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Studies on the egg parasitism in *Thaumetopoea pityocampa* over a period of four years (1991–1994) at Marikostino/Bulgaria[3]

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Abstract

In Bulgaria near Marikostino, in a forest of *Pinus nigra* Arn. 239 egg batches were sampled over four generations of *Thaumetopoea pityocampa* (Den. & Schiff.) (Lep., Thaumetopoeidae) in various seasons. Directly after collection the batches were singled in test tubes, closed with cotton stoppers and stored under laboratory conditions at 20–22 °C.

Oviposition always started from the base towards the tip of the needles. The emergence of egg parasitoids was observed daily, and the parasitoids were counted and identified.

The eggs from which parasitoids emerged were marked for further studies. After removing the adults and opening the eggs, the impact of the species of parasitoids was evaluated by identifying the meconia and other rests (left overs).

Ooencyrtus pityocampae (Mercet) was the most abundant egg parasitoid, followed by *Anastatus bifasciatus* (Fonsc.). Only few individuals of *Baryscapus servadeii* (Dom.) were collected. The hyperparasitoid *Baryscapus transversalis* Graham, was found at low densities. Only a few eggs were parasitised by *Trichogramma embryophagum* Htg. Total mortality of the host eggs varied from 34.7 to 79.7% depending on the period of parasitism and on the numbers of sterile eggs. The impact of the parasitoids was calculated to be 9.3–38.9%, depending on the period when parasitism had taken place.

The mean number of eggs per batch varied from 203 to 253. The rate of parasitism was reduced by early sampling of the egg batches. In some cases, a very high percentage (up to 29.2%) of undeveloped eggs was found. In all samples a small percentage of empty eggs was always observed.

1 Introduction

In the Bulgarian forests as well as in most mediterranean countries, the pine processionary moth, *Thaumetopoea pityocampa* (Den. & Schiff.), is one of the most frequent insect pests. Additionally to being an important defoliator, it is also harmful to local people, tourists and house animals, due to urticating hairs persessed by the caterpillars (LAMY, 1990). Since the beginning of this century, control methods were applied to reduce the populations of *T. pityocampa*. Among them, in Italy the most common method consisted in destroying the winter nests by cutting and burning or by shooting (NICOLINI, 1987). In pine stands with large trees, however this mechanical method is difficult and expensive to apply. On the other hand, spraying of microbiological or chemical pesticides by aircraft is also expensive and in some countries, like in Italy not allowed by law (MASUTTI and BATTISTI, 1990). Furthermore, for an effective application of biological preparations based on *Bacillus thuringiensis* Berl. the average day temperature should be above 15 °C (TSANKOV and MIRCHEV, 1993). In Morocco, where the aerial control was carried out with *B. thuringiensis* according to economic, ecological and touristic criteria and necessities, the effect was insufficient to reduce the populations. Therefore, over the last three decades the study of natural enemies of the pine pest increased and together with these activities the knowledge of the biology of the insect was improved (MASUTTI, 1964; HALPERIN, 1970, 1990; TSANKOV, 1972, 1990; SCHMIDT, 1988, 1990; BELLIN et al., 1990; KITT and SCHMIDT, 1993). For monitoring, the application of pheromone traps (TIBERI and NICCOLI, 1984; BARONIO et al., 1992; FREROT and DEMOLIN, 1993), collecting of egg batches and counting of the winter nests (BREUER et al., 1989) were useful. These methods allow for the evaluation of the population density and programming of control strategies in relation to the tolerance level. Concerning the control methods available none of the existing chemicals on microbiological preparations is effective and without side-effects to nature, with the exception of local application of methanolic extracts of *Melia azedarach* L. which drastically reduce the feeding activity of the caterpillars and increase mortality (BREUER and DEVKOTA, 1990).

