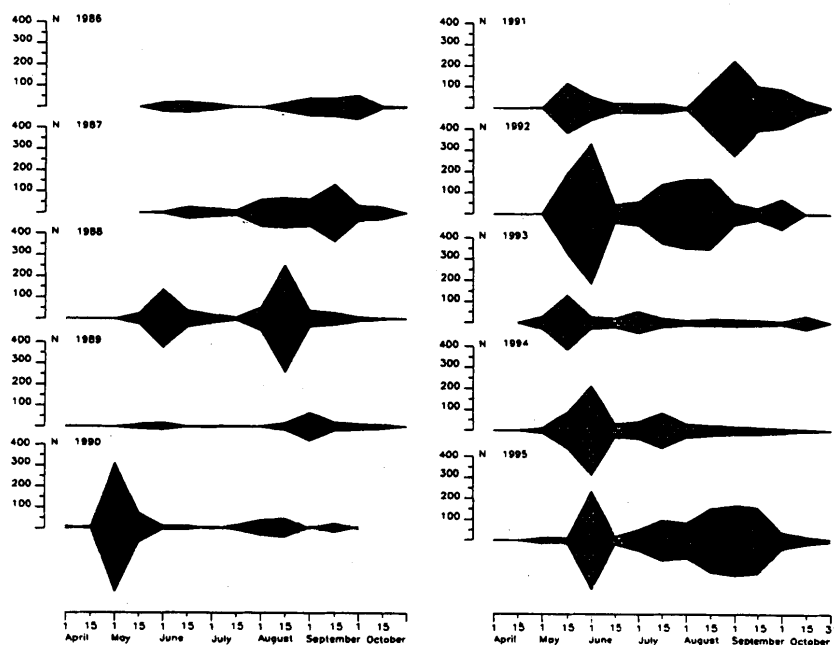


**Figure 3. Population dynamics of caterpillars in the crown fauna of birch stands (FD Sneznik, 1986-1995).**

*C. serratella*, a dominant species, showed a sudden peak in 1990. It was not collected in birch crowns from 1986 to 1989. This peak was followed by a sudden decrease of dominance in 1991 (13.4%). The absolute values of shaken caterpillars show that this peak can be characterized in the same way as *C. pusaria* (Fig. 3). A higher occurrence of this species occurred in only three localities.

*C. albipunctata* was a subdominant species, but from 1986 to 1992, we did not collect specimens of this species in birch crowns. In 1993, it appeared as a dominant species. Its representation continually grew to 19.1% (1995); peak absolute values were recorded as well (Fig. 3). *B. betularius*, a receding species, showed long-term, well-balanced dominance within the crown fauna with a slight increase recorded in 1986, 1989 and 1995. *O. undulana* was not found in birch crowns until after 1990; its dominance gradually increased to 7.5% (1994). *T. proximellus* was a dominant species, its dominance gradually decreasing from 1986 (12.2%) to 1991 (0.1%) and then slightly increasing until 1995 (2.4%) (Fig. 3).

During the 10-year period, the dynamics and phenology of the complex butterfly fauna in the caterpillar stage formed two significantly separated stages in FD Sneznik. The spring period, which culminated with a peak at the end of May and June, occurred from the beginning of May until mid June. The second caterpillar wave started in mid June and culminated in mid August. The period between the spring and late summer waves of caterpillars was characterized by low, well-balanced caterpillar representation. From phenological point of view, caterpillars were on birch trees from the beginning of leaf flush (last half of April) until the end of the vegetative period at the end of October (Fig. 4).



**Figure 4. Phenology and seasonal dynamics of crown caterpillars of birch stands (FD Sneznik, 1986-1995).**

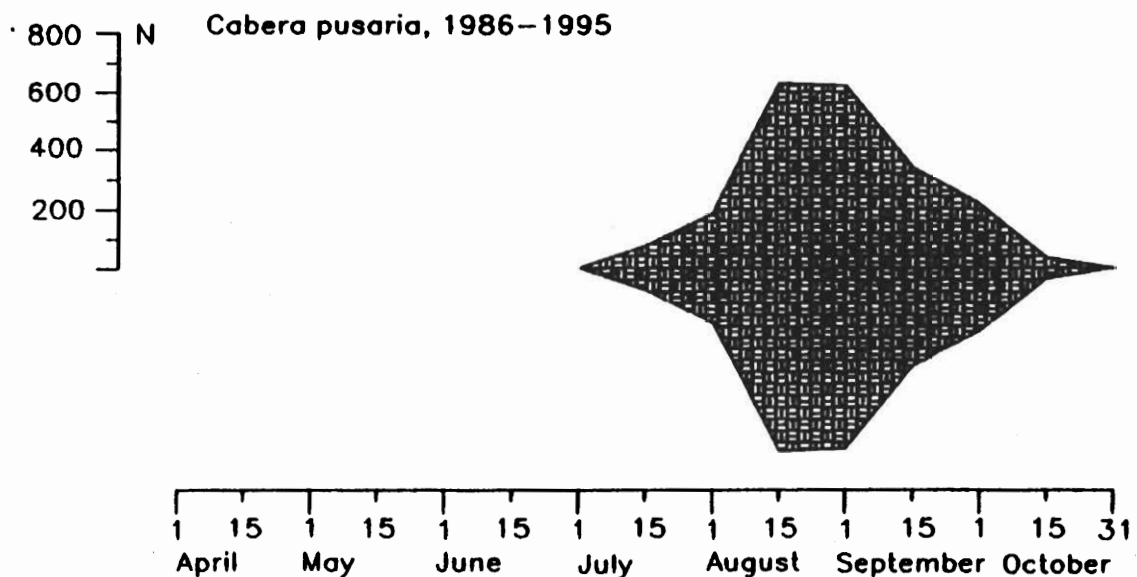
*O. fagata* which occurred in birch crowns from the beginning of May, was among the important spring species. Its occurrence peaked at the beginning of June and its development was finished by the end of June. The looper, *O. brumata*, occurred at the same time, but it did not reach as high a level of population density.

*O. undulana* caterpillars were active in mid April with a peak in mid May and sporadic presence in samples at the beginning of June. *Pandemis cerasana*, another spring species, started feeding on birch at the beginning of May; its peak population density occurred at the beginning of June, and a month later it disappeared from birch crowns.

One summer representative is *P. similaria*, which occurred from mid June to mid October with estimated peak abundance at the beginning of August and probable second generation in September. *T. proximellus* was collected from mid July until the end of October, with peak abundance occurring in mid August. Two quite evident generations characterized *C. albipunctata*, which was collected in tree crowns from mid June until the end of October with peaks in mid July and mid September. The occurrence of the second caterpillar generation was recorded in mid August. Two generations with a relatively low caterpillar abundance on birch was characteristic of *S. notata*, which appeared in crowns at the beginning of June and remained until the end of September. The first generation peaked in mid July; the second generation appeared in the second half of August and peaked in mid September.

*C. pusaria* was a late summer species with only one generation. The caterpillars hatched at the beginning of July, a peak lasted from mid August until the beginning of September, and caterpillars were found in birch crowns even at the end of October (Fig. 5).

Phenology and seasonal population density of *C. pusaria* in the 10-year study period showed significant variation in population dynamics, with peaks always occurring after two-year decreasing population density. The timing of caterpillar occurrence from July to October varied among years. In 1992, caterpillars were already developing on birch trees in June. In 1987, 1991, 1993, 1994 and 1995, caterpillars were in the crown at the beginning of July. From mid August in 1990 and from the beginning of August in 1987 and 1989, we found caterpillars of this species in birch crowns.



**Figure 5. Phenology and seasonal dynamics of *Cabera pusaria* caterpillars (FD Snezník, 1986-1995).**

Despite the collection of 119 caterpillar species from birch crowns only a few of these are significant due to their severity of damage (*O. fagata*, *C. pusaria* and others). A substantial number of species developing in birch crowns do not endanger this tree species; these species often occurred only in limited numbers during the 10-year period. The results discussed above are in agreement with the data of Kutenkova (1986).

Wiackowski et al. (1976) collected 401 species of birch crown fauna, 73.5% of which were made up of phytophagous species and 67 species of butterflies. The highest density of caterpillars, *Macrolepidoptera*, appeared in the study area less affected by air pollution. Under these conditions, *O. fagata* outbreaks appeared.

The 119 species of caterpillars which we collected ranged from butterflies to the most significant forest pests of birch. An outbreak of *Erannis defoliaria* occurred in birch stands in the first half of the 1980s (Badalik 1988) and a casebearer (*C. serratella*) outbreak center appeared in FD Snezník (Kula and Vaca 1995). While *E. defoliaria* showed slight

representation, *O. fagata* had a high population density accompanied by severe damage of birch crowns. Because this species is widely polyphagous, other broadleaved trees in forest stands in polluted areas are endangered. *O. undulana*, which also attacked broadleaved woody plants, also occurred at high population densities. Information about its outbreak potential is lacking. The casebearer *C. serratella*, which prefers birch, is also a monophagous species. Its local outbreak occurred from 1990 to 1991 (Kula and Vaca 1995). Outbreaks of this species are common in Canada (Raske 1976). This species appeared in the entire Krušné hory mountain area (Kula 1995b).

The occurrence of eudominant and dominant species and their joint seasonal dynamics may be significantly influenced by weather. With the looper *O. fagata*, a 10-year cycle of enhanced population density may occur. If an outbreak appeared at the beginning of the 1980s, it may last until the beginning of the 1990s.

According to Wiackowski et al. (1976), crown fauna and their abundance and species composition can be influenced by the degree of air-pollution. For instance, moths of the species *Acleris ferrugana*, *T. proximellus*, *Hedya atropunctata* and *Spilonota ocellana* occurred more often in moderately polluted localities. The dominant position of *O. fagata*, *Achlya flavicornis* and *Coleophora fuscedinella* was closely associated with the severely polluted area and their occurrence decreased with a decreasing degree of pollution. On the contrary, miners of *Eriocrania* sp., *Heliozela betulae* and *Lyonetia clerkella* preferred a clean environment. Caterpillars of *Lithocolletis* sp. and *Incurvaria* sp. seemed unaffected by pollution levels.

Study areas in FD Snezník are part of a larger area which has been affected by air pollution over a long time. Individual localities do not have very different air pollution histories so they could not be evaluated within an air pollution gradient. Our *C. serratella* outbreak data may support the results of Wiackowski et al. (1976), but on the other hand, *O. brumata* reacted population density in the same conditions.

### Conclusions

In the crown fauna of birch stands, we identified 119 caterpillar species (6,068 individuals). Species classified as eudominant included *C. pusaria*, *C. serratella* and *O. fagata*; dominant species included *C. albipunctata* and *T. proximellus*; and subdominant species included *B. betularius*, *O. brumata* and *O. undulana*. One hundred and eleven species were classified as subreceding.

Evaluation of caterpillar abundance over 10 years suggested that a 3- to 4-year outbreak pattern of crown dwelling butterfly caterpillars exists (1988, 1992 and 1995 were outbreak years) which was decidedly influenced by eudominant and dominant species. These species changed their dominance within the outbreak cycle. *C. pusaria* exhibited a population cycle characterized by a 2-year outbreak every 3 years. *O. fagata* populations appeared to peak every 10 years. Outbreaks of *C. serratella* appeared to be as short-lived.

The birch crown fauna caterpillar complex exhibited two distinct periods within each growing season. The spring peak culminated at the end of May and June, and the summer population reached its peak in mid August. The emergence of spring and summer caterpillar populations, as well as their culmination and duration, were heavily influenced by weather conditions in individual years. *C. pusaria*, *C. serratella* and *O. fagata* were among the most



significant butterfly species with outbreak potential in birch stands. Species identified as pests that occur in the caterpillar complex, but have not yet had outbreaks, include *Agriopis aurantiaria*, *Alcis repandata*, *Archiearis parthemias*, *Biston betularius*, *Campaea margaritata*, *Colotois pennaria*, *Cyclophora albipunctata*, *Ochropacha duplaris*, *Orthotaenia undulana*, *Pandemis cerasana*, *Semiothisa notata* and *T. proximellus*.

### Acknowledgements

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# The Use of *Bacillus thuringiensis* Against *Thaumetopoea pityocampa* Schiff. (Lepidoptera:Thaumetopoeidae) in Greece

NIKOLAOS D. AVTZIS

Technological Education Institute (TEI) of Kavala, Dep. of Forestry in Drama, Lab. of Forest Entomology, 66  
100 Drama-GREECE

**ABSTRACT** In the pine forests of Greece one of the most common needle-eating insects is *Thaumetopoea pityocampa* Schiff. It attacks all pine species at different levels of intensity and occurs almost everywhere, from sea level to an altitude of 1800 meters.

In the past, *T. Pityocampa* was controlled efficiently by using chemicals such as DDT, BHC, Malathion, Systox and Metasystox. In 1965, *Bacillus thuringiensis* was used against *T. pityocampa* in Greece for the first time by Kailidis. The formulations that were used during that period for aerial application were Thuricide, H.P. (15.8 billion I.U./Kg), Dipel (16.000 I.U./mg) and Bactospeine (16.000 I.U./mg) at a dose of 700-750 g in 50 l of water/ha. In the 1980's pesticides such as Dimilin, which are chitin inhibitors, were preferred over Bt for control of *T. pityocampa*.

More recently, however, improvements in *B. thuringiensis* formulations, such as their improved stability at a range of temperatures, improved deposit characteristics and environmental safety, have led to the renewed widespread use of Bt products against *T. pityocampa*.

