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RESEARCH ARTICLE

Flight characteristics of the summer and winter phenological form of the pine processionary moth (*Thaumetopoea pityocampa*) according to data from automatic pheromone traps near Asenovgrad in Bulgaria

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Abstract

The winter and summer phenological forms of the pine processionary moth (*Thaumetopoea pityocampa*) were recorded in two adjacent sites in the region of Asenovgrad, South Bulgaria. In the land of Muldava village (at the foot of the Rhodopes at 290 m a.s.l.), the typical winter phenological form was established, while the summer form was found in the vicinity of Dobrostan village (on the northern slopes of the Rhodopes at 1200 m a.s.l.). The calculated air fly distance between both villages is 9.3 km. In 2019, funnel automatic pheromone traps – CapTrap® were installed in Dobrostan and Muldava villages to obtain information about the flight dynamics of the male butterflies of *T. pityocampa*. For the studied period (June-September), hourly data on the humidity and air temperature in both sites were also analysed. In both sites, the flight started at the beginning of July and ended in the first decade of September. Two clear peaks in the flight of male butterflies were observed on 7-8 and 23-28 July, which was associated with catches of both phenological forms due to the proximity of the traps. In Dobrostan, no high maximum temperatures above 30-32 °C were measured in the days during the period July-September. In Muldava, critical temperatures for the survival of *T. pityocampa* were recorded in this period: July (61.3% of the days), August (87.1%) and September (23.3%).

Keywords
Thaumetopoea pityocampa, phenological forms, CapTrap®, critical temperatures, South Bulgaria

Introduction

The pine processionary moth, *Thaumetopoea pityocampa* (Denis & Schiffermuller, 1775) (Lepidoptera, Thaumetopoeidae), is a Mediterranean species distributed in pine (*Pinus* spp.) ecosystems in countries of the region. The northern border of the species passes through Bulgaria. In the first half of the 20th century, it was reported that the early (summer) phenological form of the species also occurs in our country, which completes its larval development in autumn and overwinters in the soil (Zankov, 1960). In studies on the distribution zones of the both phenological forms (Mirchev et al., 2017; Georgieva et al., 2018; 2019), it was indicated that in the region of Asenovgrad, the summer form occurs near Dobrostan village, and the typical winter form – near Muldava village (Mirchev et al., 2019).

Temperature is the main limiting factor determining the ecological niche of the distribution areas of *T. pityocampa*. For the summer months, Santos et al. (2011a,b) determined such a temperature limit of 32 °C.

Sex pheromones are multifunctional in their application. They are used as: a modern method to clarify aspects of the biology, ecology and phenology of the species (Athanassiou et al., 2007; Subchev et al., 1994a,b); to regulate the number of male butterflies (Einhorn et al., 1983), and from there on the size of the population (Trematerra, Colacci, 2019); potentially for forecasting purposes (Jactel et al., 2006), etc. Chenchouni et al. (2010) identified them as an important tool for estimating population density, adult imaginal time and, by disrupting mating through mass capture, to limit future outbreaks. These possibilities of pheromone traps are expanded with modern automatic traps that collect hourly data on the flight dynamics of male butterflies, air temperature and humidity and transmit them online.

The aim of this study was to present information about the flight characteristics of the male butterflies of the early and late forms of *T. pityocampa* in the area of Dobrostan and Muldava villages based on data on automatic pheromone traps, and to analyse the climatic conditions during the appearance of the pest.

Material and methods

The studies were conducted in 2019 in the area of Muldava village, where the winter phenological form of *T. pityocampa* develops, and Dobrostan village where the summer form occurs. The air fly distance between both sites is 9.3 km (Fig. 1).

Muldava is located 2 km away from the 'Chetirideste Izvora' dam at the foot of the Rhodopes at 290 m a.s.l. and the Muldavska River passes through the village. Dobrostan village is located on the northern slopes of the mountain at 1200 m a.s.l.

Fig. 1. Investigated localities of *Thaumetopoea pityocampa*

In this study, two funnel automatic traps CapTrap® were installed at a height of 4 m on 1 June 2019. The solar panels were oriented to be well exposed to the sun, so that during most of the day there was direct light. Both studied sites have GSM coverage, a necessary condition for hourly data transmission to the server. The CapTrap® service continuously provides counting data to send out real time notifications and charts to users via a Web interface. Dispensers in the pheromone traps were replaced monthly. The data were statistically processed using MS Excel (2016).