This study of the egg parasitoides showed that a high parasitism rate was present in various parts of the infested areas (MASUTTI, 1964; BATTISTI, 1986 and TIBERI, 1990 for Italy; SCHMIDT and DOUMA-PETRIDOU, 1989; BELLIN et al., 1990 and SCHMIDT, 1990 for Greece; HALPERIN, 1990 and KITT and SCHMIDT, 1993 for Israel; TSANKOV, 1990 and TSANKOV et al., 1996 for Bulgaria; BILIOTTI, 1958 for France and GERI, 1984 for Corsica). The mean rate of parasitism encountered was about 40%. *Ooencyrtus pityocampae* (Mercet) and *Baryscapus servadeii* (Dom.) were the most abundant parasitoids. The presence of *Anastatus bifasciatus* (Fonsc.) and *Trichogramma embryophagum* (Htg.) was always limited, these species being irregularly distributed in the regions sampled. In Greece and Bulgaria, *Baryscapus transversalis* was found as hyperparasitoid

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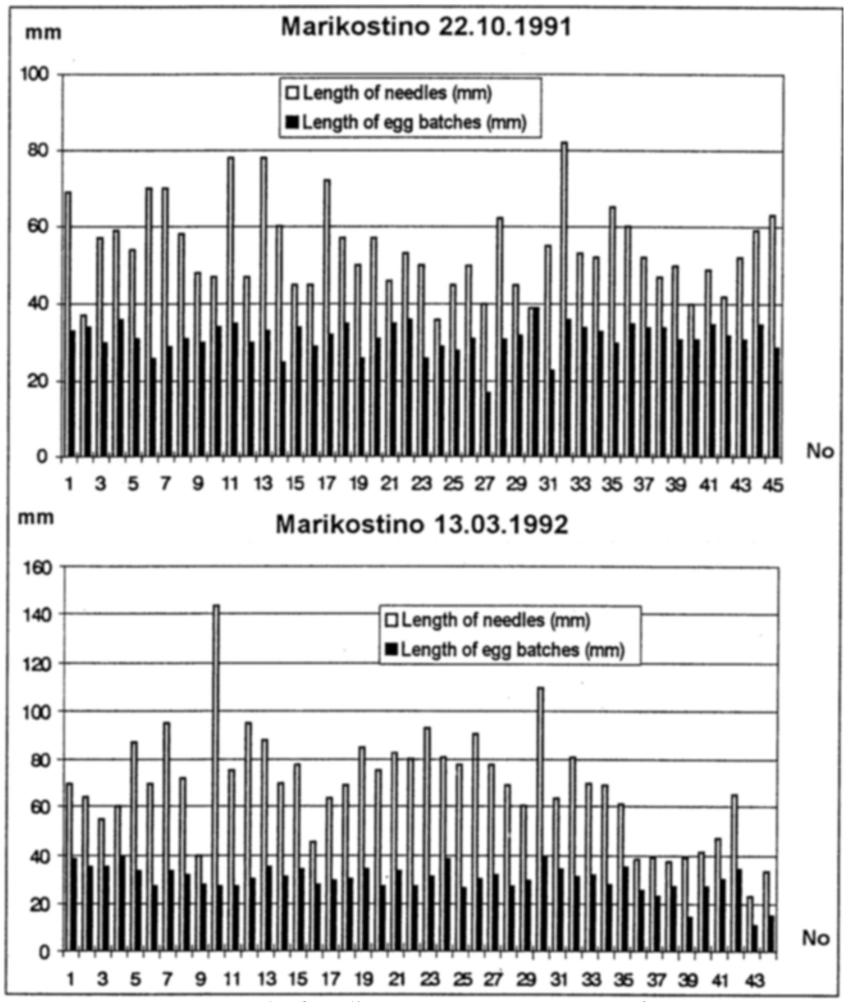

Fig. 1. Variation in the length of needles (*Pinus nigra* Arn.) selected for oviposition by the females of *T. pityocampa* at Marikostino and length of the egg batches deposited

(BELLIN et al., 1990; BELLIN, 1995; TSANKOV et al., 1996). The last mentioned authors showed that 90% of the mortality in the egg stage was due to parasitism, thus leaving 10% unaccounted for. In the literature, this issue has rarely been discussed and no studies were conducted on the impact of egg parasitism upon the life-cycle of *T. pityocampa*. We therefore, studied the egg parasitoids throughout four generations of the host in the area of Marikostino, in Bulgaria, considering several aspects.

2 Materials and Methods

The egg batches of *Thaumetopoea pityocampa* (Den. & Schiff.) were collected between 1991 and 1994 in the South-West of Bulgaria, near Marikostino, 180 m NN, a region influenced by mediterranean climate. Four consecutive generations of the host and its egg parasitoids were studied. All egg batches were taken from needles of *Pinus nigra* Arnold in different seasons. After collection, the batches were singled in test tubes with cotton stoppers and transported to the laboratory of the Forest Research Institute at Sofia. Laboratory studies were carried out at room temperature (20–22 °C).