In an effort to confirm the improvements of the new Bt product, Foray 48B, an experiment was conducted using three pesticides: Foray 48B, Dimilin 25wp, and Bactospeine wp (16.000 IUAK/mg) in 10 to 14-year-old plantations of *Pinus brutia*. All three products that were used, under the particular climatic conditions, provided good foliage protection and a high level of mortality (96.1-100%).

IN GREECE, CONIFEROUS forests occupy over 3,359,186 ha, and 25.4% of this land is categorized as commercial forest. The annual increment is estimated to be 1 m<sup>3</sup>/ha. In the pine forests, the most significant species are *Pinus halepensis*, *Pinus brutia*, *Pinus nigra* and *Pinus silvestris*, which cover about 870,378 ha (60.9%) of the coniferous forests (Eleftheriadsis 1996).

In Greek pine forests, one of the most common insects is the needle-eating *T. pityocampa*, the processionary pine caterpillar. It appears in all pine species at different population intensities (Avtzis 1983c, 1986, Schopf and Avtzis 1987) and occurs almost everywhere in Greece from sea level to 1800 meters altitude on Mt. Olympus. It is absent in only a few areas of Central Greece (e.g. in Karpenisi) because of the unsuitable weather conditions as well as on some islands of the Aegean Sea, possibly because of their geographical isolation (Avtzis 1983b). The problems caused by this insect can be placed into three categories:

- a. Health problems to humans (e.g. allergenicity).
- b. Aesthetic problems (nests on the trees, defoliation etc.).
- c. Economic problems due to the loss of growth caused by the defoliation.

Because of these effects, individually or collectively, there is a need to control this harmful insect and to minimize its damage. The control of *T. pityocampa* in Greece has passed through many stages, following progress in development of chemical pesticides and then concerns about ecology and the benefits of more environmentally safe approaches in recent years.

Initially, *T. pityocampa* was controlled efficiently by using chemicals such as DDT, BHC, Malathion, Systox and Metasystox (Kailidis 1962b, 1965). Then in 1965, Kailidis used *Bacillus thuringiensis* (Bt) against *T. pityocampa* (Kailidis 1967, from Kailidis 1986). The Bt-based products which were used during that period for aerial applications included Thuricide-HP. (15.8 billion I.U./kg), Dipel (16.000 I.U./mg) and Bactospeine (16.000 I.U./mg) at a dose of 700-750 gr in 50.1 of water/ha (Kailidis et al. 1977). In subsequent years, *B. thuringiensis* was used in Greece by other researchers for control of *T. pityocampa* with very good results (Vankova and Svestka 1976, Kailidis et al. 1977, Georgevits 1979, Avtzis 1983a, 1984).

Frequently, the Greek Forest Service was unable to provide airplanes at the proper time, and therefore applications of Bt were delayed, especially under difficult weather conditions in Northern Greece.

We obtained experimental confirmation about the effectiveness of Diflubenzuron for the control of *T. pityocampa* in Greece (Georgevits 1979, Avtzis 1981). However, because of the environmental safety of Bt and substantial improvements of new Bt products, including their improved efficacy at low temperatures, we were interested in evaluating their use against *T. pityocampa* in a Greek forest. In this paper, we report on the experimental application of Foray 48B against *T. pityocampa* in Northern Greece.

We obtained information suggesting that low temperatures have a negative effect on the effectiveness of *B. thuringiensis* (Vankova and Svestka 1976, Kailidis et al. 1977). Spectacular results were obtained with Diflubenzuron for the control of harmful insects (Skatulla 1975a, 1975b, Salama et al. 1976).

### Materials and Methods

Foray 48B was applied along with Dimilin 25wp. and Bactospeine wp. (16.000 IUAK/mg) in young plantations of *Pinus brutia* (10-14 years old) in the general area of Kedrinis Lofos near Thessaloniki. To check the effectiveness of the three substances mentioned above as compared to the control, materials were applied twice: first on October 17th, 1993, and then on November 27th, 1993 (Tables 1 and 2). It is significant to point out that on the day of the first spraying (Oct. 17th, 1993), 90% of the larvae had hatched. The effectiveness of the treatment, based on larval mortality, was estimated on the 7th, 14th, 21st, 28th and 35th day after the first spraying. After the second application, larval mortality was estimated on the 7th, 14th and 21st day.

The persistence of the three substances was investigated 7, 14 and 21 days after the first spraying. At each treatment area, three larvae were placed on five trees (3X5=15 in each area). The larval mortality on the control plots at the times the trees were artificially infested was 12.5%, 32.1% and 36.9%, respectively. On December 4, 1993, we estimated the final mortality of larvae and thus the persistence of the products.

**Table 1. Technical data of the first application<sup>1</sup> (October 17, 1993)**

Characters	Dimilin	Bactospeine	Foray 48B
Dose	250 gr/ 25 L water/ha	700 gr/ 25 L water/ha	1,5 L formulation/ha
Nozzles (number and type)	28 nozzles D-12-45 (pressure 50 PSI)	28 nozzles D-12-45 (pressure 50 PSI)	14 nozzles D- 2-45 (pressure 30 PSI)
Drop Size ( $\mu$ )	300	300	100
Application Time	12:30	13:00	14:30
Drop number per cm <sup>2</sup>	20	20	30
Nozzle positions relative to the airstream	90°	90°	45°

<sup>1</sup> Data of flying:

- Aircraft: CESSNA AG WAGON
- Flight Speed: 100 miles/hr
- Spraying height: 5-10 m above the top of the trees
- Speed of the wind: 5-6 miles/hr

**Table 2. Technical data of the second application<sup>2</sup> (November 27, 1993)**

Characters	Dimilin	Bactospeine	Foray 48B
Dose	250 gr/ 25 L water/ha	700 gr/ 25 L water/ha	3,0 L formulation/ha
Nozzles (number and type)	28 nozzles D-12-45 (pressure 50 PSI)	28 nozzles D-12-45 (pressure 50 PSI)	14 nozzles D-4-45 (pressure 40 PSI)
Drop Size ( $\mu$ )	300	300	150
Application Time	9:30	10:00	10:45
Drop number per cm <sup>2</sup>	20	20	20
Nozzle positions relative to the airstream	90°	90°	45°

<sup>2</sup> Data of flying:

- Aircraft: CESSNA AG WAGON
- Flight Speed: 100 miles/hr
- Spraying height: 5-10 m above the top of the trees
- Speed of the wind: 6-7 miles/hr

In addition to determining the effectiveness and the length of protection offered by the three substances, we also estimated the possible effect on egg hatching and parasitism.

### Results and Conclusion

As shown in Table 3, there was no apparent effect of any of the three insecticides during the first or second application on hatching, parasitism and infertile egg percentages when compared to the control. Larval mortality after the first, as well as after the second spraying, is provided in Table 4. From that table, it appears that the final mortality in both applications was very high, not only in the case of Dimilin, but of the Bt-based products as well. It varied from 96.1% (Bactospeine in the second spraying) to 100% (Dimilin, Bactospeine and Foray 48B in the first spraying). We estimated that the mortality in the control area reached 47.5% after the first application and 52.4% after the second (Table 4).

**Table 3. Hatching, parasitism and non-fertile egg percentages (%) after measurements of eggs of the 686 cylinder (batches) used in the experiment**

Application	Treatment	Hatching	Parasitism	Non-fertile
1 <sup>st</sup>	Control	65.1	10.4	24.5
1 <sup>st</sup>	Dimilin	58.8	11.2	30.0
1 <sup>st</sup>	Bactospeine	62.5	11.2	26.3
1 <sup>st</sup>	Foray 48B	58.5	13.3	28.2
2 <sup>nd</sup>	Control	65.7	11.0	23.3
2 <sup>nd</sup>	Dimilin	65.4	9.5	25.1
2 <sup>nd</sup>	Bactospeine	73.8	8.7	17.5
2 <sup>nd</sup>	Foray 48B	68.6	10.1	21.3

**Table 4. Larval mortality (%) during the two applications**

Application	Day	Control	Dimilin	Bactospeine	Foray 48B
1 <sup>st</sup>	7	12.5	80.0	91.2	88.8
1 <sup>st</sup>	14	32.1	100.0	98.6	98.6
1 <sup>st</sup>	21	36.9	100.0	99.9	98.8
1 <sup>st</sup>	28	40.5	100.0	100.0	100.0
1 <sup>st</sup>	35	47.5	100.0	100.0	100.0
2 <sup>nd</sup>	7	48.2	64.5	81.8	85.4
2 <sup>nd</sup>	14	52.3	82.7	90.5	98.5
2 <sup>nd</sup>	21	52.4	98.4	96.1	99.5

Regarding the persistence of products after the plots were re-infested with larvae, larval mortality in the Dimilin plots fluctuated between 95.8% and 100% even three weeks after treatment (Table 5). In contrast, mortality of larvae caused by the Bt products was somewhat reduced but still remained at high levels.

**Table 5. Total larval mortality (%) after the three new infestations, which took place 1, 2 and 3 weeks after the first application. (Time of the measurements: one month after the 3rd new infestation)**

Day of the new infestation	Dimilin	Bactospeine	Foray 48B	Control
1 <sup>st</sup> week (Oct. 23 <sup>th</sup> )	95.80%	99.00%	96.60%	
2 <sup>nd</sup> week (Oct. 30 <sup>th</sup> )	99.40%	99.55%	91.65%	48.20%
3 <sup>th</sup> week (Nov. 6 <sup>th</sup> )	100.00%	89.70%	90.00%	

Our conclusions are as follows:

1. None of the insecticides had an effect on egg mortality, percent egg hatching or egg parasitism. Regarding the species of egg parasites that we identified in the experimental areas, we found *Tetrastichus servandei* Dom L. and *Ooencyrtus pityocampae* Mercet. These

species coincide with those described by other authors (Kailidis 1962a, 1986, Schmidt 1988). Our results of measuring egg parasitism on our experimental areas are similar to those found by others (Table 3). For example, Biliotti (1958) found that *T. pityocampa* egg parasitism in N. France in 1956 fluctuated between 3 and 28%. Kailidis (1986) determined that egg parasitism near Thessaloniki during 1960-61 was approximately 11.2%.

2. All three pesticides used gave acceptable results, as far as larval mortality was concerned, even 20 days after spraying. This result, which is considered to be very significant for the Bt-based products, is comparable to the results of other authors (Baranova 1972, Avtzis 1983b).

3. Of the Bt-based products used under the particular climatic conditions of both experiments, Bactospeine and Foray 48B provided very high larval mortality. This fact is significant and suggests that we should reconsider the use of Bt products in October and November for the control of *T. pityocampa*, especially in N. Greece. Because we obtained larval mortality ranging from 96.1-99.5% under conditions in which the air temperature was 3.6° C at the time of the second application, and never exceeded 8.3° C during the first 10 days after spraying, concerns about the use of Bt at low temperatures should be reviewed. It was the opinion of many authors, based solely on experimental data, that the effectiveness of Bt-based products was adversely affected by climatic conditions that occurred during and after spraying; this led to the limited use of those products for the control of *T. Pityocampa*, especially in N. Greece (Burdajewa and Schtscheblanov 1975, Korchagin 1980, Molloy et al. 1981). With the development of improved strains and superior formulations, it is now possible that Bt products can be used even during the third and fourth larval instar and will provide very good results.

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# Activity of Several Contact Insecticides Against Selected Forest Insect Pests in Poland

D. WORETA AND H. MALINOWSKI

Forest Research Institute, Department of Forest Protection, Bitwy Warszawskiej 1920 r. nr 3, 00-973 Warsaw, Poland

**ABSTRACT** Four pyrethroids (alphamethrin, deltamethrin, lambda-cyhalothrin, and zeta-cypermethrin) and etofenprox were tested against several insects that are severe pests in Polish forests. Results of the evaluations indicate that adults of the common cockchafer are equally susceptible to the four pyrethroids but 10 times less susceptible to etofenprox. Larvae of the pine moth, nun moth, and Diprionid sawflies were slightly less susceptible than cockchafer adults to the pyrethroids; however, in all cases the LD<sub>50</sub> was less than 0.3 µg/g. Adults of the large pine weevil were not very susceptible to the pyrethroids used in these evaluations.

IN RECENT YEARS, populations of the common cockchafer (*Melolontha melolontha* L.) and other soil insect pests have increased significantly, especially in agricultural lands that have been abandoned (Woreta 1994). This results in serious problems when attempts are made to reforest these lands by planting trees because of the damage caused to the roots by larvae of *M. melolontha*. The adults of this species also cause significant defoliation of deciduous stands that are adjacent to the plantations.

Application of chemical insecticides is necessary to protect the roots of trees and shrubs that are planted in these areas. These products are applied to the surface of the seedlings prior to planting or injected into the soil after planting (Malinowski et al. 1996). Sometimes, insecticides are applied aerially to deciduous stands to prevent defoliation and to reduce the number of adult beetles that are available to produce progeny for the next generation. In Poland, four pyrethroid insecticides (alphamethrin, deltamethrin, lambda-cyhalothrin, and zeta-cypermethrin) and etofenprox are recommended for control of the common cockchafer, the large pine weevil (*Hylobius abietus* L.), and larvae of other leaf-feeding insects (Glowacka 1995). The objective of this study was to evaluate the activity of these five insecticides against adults of the common cockchafer and large pine weevil as well as against larvae of three defoliating species.

