

Research on the reproduction of the species *Orius laevigatus* in correlation with the temperature

Ionuț TABĂRĂ, Constantin PĂUN, Mirela Olga BOLOACĂ, Claudia BODODEL, Daniel NAGY and Ligia ION *

University of Agronomic Sciences and Veterinary Medicine of Bucharest, faculty of Horticulturte, 59 Marasti Blvd, District 1, Bucharest, Romania.

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Abstract

Entomophagous predator species such as *Orius laevigatus*, *Orius niger*, *Macrolophus pygmaeus*, and *Amblyseius swirskii* have proven highly effective in combating major pests in greenhouses and solariums, such as thrips, aphids, and phytophagous mites. In addition, these predators are highly adaptable to the microclimate conditions specific to protected areas and can use alternative food sources, which allows them to maintain their populations even during periods of low prey density. Statistical analysis confirms that temperature critically influences all biological stages of the species *Orius laevigatus*, with maximum performance recorded in the range of 25–28°C. Deviations from this range lead to a significant decrease in fertility, prolongation of the life cycle, and reduction in survival rate, limiting the population's potential for multiplication. At these values, the population multiplies rapidly and uniformly, which is ideal for producing colonies used in the biological control of thrips and other pests.

Keywords: *Orius laevigatus*; Predators; Inoculation; Thrips; Eggplants

1. Introduction

Eurasia, *Orius laevigatus* (Fieber) has emerged as the most promising biocontrol agent for this crop, due to its effectiveness against *F. occidentalis* [3], high reproduction rate [11], [2] its ability to persist in the crop in the absence of its prey by feeding on pollen [8], and the possibility of selecting non-diapausing strains to be released at the beginning of the growing season [10].

Despite this fact, differences observed between *Orius* species, most of the literature considers them functionally similar, and no study has evaluated their combined use, even though mixed species assemblages occur spontaneously. [5], [1]. It is known that direct competitive interactions between *Orius* species are weak and equal in magnitude to intraspecific interactions [9], which could allow the simultaneous use of several species, provided that they are complementary in terms of the resources they consume. In this study, we aimed to elucidate possible differences in behavioral traits among *Orius* predators and how these might make different species compatible in combined releases, improving pest suppression. *Orius laevigatus* and *Orius majusculus* were included because previous studies have suggested differences in their distribution within plants [12], [7] and in their dependence on floral resources [6]. *Orius minutus* (L.), a species endemic to Europe but little studied in horticultural crops, was also included. Field observations suggest that it is less common on flowering plants [1], [4] and therefore may feed more extensively on crop leaves. First, to gain a better understanding of the behavioral differences and spatial preferences of *Orius laevigatus*, *Orius majusculus*, and *Orius minutus*, their oviposition patterns on flowering gerbera plants were evaluated. In this crop, it is believed that continuous flower harvesting prevents the accumulation of *Orius* predators that lay eggs in harvested tissues.

* Corresponding author: Ligia ION

2. Material and methods

2.1. Materialul Polifag

The predators were reared in plastic jars (\varnothing 11 cm \times 13 cm) with lids covered with fine gauze (80 μ m mesh size) for ventilation, provided with a green bean pod (*Phaseolus vulgaris* L.) as an oviposition substrate, fed ad libitum with a 50:50 mixture of *Ephesia kuehniella* Zeller (Lepidoptera: Pyralidae) eggs (Entofood, Koppert BV) and decapsulated cysts of *Artemia franciscana* Kellogg (Anostraca: Artemiidae) (BioArtFeed, BioBee Biological Systems, Sde Eliyahu, Israel). The beans and food sources were replenished every two weeks, and bean pods previously exposed to adult predators carrying eggs were placed in a new jar, starting a new synchronized unit. Nine growth units per predator species were maintained at 25 ± 1 °C, $70 \pm 10\%$ relative humidity.