Depending on the collection date of the egg batches, many egg parasitoids emerged before the winter period from eggs oviposited in the autumn of the same year. Therefore, observations of emergences have to be made all year around. At the end of December following egg deposition, the scales of the egg batches were removed, the parasitoids that emerged were counted daily, eliminated from the test tube, and transferred to smaller ones, for identification. This was done throughout the whole period of emergence. From the egg batches collected in March, the scales were removed immediately and observations performed on an almost daily basis. From the egg batches collected in June of the year after deposition, the scales could not be removed without destroying the batch and quantitative counting of egg parasitoids was not possible. Therefore, they were put in test tubes with scales and the emergence of parasitoids recorded. The unscaled eggs from which the parasitoids emerged were marked blue and the date of emergence was quoted in a table for either egg batch separately, presented by TSANKOV et al., (1996). The identification of the species was carried out using a WILD stereomicroscope (×40 magnification).

The last observation of all egg batches was made in Oktober 1995 at the Section of Entomology, University of Hannover. The eggs without a hole in the shell and all parasitised eggs were opened carefully and the meconia and remains of the dead and emerged insects were determined. The meconia were identified after SCHMIDT and KITT (1994) and TANZEN and SCHMIDT (1995). Additionally, the developing stages of the egg parasitoids were registered, whether dead or alive.

3 Results

3.1 Characterization of the egg batches

A total of 239 egg batches, oviposited over 4 generations in the area of Marikostino were studied. Almost all egg batches were wrapped around two needles of *Pinus nigra,* two egg batches only involved four needles, and oviposition took place from the base towards the tip of the needles, mostly leaving some distance from the base. The length of the egg batches depended mainly on the number of eggs which were deposited in 7 to 11 rows. The diameter of the egg batches varied from 3–4.5 mm. In some cases (3.8%) the length of the egg batches was equal, or longer than the shortest needles used for egg deposition (Fig. 1). When short needles were used, the resulting egg batch was relatively short. Generally, no clear relation was found between length of needles and length of egg batches. The number of eggs per batch varied from 51–327, throughout the years means of 203–253 eggs were obtained. There was no indication that these numbers would be influenced by the number of egg batches

Table 1. Characterization of the egg-batches of *Thaumetopoea pityocampa* (Den. & Schiff.) sampled at Marikostino in Bulgaria

T. pityocampa generations	I		II			III	IV
Date of sampling	22. 10. 1991	13. 3.1 992	8. 8. 1992	10. 9. 1992	2. 6. 1993	14. 8. 1993	2. 11. 1994
Number of egg batches	45	44	27	31	44	11	37
Length of needles *(P. nigra)*	36–78 mm	23–143 mm	45–130 mm	40–115 mm	45–95 mm	47–97 mm	42–123 mm
Length of egg batches	18–37 mm	10–39 mm	10–37 mm	10–38 mm	10–39 mm	24–32 mm	17–40 mm
Diameter of egg batches	3.5–4.5 mm	3.0–4.5 mm	3–4 mm	3–4 mm	3–4 mm	4–4.5 mm	3–4.2 mm
Number of egg rows per batch	7–9	7–10	8–11	7–10	7–10	8–11	7–10
Distance of egg batches to base of needles	0–43 mm	0–10 mm	0–28 mm	0–15 mm	0–30 mm	0–22 mm	0–12 mm
Number of eggs per batch	133–303	68–310	90–315	69–300	51–306	171–243	155–327
Total number of eggs	11373	10217	6629	7436	8946	2357	8252
mean per batch	253	232	245	240	203	214	223

studied, neither by the season in which they were collected (Tab. 1). From the eggs collected on 8. 8. 1992 and 14. 8. 1993 the caterpillars had not yet hatched.

3.2 Species of egg parasitoids found

At Marikostino, *O. pityocampae* was the most abundant species of egg parasitoids in all egg batches sampled. The highest number of live adults was obtained in the batches sampled in March 1992 (corresponding to 27.6% of all eggs studied). Most of the individuals were females found alive after collection; a maximum of 1.0–4.8% males were counted (Tab. 2). *Anastatus bifasciatus* was far less abundant; in both samplings of the first generation studied the percentage of adults, pupae and larvae was 11.1% and 10.7%, respectively, related to the number of eggs. In the second generation only 17 individuals of this species could be registered, only one in the third, and 19 in the fourth generation. All specimens were males, only four females appeared in the first generation.