## Materials and Methods

The following insect species were tested:

- (a) common cockchafer (*M. melolontha*) adults (ca. 7 days old)
- (b) nun moth (*Lymantria monacha* L.) second and third instar larvae
- (c) pine moth (*Dendrolimus pini* L.) third and fourth instar larvae
- (d) conifer sawflies (Diprionidae) second instar larvae (mixed species)
- (e) large pine weevil (*H. abietis* L.) adults of undetermined age



The insecticides studied were:

Pyrethroids

- (a) alphasmethrin as 100 g/l EC (Fastac 100 EC, Shell)
- (b) deltamethrin as 25 g/l EC (Decis 2,5 EC, Roussel-Uclaf)
- (c) lambda-cyhalothrin as 25 g/l EC (Karate 025 EC, Zeneca)
- (d) zeta-cypermethrin as 100 g/l EC (Zorro 100 EC, FMC)

Arylpropylether

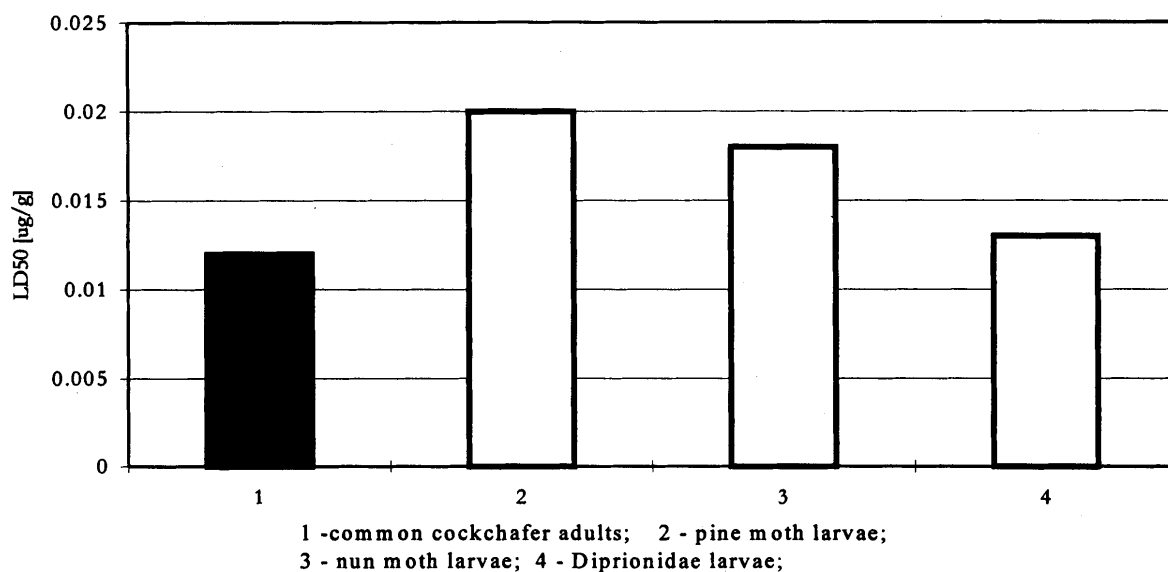
- (a) etofenprox as 100 g/l EC (Trebon 10 SC, Mitsui Toatsu)

The activity of the insecticides was determined by topically applying a 1  $\mu$ l drop of the product dissolved in acetone with a micro-applicator. The droplet was applied to the ventrum of the abdomen of adult beetles and to the dorsum of the larvae. All insecticides were diluted in acetone to produce six to eight different concentrations. Two or three replications of 15 individuals were treated at each dosage and percent mortalities were calculated at 48 or 76 hours after treatment. Controls were treated with acetone alone. Mortality data were subjected to probit analysis (Finney 1952) and LD<sub>50</sub> values were calculated using four to six doses that caused mortality between 10% and 90%.

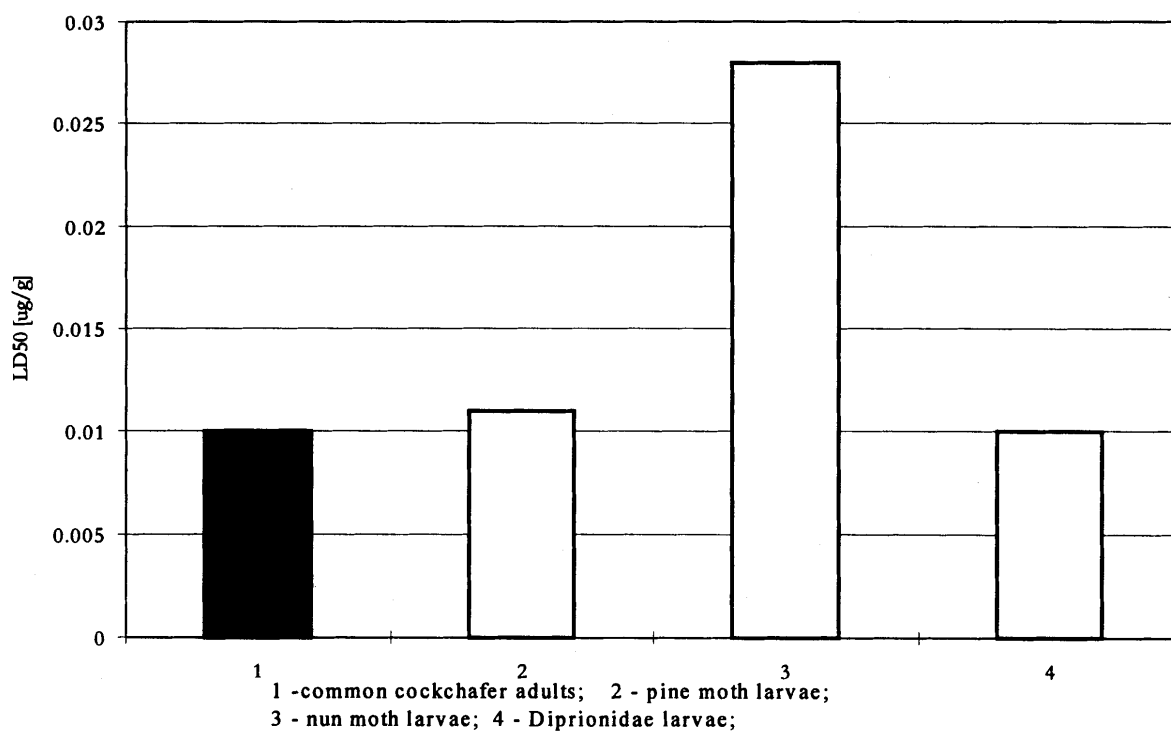
### Results and Discussion

Adults of the cockchafer were equally susceptible to the four pyrethroids tested (LD<sub>50</sub> = 0.008-0.012  $\mu$ g/g) and in most cases were more susceptible to the pyrethroids than were larvae of the nun moth, pine moth, and sawflies (Figs. 1-4). Conversely, the cockchafer was less susceptible than the other species to etofenprox, the LD<sub>50</sub> being 10 times higher than that calculated for pyrethroids. The LD<sub>50</sub>'s for all species tested against etofenprox were higher than those calculated for pyrethroids (Fig. 5). Adults of the large pine weevil were 500-fold less sensitive to the pyrethroids than were adults of the cockchafer. Etofenprox was not tested against *H. abietis*.

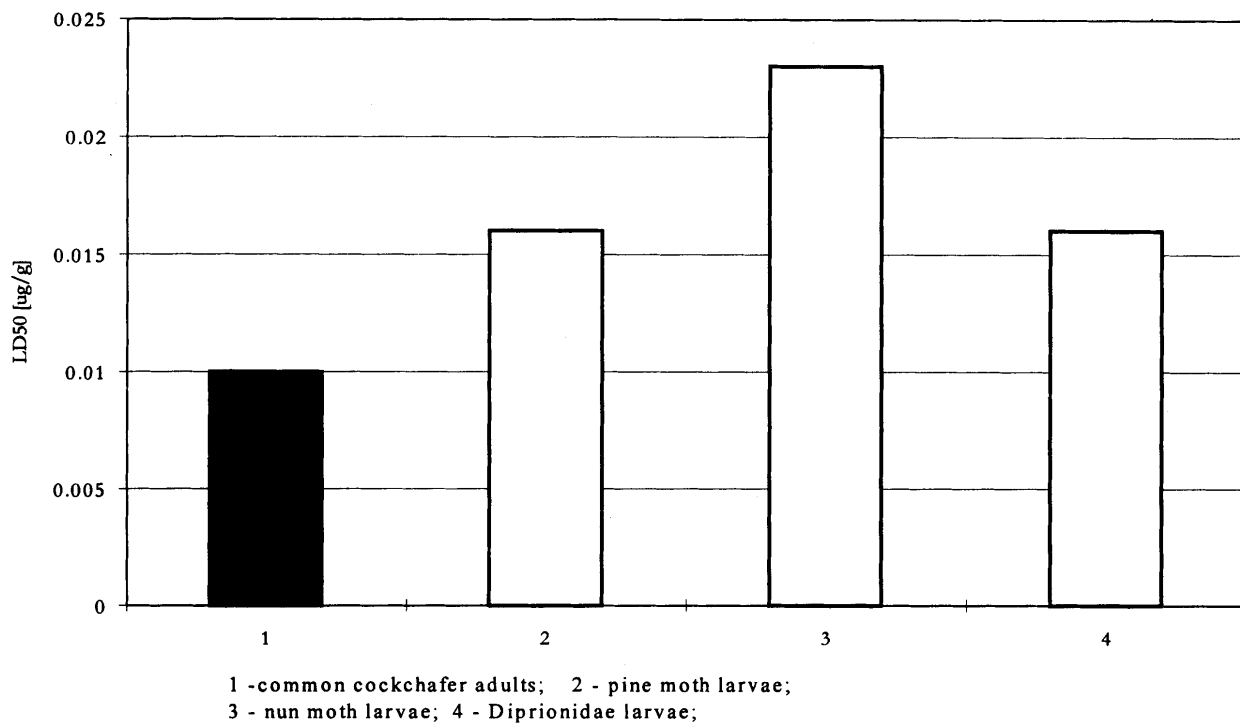
With the exception of alphasmethrin (Fig. 1), larvae of the nun moth were slightly less susceptible to the pyrethroids than were larvae of the pine moth and sawflies, though these differences probably are not significant.



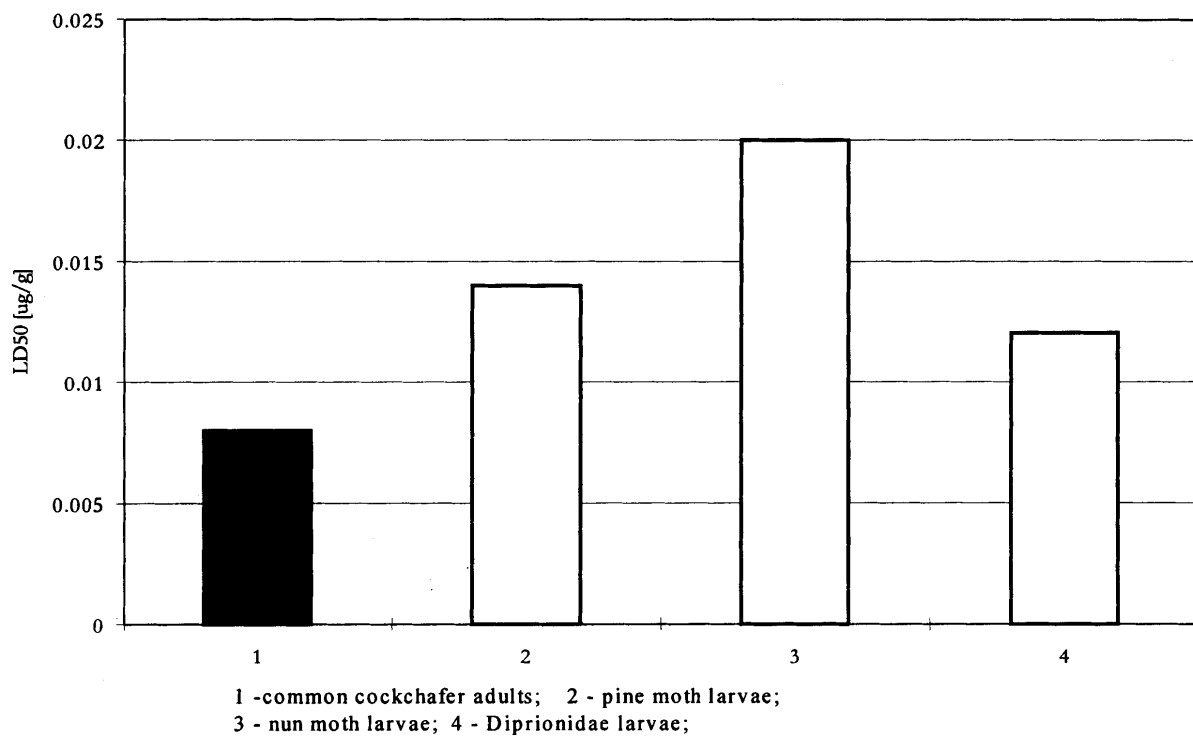
**Figure 1. Comparative activity of alphamethrin against the common cockchafer and some important leaf-feeding larvae.**