Results

Air temperature

Both surveyed sites in Dobrostan and Muldava are close to each other at 9.3 km in a straight line, but have significant differences in the altitude. Dobrostan's altitude is 910 m higher than that of Muldava. This determines the temperature regime in both sites. During the three months of measurements, the average monthly temperatures were about 6 °C higher in Muldava as follows: in July it was 6.8 °C, in August – 5.9 °C, and in September – 5.4 °C (Table 1). The differences in the daily average temperatures in the two locations were also of this order (Fig. 2).

Table 1. Average monthly temperatures and air humidity in Dobrostan and Muldava

Month	Average monthly temperature, °C		Average monthly air humidity, %	
	Dobrostan	Muldava	Dobrostan	Muldava
July	17.2±2.9	24.0±3.4	82.0±14.0	78.8±12.4
August	19.1±1.9	25.0±1.6	78.6±10.0	74.1±7.8
September	15.7±2.8	21.1±2.8	87.9±12.0	80.8±10.8

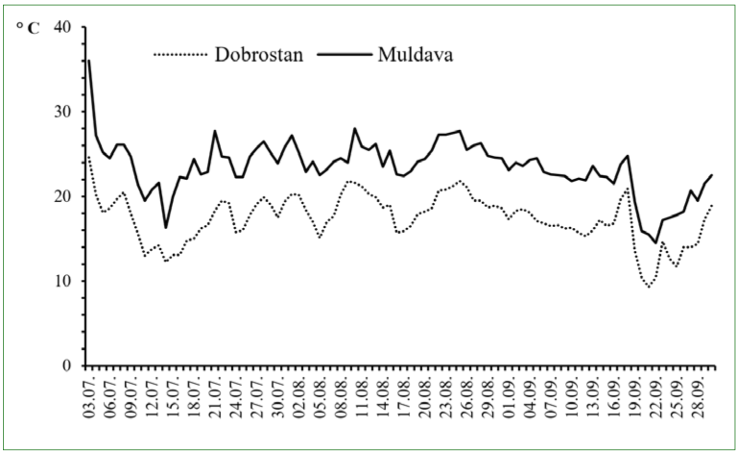

Fig. 2. Average daily temperatures in Dobrostan and Muldava

In Dobrostan, high maximum temperatures exceeding 30-32 °C were not measured in July-September. In Muldava, critical temperatures for the survival of *T. pityocampa* were recorded in July (61.3% of the days), August (87.1%) and September (23.3%) (Table 2).

Table 2. Days with critical maximum temperatures in Muldava

Month	Temperatures, 30-32 °C		Days with critical maximum temperatures			
			Temperatures above 32 °C		Σ Days with critical temperatures	
	Number	%	Number	%	Number	%
July	8	25.8	11	36.7	19	61.3
August	7	22.6	20	64.5	27	87.1
September	4	13.3	3	10.0	7	23.3

Air humidity

Unlike the air temperature, the differences in the air humidity values were not so significant in both locations. The predominant case was with higher humidity in Dobrostan (Fig. 3). The close values of this parameter in Muldava, which is located at a significantly lower altitude, were compensated by the conditions created by the river

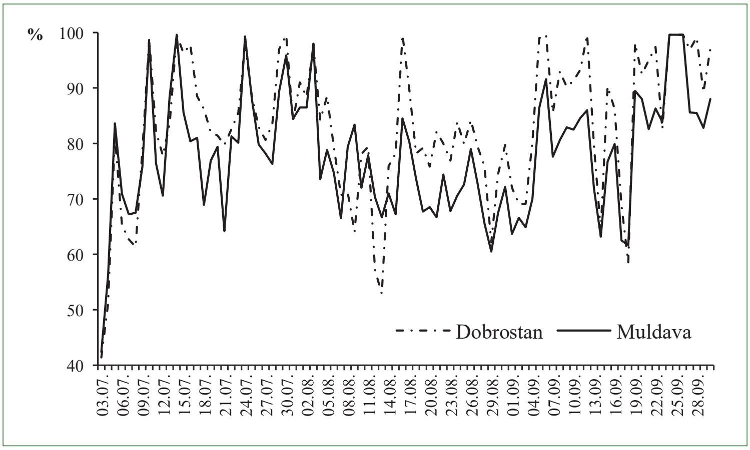

Fig. 3. Average daily air humidity in Dobrostan and Muldava

passing through the village and especially by the relatively large water basin located nearby, the 'Chetirideste Izvora' dam.

The conclusions about significant differences in the temperature regime in the two objects and not so significant in the values of air humidity are confirmed by the statistical processing of the data, for example, according to the second indicator, they are significantly smaller or for the first month of measurements – July, there are none at all (Table 3).

