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**EFFECTIVENESS OF THE PREDATORY BUG *MACROLOPHUS PYGMAEUS*  
(HEMIPTERA: MIRIDAE) IN THE CONTROL OF THE GREENHOUSE  
WHITEFLY – *TRIALEURODES VAPORARIORUM* (HEMIPTERA:  
ALEYRODIDAE)**

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**Abstract**

This study evaluates the efficacy of the predatory bug (*Macrolophus pygmaeus*) as a biological control agent against the greenhouse whitefly (*Trialeurodes vaporariorum*) in greenhouse-grown pepper (*Capsicum annuum*). The experiments were conducted under controlled greenhouse conditions in six experimental cages, involving two treatments: *M. pygmaeus* + *T. vaporariorum*, and an untreated control (without predatory insects).

The results demonstrate that the population density of *T. vaporariorum* remained significantly lower in the cages with *M. pygmaeus* compared to the untreated control. Specifically, the average number of whiteflies was 45.7% lower within the first 14 days and decreased by over 90% in the later stages.

The results underscore the potential of *M. pygmaeus* as an effective biological control agent for the management of *T. vaporariorum* in greenhouse-grown pepper crops.

**Keywords:** *Macrolophus pygmaeus*, *Trialeurodes vaporariorum*, Biological control, Greenhouse crops, Pepper (*Capsicum annuum*), Pest management

**INTRODUCTION**

Greenhouse crops are frequently attacked by numerous pest species, including thrips, aphids, and whiteflies (Pedley, 2010). Among them, the greenhouse whitefly *Trialeurodes (Asterochiton) vaporariorum* (Westwood, 1856) (Hemiptera: Aleyrodidae) is considered one of the most economically important pests, particularly in the production of tomatoes, peppers, lettuce, melons, watermelons, beans, pumpkins, and other crops, both in greenhouses and in open-field conditions (Van Lenteren and Noldus, 1990; Zabel et al., 2001; Alborno et al., 2023). The species has become invasive and is currently reported in more than 75 countries worldwide (Byrne et al., 1990; Evans, 2007; EPPO, 2024). Its high reproductive capacity and

efficient dispersal ability enable infestation of a broad range of host plants (Smith, 2009). Moreover, *T. vaporariorum* readily develops resistance to insecticides (Sood et al., 2003; Gorman et al., 2007), making chemical control increasingly unreliable and costly. As a result, biological control strategies have emerged as an effective and environmentally sustainable alternative, demonstrating high efficacy against *T. vaporariorum* in recent decades.

*T. vaporariorum* is one of the most widely distributed species within the family Aleyrodidae and is a typical polyphagous pest with more than 850 recorded host plant species belonging to 469 genera and 121 families (Evans, 2007; Lee & Zhang, 2018; CABI, 2021). Host plants include economically important agricultural and ornamental crops grown primarily in protected cultivation systems. Damage caused by the greenhouse whitefly is both direct - through feeding on plant sap - and indirect - by transmission of phytopathogenic viruses (Español & Corredor, 1988; Navas-Castillo et al., 2011; Scotta et al., 2014). Economic losses associated with this pest are estimated to exceed USD 1 billion annually (Gonzalez et al., 1992; Legg et al., 2006; Aregbesola et al., 2019).

The predatory mirid bug *Macrolophus pygmaeus* (Rambur, 1839) (Hemiptera: Miridae) is a natural enemy of *T. vaporariorum* and is widely used in Europe for biological control in greenhouse crops (Trottin-Caudal and Millot, 1994; Malézieux et al., 1995; van Schelt et al., 1995; Lucas and Alomar, 2002). Its capacity to establish and maintain high populations and its ability to survive on alternative food sources in periods of low prey availability make it particularly valuable biocontrol agent (van Lenteren, 2012; Messelink and Janssen, 2014). As a zoophytophagous species, *M. pygmaeus* feeds on both plant tissues and prey, and has been shown to effectively reduce populations of *Myzus persicae*, and other major pests, including the greenhouse whitefly (Messelink et al., 2011). The predator is especially suitable for deployment in pepper crops, where it can successfully develop and reproduce (Perdikis and Lykouressis, 2000).