Baryscapus servadeii was found in very low numbers, about 1% of the eggs were attacked in either generation. A total of 46 adult females and 10 males were counted, mainly after sampling of the egg batches.

The hyperparasitoid, *Baryscapus transversalis* Graham, was observed in low numbers and was not found in all samples (Tab. 2). Males and females were counted.

Trichogramma embryophagum was registered in two eggs of the first generation sampled on 22. 10. 1991; respectively 7 and 27 individuals were found dead inside the eggs, without opening. In the second generation 12 eggs, sampled on 8. 8. 1992, were parasitised. The number of parasitoids could not be recorded, because they emerged before the egg batches were sampled. Additionally, one male of *Macroneura vesicularis* (Retzius) emerged after sampling of the batches, on 14. 8. 1993.

Most of the egg parasitoids emerged through a hole made on the top end of the egg, but a considerable number of all species found its way out through a hole made on the side of the egg shell.

3.3 Parasitism rate and emergence dynamics

The rate of parasitism was considered to express the impact of the egg parasitoids. It depended on the date of egg batch collection after host oviposition. Early sampling of the egg batches, after oviposition, reduced the impact to 9.3% on 8. 8. 92, on which the caterpillars had not hatched. The hatching period was observed between 31. 8.–12. 9. 92, thus parasitism could only have taken place during one or two weeks, starting at the beginning of host oviposition. This agrees with the observation that no remains of the host could be found in parasitised eggs. A similar observation was made for the sample of 14. 8. 93 with a parasitism rate of 19.3%; again on this date the caterpillars had not hatched. These egg batches could have been parasitised over a week longer than the previous ones, and a 10% higher impact of the egg parasitoids resulted.

During our research period of four years, the impact of egg parasitoids decreased from 38.9% to 32.7%, considering that parasitism could take place during the whole embryonic development of the host (Tab. 3). This was correlated with a remarkable reduction of the population density of the host.

The total mortality of the host eggs was always higher than the rate of parasitism, because not all caterpillars were able to hatch. Many of them opened the egg shell, but died without leaving. On the other hand, a variable percentage of eggs remained undeveloped (sterile eggs) and some eggs were totally empty (Tab. 3).

Concerning *O. pityocampae*, the adults emerged up to November of the year in which host oviposition took place and during the following year up to September from egg batches laid about one year before. The number of emerged adults varied widely. From egg batches sampled on 8. 8. 92, 388 individuals emerged in 1992 and 86 in 1993, while from those collected on 10. 9. 92 a number of 213 specimens emerged in 1992 and 469 in 1993, all belonging to the same host generation. From egg batches collected on 14. 8. 1993, 313 adults emerged in 1993 and 55 in 1994, remains of the host were found in one parasitised egg, only. From these data it seems that parasitoids developed faster and emerged earlier in yolky eggs than in those with advanced developed embryos.

The percentage of *O. pityocampae*, emerged before and after collection of egg batches, did not depend on the sampling date. The highest rate (62.1%) emerged after the collection date of 13. 3. 92, but in all other cases the rate of parasitism was higher before collection of the egg batches, if the parasitoids could parasitise the eggs during the whole embryonic development of the host (Tab. 4).

Normally 3–5% of adult, pupal and larval parasitoids died inside the eggs. Therefore, it was surprising that in the egg batches collected on 10. 9. 92, 14. 8. 93 and 2. 11. 94, dead and live pupae, and larvae, were found in much higher numbers (39.7, 14.9 and 16.8%, respectively), when the dissection of the eggs took place after more than one year. Normally all developing stages found alive should be adults by then.