Table 3. Comparison of environmental parameters measured in Dobrostan and Muldava, by means of T-Test, significance level α=0.05

Parameters		T-Test			
		3-31 July	1-31 August	1-30 September	3 July-30 September
Temperature	Average	1.47943E-11	8.34825E-20	2,52435E-10	3,65767E-29
	Minimal	0.00054501	6.44418E-07	0,000386812	8,32849E-09
	Maximum	3.87751E-12	3.48402E-20	2,82268E-10	2,44738E-30
Air humidity	Average	0.186636078	0.027333753	0,010992907	0,002868686
	Minimal	0.033067558	0.001352376	0,001902222	1,54332E-05
	Maximum	0.086316651	0.006512003	0,07537451	0,003371969

Flight of the pine processionary moth

In Dobrostan, the amount of male butterflies caught are nearly 5 times (4.6) more than in Muldava (Table 4). In both sites, the flight starts practically at the beginning of July and ends in the first ten days of September. In Dobrostan, a butterfly caught on November 15th can be considered an extraordinary exception. There are two peaks in

Table 4. Flight of male butterflies of *T. pityocampa* in Dobrostan and Muldava

Site	Caught butterflies, n	Start of the flight	End of the flight	Time median, T50	Time percentile, T90
Dobrostan	421	3 July	15 November	23 July	13 August
Muldava	91	7 July	10 September	28 July	17 August

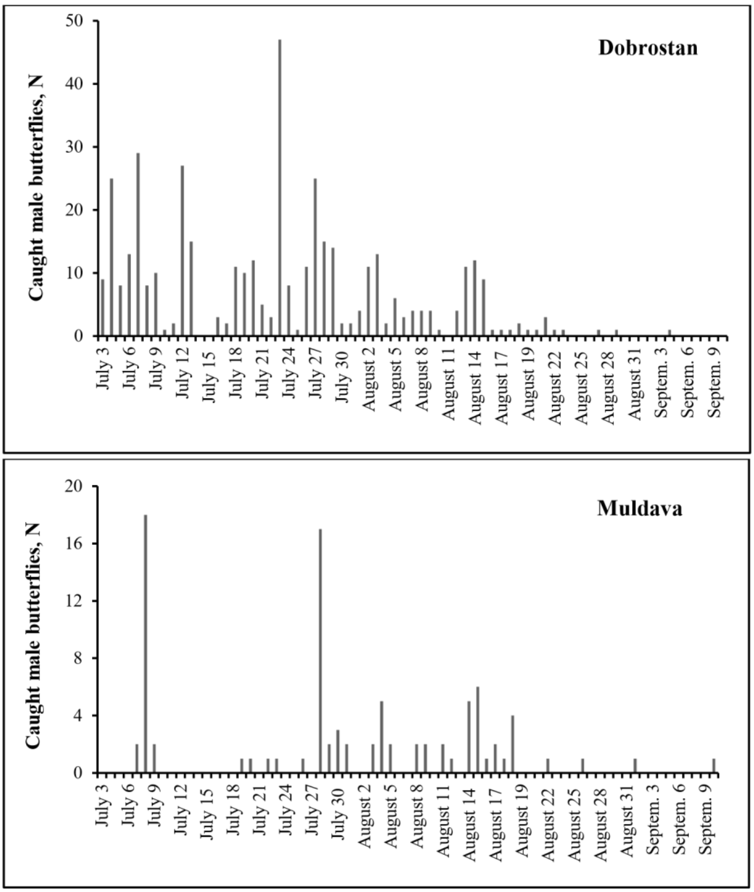

Fig. 4. Flight dynamics of male pine processionary moths in Dobrostan and Muldava

the flight of male butterflies – on July 7-8 and July 23-28 (Fig. 4), which is most likely related to catches of the two phenological forms due to the proximity of the two traps.

In both sites, half the number of butterflies flew out till the end of July, with a difference of 5 days between them. In Dobrostan, the limit of 50% was reached on July 23rd, and in Muldava on July 28th. In the time percentile (T_{90}), the difference was 4 days, August 13th in the first object, and, respectively - August 17th in the second (Table 4, Fig. 4).

The flight takes place at night, between 9 pm and 6 am. The time median (T_{50}) in Dobrostan is reached half an hour after midnight, and T_{90} – after 2 hours (Fig. 5). An interesting phenomenon was observed in Muldava – 20.1% of butterflies were caught early in the morning at 6 am and T_{50} at 2 am.