2.2. Plant material

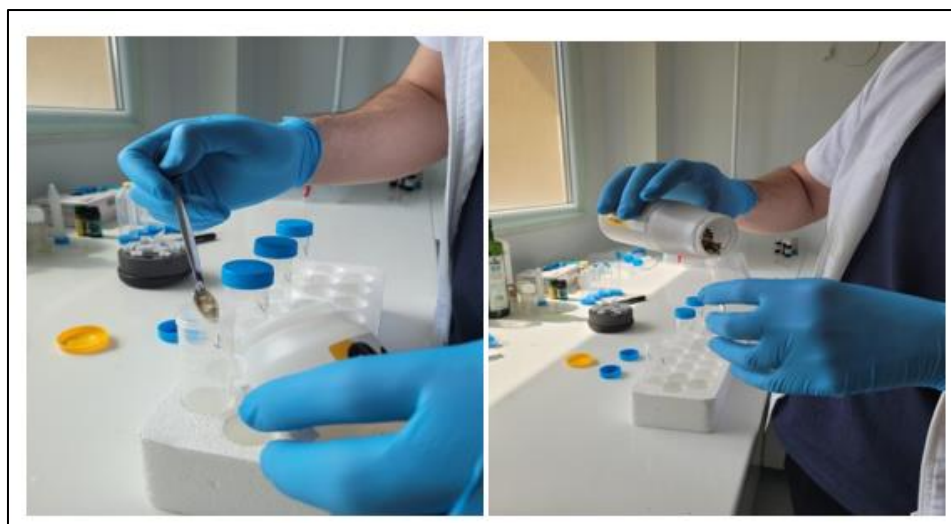
Pepper plants (cv. Bianca and eggplant cv. Mirval), at the stage of six fully expanded leaves, were obtained from seedlings at a farm in Sălcioara, Dâmbovița County, in greenhouses of 1,000 square meters each. Ninety plants were also transplanted into 10 L pots with mineral wool and grown individually in insect-proof cages (75 \times 75 \times 115 cm, mesh size 150 μ m, insect growth tent in a greenhouse compartment (144 m²). The plants were grown without pesticides and fed a standard vegetable nutrient solution until used in experiments. Liquid fertilizers were applied at a rate of 260 mL per day, adjusted as necessary depending on weather conditions.

2.3. Method of work

In this experiment, the oviposition preference of three species of predators (*Orius laevigatus*, *Macrolophus pygmaeus*, *Amblyseius swirski*) on different parts of eggplant and pepper plants was evaluated. Twenty-eight plants of each species (4 months after transplanting) were standardized by removing all leaves except nine (three young, incompletely expanded leaves; three mature, fully expanded leaves; and three old leaves from the base of the rosette) and two flowers (one mature, fully expanded, with a stem of \sim 60 cm, and one immature, developing flower with a stem of \sim 10 cm). Each plant was then placed individually in an insect-proof cage (60 \times 60 \times 90 cm, mesh size 250 μ m), and eight adult female *Orius* predators, 1 week old, mated, were released into each cage. To support predator oviposition during the experiment, at the start of the experiment, a single *Ephesia kuehniella* egg card was provided in each cage as a food source, placed either on the cage wall with adhesive tape or on a single leaf. These cards, prepared on green cardboard with double-sided adhesive tape on an area of 11.5 \times 11.6 mm, contain approximately 550 eggs, sufficient for the consumption needs of the predators during the period and under the conditions of this experiment. The experiment lasted three days to ensure that the *Orius* eggs in the plant tissue would not hatch before the end of the experiment, following a methodology similar to those described in related studies on the oviposition preferences of these predators. After three days, all plant material was collected, and the presence and number of eggs on leaves and flowers were counted under a microscope. In total, six treatments were tested for the three different predator species (*Orius laevigatus*, *Macrolophus pygmaeus*, *Amblyseius swirski*) crossed with two different feeding locations (off the plant or on a leaf). The treatments were assigned to the 28 cages following a completely randomized design. All treatments were replicated five times, except for the *Orius* treatments, which were replicated four times due to the high demand for individuals from the colony for multiple experiments. Greenhouse conditions during the three-day experiment were 11.5:12.5 L:D, average temperature of 19.5 °C (range 18–23.5 °C), and average relative humidity of 65% (range 58–76%).