The aim of the present study is to evaluate the effectiveness of *M. pygmaeus* in suppressing *T. vaporariorum* populations under controlled greenhouse conditions using experimental cages.

## MATERIAL AND METHODS

The study was conducted in 2022 and 2023 at the experimental facility of "Opora Zaden Bulgaria" Ltd., in Tsalapitsa, Plovdiv Province. The polyethylene greenhouse was equipped with frontal and roof mesh ventilation openings, drip irrigation, screens, a misting system, and an automated meteorological station. Temperature control was managed by an ONDO system that automatically activated the ventilation openings at 27°C. No heating or supplemental lighting was used. Temperature and relative humidity were recorded at one-minute intervals.

Two independent zones were used:

Zone A: Potted pepper plants (*Capsicum annuum* L., cv. Amaretta) were maintained under uniform conditions, irrigated and fertilized as needed. No pesticides, fungicides, pollinators, or prior crops had been introduced. Environmental conditions were maintained at  $24 \pm 1^\circ\text{C}$  and  $65 \pm 5\%$  RH.

Zone B: This section also housed pepper plants, no plant protection treatments were applied. Its purpose was to maintain populations of *T. vaporariorum* to ensure sufficient biological material for observations and experiments involving *M. pygmaeus*.

To assess the effectiveness of the predatory bug *M. pygmaeus* in regulating populations of *T. vaporariorum*, experiments were carried out in six experimental cages (0.5 m × 0.9 m × 1.6 m) located in Section A of the greenhouse. Each cage contained four potted pepper plants. The cages were fully enclosed with fine mesh (250 µm) on all sides to prevent insect intrusion. Access to the plants was provided through a small zippered opening integrated into the mesh.

The experiments included two treatments: *M. pygmaeus* + *T. vaporariorum*, and a control (no predator added). Each treatment consisted of three replicates, with each replicate housed in a separate cage to enhance the reliability of results and minimize random variation.

Adult *M. pygmaeus* specimens were supplied by Biobest Group N.V., Bulgaria, in specialized transport boxes with cooling elements, packaged in plastic containers holding 500 individuals with adjustable openings. A total of 80 adults (20 individuals per plant) were introduced onto the four plants in each cage using a fine brush.

Adult *T. vaporariorum* individuals were collected from a colony maintained on pepper plants in Section B. They were introduced into the cages via infested leaf material and transferred onto the experimental plants using a brush and a handheld ×15 magnifying lens. The whiteflies were introduced 24 hours after predator release. A total of 200 adult whiteflies were added per cage (50 individuals per plant).

The population density of *T. vaporariorum* was expressed as the number of individuals (nymphs and adults) per plant. Counts were performed directly on one randomly selected plant per cage using a handheld ×15 magnifying lens. Counting was carried out at weekly intervals (every seven days) after the introduction of the whiteflies and continued until the end of the experiment. Statistical analysis was conducted using one-way analysis of variance (ANOVA), followed by Student's t-test to determine statistically significant differences between treatments at a significance level of  $P < 0.05$ .

## RESULTS AND DISCUSSION

The results of the studies conducted in 2022 and 2023 to determine the effectiveness of the predatory bug in regulating *T. vaporariorum* populations are presented in Table 1 and Table 2.

Observations indicate that in the experimental cages where *M. pygmaeus* was introduced, the population of the greenhouse whitefly remained significantly lower compared to the control cages without the predator. This confirms the effectiveness of *M. pygmaeus* as a biological control agent for reducing greenhouse whitefly populations in greenhouse-grown pepper.