Table 2. Analysis of the main egg parasitoids of *Thaumethopoea pityocampa* (Den. & Schiff.) and their developmental rates in batches of various generations sampled at Marikostino. The egg batches sampled on 2. 6. 1993 were not analysed in the same way as the others (see Material and Methods). A total of 396 (7 males) of *O. pityocampae*, 2 males of *A. bifasciatus*, 18 (7 males) of *B. servadeii*, and 46 (16 males) of *B. transversalis* emerged after collection of the batches

Date of collection	22.10.1991		13.3.1992		8.8.1992		10.9.1992		14.8.1993		2.11.1994	
Ooencyrtus pityocampae (Mercet)	all females		all females		all females		2.5% males		1.0% males		4.8% males	
emerged before collection of egg batches with												
opening on top of egg shell	1163		900		103		730		198		1131	
opening on side of egg shell	55		54							368	42	
emerged after collection of egg batches with		2136 (18.8%)		2820 (27.6%)		577 (8.7%)		1412 (19.0%)		(15.6%)		2048 (24.8%)
opening on top of egg shell	851		1777		430		682		170		806	
opening on side of egg shell	67		89		44					455	69	
adults died in eggs with opening	62		27				110				0	
adults died in egg without opening	213	502	63	183	25	123		1113	19		126	566
pupae died in the eggs	108		37		45		502		11 (3 alive)		104 (1 alive)	
larvae died in the eggs	119		56		53		501		57 (40 alive)		336 (47 alive)	
pupae parasitised by *B. transversalis*	3		0		0		0		0		0	
Anastatus, bifasciatus (Fonsc.)	all males		4 females		only 1 ♂		only 15 ♂♂		only 1 ♂		all males	
emerged before collection of egg batches with												
opening on top of egg shell	11		5									
emerged after collection of egg batches with		277		335 (4 ♀♀)								
opening on top of egg shell	221		290								11	
opening on side of egg shell	45		40								2	
adults died in eggs without opening	36		18								3	
pupae died inside the eggs	12	60	1	27							1	
larvae of different stages died inside the eggs	12		8								2	
Baryscapus servadeii (Dom.)	all females		all females		only 7 ♀♀		only 5 ♀♀					
emerged before collection of egg batches with												
opening on top of egg shell	3		0								0	
emerged after collection of egg batches with												
opening on top of egg shell	9		10								3 ♂♂	
opening on side of egg shell	1		0									
adults died in eggs without opening	1		0								1	
pupae died inside the eggs	1		0								4	
pupae parasitised by *B. transversalis*	0		0								2 + 23 larvae (4 with remains)	
Baryscapus transversalis Graham												
emerged before collection of egg batches	2		0									
emerged after collection of egg batches with												
opening on top of egg shell	9♀♀, 7♂♂		15♀♀								6♂♂	
opening on side of egg shell	6♀♀		0									
adults died in eggs without opening	5♀♀, 8♂♂		1♀								1♂	
undetermined larvae of parasitoids died in various stages	1338		446									

No specimen of *A. bifasciatus* emerged before collection of the egg batches. Thus, all adults and developmental stages were counted. All *B. transversalis*, except two specimens, and *B. servadeii*, except three individuals, emerged after sampling of the egg batches, independently from the collection date.

The egg batches collected on 2. 11. 94 contained two eggs in which two pupae were observed, one of *O. pityocampae* and one of *B. servadeii;* in two other eggs several small larvae were found.

3.4 *Sterile host eggs*

In most of the batches, a variable number of eggs was sterile; in the first generation studied 3% did not develop. In these eggs, dried-up yolk was found. The percentage increased up to 29.2% in the sample from 10. 9. 92, and was 4.9% in the sample collected one month before (Tab. 3). Also in the following years the amount of sterile eggs in the batches was relatively high (9.4 and 20.6%). A close observation of the sterile eggs showed that a small number of eggs was found in both samples, which were totally empty (without any remains).

In the sample from 14. 8. 93 two egg batches were found in which almost all eggs were sterile (yellow coloured): one of these batches (with 181 eggs) contained one egg from which a female of *O. pityocampae* emerged; from the other batch with a number of 160 eggs, three caterpillars hatched and one caterpillar died in the egg with an opening, additionally three eggs showed an opening through which *O. pityocampae* emerged, the identification being achieved by the characteristic meconia.

From the eggs collected on 2. 11. 94 seven batches contained numerous sterile eggs: four egg batches were found from which no caterpillar hatched and from 3 egg batches very small numbers (9–10) of caterpillars left the eggs, while in all of these egg batches many sterile eggs (up to 45%) were parasitised.