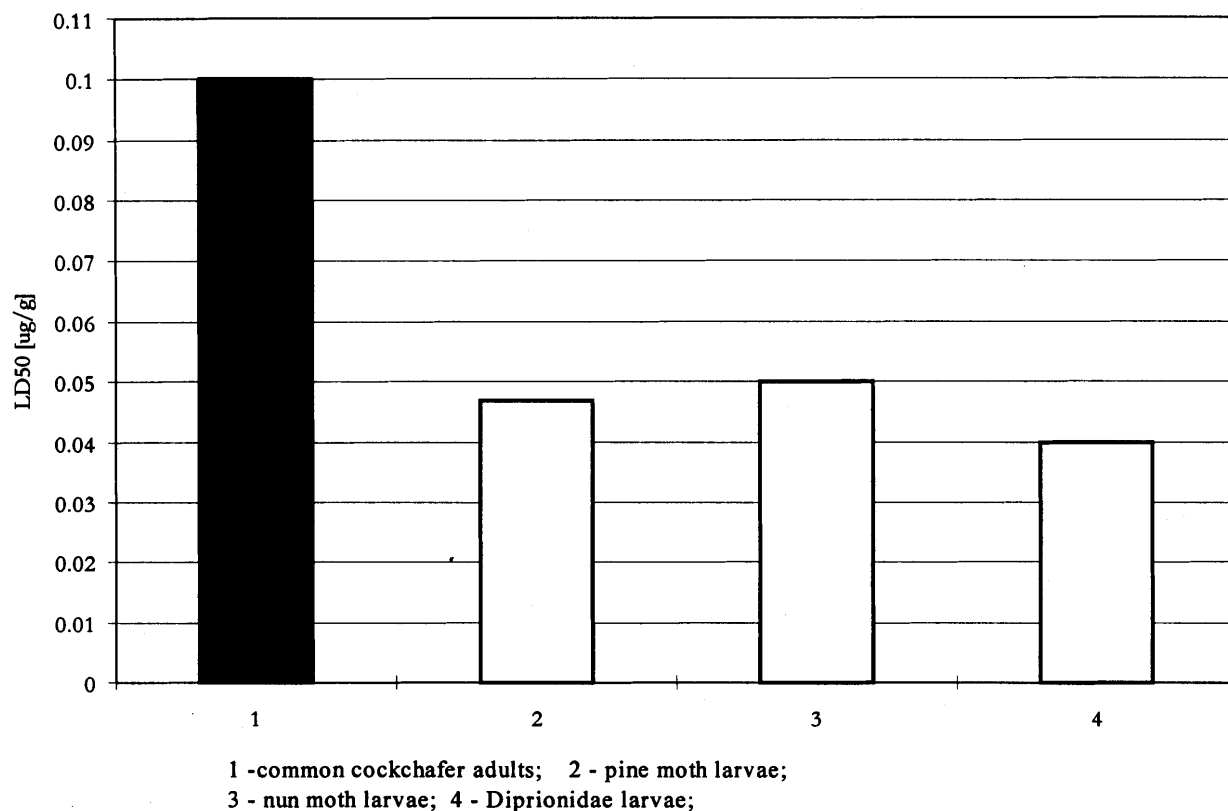
**Figure 2. Comparative activity of deltamethrin against common cockchafer adults and some important leaf-feeding larvae.**



**Figure 3. Comparative activity of lambda-cyhalothrin against common cockchafer adults and some important leaf-feeding larvae.**



**Figure 4. Comparative activity of zeta-cypermethrin against common cockchafer adults and some important leaf-feeding larvae.**



**Figure 5. Activity of etofenprox against common cockchafer adults and some important leaf-feeding larvae.**

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# Pathogens of the Gypsy Moth in Central Europe: Host Range and Interactions

JAROSLAV WEISER

Insect Pathology, Institute of Entomology, Czech Academy of Sciences, Branisovska 31, 370 05 Ceske Budejovice, Czech Republic

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**ABSTRACT** In Europe outbreaks of the gypsy moth are limited by pathogens which cause high mortality and subsequent collapse of peak gypsy moth populations. The most efficient of these pathogens is the very pathogenic nuclear polyhedrosis virus, but the more chronic cytoplasmic virus acts as a complicating factor. Pathogens such as the entomophilic fungi, bacteria and nematodes are of only marginal importance. Several species of microsporidia also influence gypsy moth outbreaks. At specific locations several different microsporidian species, often occurring simultaneously in gypsy moth populations, are involved. The following review summarizes the microsporidian species described from the gypsy moth and provides some new details on the ultrastructure of some of these microsporidia.

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PATHOGENS OF THE gypsy moth were first investigated in Central Europe. In the 1920s, Escherich and Breindel first demonstrated that the causative agent of the nuclear polyhedrosis was a filterable virus and in 1947 Bergold produced electron micrographs demonstrating the virus rods inside the polyhedra. In 1927, Zwölfer isolated *Plistophora schubergi*, from the brown tailed moth. This microsporidian was also infectious to *Lymantria dispar* and *Malacosoma neustrium* and was the first report of a microsporidian infection in *L. dispar* (Table 1). Zwölfer also made field applications of *P. schubergi* in an attempt to control these three forest pests. Weiser (1957a) described additional microsporidia obtained from gypsy moths collected during 1956 outbreaks in Slovakia. These species included *Nosema muscularis* and *Nosema lymantriae* together with *Thelohania similis*, recovered originally from the browntail moth. In addition, Weiser (1961a) described *Nosema serbica* from the gypsy moth. Timofeeva (1956) isolated *Thelohania disparis* from gypsy moths collected during an outbreak in the Ukraine. One inter-outbreak period later, *Plistophora schubergi* reappeared in gypsy moth populations but since that time has rarely been found.

Most of the microsporidia listed above were described before ultrastructural information was routinely included as a part of their description. Therefore, spore ultrastructure was not available for use in identifying these microsporidian species. Because ultrastructural information was not available, it has been difficult to identify microsporidian species isolated from gypsy moth outbreaks in different parts of Europe. A study organized by McManus et al. (1989) brought new isolates of microsporidia from gypsy moths collected from several additional European locations. Living isolates of these microsporidia are maintained in liquid nitrogen (Solter et al., 1996). Studies on these isolates will contribute to a better understanding of microsporidia as naturally occurring control agents of the gypsy moth.

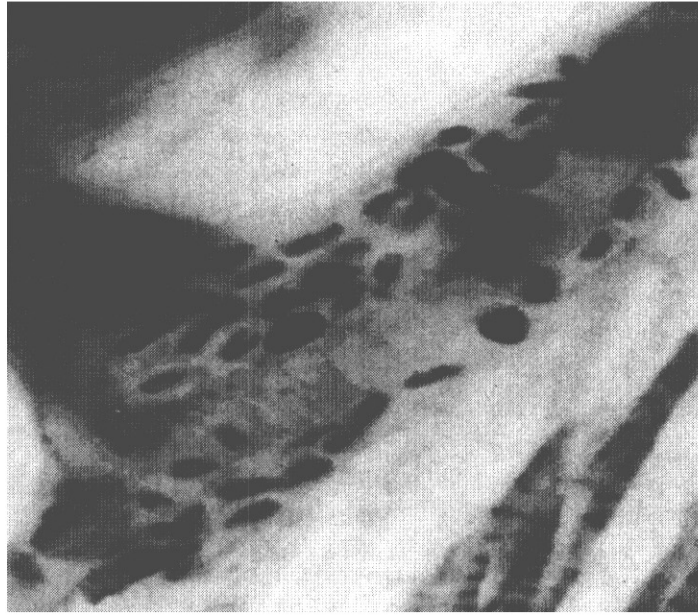
## Materials and Methods

Type material relative to Weiser's 1957 and 1961 descriptions are available in the Type collection of microsporidia maintained in the author's laboratory. They consist of dry Giemsa-stained smears and histological sections of infected caterpillars and include the following species isolated from *L. dispar*: *Nosema muscularis* Weiser (1957a), *N. lymantriae* Weiser (1957a), *Thelohania similis* (Weiser 1957a), *N. serbica* Weiser (1961a), and *Plistophora schubergi* Zwölfer (1927). These microsporidia, imbedded in paraffin blocks, also are included in this collection. Material in paraffin blocks, which was fixed in Bouin's, were transferred to xylol to remove the paraffin. After a final washing in water the material was fixed again in gluteraldehyde and osmic acid according to the procedure of Weiser et al. (1995). After embedding in Vestopal W the material was sectioned with an ultramicrotome and examined with an electron microscope. Some of the material extracted from paraffin was not suitable for observation but additional material from 1957 experimental infections will also be examined ultrastructurally.

## Results

Individual species were characterized using the 1957 protocols and materials. The paraffin-imbedded material, which was processed for the electron microscope as described above, produced ultrathin sections in which spore ultrastructure of some of the microsporidian species could be observed.

***Nosema muscularis* Weiser (1957a)** This microsporidian was isolated from L3 caterpillars of *Lymantria dispar* near Ziar n.H. The infection was localized in the circular and longitudinal muscles of the midgut where the pathogen occurred in longitudinal "galleries" in the muscle strands. The schizonts were short, binucleate stages, some with hypertrophic nuclei of the meiotic series. Oval spores with blunt ends, 6 x 3  $\mu\text{m}$ , contained a visible vacuole at the posterior end. Mature spores of the same shape were 4.8-6 x 3  $\mu\text{m}$ . The midgut muscles were progressively destroyed as the infection entered the tracheal matrix (Fig. 1), especially the terminal asteroid cells adhering to the midgut wall. Later the infection appeared in the secretory cells of the Malpighian tubules. Infection in the gut epithelium did not extend from the primary centers of entry that occurred immediately after ingestion of spores (peroral infection), but when infected Malpighian tubule cells ruptured, they released spores into the feces. Spores appeared in the muscles of the gut on the 5<sup>th</sup> day after spores were ingested and continuous mortality occurred over the next 20-25 days. Infected larvae appeared emaciated and became desiccated as they died. Apparently the midgut infection inhibited midgut peristalsis and thus inhibited feeding by infected larvae. Bodies of infected larvae were c- or s-shaped with a liquid diarrhea. Dying larvae had no fat body and the midgut was brown in color. Spores of *N. muscularis* were fed to other Lepidopteran hosts by placing spores on leaves of their host plants. The browntail moth and the fall webworm, *Hyphantria cunea*, were infected by *N. muscularis* while *Malacosoma neustrium*, *Aporia crataegi*, *Eriogaster lanestris* and *Bombyx mori* were refractive. In susceptible hosts the infection was localized in the muscles of the midgut. Material for EM analysis was not available.



**Figure 1. Tracheal matrix infected with *Nosema muscularis*. Magnification 1200x.**

*Nosema lymantriae* Weiser (1957a, 1957 b) *Nosema lymantriae* was isolated from infected gypsy moth caterpillars and maintained in laboratory rearings. Fat body and silk glands were infected (Fig. 2). Long ribbonlike merozoites with a longitudinal row of nuclei  $30 \times 2.5\text{-}3.5 \mu\text{m}$  were formed during schizogony. Young spores,  $6 \times 2.5 \mu\text{m}$  had an apparent vacuole at the posterior end. Fresh mature spores (Fig. 3) measured  $5\text{-}6 \times 2\text{-}2.5 \mu\text{m}$  and were spindle-shaped. Polar filaments,  $125 \mu\text{m}$  long, were occasionally extruded. Eleven days after ingestion of spores (20-30,000 spores per larva on leaves) small pockets of infection were present in the silk glands and fat body. Fifteen days after ingestion of spores, two-thirds of the silk gland was filled with spores and the fat body was infected. Infected larvae lived up to 30 days in controlled laboratory rearings. The original isolate did not contain any octospores. Spores from infected silk glands were used to initiate laboratory infections; feces from infected caterpillars was not infectious. Infected larvae exhibited reduced feeding and less movement than was observed in non-infected caterpillars. Infected larvae had a well developed fat body and demonstrated no symptoms of starvation. Dead infected caterpillars were desiccated. Spores of *N. lymantriae* were fed to other Lepidopteran hosts by placing spores on leaves of their host plants. *Euproctis chrysorrhoea*, *Aporia crataegi*, *Malacosoma neustrium*, *Hyphantria cunea*, *Eriogaster lanestris*, and *Bombyx mori* were all refractive to infection using the same spore suspension that produced infections when fed to *L. dispar*.



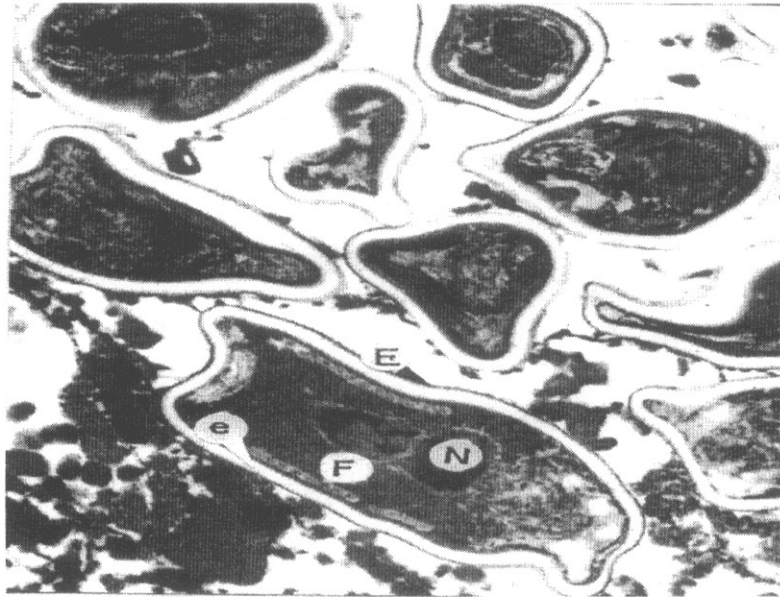
**Figure 2.** Silk gland of *L. dispar* infected with *Nosema lymantriae* (black spots). Magnification, 100x.



