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RESEARCH ON THE APPEARANCE AND EVOLUTION OF *MONARTHROPALPUS BUXI* (LABOULBÈNE) IN THE BOXWOOD PLANTATIONS OF IASI COUNTY

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Abstract

Boxwood species, valued for their ecological adaptability, face growing threats from pests, notably the boxwood leafminer (Monarthropalpus buxi Laboulbène), which has severely impacted landscapes and nurseries in recent years. The aim of this study was to monitoring the evolution and ecology of Monarthropalpus buxi according to the local biotope and testing two insecticides in order to control the pest in North-Eastern part of Romania. The life cycle of boxwood leafminer was described in relation to GDD. The appearance of the first adults in 2022 was recorded at an accumulation of 196.2 GDD on 09th May, and the appearance of adults began at an accumulation of 191.7 GDD on 02nd May in 2023. The maximum flight curve was recorded at accumulations between 262.7 GDD (2022), respectively 246.3 GDD (2023). Last appearance of adults was observed on 28th May (2022) and 25 Mayth (2023). The larval stages in 2022 developed at accumulations between 675.7 - 1298.3 GDD and in the following year the larvae developed between 670.4 - 1317.1 GDD. Statistical methods were used to interpret the obtained results.

Key words*: buxus, boxwood leafminer, Growing Degree - Days, monitoring, temperature.*

INTRODUCTION

Boxwood (*Buxus* spp.) are valued in landscape design for the characteristics of its foliage and versatility. They are used in landscaping in a range of sizes and shapes, from low-growing varieties suitable for hedges and edging to taller specimens that can be used as focalized points or izolated plants. This versatility allows them to be used in various ways to create different effects in the landscape. The permanent interest in this plant has increased the production of boxwood in nurseries all over the world (Weiland et al., 2022).

Ever since the last century, the cultivation and production of boxwood has been challenged by the attack of several harmful arthropods and the threat of pathogens with negative effects on the general appearance of the plant (Brewer et al., 1980; Baylac & Daufresne, 1996; Ivors et al., 2012; Dhakal et al., 2022).

Boxwood leafminer, known scientifically as *Monarthropalpus buxi* or *Monarthropalpus flavus* (Diptera: Cecidomyiidae), is a serious key pest of boxwood, both in the landscape and especially in nurseries (d`Eustachio & Raupp, 2001). This monophagous small fly is native to Europe, but widespread in all buxus cultivation areas distributed including in United States of America. In Romania, *Monarthropalpus buxi* was described by Perju et al. (2001) and it was reported in the urban areas of Cluj County and in the parks of Bucharest (Soporan et al., 2012; Soporan et al., 2015; Ciceoi et al., 2017).

The larvae of this pest cause significant damage to the boxwood at the level of the leaves, feeding exclusively on the leaf parenchyma. The presence of *Monarthropalpus buxi* larvae and the way of feeding causes the appearance of discolored galls on the surface of the leaf, mostly located near the midrib. The development dynamics of the larvae during the growing season is not uniform. This differs from one stage to another and is primarily influenced by the atmospheric temperature. studies conducted on this species showed that the egg stage of the boxwood leafminer was at an accumulation between 352-748 GDD (F), while the larval stages developed at accumulations between 679 and 2442 GDD (F) (d'Eustachio & Raupp, 2001). On the other hand, the development and survival of the