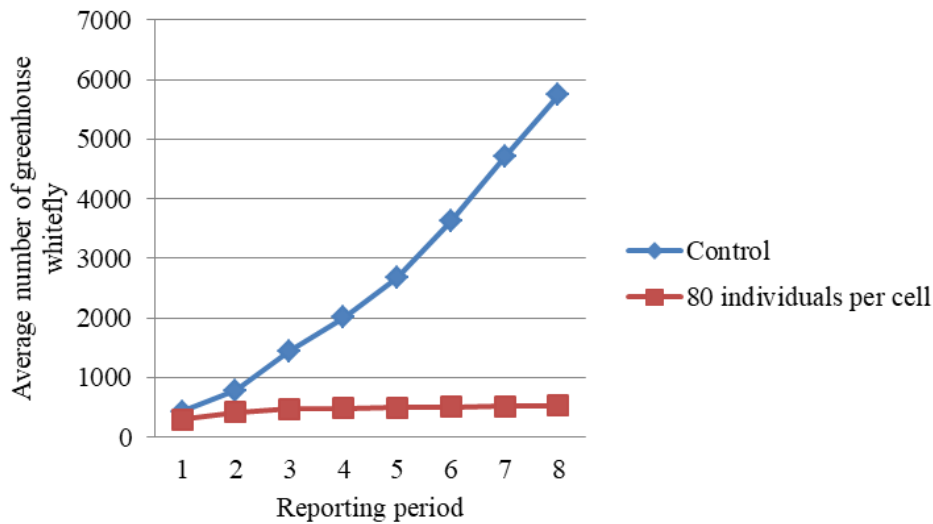
Data presented in Table 1 show that the density of *T. vaporariorum* populations did not differ substantially during the first two weeks following the introduction of *M. pygmaeus*, reaching an average of  $297.18 \pm 0.43$  and  $421.32 \pm 0.18$  individuals in 2022. Compared with the control cages lacking the predator, the

mean number of whiteflies was 30.9% and 45.7% lower at 7 and 14 days, respectively, after the introduction of the predatory bug (Figure 1).

**Table 1.** Average number of *T. vaporariorum* on pepper plants in the presence and absence of *M. pygmaeus* under greenhouse conditions for 2022.

Reporting period, weeks	2022			
	<i>T. vaporariorum</i> + <i>M. pygmaeus</i>		Control	
	Average value (M) ± SE	Confidence intervals P=0.05%	Average value (M) ± SE	Confidence intervals P=0.05
I	297.18 <sup>d</sup> ± 0.43	0.026-0.083	430.21 <sup>a</sup> ± 0.23	0.019-0.038
II	421.32 <sup>d</sup> ± 0.18	0.239-0.389	775.38 <sup>a</sup> ± 0.84	0.256-0.844
III	473.29 <sup>c</sup> ± 0.73	0.269-0.283	1441.85 <sup>a</sup> ± 0.40	0.297-0.496
IV	485.31 <sup>c</sup> ± 0.14	0.486-0.945	2002.09 <sup>a</sup> ± 0.78	0.239-0.778
V	497.23 <sup>b</sup> ± 0.58	0.503-0.734	2674.46 <sup>a</sup> ± 0.12	0.437-0.989
VI	504.25 <sup>b</sup> ± 0.71	0.635-0.928	3620a.22 <sup>a</sup> ± 0.36	0.570-0.882
VII	512.31 <sup>b</sup> ± 0.27	0.472-0.949	4708.49 <sup>a</sup> ± 0.17	0.433-0.248
VIII	528.33 <sup>b</sup> ± 0.68	0.481-0.498	5740.19 <sup>a</sup> ± 0.31	0.450-0.265

Differences in values followed by the same letter are not statistically proven a,b,c,d Level of evidence by Student t-test at significance level p=0.05