**Figure 3.** *Nosema lymantriae*, fresh spores. Magnification 1000x.



In ultrathin sections of material restored from paraffin, (Fig. 4) spores of *N. lymantriae* have a thick, rigid electron-negative endospore and a thin granular electron-positive exospore. The spore contains a pair of nuclei and a polar filament coiled in a single row directly under the spore wall in 10/11 or 12/13 isofilar coils.



**Figure 4.** *Nosema lymantriae*, type of 1957. Spores with thick endospore (e), thin exospore (E), and the polar filament in single row with 12/13 coils (F). Diplokaryon (N) in the center of the spore. Magnification, 12,000x.

***Nosema serbica* Weiser (1961a)** *Nosema serbica* was collected in Serbia by L. Vasiljevic. The same microsporidian also infected eggs of the gypsy moth from Varna, Bulgaria and from Cherson, Crimea, USSR. Larvae hatching from infected eggs were infected and contained broad oval spores, 5-6 x 4 and 6-8 x 3-4  $\mu$ m, in all tissues of the body. The muscles of the midgut, the silk glands, and the fat body were first infected and later the infection moved to the Malpighian tubules and segmental muscles. In adults, the microsporidian enters the nutritive cells of the gonads and infects the developing egg follicles. The earliest eggs laid by the infected female have a lower percent infection than eggs laid later. The eggs oviposited latest die and desiccated. Eggs that do hatch produce larvae that are infected. Octospores were not mentioned in the original description of *N. serbica*. Spores of *N. serbica* were fed to other Lepidopteran hosts by placing spores on leaves of their host plants. *Malacosoma neustrium*, *Hyphantria cunea*, and *Euproctis chrysorrhoea* were successfully infected by *N. serbica*. No difference in the feeding behavior and larval activity was noticed in infected larvae. The original type material in paraffin blocks is not available.

***Thelohania similis* Weiser (1957a, 1957b)** *Thelohania similis* was not originally a pathogen of the gypsy moth but invaded gypsy moth populations when an outbreak of the browntail moth occurred contiguous to an outbreak of the gypsy moth in Slovakia. *T. similis* spores infected gypsy moth larvae and the infection occurred in the fat body as was the case

in the browntail moth. Schizogony was with stages with only a few nuclei with some stages of the meiotic sequence. During sporogony, spherical plasmodia with 2, 4 and 8 nuclei were formed. These developed eight sporoblasts and eight spores. Fresh spores were broadly oval, 5-6 x 2-2.5  $\mu\text{m}$ , but in dry smears, spores measured 5 x 3  $\mu\text{m}$ . In the original material only fat body was infected. Silk glands, Malpighian tubules, and muscles of the midgut were not infected. Development of the infection in the gypsy moth was slow and with no apparent symptoms. *Euproctis chryorrhoea*, *Lymantria dispar*, and *Stilpnotia salicis* were all susceptible to infection by *T. similis*. When the three microsporidia, *N. lymantriae*, *N. muscularis*, and *T. similis* occurred as mixed infections, each microsporidian species infected only the host tissues typically infected by that species. *Nosema muscularis* infected muscles of the midgut, *N. lymantriae* the fat body and silk glands, and *T. similis* the fat body. Although *N. lymantriae* did not infect the browntail moth, mixtures of spores developed in the browntail moth. The mixture of *N. lymantriae* with *N. muscularis* did not infect *Euproctis*, however, the mixture of *T. similis* with *N. lymantriae* did infect that species.

In ultrathin sections, spores of *T. similis* (Fig. 5) have a thick, electron-positive exospore and a rather thin electron negative endospore. The spore content is less distinct. The polar filament is coiled in an irregular double row in 24/25 coils. The pansporoblast membrane is formed of a granular layer and inside is a dense mass of secretion granules, sometimes organized in long tubules, without evident fixation on the surface of the spores or the pansporoblastic membrane.



**Figure 5.** *Thelohania similis*, type 1957. Thick foamy exospore (E) and thin endospore (e) of mature spores. Polar filament (F) in double row of 24/25 coils. Pansporoblast membrane amorphous, granular (p), inside the pansporoblast are secretion granules and tubules (T). Magnification, 12,000x.

*Nosema lymantriae* (David and Weiser 1989) The material on which this study is based was collected in 1988. Both binucleate single spores and octospores were present in ultrathin sections. In fresh material, binucleate single spores were 5-6 x 2.5-3 $\mu$ m and octospores were 4.2-4.5 x 2.5-3  $\mu$ m. The pansporoblastic membrane surrounding the octospores was a unit membrane with a fine granular deposit on its outer surface (Fig. 6).

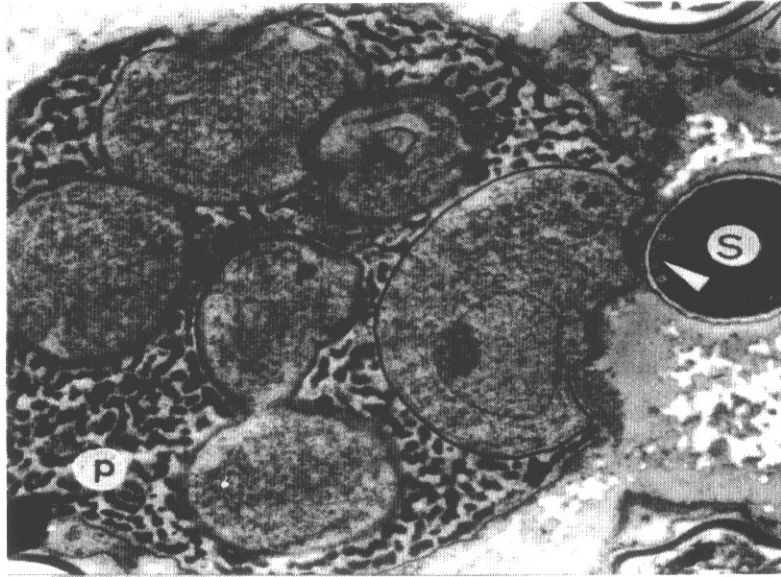


Figure 6. "*Nosema*" -*Vairimorpha lymantriae* of David and Weiser 1989. Octosporous pansporoblast with sporoblasts and secretion granules. Thin pansporoblastic membrane (p). On the right, a mature spore (S) with 5 cross-sections of coils of the polar filament (F). Magnification, 2000x.

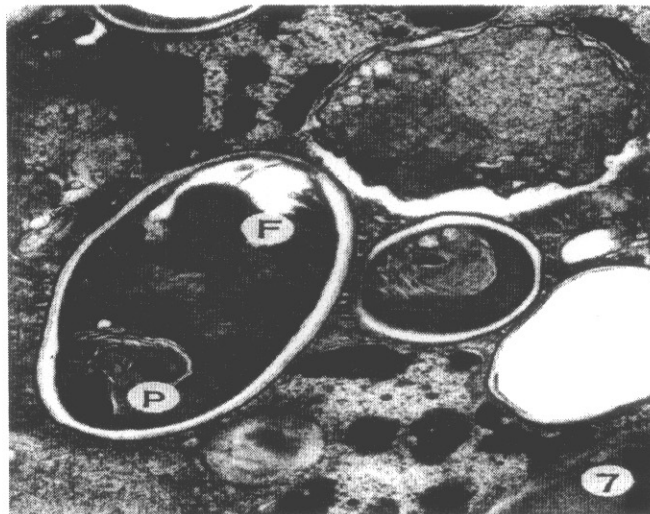


Figure 7. Final spore and sporont of the binucleate sereas of *N.-V. lymantriae*. Polar filament in 5 coils (F), polaroplast activated (P) with anchoring disc. Magnification 10,000x.

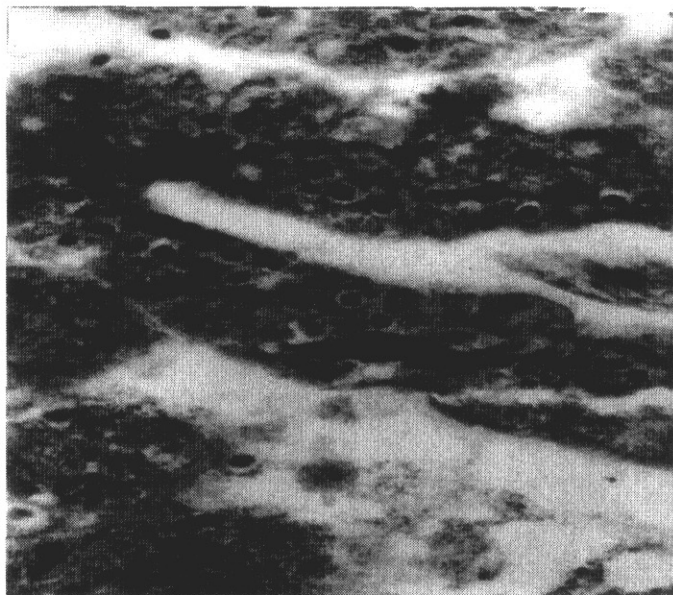
Secretion granules and tubules typical for *Vairimorpha* species fill the space between the spores. Binucleate spores have a rigid endospore and a very thin, smooth exospore. The polar filament is isofilar with a minimum of 5/6 coils and is attached to a knoblike anchoring disc (Fig. 7). The structure of the polaroplast, which is not visible, is activated during fixation. Octospores, produced in an octosporous sporoblast, have a rather thick exospore and a thin endospore. The polar filament (Fig. 8) has five coils, but in young spores only three coils.



**Figure 8.** Part of octosporous pansporoblasts with young spores of *N.-V. lymantriae* with cross-section of 5 coils (arrow). Magnification 2000x.

*Plistophora schubergi* Zwölfer (1927) (Weiser 1961a) *Plistophora schubergi* infections occur when infected *Euproctis* come in contact with *Lymantria*. The infection is confined to the midgut. Only the epithelial cells of the midgut are infected and, when heavily infected, these cells are filled with spores (Fig. 9). Infected cells, filled with spores, separate from the midgut and are eliminated in the feces. Regeneration nodules produce replacement midgut epithelial cells. Spores develop in compact pansporoblasts with multinucleate plasmodia. These divide within spherical sporoblasts and mature into small oval spores 2.5 x 1.5µm. Later, in disorganized tissues, the pansporoblasts are dissolved and sporoblasts mature free into spores. *Plistophora schubergi* is included in the genus *Endoreticulatus*, but the type species, *Endoreticulatus fidelis* differs in some parts of its development and this transfer should be evaluated further. *Plistophora schubergi* occurs in many different Lepidopteran hosts representing several different families. Although the strains from different hosts are morphologically identical, they are not equally infectious in cross-transmission studies. The strain occurring in the gypsy moth (Weiser 1961a) also infects *Euproctis chrysorrhoea*, *Stilpnotia salicis*, *Hyphantria cunea*, and *Barathra brassicae*. In *B. brassicae* large quantities of spores can be produced in the laboratory. The strain isolated from *Choristoneura fumiferana* in Canada will not infect *L. dispar*. *Plistophora schubergi* is not commonly found even in outbreak populations of *L. dispar*. Spores are less resistant to storage, however, they survive in hibernating L1 larvae of *E. chrysorrhoea*. Spores first appear in the feces of infected *L. dispar* larvae 6-8 days after oral infection. For the next 15-

20 days, the number of spores in the feces increases constantly. Infection of the progeny occurs when the egg mass is contaminated with feces. The feces are rich in proteins from the infected epithelial cells of the midgut and are attractive as food when larvae hatch from the egg mass.



**Figure 9.** *Plistophora (Endorectulatus) schubergi*, masses of spores in the midgut (epithelium of *L. dispar*. Magnification, 450x.

### Discussion

Outbreaks of the gypsy moth develop from low level populations which cover all of Europe and these are detectable by pheromone traps. The build-up of gypsy moth populations is influenced primarily by climatic conditions, the physiological status of the population including resistance to pathogens, and by the quantity and availability of infectious agents in the forest. The eleven year inter-gradation period indicated in older textbooks is today more variable, usually 6 years or less. The use of biological means such as *Bacillus thuringiensis* (in Bulgaria and Slovakia) breaks the periodicity of outbreaks. There is evidence of a wave-like migration from outbreak frontiers in the Crimea (Tchugunin, 1958) and in Yugoslavia. Gypsy moth populations at the border of the Black Sea or Adriatic are consistently higher, but do not necessarily result in outbreaks. In this region, gypsy moth populations contain a high percentage of the eggs infected with microsporidia and parasitized with egg parasites (these observations may be related).

Among the factors affecting outbreak populations of the gypsy moth, the nuclear polyhedrosis virus is the most important and is usually responsible for the final collapse of populations. After one year with high infestations of late instar caterpillars, the "Wipfeln" syndrome is evident and eggs deposited by infected females hatch into larvae that die in the early larval instars. There is no evidence of more or less virulent strains of NPV in nature, but in artificial introductions of NPV in early phases of the outbreak, there are strains of NPV

that cause immediate infections and other strains that produce only protracted, inapparent, egg-transmitted infections. Active virus strains can be selected by using for recycling virus produced from the first caterpillars that appeared infected. In order to acquire NPV material that is highly active, this process must be repeated 4 to 6 times.