larvae is also influenced by the morphological or physiological aspects of the leaf.
Soporan et al. (2012) showed that the larvae were smaller on the leaves with less leaf surface. also, the positioning of the infested leaves in the crown of the boxwood directly influences the size of the larvae, also increasing the duration of the larval stages. Thus, in poorly lit leaves or placed in the lower part of the branches, the smallest larvae develop and have a long development period. This highlights the relationship between the physiological processes of the plant and the development of *Monarthropalpus buxi* larvae. The intense chlorophyll assimilation provides a rich source of food for the larvae and their development takes a shorter period.
Frequent and persistent attacks caused by this pest cause the death of branches and the susceptibility of plants to biotic or abiotic stressors. Cultural management is very important in the control of this pest and involves both monitoring methods by capturing adults, as well as chemical control methods by applying systemic or contact insecticides. The most recent studies have focused on chemical control, by applying insecticides, since there are few natural enemies of this pest, and biological control is significantly reduced (Raupp et al., 2004; Dunn & Saunders, 2014; Yoder et al., 2022).
The susceptibility of *Buxus* sp. before the attack of this pest causes significant economic losses in the production of boxwood. The newly formed plants are very vulnerable, regardless of the cultural practices approached in nursery technology.
The aim of this paper was to monitoring the evolution and ecology of *Monarthropalpus buxi* according to the local biotope and testing two insecticides in order to control the pest in an ornamental plant nursery from Iași County. Being a species reported for the first time in the North-Eastern area of Romania, the results obtained will constitute an additional source of documentation for further research.

MATERIALS AND METHODS

The study was carried out in two consecutive years, 2022 and 2023 in a production subunit of National Forest Administration - ROMSILVA. The location of the ornamental plant nursery is in the North-Eastern part of Romania, in the metropolitan area of the Iași city, namely Ornamental Plant Nursery – Galata, (47°14´N and 27°55´E).
The natural environment where the research took place is located in the forest steppe of Moldova on a geomorphological unit characterized by a cambic chernoziom soil. According to the Köppen-Geiger classification, the climate of this area was considered Dfb type with an average multiannual temperature of 10.2°C and a significant rainfall of 562.6 mm, distributed evenly throughout the year. The climatic factors were monitored with the meteorological FieldClimate system, located in the experimental field.
The biological material includes production fields with boxwood varieties of different ages. All samplings were multiply vegetatively, planted in the production fields at distances of 0.5 m × 0.7 m. During the growing season, maintenance of the fields was carried out with soil mobilization, fertilisation and irrigation typical for nursery production technologies.
In order to obtain conclusive results regarding the appearance and evolution of *Monarthropalpus buxi*, the observation, recording and statistical processing of data were used as research methods. During the experimental period the appearance of adults was monitored using colored glued traps: white, yellow and blue. The traps were placed every year on April 20th. from the first appearance of the adults, they were observed and inventoried daily until the end of May.

Figure 1. The placement of traps in the experimental fields

To predicting pest activity and timing control tactics were used Growing Degree-Day models. To calculated GDD for monitoring the evolution of *Monarthropalpus buxi* was used the followed mathematical equation:

$$GDD=\frac{Tmax+Tmin}{2}-threshold\ temperature,$$

where T_{max} and T_{min} are daily max and minim temperatures, threshold temperature= 10°C (Davidson and Raupp, 1999).

The GDD calculation started on March 1, because in the monthly averages of January and February they did not exceed the biological threshold of this species. According to GDD, the development of the insect's life stages was monitored. To determine the evolution of larvae stages were analysed in the laboratory thirty infested leaves, during vegetation period.

Another aspect pursued in the study was the establishment of the effectiveness of two insecticides applied during the flight period of the adults. For testing, two insecticides were compared: acetamiprid (Mospilan 20 SG) in a concentration of 0.15% and lambda-cyhalothrin (Karate Zeon) in a concentration of 0.015%. The substances were applied to the plot of boxwood mother plants, and the effectiveness was measured taking into account the dynamics of the population level before and after the treatment. They were monitored with the help of glued traps, their number being evaluated 24 hours, 48 hours and 7 days after the treatment. The obtained results were interpreted statistically.

RESULTS AND DISCUSSIONS

Boxwood (*Buxus* spp.) is part of the category of ornamental species known for its adaptability to different growing conditions, which makes it a versatile choice for landscaping. Climate, in relation with pedological factors, represent the parameters determinants in increasing or decreasing the productivity of agro-ecosystem. The temperature influences the growth and development of boxwood plants. Different species and cultivars have varying temperature requirements and tolerances (Hanaka et al., 2021; Peng et al., 2021). While some boxwoods varieties are cold-hardy and can withstand freezing temperatures, others are more sensitive to frost and require milder climates. Extreme heat can also stress boxwood plants, especially if they are not adequately watered (Ingram et al., 2015).