Table 3. Analysis of egg development and hatching of caterpillars of *Thaumetopoea pityocampa* (Den. & Schiff.) from batches sampled at Marikostino/Bulgaria in various generations

Date of collection	22. 10. 1991	13. 3. 1992	8. 8. 1992	10. 9. 1992	14. 8. 1993	2. 11. 1994
caterpillars died without opening	237 } 2.6%	319 } 3.8%	174 } 18.8%	783 } 14.2%	289 } 17.1%	415 } 6.4%
caterpillars died with opening	58	70	1074	272	113	111
undeveloped eggs with dried-up yolk	346 (3.0%)	318 (3.1%)	324 (4.9%)	2169 (29.2%)	222 (9.4%)	1705 (20.6%)
caterpillars hatched	6193 (54%)	5311 (52%)	4329 (65.3%)	1506 (20.3%)	1275 (54.1%)	3311 (40.1%)
eggs with undeveloped caterpillars	9	43				
eggs destroyed by predators						22
eggs totally empty, without any remains	162 (1.4%)	319 (3.1%)	111 (1.7%)	158 (2.1%)	2 (0.1%)	15 (0.2%)
total number of eggs from which no caterpillar hatched without influence of parasitoids	812 (7.1%)	1069 (10.5%)	1683 (25.4%)	3382 (45.5%)	626 (26.6%)	2246 (27 2%)
Impact of egg parasitoids	38.9%	37.5%	9.3%	34.2%	19.3%	32.7%

4 Discussion

4.1 Egg batch deposition and number of eggs

On *Pinus halepensis* Miller the female moth of *T. pityocampa* can form an egg batch using 1 to 6 needles, as well as small twigs (DEMOLIN, 1969; SCHMIDT, 1988, 1990; BELLIN et al., 1990; KITT and SCHMIDT, 1993). On *P. nigra* the egg batches found were wrapped almost exclusively on two needles, and the eggs were laid from near the base towards the tip of the needles, as found in Bulgaria in a former study (TSANKOV et al., 1996). The same behaviour was found in populations of *T. wilkinsoni* Tams in Israel on *P. halepensis*, by KITT and SCHMIDT (1993), in contrast to Greek populations of *T. pityocampa* which also oviposited on *P. halepensis* needles (BELLIN et al., 1990; SCHMIDT, 1990).

The mean numbers of eggs per batch, found at Marikostino in various years, varied from 203–253, which is in accordance with the records of TSANKOV (1972) and TSANKOV et al. (1996), where mean numbers of 223 and 233 eggs were calculated. The mean numbers of eggs per batch in Bulgaria are the highest ones reported in the literature.

Regarding the correlation of the length of the needles to the length of egg batches, it seems to be obvious that short egg batches were not found predominantly on short needles. The female moth may have the capacity to measure needle's length before producing an egg batch, but no experimental work has been done to test this hypothesis.

4.2 Egg parasitoid impact

At Marikostino, *O. pityocampae* was the only egg parasitoid having an important impact (23.2–34.0%) on the mortality of the host, when parasitism could take place during the whole embryonic development. In collections made in August during host embryonic development, the impact was lower (10.6% on 8. 8. 92 and 19.3% on 14. 8. 93), which is in accordance with the observations of BELLIN et al. (1990). From 22. 10. 91 to 13. 3. 92 the impact increased from 23.2% to 29.4%, however, during this cold period no additional parasitism would have been possible. The reason for the difference of the impact recorded before and after hibernation was the high number of undetermined larvae of parasitoids which died in various stages, in the autumn sample. When the latter were included, percentages of 35% and 33.8% were counted, and the impact of *O. pityocampae* has to be considered as being equal in both sampling periods. A similar impact (34%) was found in the samples of 10. 9. 92, and 31.7% on 2. 11. 94.

The impact of *A. bifasciatus* reached a maximum of 3.5% in the first generation studied and decreased to almost zero (0.2%) in the second and fourth generations. It was very low when the samples were collected before hatching of the caterpillars. This indicates that *A. bifasciatus* parasitised predominantly older embryonic stages of the host than *O. pityocampae*. This agrees with the observation that in only very few eggs parasitised by *O. pityocampae* remains of the host (sclerotised head capsules and larval leg parts) were found. For the first time, four females of *A. bifasciatus* were observed developing in *T. pityocampa* eggs in Bulgaria. HALPERIN (1990) found *A. bifasciatus* females only in *Thaumetopoea* eggs sampled in Cyprus.