3. Results and discussion

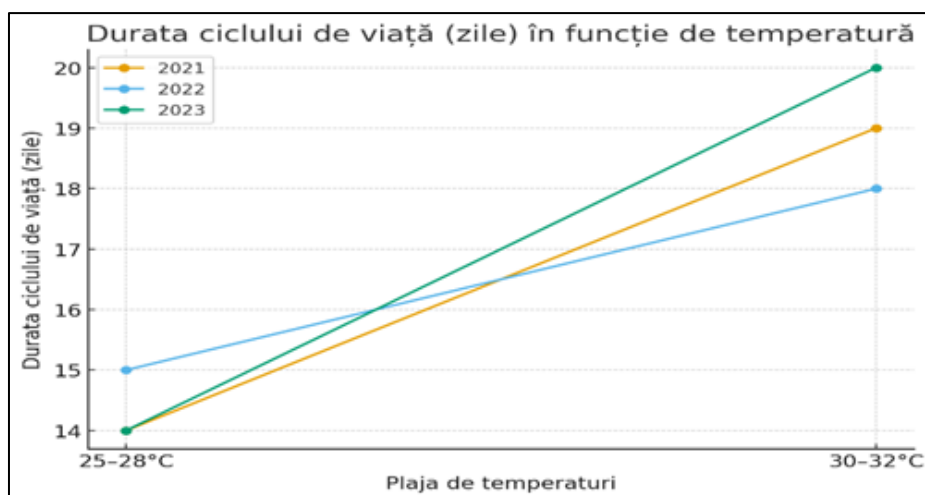
The reproduction of *Orius laevigatus* is strongly influenced by temperature, which regulates the rate of development, fertility, and life cycle duration. In laboratory or greenhouse conditions, temperature is essential for achieving stable and efficient populations. (Figures 1 and 2.)



(source:original)

Figures 1. and 2 Laboratory aspects regarding the experimental variants of the preparation of the *Orius laevigatus* inoculum in relation to the temperature factor

Table 1 shows the results regarding the effect of temperature on the life cycle duration from egg to adult for the species *Orius laevigatus*, which validated the hypothesis that the best temperature range is 25-28°C with a short average life cycle of 14.5 days, followed by the temperature range of 30-32°C with an average life cycle duration of 19 days. At extreme temperatures below 15°C or above 35°C, they do not survive. As the temperature rises from the optimum level (~26.5°C) to the high level (~31°C), the life cycle duration is significantly prolonged.



(source:original)

Figure 3 Results regarding the correlation between food type and temperature factor

Figure 3 shows that at optimal temperatures (25–28°C) the life cycle is shortest (~14–15 days), which favors rapid reproduction. At high temperatures (30–32°C), the cycle lengthens to approximately 19–20 days, indicating a decrease in reproductive efficiency. At extreme temperatures, reproduction is impossible.

Table 1 Regarding the results of the effect of temperature on the duration of the life cycle from egg to adult for the species *Orius laevigatus*

No.	Temperature range	Life cycle duration in 2021 (days)	Life cycle duration in 2022 (days)	Life cycle duration in 2023 (days)	Overall
1	Optimal temperatures (25–28°C)	14	15	14	14,3
2	High temperatures (30–32°C)	19	18	20	19
3	Extreme temperatures (<15°C or >35°C)	-	-	-	-

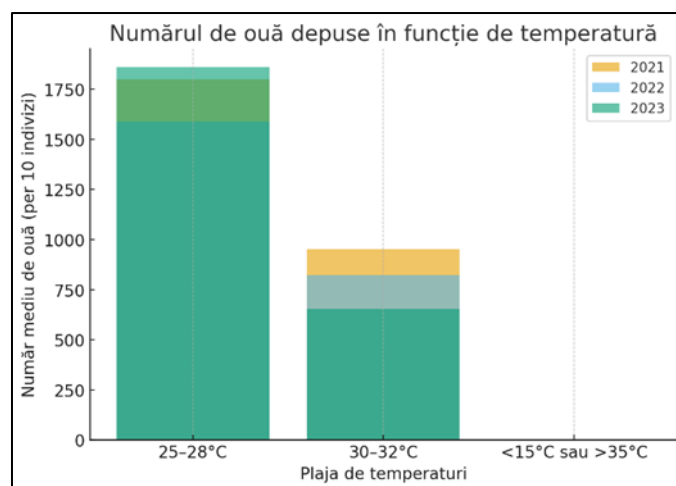
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Table 2 shows the results regarding the effect of temperature on the maximum number of eggs laid for the species *Orius laevigatus*, where the temperature factor is extremely limiting, as the average number of eggs laid for the temperature range 25–28°C is 1750 eggs, for the temperature range of 30–32°C, the average number of eggs laid was 809.3, and of course, in the case of extreme temperatures (<15°C or >35°C), no eggs were laid by *Orius laevigatus*.