Adequate moisture is essential for the growth of boxwood, especially during the establishment phase. Boxwood plants prefer consistently moist but well-drained soil. In regions with low rainfall, supplemental watering may be necessary to maintain soil moisture levels, particularly during dry periods. However, excessive waterlogging can lead to root rot and other problems, so it's essential to ensure proper drainage (Helaly, 2017).

In first table (Table 1) were presented the monthly average temperature and total rainfall during the studied period. The deviation from the thermal multiannual average was also calculated. Thus, in the investigation period (2022-2023) the average annual temperature was 12.0°C with a positive deviation of 1.5°C compared to the multiannual average (10.5°C). The accumulated monthly precipitation during the studied period recorded a value of 363.6 mm, 154.0 mm less than the multiannual limit. In general, rainfall was unevenly distributed throughout the year, with a minimum in January of 9.4 mm and a maximum in July of 59.7 mm.

Table 1. Monthly and annual average of the thermal and rainfall (Ornamental Plant Nursery - Galata, Iași, 2022-2023)

Month	Air temperature (°C)			Precipitation (mm)		
	Av.	Multiannual	Deviation	Sum	Multiannual	Deviation
I	1.6	-1.9	+3.5	9.4	35.5	-19.6
II	2.7	-1.2	+3.9	16.9	32.1	-10.6
III	5.0	4.7	+0.3	31.4	71.2	+3.3
IV	9.9	11.4	-1.5	46.5	51.4	+6.2
V	16.4	17.0	-0.6	11.2	71.1	-41.3
VI	21.3	20.5	+0.8	17.3	82.9	-57.8
VII	23.3	22.4	+0.9	59.7	64.7	-9.6
VIII	24.0	21.9	+2.1	38.6	50.8	-19.0
IX	17.7	16.3	+1.4	40.3	36.5	-0.5
X	13.5	10.1	+3.4	19.3	34.4	-15.1
XI	6.7	5.4	+1.3	55.1	33.4	20.5
XII	1.7	0.1	+1.6	18.1	30.6	-10.8
Av./Sum	**12.0**	**10.5**	**+1.5**	**363.6**	**517.6**	**-154.0**

Even if in the experimental fields of the nursery this water deficit was regulated with the help of irrigation, recent studies show that deficit irrigation can be a strategy for controlling the growth of ornamental plants and increasing their ability to adapt to drought conditions (Sánchez-Blanco et al., 2019).

From another perspective, climatic factors also influence specific pests of boxwood plants. The climatic factors interact with various ecological variables to shape the evolution and population dynamics of *Monarthropalpus buxi*. Seasonal variations in temperature and day length influence the phenology of *Monarthropalpus buxi.* The insect's activity peaks during the warmer months when host plants are actively growing and producing new foliage. Winter dormancy or diapause may occur in colder climates, allowing the insect to survive adverse conditions until favorable conditions return (Raupp et al., 2002).

Figure 2 highlight the flight season adults of *Monarthropalpus buxi* in 2022 and 2023, monitored and captured using colored traps. Under the conditions of 2022, the first adults were captured on 9th May on the white panels, and in the following days, colored traps were also occupied. The maximum flight curve was on 17th May when 1108 adults were captured (723 adults on white traps; 288 adults on yellow traps and 97 adults on blue traps). After that date, the number of adults gradually decreased, and the last ones captured were on 28th May. In 2023, spring temperatures were higher, causing the pest to appear earlier. The first adults were captured on 2nd May on the white panels, on 4th May on the yellow panels and on 5th May on the blue panels. The maximum flight curve was on 13th May, when the largest number of captures was obtained – 1471 adults. Their last flight was recorded on 22th May 22 on the blue panels, and adults were captured on the other two until 27th May. During the study period, a total of 12999 adults were captured.