The cytoplasmic polyhedrosis virus only appears in populations of the gypsy moth that are in close contact with the browntail moth or the fall webworm. The CPV has an incubation period of 8 days in the gypsy moth but it is efficient only in combination with other pathogens. No other virus is transmitted to *L. dispar* from other Lepidoptera that attack oak trees. This includes the NPV and granulosis virus of *H. cunea* and the poxvirus of *Operophtera*.

*Bacillus thuringiensis* does not cause the collapse of gypsy moth outbreaks and does not typically occur in Lepidopteran defoliators of oak. Its use as a microbial insecticide reduces defoliation and, in an artificial way, breaks the periodicity of gypsy moth outbreaks. *Bacillus thuringiensis* does not protect the oak flowers from destruction by the gypsy moth and thus is not effective in protecting the acorn crop.

Fungus infections develop only in caterpillars or pupae that have been damaged or killed by other factors. *Entomophthora aulicae* is primarily a pathogen of smooth caterpillars (Tortricids, Noctuids, etc.). In Europe, gypsy moths (primarily early instars) are infected with this fungus only from outbreaks of smooth caterpillars. Nematodes and mermithids sometimes occur in gypsy moth caterpillars in young stands, close to the soil.

Several species of microsporidia infect the gypsy moth, more than any other group of insect pathogens. At the time when most of the microsporidia were described from the gypsy moth, species with similar spore sizes and shapes were differentiated mainly on the basis of host range and development in the host. These are no longer the most important criteria used for identifying and classifying microsporidia and it is appropriate to reevaluate these microsporidian species using ultrastructural characteristics and, eventually, molecular characteristics. In recent studies some authors have provided details of the internal structure of the spores.

*Nosema* sp. studied by Sidor (1976) had a polar filament with 10/11 coils, a thin exospore and a solid endospore.

*Nosema* sp. from Romania collected by Saftoiu (1976) had a polar filament with 6/7 or 7/8 coils and a thin exospore. Other spores in this material had a polar filament with 10/12 coils.

*Nosema* from Bulgaria studied by Pilarska and Vavra (1987) had binucleate spores and a polar filament with 5/6 coils.

Since the first microsporidian was described from the gypsy moth over 40 years ago, several complicating factors have developed. These factors must be considered when evaluating and comparing the older microsporidian descriptions with the newer descriptions. One of these factors was the creation of the genus *Vairimorpha*. This genus has a diplokaryotic single spore sporogony as well as a sporogony with eight sporoblasts and spores. The sporogony with eight sporoblasts and spores copies the development of the genus *Thelohania* in the old concept. Another complicating factor is the formation of two types of spores in the host tissue, early spores for autoinfection and distribution of the microsporidian in host tissues, and the persistent spores for transmission and infection of other hosts. Except for differences in spore size and in the structures of the spore wall, there are, in some cases,

no simple method for identifying these types of spores. These differences are reflected in the old descriptions by indications of the range of spore sizes. The development of infections in different hosts, the ranges of host tissues, and the development from larval to adult stages and eventual transovarian transmission is, according to actual knowledge, variable to some extent and influenced by the method of transmission from host to host.

The project supported by the US Forest Service, and implemented by M. L. McManus and J. V. Maddox, has produced a large collection of microsporidia from gypsy moths collected from different parts of Europe. This collection of microsporidia, viable spores of which are stored in liquid nitrogen, will allow the old observations to be compared with modern evaluations of existing species. This report was prepared in support of this effort.

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# Oak Defoliating Insects in Hungary

GYÖRGY CSÓKA

Forest Research Institute, Hegyalja u. 14., 3232 Mátrafüred, Hungary

**ABSTRACT** Oaks are the most important species in Hungarian forests both from an ecological and economic point of view. Of the total forested area in Hungary (1.7 million hectares), ca. 35% is occupied by oak forests. The indigenous oak species are *Quercus petraea*, *Q. robur*, *Q. cerris*, *Q. pubescens*, *Q. farnetto* and *Q. virgiliana*. In addition to the native species, the North American *Q. rubra* is planted widely.

The insect folivore guild of oaks is extremely rich in species. Four hundred and forty-five species of herbivorous insects have been found to feed on the foliage of oaks in Hungary. Folivores represent 72% of the 629 species of all known oak herbivorous insects. Sixty-two percent (280 species) of all folivore species (455) are Lepidoptera. The proportion of species that are found only on oaks is very high; therefore, the oaks play an extremely important role in maintaining the diversity of herbivorous insects.

Of the 455 folivore species, 74% are leaf-feeders, 10% are leafminers, 10% are gall-makers, and 6% are sap suckers. The monthly abundance of folivore species shows a characteristic seasonal pattern, whereby they peak in early June, then decline continuously. The spring/early summer fauna is dominated mainly by polyphagous larvae of the Geometrid/Noctuid group in hilly and mountainous regions, and by Lymantriids in lowland oak forests. The late season fauna is dominated mainly by more specialized groups such as leafminers, leafgallers, Notodontid larvae, etc.

The Forest Research Institute has maintained a forest damage monitoring system since 1961. All forest owners and forest companies are required to report abiotic and biotic damage that occurs in their forests, including that caused by all of the major pests. This long-term database provides us with an excellent opportunity to investigate the trends of different pests and their characteristic damage. Based on this database, the Geometrid/Noctuid group dominates the spring/early summer folivore fauna, especially in sessile oak (*Q. petraea*) stands in the hilly and mountainous regions. This group has caused defoliation on an average of 9,138 ha/year during the past 35 years. The most common and abundant species of Geometrids are *Operophtera brumata*, *Erannis defoliaria*, *Colotois pennaria*, *Alsophila aescularia*, *Agriopis aurantiaria*, and *A. marginaria*. In addition, several Noctuids, such as *Orthosia gothica*, *O. incerta* and *O. stabilis*, play a significant role in this group. *Lymantria dispar* is by far the most significant defoliator of forests in lowlands and on hills at lower elevations. *L. dispar* prefers *Quercus robur* and *Q. cerris*, but seldom causes severe damage in sessile oak (*Q. petraea*) stands. Outbreak areas can be found all over the country, though the average area defoliated is around 6000 ha/year.

Tortricid leafrollers cause damage on ca. 1200 ha/year, though significant increases in damage have occurred over the last 35 years. The most important species are *Tortrix*

*viridana* and *Archips xylosteana*. *Euproctis chrysorrhoea* and *Malacosoma neustria* defoliate mainly lowland pedunculate oak forests, especially younger stands.

*E. chrysorrhoea* causes damage on ca. 974 ha/year, although the area defoliated by this pest has increased significantly over the last 35 years. Defoliation by *M. neustria* occurs on 560 ha/year.

The largest areas defoliated were recorded in the early sixties and in the early nineties. The Geometrid species group caused the most damage in 1963 (ca. 71,000 ha), whereas *L. dispar* damaged more than 34,000 ha in 1994. The weather during both periods was extremely dry and warm. It has been demonstrated statistically that drought played a key role in provoking more serious outbreaks of several forest insects (i.e. oak defoliators), leading to a significant expansion of territory damaged. In addition to the increasing damage caused by oak defoliators, several other species that are favored by warm and dry conditions appeared as new pests in the last two decades in Hungarian oak forests. These include *Tischeria* leafminers, *Caliroa* sawflies, and the oak leaf beetle, *Haltica quercetorum*.

# Effect of the Mimic Prototype, Rh 5849, on 20-Hydroxyecdysone Titters in Defoliating Lepidopteran Larvae

MACIEJ A. PSZCZOLKOWSKI

Department of Forest Protection, Forest Research Institute, Bitwy Warszawskiej 1920r. nr. 3, 00-973 Warsaw, Poland

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**ABSTRACT** The first nonsteroidal ecdysteroid agonist, RH 5849 (1,2-dibenzoyl-1-tert-butylhydrazine), is a prototype of MIMIC, a novel insect growth regulator that can induce a lethal premature moult in larval Lepidoptera. The effects of RH 5849 applied to Lepidopteran larvae on changes in levels of endogenous ecdysteroids titers are reported here. Larvae of *Spodoptera littoralis* (Noctuidae), a defoliating pest of cotton, were used as the model insect in this study.

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LARVAE WERE TREATED with RH 5849 at varying times before the onset of pupation. Non-lethal doses of RH 5849 (5 µg), dissolved in acetone, were applied topically. 20-hydroxyecdysone (20E) levels were determined by means of an enzyme immunoassay using acetylcholinesterase as label.

During the day of pupation, the concentration of ecdysteroids in the haemolymph of untreated larvae increases, reaches its peak (so called transient peak), and then decreases. As in many other insect species, a rapid increase in the rate of pupation is observed in *S. littoralis* during the decrease of 20E level. Application of RH 5849 before the transient peak resulted in a remarkable increase in endogenous ecdysteroid titre. The decrease in ecdysteroid levels occurred about 12 hours later in insects treated with RH 5849 than in larvae which received solvent alone. When the concentration of endogenous ecdysteroids reached the transient peak, application of RH 5849 had an opposite effect. The level of endogenous ecdysteroids increased and then rapidly dropped, attaining its minimum about 12 hours earlier than in solvent-treated controls. As a consequence of treatment with RH 5849, larvae delayed or precipitated the time of pupation depending on the time of treatment.

It is noteworthy that the doses of RH 5849 (5 µg) used in this study were non-lethal. Even a dose of 50 µg of RH 5849 did not cause any lethal effect in *S. littoralis*, when applied during the day of pupation. However, low doses of RH 5849 caused shifting of pupation time and treated insects pupated in daylight whereas untreated controls pupated during the night. This means that although insects treated with non-lethal doses of RH 5849 do not die, they can change their circadian rhythms of pupation (i.e. they can change their ecological niche). Consequently, insects treated with non-lethal doses of RH 5849 may be eliminated by day active predators (e.g. by birds).

The data presented on RH 5849 effect indicate that non-steroidal ecdysteroid agonists can interfere differently with ecdysteroid-dependent physiological processes, depending upon the time of treatment. Thus, potential users of ecdysteroid agonists probably should consider not only the proper dose of these insecticides, but also the proper time of application, particularly with respect to those species that develop synchronously in the field.

## The Stone Pine Needleminer (*Ocnerostoma copiosella* Heyd.) in the Engadine Valley, Switzerland

BEAT FORSTER

Forest Insect and Disease Survey, Swiss Federal Institute for Forest, Snow and Landscape Research WSL/FNP, CH-8903 Birmensdorf, Switzerland

**ABSTRACT** During the last two decades, outbreaks of the stone pine needleminer *Ocnerostoma copiosella* have occurred more frequently than before. The affected Swiss stone pines (*Pinus cembra* L.) show needle loss and are more susceptible to secondary pests.

*Ocnerostoma copiosella* is a small, silver-greyish moth with a wing span of approximately 5 mm that completes two generations per year. The young larvae feed inside last year's and older needles which turn yellow and later brown. Then the larvae leave the needles through oval holes and spin their cocoons inside the five-needled fascicles. Adults emerge in June and July. The second generation overwinters as larvae within the needles.

The stone pine needleminer is common in the Central Alps within the natural distribution of Swiss stone pine. In the Upper Engadine Valley, between 1700 and 2000 m in elevation, the insect has optimum conditions. Trees that are attacked lose a high proportion of their older needles. Typically, stone pines that are attacked experience only a small growth reduction and recover in subsequent years. Since the needleminer has been attacking the pines more frequently, the crowns have become thinner and the trees more susceptible to secondary pests. Sometimes weakened trees die due to subsequent attacks by bark beetles, such as *Ips amitinus*, *Pityogenes conjunctus* or *Polygraphus grandiclava*.

There is concern that, in the near future, an outbreak of the stone pine needleminer will occur simultaneously with an outbreak of the larch bud moth (*Zeiraphera diniana* Gn.). Larch bud moth larvae also affect stone pines; however, they attack the youngest needles, which are normally spared by the needleminer. Until now, the outbreaks of the two species have never occurred at the same time.

Occasional outbreaks of this needle-mining moth have been recorded in the last century; however, over the past two decades, outbreaks have occurred more frequently. Since 1989, outbreaks have occurred every two years in the odd numbered years. To date, we have not been able to demonstrate a correlation between the mass attacks and yearly weather conditions. But since 1983, the average temperature in July has been 0.8°C higher than before, and there have been deficits of precipitation. The causes of the 2-year cycles are still unknown and require further investigation.

## Wind and Bark Beetles - the Most Serious Harmful Agents in the Norway Spruce Stands of Slovakia

BOHDAN KONÔPKA

Forest Research Institute, Masarykova 22, 960 92 Zvolen, Slovak Republic

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**ABSTRACT** Wind is the most serious harmful abiotic agent in the forests of Slovakia while bark beetles are the most serious biotic agent. The volume of timber removed due to wind damage was recently approximately 800,000 m<sup>3</sup> annually, and timber removal due to bark beetle damage was approximately 200,000 m<sup>3</sup> annually, which together represented almost 20% of total annual fellings. However, the total damage in forest ecosystems caused by wind and consequently by bark beetles is much higher. Heavy damage to ecological conditions in forest stands caused by wind and bark beetle outbreaks create suitable conditions for other pests. Wind storms and subsequently bark beetles, cause great economic losses due to the premature decline of stands, increased costs of tree removal, worsened timber quality and sudden deforestation of land with all its unfavorable consequences (e.g., soil erosion). Late timber processing and replanting of forest plantations on gale-disaster areas results in higher costs of weed control and soil preparation.