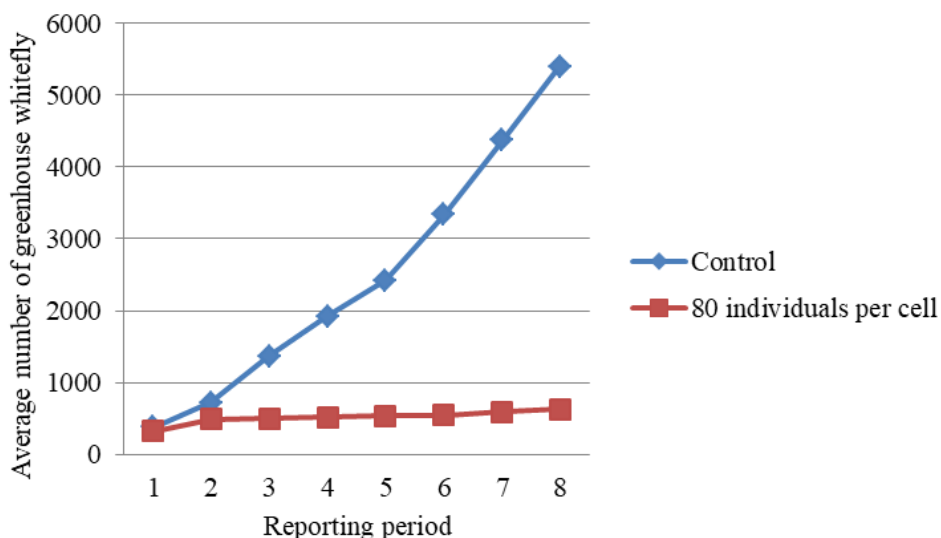
**Fig. 1.** Rate of population decline of *T. vaporariorum* in the presence and absence of *M. pygmaeus* for 2022.

The data show that in the experimental cages containing predatory bugs, the mean number of whiteflies in 2023 was 321.49 ± 0.52 and 484.24 ± 0.50 during the first two weeks after predator introduction (Table 2). The reductions in *T. vaporariorum* population were 14.4% and 32.9% at seven and fourteen days after predator release, respectively (Fig. 2), compared with the cages without added predators during the same periods.

**Table 2.** Average number of *T. vaporariorum* on pepper plants in the presence and absence of *M. pygmaeus* under greenhouse conditions for 2023.

Reporting period, weeks	2023			
	<i>T. vaporariorum</i> + <i>M. pygmaeus</i>		Control	
	Average value (M) ± SE	Confidence intervals P=0.05%	Average value (M) ± SE	Confidence intervals P=0.05
I	321.49 <sup>d</sup> ± 0.52	0.217-0.452	375.16 <sup>a</sup> ± 0.24	0.114-0.224
II	484.24 <sup>d</sup> ± 0.50	0.121-0.367	721.43 <sup>a</sup> ± 0.13	0.438-0.138
III	495.2 <sup>c</sup> ± 0.95	0.157-0.359	1368.18 <sup>a</sup> ± 0.32	0.320-0.109
IV	511.22 <sup>c</sup> ± 0.48	0.156-0.442	1925.17 <sup>a</sup> ± 0.54	0.574-0.182
V	533.50 <sup>b</sup> ± 0.33	0.169-0.503	2417.41 <sup>a</sup> ± 0.27	0.654-0.176
VI	543.28 <sup>b</sup> ± 0.74	0.158-0.513	3334.76 <sup>a</sup> ± 0.69	0.283-0.147
VII	586.62 <sup>b</sup> ± 0.36	0.197-1.646	4377.84 <sup>a</sup> ± 0.72	0.419-0.127
VIII	627.19 <sup>b</sup> ± 0.53	0.207-1.543	5401.24 <sup>a</sup> ± 0.57	0.422-0.157

Differences in values followed by the same letter are not statistically proven a,b,c,d Level of evidence by Student t-test at significance level p=0.05