The impact of *B. servadeii* was very low (only 0.2%). All individuals of this species emerged after collection of the egg batches, mainly in the following year. Thus, they could only parasitise the next generation of the host. Almost the same rate of emergence was observed in *B. transversalis*, a hyperparasitoid of *B. servadeii*. It seems obvious that the population of the latter was depressed by that of the former.

Trichogramma embryophagum was found only in a few eggs, and this parasitoid played no role in host control.

It can be stated that parasitism is going on during the whole embryogenesis of the host, and after hatching of the caterpillars from the egg batch the impact of the egg parasitoids cannot increase. Successful parasitism decreases during the last third of host embryonic development (KITT and SCHMIDT, 1993).

The decrease of the impact of the parasitoids, from 38.9 to 32.7% throughout the four generations studied, seemed to be influenced by the reduction of host population density, since it was much far more difficult to find egg batches after the period of egg laying and fewer winter nests were formed. The reduction of the host population density may be related to the use of pheromone baited traps in the summer of 1992 (BALDASSARI et al., 1994).

The impact of the parasitoids is not the only factor which reduced the caterpillars' viability. In the first generation studied, all other factors together caused between 7 to about 10% mortality. If the parasitoids could not attack the eggs throughout the whole period of development, the hatching rate of caterpillars was relatively high. In the samples, especially the ones from 10. 9. 92 and 2. 11. 94, it was remarkable that the hatching survival of larvae was considerably reduced to respectively 20.4 and 40.1%. From some of

Table 4. Percentage of *Ooencyrtus pityocampae* that emerged and found in closed eggs of *T. pityocampa* between 1991–1994 at Maricostino. In the August samples, the egg parasitoid was allowed to parasitise the host eggs for one or two weeks only, after oviposition

	collection dates of egg batches					
O. pityocampae	22. 10. 1991	13. 3. 1992	8. 8. 1992	10. 9. 1992	14. 8. 1993	2. 11. 1994
emerged before collection	1218 (46.1%)	954 (31.8%)	103 (14.7%)	730 (28.9%)	198 (43.5%)	1173 (44.9%)
emerged after collection	918 (34.8%)	1866 (62.1%)	474 (67.7%)	682 (27.0%)	170 (37.4%)	875 (33.4%)
total alive	80.9%	93.9%	82.4%	55.9%	80.9%	78.3%
adults died in eggs	275 (10.4%)	90 (3.0%)	25 (3.6%)	110 (4.4%)	19 (4.2%)	126 (4.8%)
pupae + larvae in eggs	230 (10.4%)	93 (3.1%)	98 (14%)	1003 (39.7%)	68 (14.9%)	440 (16.8%)
Total number	2641 (100%)	3003 (100%)	700 (100%)	2525 (100%)	455 (100%)	2614 (100%)

those egg batches no caterpillar hatched. Since no eggs are laid without mating, it can be assumed that the eggs were not fertilized. This may be linked to the application of pheromones, that removed only males from the population and thus left with a shortage of sexual partners.

4.3 Phenology of the egg parasitoids

TSANKOV et al. (1996) showed that in Bulgaria all parasitoid species are able of hibernating inside the eggs and of emerging in the next spring and summer. Only one fraction of the populations emerged in the same year of oviposition. As KITT and SCHMIDT (1993) demonstrated, *O. pityocampae* may be bi- or multivoltine in Bulgaria. If parasitism can take place very soon after egg deposition, the same egg batch may be attacked again as HALPERIN (1990) mentioned for Israel, but no experiments were done to prove it.

At Marikostino, *O. pityocampae* appeared throughout the whole year, except during the cold period (November, December, January). At room temperature, some parasitoids emerged also during the winter period (TSANKOV et al., 1996). Tbe highest rate of emergence was found in May. KITT and SCHMIDT (1993) found that this parasitiod can survive up to 10 weeks, under laboratory conditions, and can thus reach the next oviposition period of the host taking place in July–August. Under field conditions, the peak of emergence of *O. pityocampae* was shifted to the summer months. At Marikostino, a larger proportion of the diapausing population contributed to the parisitism of the egg masses of the host in the following season than reported for Israel by HALPERIN (1990).

A. bifasciatus emerged from December to June of the following year under laboratory conditions, and in the field emergence was observed mainly from April to June (TSANKOV et al., 1996).