In cases of large wind damage, there is a threat of bark beetle outbreak which could last during the following years. One example of a bark beetle outbreak due to wind destruction was a situation in Slovakia in the mid-1960's. In that case, the volume of timber removal caused by bark beetles tripled after extraordinary high wind disaster. Recently, emissions, weather extremes, wind storms and total weakening of forest ecosystems (especially in Norway spruce stands) in Slovakia have created optimum conditions for further outbreaks of bark beetles. For this reason, the significance of forest protection against this group of injurious agents will increase continually.

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## Defoliating Insects: History of Outbreaks in the Coniferous Forests of Russia

ANDREY V. SELIKHOVKIN, DIMITRY L. MUSOLIN, AND TAMARA E. SERGEEVA

Dept. of Zoology, St. Petersburg Forest Technical Academy, Institutskiy per., 5, 194021, St. Petersburg, Russia

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**ABSTRACT** Moths (*Dendrolimus superans sibiricus*, *D. pini*, *Lymantria monacha*, *Bupalus piniarius*, *Panolis flammea*) and sawflies (*Diprion pini*, *Neodiprion sertifer*, *Acantholyda spp.*) are usually considered as the most important defoliating pests in Russian taiga forests.

The history of outbreaks of these species have been analyzed based upon statistical data (calculations and estimates according to reviews of the Federal Forest Service of the Russian Federation, 1977-1994) and publications in forestry periodicals (1870s - present).

We determined that there are no common trends in the distribution of outbreak areas of these insect pest species, and that there are no common environmental factors that either regulate the dynamics of outbreaks of these species or that occur simultaneously in all of the Russian forests. It is very probable that there is a periodicity to outbreaks in nature, but that they occur only in localized populations.

The appearance of outbreaks of *D. superans sibiricus* and some other pests in more northern regions (in comparison with the records examined in several previous decades) may be related to trends in global climate change. However, we suggest that this appearance is possibly caused by the absence of accurate information about historical outbreaks in the sparsely populated regions of Siberia.

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## Damage Caused by Cockchafers (*Melolontha* spp.) in Hungary during the Last 30 Years

JÓZSEF TÓTH

Forest Research Institute, Frankel L. 42-44., 1024 Budapest, Hungary

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**ABSTRACT** There are seven tribes of *Melolontha melolontha* that occur in the Carpathian Basin, and three of the seven tribes occur in Hungary. The distributions of the tribes are different, but at some locations two or even three tribes are sympatric. In Hungary, *Melolontha* completes its development in 3 years.

<u>Tribe</u>	<u>Years of swarming</u>
V.	1989-1992-1995-1998
VI.	1990-1993-1996-1999
VII.	1991-1994-1997-2000

The cockchafers react positively to artificial light; therefore, they can be collected easily using light traps. We have been using a network of 25 light traps in Hungary since 1962 to monitor cockchafer populations.

The cockchafer causes serious damage in forests. The adults can totally defoliate trees, and the larvae can kill even older trees by feeding on the roots. In the last 34 years, the average yearly defoliation by adult cockchafers has been 15,095 hectares. In the same period, the larvae caused damage on 1,947 hectares. *Melolontha hippocastani*, which occurs simultaneously with *M. melolontha*, also causes damage. This species also requires 3 years to complete its development; however, distributions of its tribes are not well understood. Other common and abundant cockchafer species include: *Anomala dubia*, *Anomala vitis*, *Anoxia orientalis*, *Melolontha pectoralis*, *Polyphylla fullo*, *Rhizotrogus aequinoctalis* and *Rhizotrogus vernus*.

Helicopter spraying of pesticides against the adults and soil treatments against the larvae have been recommended.

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## Reaction of *Cyzenis albicans*, *Ernestia rudis*, and *Parasetigena silvestris* (Diptera:Tachinidae) to Insecticides Used in the Control of the Nun Moth

CEZARY BYSTROWSKI

Department of Forest Protection, Forest Research Institute, Bitwy Warszawskiej 1920 r., no 3, 00-973 Warsaw, Poland

**ABSTRACT** The nun moth is the most destructive defoliator of coniferous trees in Europe. Its outbreaks have been recorded in Scots pine and spruce stands in Poland since the 19th century. During the period 1946-1984, approximately 6 million ha of infested coniferous stands were treated with organochlorine and pyrethroid insecticides to control the nun moth. Beginning in 1991, increases in the density of nun moth populations were observed. In 1994, the project "The Integrated Control of Nun Moth in Polish Forests" was developed and financed by Polish and foreign funds. The use of pyrethroids was reduced because of environmental concerns and consequently Foray 02.2 UL and Dimilin 480 SC were used on 90% of the treated area. A study of the reaction of tachinids to these insecticides was initiated by the Forest Research Institute as a part of research to determine the impact of the control treatments on the forest environment.

In 1994, four nun moth-infested experimental plots were established in the Ostrów Mazowiecka forest district in the eastern part of Poland within even-aged Scots pine stands. The insecticides were applied on the 22nd of May. Two experimental plots were treated with contact insecticides (Decis 2.5 EC and Trebon 10 SC) and two were treated with stomach insecticides (Dimilin 480 SC and Foray 02.2 UL). Six Moericke's traps were placed in the tree crowns in each experimental plot. The traps were checked every two weeks from April to October in 1994 and 1995.

Six thousand individuals representing 42 tachinid species were collected in 1994, and about three thousand individuals representing 25 species were collected in 1995. In 1994, the most numerous species recorded were *Cyzenis albicans*, *Ernestia rudis* and *Parasetigena silvestris*. In 1995, the numbers of two species, *Parasetigena silvestris* and *Ernestia rudis*, were drastically reduced apparently due to both the control treatment and unfavorable weather conditions during the development period of their hosts. However, the number of *Cyzenis albicans* increased in 1995 apparently due to its early emergence and rapid development of both this fly and its host.

## Growth and Survival of *Chrysomela tremulae* (Coleoptera, Chrysomelidae) on Transgenic Poplar Containing a *Bacillus thuringiensis*-Endotoxin Gene

S. AUGUSTIN<sup>1</sup>, A. DELPLANQUE<sup>1</sup>, J.C. LEPLE<sup>2</sup>, G. PILATE<sup>2</sup> AND L. JOUANIN<sup>3</sup>

<sup>1</sup>INRA, Station de Zoologie Forestière, 45160 - Ardon, France

<sup>2</sup>INRA, Station d'Amélioration des Arbres Forestiers, 45160 - Ardon, France

<sup>3</sup>INRA, Laboratoire de Biologie Cellulaire, 78026 Versailles Cedex, France

**ABSTRACT** In France, the chrysomelid beetle *Chrysomela tremulae* is the main defoliating pest in both young plantations and short rotation stands of poplars. Larvae and adults of *C. tremulae* feed on leaves of young poplars, leaving only the vein, and can be a limiting factor for biomass production. They also attack the bark when foliage is no longer available, frequently killing the seedlings.

In view of the problems concerning the use of insecticides (i.e., prohibitive cost and environmental safety), the introduction into plants of genes conferring resistance to this insect is a new promising strategy.

The *Bacillus thuringiensis*-endotoxin gene Cry III A was introduced via cocultivation with *Agrobacterium tumefaciens* in a Leuce hybrid clone (*Populus tremula* x *P. tremuloides*) that is very sensitive to *C. tremulae*. Three transformants were tested for their effects on survival and development of *C. tremulae*.

*C. tremulae* survival was significantly reduced on transgenic foliage tested compared to non-transformed foliage. Larval development time was also affected by foliage: it was longer on leaves of the three transgenic than on the non-transgenic control leaves. Male and female pupal wet-weights, measured as an estimate of reproductive potential, were lighter on leaves of the three transgenic foliages.

The three transformants assayed on *C. tremulae* survival and development provided protection to poplar, but this protection was not absolute. The results on mortality suggested that the level of expression of the endotoxin was moderate, but sufficient to induce effects on insect development that will enhance the teratological action.

## Foliage Palatability of Pine Species and Clones to the Rare Subspecies *galliegria* of the Beauty Moth, *Graellsia isabellae* Oberthür (Lepidoptera: Attacidae)

CLAUDE GÉRI<sup>1</sup>, FRANCIS GOUSSARD<sup>1</sup>, MARIE-ANNE AUGER-ROZENBERG<sup>1</sup>,  
CATHERINE BASTIEN<sup>2</sup> AND FLORENCE PASQUIER<sup>1</sup>

<sup>1</sup>INRA, Station de Zoologie Forestière, 45160 Ardon, France

<sup>2</sup>INRA, Station d'Amélioration des Arbres Forestiers, 45160 Ardon, France

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**ABSTRACT** The suitability of foliage from various Scots pine clones to the sawfly *Diprion pini* L has previously been shown. We studied their suitability to the beauty moth *Graellsia isabellae*. Throughout its range in France and Spain, *G. isabellae* Graells feeds principally on Scots pine, *Pinus sylvestris*. It is the only endemic Attacidae in Europe. The French subspecies *galliegria* Oberthür only exists in specific alpine valleys, and is protected. The vividly colored caterpillars feed individually on foliage during 5 instars before spinning a cocoon. Pupae overwinter in diapause.

First we established a laboratory strain in order to conduct experiments without collecting insects from the field. The colony was founded from 150 eggs in 1990 with authorization from the French Department of Environment. Batches of 5 to 10 caterpillars were fed cut pine shoots in field and laboratory conditions. In order to avoid inbreeding, females were placed in small open cages in the Alps and mated with wild males. The colony produced several thousand insects per year. Simultaneously, experiments were carried out on the insect's biology and on the range of its host plants in order to identify its potential range and improve rearing methods. The diapause of *G. isabellae* could not be prevented by varying photoperiod, temperature or food. However, diapause was successfully broken at any moment by injection of  $\beta$ -ecdysone. Adults emerge 20 days following injection and insects can thus be obtained during the entire year. This species can mate, oviposit and feed in field conditions at low elevation. Feeding bioassays described above were conducted using cut shoots of the foliage of different pine species and confirmed that *P. sylvestris* was the most preferred. *Pinus uncinata* and *P. contorta* were nearly as suitable, but Austrian, Salzman and Jeffrey pines were not fed upon at all. Bioassays with the foliage of 13 clones, previously ranked as favorable (G) or unfavorable (B and B+) for *D. pini*, were also performed in 1992, 1993, 1994 and 1995. They showed significant differences in mortality, development rate and pupal weights. The worst performance was observed on the B+ clones; the G clones were highly suitable. Performances was found to be better on these clones than on naturally occurring trees. The high performance of insects fed on certain clones indicates that the selection of trees for silvical properties alone may be problematic.

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## Biotic Factors Affecting *Stereonychus fraxini* Populations in the Longoza Forest, Bulgaria

GALINA MARKOVA

Forest Research Institute, K1. Ochriski 132, 1756-Sofia Bulgaria

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**ABSTRACT** The ash weevil, *Stereonychus fraxini* Deg., is a major insect pest of ash trees in Bulgaria and other European countries. At very high infestation levels, some damaged stands in the Longoza Forest in Bulgaria looked like winter, because the trees did not produce leaves. Because of environmental concerns associated with the use of chemical control, there is interest in alternative control measures, such as biological control, for maintaining this pest at low densities.

A study was conducted to estimate the natural biotic factors that regulated ash weevil populations during the period 1988-1995. Examination of the adult *S. fraxini* population for entomopathogens indicated that the fungus *Beauveria bassiana* was quite abundant. This fungus reduced the population density of adults by 58% in 1988. Laboratory experiments demonstrated that this fungus strain was very virulent against adult weevils. Although the epizootic occurred only during the period of high host density, infected beetles were observed in subsequent years.

After the collapse of *S. fraxini* populations in 1988, three species of parasitoids were consistently recovered in rearings: *Mesopolobus mediterraneus*, *Pteromalus cioni* and *Entedon cionobius*. Combined parasitism by these species varied from 35.7-65.1% each year, though there appeared to be no relationship between richness of the parasitoid complex and host population density. It is concluded that these parasitoids play an important role in maintaining *S. fraxini* populations at low densities.

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## Significance of *Glyptapanteles liparidis* (Hym., Braconidae) as a Regulator of Gypsy Moth, *Lymantria dispar* (Lep., Lymantriidae) in Different Host Population Densities

AXEL SCHOPF AND GERNOT HOCH

Institute of Forest Entomology, Forest Pathology and Forest Protection, University of Bodenkultur Vienna, Hasenauerstrasse 38, A-1190 Wien, Austria

**ABSTRACT** The significance of the endoparasitic braconid *Glyptapanteles liparidis* was studied from 1993 to 1995 at three experimental sites in Austria with high, medium and low density *Lymantria dispar* populations. To ascertain rates of parasitism and acquire data on the bionomics of *G. liparidis*, gypsy moth larvae were collected at specific stages at the sites and reared in the laboratory until parasitoids emerged, larvae died, or they pupated. At low density sites, we had to supplement the native population with *L. dispar* eggs and young larvae in order to perform the studies. Additionally, we tested the feasibility of calculating rates of parasitism retrospectively from densities of parasitoid cocoon clusters and gypsy moth egg masses.