**Fig. 2.** Rate of population decline of *T. vaporariorum* in the presence and absence of *M. pygmaeus* for 2022.

The statistical analysis of the results from the two experimental years showed that the population density of *T. vaporariorum* during the third and fourth weeks after the introduction of *M. pygmaeus* reached mean values of  $473.29 \pm 0.73$  and  $485.31 \pm 0.14$  individuals in 2022, and  $495.27 \pm 0.95$  and  $511.22 \pm 0.48$  individuals in 2023. Compared with the control cages without predators, the number of whiteflies was 67.2% and 75.8% lower in 2022 and 63.8% and 73.4% lower in 2023 in the predator-treated cages during these periods.

Across both years of the study, a substantial reduction in the greenhouse whitefly population was observed in the cages containing predators in the subsequent weeks until the end of the experiment. Data from 2022 indicate that population suppression reached 81.4%, 86.1%, 89.1%, and 90.8% on the 35th, 42nd, 49th, and 56th day after predator introduction, respectively. During these periods, mean values ranged from  $497.23 \pm 0.58$  to  $528.33 \pm 0.68$  whiteflies in the cages with *M. pygmaeus*.

In the second year, reductions of 77.9%, 87.6%, 86.6%, and 88.4% were recorded on the 35th, 42nd, 49th, and 56th day after predator release, with corresponding mean values of  $533.50 \pm 0.33$ ,  $543.28 \pm 0.74$ ,  $586.62 \pm 0.36$ , and  $627.19 \pm 0.53$  individuals in the predator-present cages.

The statistical analysis confirmed a significant reduction in greenhouse whitefly populations following the release of the predatory bug in both years of the experiment. Student's t-test further indicated significant differences between the mean values of predator-treated and control cages, demonstrating clear separation into distinct groups and confirming the consistent impact of predator introduction.

The results show that the effectiveness of the predatory bug in suppressing the greenhouse whitefly population peaked after the fourth week (day 28), with nearly identical reduction rates in both years (approximately 80–90%).

The study confirms the capacity of *Macrolophus pygmaeus* to reduce greenhouse whitefly populations. In the study by Lykouressis et al. (2009), the predation rate of *M. pygmaeus* on eggs and second-, third-, and fourth-instar nymphs of *T. vaporariorum* was evaluated using tomato leaf discs in Petri dishes. The authors established prey densities of 30, 60, 90, 120, and 150 eggs; 20, 40, 60, and 80 second-instar nymphs; 10, 20, 30, and 40 third-instar nymphs; and 5, 10, 15, and 20 fourth-instar nymphs. Their results showed that predation increased with prey density; however, no significant differences in predation rates were observed at the highest densities of eggs and third- and fourth-instar nymphs. The highest mean daily consumption consisted of 94 eggs and 56.5, 24.4, and 11.8 nymphs of the second, third, and fourth instars, respectively. These findings highlight the high potential of *M. pygmaeus* as a biological control agent against the greenhouse whitefly. Moreover, such knowledge is useful for optimizing biological control strategies involving this predator.

## CONCLUSIONS

*M. pygmaeus* demonstrated high effectiveness as a biological control agent against *T. vaporariorum* in greenhouse-grown pepper (*C. annuum*), reducing pest populations by more than 80% after the fourth week following its introduction. In both years of the study, a consistent trend of substantial population suppression was observed in predator-treated cages, with statistically significant differences compared with untreated controls at  $P < 0.05$ . The early effect of predator introduction was moderate (30–45% reduction by the second week) but increased over time, exceeding 90% reduction in the later stages of the experiment.

The data confirm the ability of *M. pygmaeus* to adapt and maintain stable populations even under limited prey availability, demonstrating its suitability for long-term integration into biological control systems in greenhouse pepper production.

The findings are consistent with previous research reporting high predation rates of *M. pygmaeus* on various developmental stages of *T. vaporariorum*, supporting its applicability as part of integrated pest management strategies compatible with organic farming principles.

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