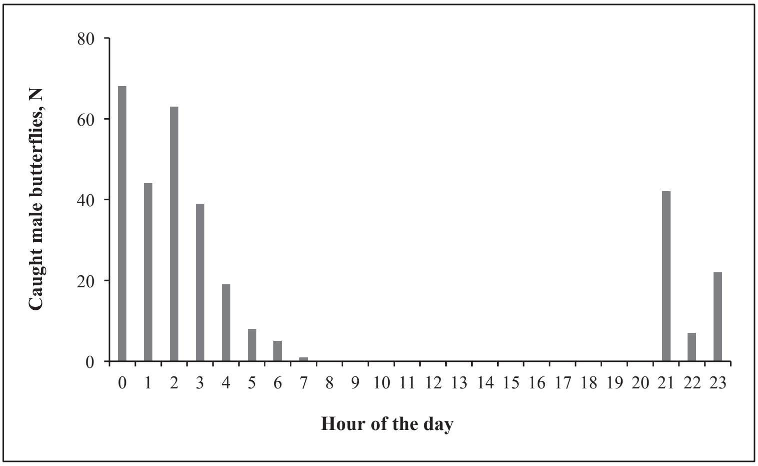

Fig. 5. Diurnal flight dynamics of the male butterflies of *T. pityocampa* in Dobrostan

Discussion

Climatic conditions are the main factors determining its suitability for the successful development of the pine processionary moth. Summer temperatures above 32 °C (Huchon, Demolin, 1970; Santos et al., 2011a,b) or 40 °C (Robinet et al., 2013) are considered lethal to eggs and newly hatched caterpillars. The impact of temperature should not be considered in isolation, but in a complex, in combination with other elements of the environment. Mirchev et al. (2016) reported that under low winter temperatures, caterpillar mortality increased with increasing air humidity. In this aspect, rather than as a fixed limit, the threshold of 32 °C should be accepted. In a large-scale study on this issue in Bulgaria, it was established that in both phenological forms, the flight of the pine processionary moth takes place during the hottest months of the year, when these air temperatures are exceeded. In this period, in the range of the species in the country, temperatures above 40 °C were recorded at the end of July and the beginning of August only in the vicinity of the same research area (Asenovgrad) and Sandanski in Southwestern Bulgaria (Zaemdzhikova et al., 2020).

The flight perimeter of male butterflies is relatively large, reaching up to 50 km (Mirchev et al., 2013). In the two phenological forms, there are no morphological differences in the butterflies and considering the close distance, about 10 km between the two sites, it is not possible to determine in each of the samples whether and what part of the butterflies are from the other phenological form. In this aspect, the data on the flight dynamics of adults and their numbers are too conditional.

An early phenological form of *T. pityocampa* was also found in Portugal (Santos et al., 2013). The evolutionary development of the pine processionary moth allows it to develop in an ecological niche in a certain range of environmental temperature conditions. Huchon, Demolin (1970) and Robinet et al. (2013) reported that high temperatures were lethal to eggs and newly hatched larvae.

The close coexistence of the two phenological forms of the pine processionary moth in the region of Asenovgrad raises the question of possible hybridization between them, an indication of which is the presence of two peaks in the catches of male butterflies in the traps in Dobrostan and Muldava. For sympatric Portuguese populations, adults of the two forms were found to remain reproductively isolated despite inhabiting the same locality (Santos et al., 2007). Research so far has not proven the existence of a genetic barrier between the two forms. On the contrary, mating has been shown to be possible in a laboratory experiment (Branco et al., 2017; Servedio et al., 2011). Apart from phenological differences, sympatric Portuguese populations also differ in some adaptive, morphological and biological characteristics (Santos et al., 2011a,b; 2013).

Similar to the situation in Leiria, Portugal (Pimentel et al., 2006), the studied areas in Bulgaria are also inhabited by sympatric populations of *T. pityocampa* – summer and winter phenological forms with an overlapping range. Higher adaptability of the summer form to high temperatures (Santos et al., 2011a), differences in egg size and eggshell color, and fecundity of female butterflies have been found in Portuguese sympatric populations (Santos et al., 2013).

In conclusion, it could be noted that the long-term monitoring showed the presence of 'pure' phenological forms of *T. pityocampa* in the Asenovgrad region: a summer form in the vicinity of the village of Dobrostan and a winter form in the village of Muldava, despite the obvious possibilities of hybridisation between them. In this regard, it is desirable to conduct genetic studies of the pine processionary moth in Bulgaria, especially of the two phonological forms, as a first step to study the mechanisms of differentiation of the flight period of the species.

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