The highest percent parasitism by *G. liparidis* (47.6%) occurred among larvae collected as 3<sup>rd</sup> and 4<sup>th</sup> instars from a latent population in 1993. This braconid species was the dominant species at this low density site where we artificially augmented host populations. At sites with high gypsy moth densities, parasitism by *G. liparidis* did not exceed 3% parasitism in 1993. In 1994, after the medium to high gypsy moth populations had collapsed, parasitism by *G. liparidis* increased at these localities. In the third year, the studies were performed only at the former high and low density sites, where it was necessary to artificially augment gypsy moth populations. In both locations, rates of parasitism were estimated to be at about 15%. Apparently, due to its excellent searching capacity, this parasitoid species is believed to be especially effective in low density gypsy moth populations and in focal areas where populations increase.

Rates of parasitism by this braconid species were also favoured by retarded host larval development in 1994 and 1995, due to low temperatures in the spring.

Most of the *G. liparidis* larvae emerged when hosts were in their 4<sup>th</sup> instar. However, in 1994, a higher percentage of parasitoids emerged from 3<sup>rd</sup> instars than the previous year; this may have been caused by early hatch of *L. dispar* larvae in 1994, and then their retarded development. This also influenced the pattern of emergence of the parasitoids over time, which was bimodal in 1994. In 1993, there was only a single dominant peak of emergence. Rates of parasitism calculated from estimates of parasitoid cocoon cluster density and gypsy moth egg density, did not accurately reflect actual rates of parasitism. Thus, counts of cocoon clusters of *G. liparidis* should not be used to retrospectively estimate parasitism by this species.

## Weevils Affecting Reforestation in Poland

IWONA SKRZECK

Forest Research Institute, Bitwy Warszawskiej 1920r No. 3, 00-973 Warsaw, Poland

**ABSTRACT** In Poland, even-aged forest plantations that are established on clearcut areas and in which Scots pine (*Pinus sylvestris* L.) occupies more than 60% of the reforested areas, are infested by pest insects of the family *Curculionidae*. Conifer stumps on the clearcuts favor the development of the large pine weevil (*Hylobius abietis* L.) and the spruce pine weevil (*Hylobius pinastri* Gyll.), which are the most serious pests of Polish forest plantations. Both species oviposit in roots of fresh conifer stumps and the larvae develop under the bark of roots. Adult weevils feed on the bark of seedlings, which eventually leads to seedling death. Serious economic loss is caused primarily by *H. abietis*, which is much more abundant than *H. pinastri*.

Forest plantations and natural stands, especially those established on poor plagiatous sandy soils and on burned areas, are damaged by such species as *Cneorhynchus plagiatus* Shall and *Strophosomus capitatum* Deg. The most serious damage is caused by the adult beetles of those species which feed on pine needles, which leads to their senescence. Premature shedding of young pine tree needles is also caused by the adult beetles of *Brachyderes incanus* L. and *Brachonyx payk.*

At present, there is no successful method to predict the amount of tree damage caused by weevils. Our estimation of the threat to forest plantations by the weevils is based on the number of adults captured in traps baited with pine billets, pieces of fresh pine bark, or IBL-4 artificial traps containing food attractants.

The abundance of weevils in forest stands is determined by estimating the extent of needle damage caused by the pests.

The following control methods are used to protect reforested areas against pest weevils:

1. mechanical methods with the use of traps (pine billets, artificial traps) for capturing adults;
2. chemical methods that consist of dipping seedlings in contact insecticides prior to planting or spraying young trees after planting;
3. biological methods based on using the Julich fungus *Phlebiopsis gigantea* (Fr.: Fr.) to reduce colonization of stumps by *H. abietis*.

## Effect of Litter and Soil-Borne Entomogenous Fungi on Mycoses of Pine Sawfly (*Diprion pini* L.) During Hibernation Period

CEZARY TKACZUK, RYSZARD MIĘTKIEWSKI AND JAROSLAW GRODZICKI

Agricultural and Pedagogical University, Department of Plant Protection, ul. Prusa 14, 08-110 Siedlce, Poland

**ABSTRACT** At the beginning of June, 1994 and again in 1995, 20 samples of litter were selected in a pine forest (stand age about 35-40 years) situated in Lysow. From each sample (0.25 m<sup>2</sup> in area), all cocoons of the pine sawfly were removed along with samples of litter and soil down to a depth of 10 cm. The same methods of investigation were also used in another forest near Chotyłow; however, at this location, cocoons were collected in mid-November 1995 rather than in June. The causes of mortality of pine sawfly larvae in cocoons were determined in the laboratory. Insects infected by fungi were incubated in moist chambers at 22° C until the presence of fungal pathogens was evident, at which point we were able to identify the organism to species. In addition, we used *Galleria mellonella* larvae as "bait insects" in order to determine the spectrum of entomopathogenic fungi that occurred in the samples of pine litter and soil from which the *D. pini* cocoons were removed. The larvae were placed in plastic Petri dishes filled with either litter or soil. The dishes were incubated at 22° C in 1994, and at 18° C and 26° C in 1995.

Entomopathogenic fungi infected from 12.3% to 52.2% of cocoons containing larvae of *D. pini*. *Beauveria bassiana*, *Paecilomyces farinosus*, *P. fumosoroseus* and an unidentified species of *Entomophaga* were isolated from hibernating larvae. The fungus *B. bassiana* infected up to 85.2% of the total number of larvae that were infected by fungi in the forest city near Lysow in 1994 and 1995. Unidentified species from the genus *Entomophaga* infected 74% of *D. pini* cocoons in the forest situated near Chotyłow. *Paecilomyces farinosus* and *P. fumosoroseus* were occasionally found in dead pine sawfly larvae. *Beauveria bassiana*, *P. farinosus*, *P. fumosoroseus* and *Metarhizium anisopliae* were isolated from litter and forest soil using the "insect bait method". The fungi *B. bassiana*, *P. farinosus* and *P. fumosoroseus*, which infected hibernating larvae of the pine sawfly, were also isolated from forest soil. *Beauveria bassiana* was the dominant species that occurred in localities and at all temperatures. *Beauveria bassiana* infected relatively more *G. mellonella* larvae at 18° C and 22° C than at 26° C, whereas the fungus *M. anisopliae* was recovered only at 26° C from soil taken from the Chotyłow locality. *Beauveria bassiana* and *P. fumosoroseus* were also isolated from pine litter using the bait insect method.

## Predicting Gypsy Moth Defoliation Using Spatially Explicit Information

RONALD M. WESELOH

Department of Entomology, Connecticut Agricultural Experiment Station, New Haven, CT 06511 USA

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**ABSTRACT** A procedure has been developed to predict forest defoliation by the gypsy moth, *Lymantria dispar* L., in Connecticut, U.S.A. The procedure uses elevation, percent of soil region with poor soil drainage, average defoliation from 1969 to 1994, past defoliation (obtained from hand-sketched aerial survey maps), and egg mass counts (obtained from a regular grid throughout the state and interpolated by inverse-weighting) to fit logistic regression models that have been used to predict defoliation for 1985 and 1989-1995. When defoliation was substantial, the models predicted locations of defoliation accurately, but when defoliation was low, predicted and actual defoliation did not often correspond. However, the models accurately forecast the amount of defoliation that occurred in most cases. Thus, for years in which defoliation is moderate to heavy, the models can be expected to be useful in predicting both the amount and location of defoliation. In sparse years, the models will at least indicate that expected defoliation will be low, in which case the actual locations are not as important. The procedure should be useful in helping to make control decisions, and will be validated in future years.

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## ***Phytobia* Population Densities in European White Birch (*Betula pendula* Roth)**

TIINA YLIOJA<sup>1</sup> AND HEIKKI ROININEN<sup>2</sup>

<sup>1</sup>Finnish Forest Research Institute, Punkaharju Research Station, Finlandiantie 18, FIN-58450 Punkaharju, Finland

<sup>2</sup>University of Joensuu, Department of Biology, P.O. Box 111, FIN-80101 Joensuu, Finland

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**ABSTRACT** Larval boring holes of *Phytobia betulae* Kang. (Diptera: Agromyzidae) in birch stems constitute a record of *Phytobia* population densities in the past. A larva, which hatches from an egg laid into a growing shoot in the canopy, mines in the differentiating xylem towards the base of the host tree. The tree fills the larval tunnel with brown parenchyma cells and resin and, therefore, the larval tunnel remains visible during the tree's life span.

We randomly sampled 26 European white birches (*Betula pendula* Roth) from a 30-year-old stand. The number of late instar *Phytobia* larvae was measured from wood disks taken from the stems at a height of 1 meter. We calculated the annual mean population density in the sampled trees, which provided us with a time series of 28 years.

The number of larval tunnels per tree correlated positively with the breast height diameter of the host tree ( $r=0.69$ ,  $N=26$ ,  $P<0.0001$ ). The individual host trees and the age of the host tree explained 19% and 41% of the annual variation in population size, respectively. Some host trees maintained high populations and others sustained low populations. Our results support the plant vigor hypothesis: strong growth in host tree diameter is more suitable for larval development than is weak growth.

A trend in the population trajectory indicated nonstationarity: the population was going through a growth phase. A simple regression curve was fitted to the time series in order to remove the trend. The partial rate correlation function showed a single negative spike at lag 1, indicating a first order density-dependent process. This suggests that the population is resource regulated. Both *Phytobia* population densities and the yearly variation in the size of populations are low. The population growth follows an annually increasing food resource provided by the host tree. The close relationship of *Phytobia* and its host indicates that, most probably, the factors limiting the density of *Phytobia* populations are associated with the characters of host trees.

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# Recent Outbreak of the Nun Moth (*Lymantria Monacha L.*) in the Czech Republic

JAN LISKA AND PETR SRUTKA

Department of Forest Protection, Forestry and Game Management Research Institute, Jiloviste - Strnady, Czech Republic

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**ABSTRACT** In this century, several outbreaks of the nun moth (*Lymantria monacha L.*) have been recorded in the territory of the Czech Republic. The largest and best known outbreak was during the period of 1917-1927 when more than one-half million hectares of mostly spruce forest stands were infested. Several less severe outbreaks occurred in 1930-1932, 1938-1943, 1946-1950 and 1964-1967. The total area infested in these periods was approximately 22,000 ha. The first symptoms of the present outbreak occurred in 1993, roughly a quarter of a century since the last outbreak period had faded out. During the years 1994-1996, approximately 34,000 ha of forest stands, almost exclusively monocultures of Norway spruce (*Picea abies*), have been infested by the nun moth in the Czech Republic. It was necessary to protect most of this area by means of aerial spraying.

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## Population Dynamics

As in the past, the initiation of the current nun moth outbreak followed a period of dry and warm weather, which occurred mainly in 1989, 1990, and 1992. In most cases, single outbreak foci were located in the same sites where at least one outbreak occurred in the past (so-called historical foci). What differed from the past, however, was that most of the infestations were at higher elevations, between 600-700 m. The infestations occurred almost exclusively in spruce monocultures 50-80 years old. Centers of the single foci, however, often comprised mixed stands of Norway spruce (*Picea abies*) and European larch (*Larix decidua*). The area of these centers varied from a few hectares to a few tens of hectares.

## Pest Management

**Monitoring Methods** Prior to the most recent outbreak, nun moth populations were monitored mainly with pheromone traps. However, since 1993, the system for monitoring populations has been modified and emphasis has been placed on more conventional methods. This consists of using pheromone traps to determine the period of peak flight, and then checking the incidence of swarming moths during this period (the walk-and-watch method).

Another method being evaluated is to measure the amount of excrement (frass) produced by larval populations. In endangered localities, efforts are made to estimate the number of female moths at the bases of sample trees (the Wellenstein method). Before a

decision is made to conduct control measures in the Spring, trees are limed with sticky bands to estimate the abundance of early instar larvae.

**Control Methods** The most frequently used insecticides were Trebon 30EC and Trebon 10F used with a carrier of light mineral oil or an emulsion of oil with water. These products were applied at a dose of 1 L/ha using micronair AU 4000 rotary atomizers. Trebon was applied in those sites where it was determined that a critical population density occurred. The effect of the treatment was immediate and reliable. Even in those cases where the weather was not favorable before and after application, the effectiveness of treatment varied from 90 to 95%.

In locations where lower-density populations of nun moth occurred, the biopreparation Foray FC 48B (*Bacillus thuringiensis*) and the chitin synthesis inhibitor (Dimilin 45 ODC) were used. Their effectiveness was adequate when the weather was favorable during and after they were applied.

**Behavior of other Lymantriid Moths** The characteristic phenomenon of the recent nun moth outbreaks was its synchrony with outbreaks of other species of the family Lymantriidae. Immediately preceding the nun moth outbreak, the largest outbreak of the gypsy moth, *Lymantria dispar*, occurred in the Czech Republic. Furthermore, local outbreaks of the following related species were recorded: *Orgyia antiqua*, *Calliteara (Dasychira) pudibunda*, *Euproctis chrysorrhoea*, and *Leucoma (Stilpnotia) salicis*.

McManus, M.L.; Liebhold, A.M., eds. 1998. **Proceedings: population dynamics, impacts, and integrated management of forest defoliating insects; 1996 August 18-23; Banská Štiavnica, Slovak Republic. Gen. Tech. Rep. NE-247.** Radnor, PA: U.S. Department of Agriculture, Forest Service, Northeastern Research Station. 352 p.

This publication contains 52 research papers about the population ecology and management of forest insect defoliators. These papers were presented at a joint meeting of working parties S7.03.06, "Integrated Management of Forest Defoliating Insects", and S7.03.07, "Population dynamics of forest insects", of the International Union of Forestry Research Organizations (IUFRO). The meeting was held August 18-23, 1996, in Banská Štiavnica, Slovak Republic.

**Keywords:** Forest insect, defoliator, integrated management, population dynamics, ecology

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