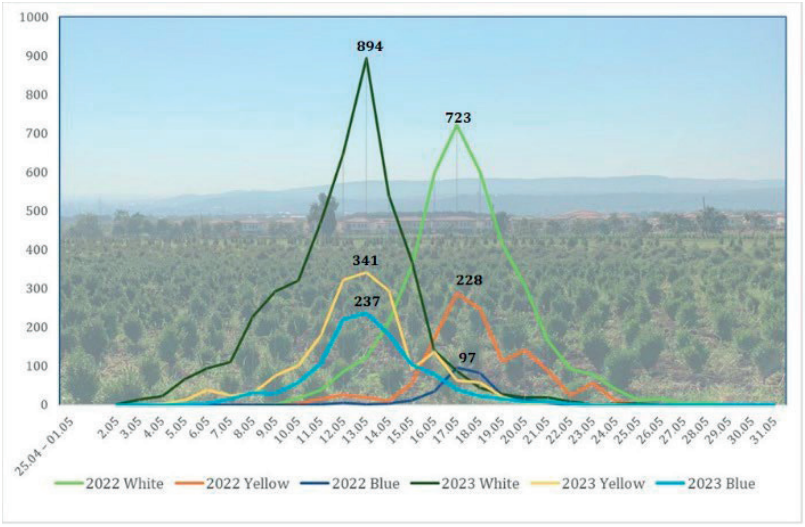

Figure 2. The flight curve of adults captured during the study period

The study on the capture of adult populations using chromotropic traps of different colors, carried out within the ornamental plant nursery, during the period 2022-2023, highlights significant variations in their effectiveness, influenced by the behavioral preferences of the studied species and by local abiotic factors (Table 2). Comparative analysis of empirical data demonstrates that, in both experimental seasons, white traps had a significantly higher attractiveness than yellow and blue ones, capturing a much higher number of individuals, with an increase from 185.9% compared to the control in 2022 to 204.5% in 2023. In contrast, yellow traps showed an intermediate effectiveness, oscillating between 58.7% of the control value in 2022 and 83.7% in 2023, suggesting a variable sensitivity of

Monarthropalpus buxi adults to this chromatic spectrum, correlated with seasonality and abundance of available trophic resources.

On the other hand, blue traps proved considerably less effective, recording only 13.2% of the control value in 2022 and a modest increase to 54% in 2023, which indicates a low receptivity of insects to this chromatic spectrum, according to previous studies on the negative phototactism of some entomological species to blue light (Otieno et al., 2018). The observed differences are supported by statistical analyses, with white traps recording highly significant differences ($p < 0.01$ in 2022 and $p < 0.001$ in 2023), while the efficiency of yellow and blue traps did not reach relevant statistical significance thresholds, being labeled as "Mt" (non-significant).

Also, comparing the results obtained, it is observed that following the application of the Duncan Test there are significant differences between the variants.

Table 2. The level of adult populations captured with colored traps (n=6, Ornamental Plant Nursery - Galata, Iași, 2022-2023)

Colour traps	Year	Samples	% of control	Difference to control	The semnificance of difference	Duncan Test [1]
White	2022	4027	185.9	+1861	**	A
Yellow		1272	58.7	-895	Mt	C
Blue		287	13.2	-1880	oo	D
White	2023	4430	204.5	+2264	***	A
Yellow		1814	83.7	+645	Mt	b
Blue		1169	54.0	-998	Mt	c
Average (control)		2167	100	-	-	-
DL (P 5 %) – 1013.82; DL (P 1 %) – 1423.07; DL (P 0.1 %) – 2009.04						

[1]different letters correspond with the significant statistical difference for $P \leq 5\%$, Duncan test.