The observations showed that temperature is one of the factors contributing to the termination of diapause undergone by many of the egg parasitoids as mature larva. In the winter season, a reduction of emergence was also observed under laboratory conditions.

Additionally, we found that many egg parasitoids emerged in the Fall of the same year of host oviposition, mainly before the host caterpillars hatched (SCHMIDT, 1988, TSANKOV et al., 1996). TSANKOV (1990) reported for Bulgaria that hatching of the caterpillars took place 5–7 days later than the emergence of most of the egg parasitoids. Another fraction of the adult population died inside the eggs, sometimes after making a hole for emergence.

In many cases, the remaining meconia of *O. pityocampae* showed that the egg parasitoids can also hibernate inside the eggs which were parasitised directly after oviposition. It is an open question, why some individuals go into diapause and while others emerge from the same egg batch and in a similar stage of development after about 2–3 weeks (HALPERIN, 1990).

Hyperparasitism reduces the effectiveness of the primary parasitoids. In Bulgaria and Greece, *B. transversalis* is an obligatory hyperparasitoid of the primary parasitoids, particularly of *B. servadeii* (BELLIN et al., 1990; BELLIN, 1995; TSANKOV et al., 1996; SCHMIDT et al., 1997). The reproduction of the hyperparasitoid lasted from the spring to the autumn, when the primary parasitoids are in diapause. At Marikostino, hyperparasitism took place, although the population density of *B. servadeii* was very low. Males and females of *B. transversalis* were observed in about equal numbers. TSANKOV et al. (1996) pointed out that *B. transversalis* is less successful in larvae of *O. pityocampae* than in those of *B. servadeii*.

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Zusammenfassung

Untersuchungen zum Eiparasitismus von Thaumetopoea pityocampa bei Marikostino/Bulgarien während einer Vierjahresperiode (1991–1994)

In Bulgarien, in der Nähe von Marikostino, wurden 239 Eigelege des Pinienprozessionsspinners *Thaumetopoea pityocampa* (Den. & Schiff.) (Lep., Thaumetopoeidae) in einem *Pinus nigra* – Forst über 4 Generationen zu verschiedenen Jahreszeiten gesammelt. Die Gelege wurden sogleich einzeln in Reagensgläser gegeben, die mit Wattestopfen verschlossen wurden, und unter Laborbedingungen bei 20–22 °C aufbewahrt. Die Eiablage fand stets von der Nadelbasis zur Spitze hin statt.

Der Schlupf der Parasitoiden wurde täglich registriert. Nach Entfernen der Gelegeschuppen wurde jedes Ei markiert, aus dem ein Parasitoid schlüpfte. Alle adulten Parasitoide wurden sogleich nach dem Schlupf entfernt und zur Determination vorbereitet. Nach Ende der Schlupfperiode (mindestens 1 Jahr nach dem Raupenschlupf) wurden alle Eier geöffnet. Der Einfluß der einzelnen Parasitoiden-Arten auf die Eimortalität wurde anhand der Mekonien und Rückstände in den Eiern bewertet.

Ooencyrtus pityocampae (Mercet) war in den Eiern die häufigste Art, gefolgt von *Anastatus bifasciatus* (Fonsc.). Von *Baryscapus servadeii* (Dom.) wurden nur wenige Individuen gefunden. Auch der Hyperparasitoid, *Baryscapus transversalis* Graham, war nur in geringer Populationsdichte vertreten. Nur wenige Eier waren von *Trichogramma embryophagum* Htg. parasitiert. Die Gesamtmortatität der Wirtstiere variierte von 34,7% bis 79,7%; sie war abhängig von der Parasitierungsperiode und der Menge an sterilen Eiern. Der Einfluß der Eiparasitoide betrug 32,7–38,9%, wenn sie die gesamte Embryonalentwicklung des Wirtes zur Parasitierung nutzen konnten.

Die mittlere Anzahl der Eier pro Gelege variierte in den Sammelproben von 203 bis 253. Die Parasitierungsrate war nach frühem Einsammeln der Gelege (nach Oviposition des Wirtes) stark reduziert. In einigen Fällen war ein sehr hoher Prozensatz (bis zu 29,2%) an nicht entwickelten Eiern zu finden. In allen Eigelegen waren einige Eier vorhanden, die total leer waren.

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