The development of boxwood leafminer (*Monarthropalpus buxi*) in relation with GDD accumulations was reported in Table 3. The boxwood leafminer adults developed at accumulations between 196.2-329.7 GDD in 2022, and in 2023 they appeared following accumulations of 191.7-324.6 GDD. The first eggs were observed at 329.7 GDD in 2022 and next year at 324.6 GDD. This identification was observed on the new growth leaves. The larval stages evolved between 587.9-1298.3 GDD in 2022. The following year accumulation degree were included between 324.6-1317.1 GDD. The transformation of 4th larvae stage into pupae requires a low temperature stage, below 0°C. Therefore, for this period was accumulated minimum 98.6-196.2 GDD (2022) and 117.9- 191.7 (2023).

Table 3. The evolution of *Monarthropalpus buxi* in relation to the accumulation of Growing Degree-Days - GDD (°C)

No.	Life stage	Year	GDD (°C)			Period
			First occurance	Peak occurance	Last occurance	
1.	Pupa	2022	98.6-196.2			April -May
		2023	117.9-191.7			
2.	Adult	2022	196.2	262.7	329.7	May
		2023	191.7	246.3	324.6	
3.	Egg	2022	329.7-587.9			May -June
		2023	324.6-602.1			
4.	1st instar	2022	587.9-675.7			June
		2023	602.1-670.4			
5.	2nd instar	2022	675.7-984.3			July- September
		2023	670.4-991.5			
6.	3rd instar	2022	984.3-1298.3			September- March
		2023	991.5-1317.1			
7.	4th instar	2022	0-98.6			March -April
		2023	0-117.9			

Figure 3 shows the life stages of the boxwood leafminer determined in the laboratory during the study period. Another aspect pursued in the study was testing the effectiveness of two insecticides to combat the pest. Table 4 shows the results obtained in the two consecutive years regarding the distribution of *Monarthropalpus buxi* adults before and after the application of the treatment. In combating boxwood leafminer (*Monarthropalpus* spp.), both systemic and contact insecticides can be effective depending on the severity of the infestation and the specific circumstances of the situation.

Figure 3. Life stages of *Monarthropalpus buxi* determined in the study period

In our study we obtained a high efficiency of the two substances: acetamiprid registering in the two years an efficiency between 95.6-100%, and the efficiency of lambda-cyhalothrin registered values between 97.8-99.7%

Table 4. The effectiveness of insecticide application in combating *Monarthropalpus buxi*

		V1				V2			
		Acetamiprid				*Lambda-cyhalothrin*			
		2022	Eff.(%)	2023	Eff.(%)	2022	Eff.(%)	2023	Eff.(%)
No of captures before treatment		478	-	623	-	365	-	557	-
No of captures after treatment	24 hours	21	95.6	18	97.1	7	98.1	12	97.8
	48 hours	13	97.2	9	98.5	3	99.1	5	99.1
	7 days	2	99.9	0	100	1	99.7	2	99.6

CONCLUSIONS

Boxwood leaminer (*Monarthropalpus buxi*, Diptera, Cecidomyidae) has become a key pest of boxwood varieties in recent years, causing damage to the general appearance of the plant and also affecting the photosynthetic activity of the leaves.

The climatic factors interact with various ecological variables in the evolution and population dynamics of *Monarthropalpus buxi*. Seasonal variations in temperature and day length influence the phenology of *Monarthropalpus buxi*. In the conditions of North-Eastern part of Romania, the life cycle of *Monarthropalpus buxi* was monitored in relation to GDD. The first adults appeared between 191.7-196.2 GDD (°C), and the maximum flight time was at accumulations between 246.3-262.7 GDD (°C). Larval stages developed between 670.4-1317.1 GDD (°C).

Combating this pest is effective by combining physical methods (colored calpans) with chemical methods (insecticides). During the study period, a total of 12999 adults were captured, the most catches being registered on the white traps. In combating boxwood leafminer (*Monarthropalpus* spp.), both systemic and contact insecticides can be effective depending on the severity of the infestation and the specific circumstances of the situation.

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