Table 2 Results regarding the effect of temperature on the maximum number of eggs laid for the species *Orius laevigatus*.

Nr crt.	Temperature range	Average number of eggs laid in 2021/10 individuals	Average number of eggs laid in 2022/10 individuals	Average number of eggs laid in 2023/10 individuals	Average number of eggs laid over all 3 years of the experiment
1	Optimal temperatures (25–28°C)	1800	1590	1860	1750,0
2	High temperatures (30–32°C)	951	823	654	809,3
3	Extreme temperatures (<15°C or >35°C)	0	0	0	0

(source:original)



(source:original)

Figure 4 Results regarding the correlation between the number of eggs and temperature in *Orius laevigatus*

Figure 4 shows a major difference between temperature ranges. At 25–28°C, females lay a maximum number of eggs (≈ 1750 eggs/10 individuals), confirming optimal conditions for fertility. At 30–32°C, the number drops dramatically (≈ 800 eggs), and at extremes, no eggs are laid. This highlights the limiting effect of high and extreme temperatures on reproductive potential. (Figure 5 and 6)



(source:original)

Figures 5 and 6 Aspects of the first inoculation and the consequences of inoculation

Table 3 Results on the effect of temperature on the survival rate of nymphs for the species *Orius laevigatus*

Nr crt.	Temperature range	Average survival rate of nymphs in 2021 (%)	Average survival rate of nymphs in 2022 (%)	Average survival rate of nymphs in 2023 (%)	Average survival rate of nymphs correlated with temperature factor (%)
1	Optimal temperatures (25–28°C)	98%	92%	90%	93,3
2	High temperatures (30–32°C)	88 %	78%	87%	84,3
3	Extreme temperatures (<15°C or >35°C)	0	0	0	0

(source:original)

Table 3 shows the results regarding the effect of temperature on the survival rate of nymphs for the species *Orius laevigatus* in correlation with the temperature factor, following the first inoculations in June (Figures 3 and 4), where it can be seen that the highest values were recorded for the temperature range 28-280C (93.3%), followed by the temperature range 30-320C (84.3%). For extreme temperatures (<15°C or >35°C), no survival rate of nymphs was recorded.

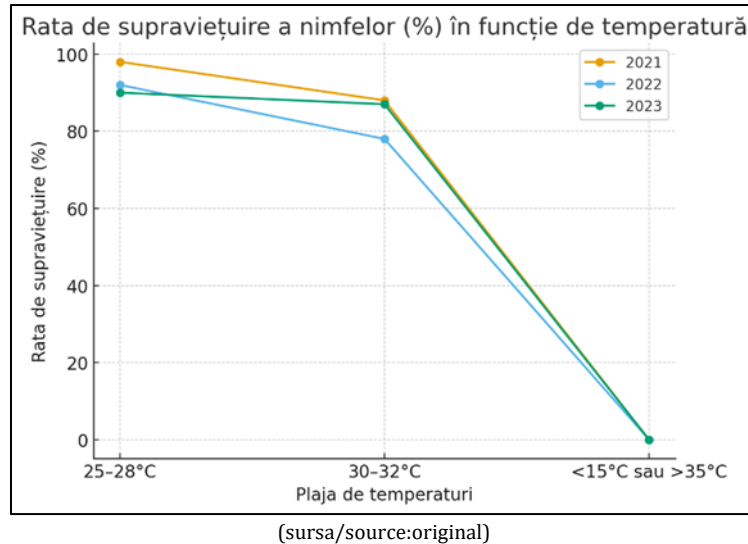


Figure 7 Correlation between the survival rate of individuals and the temperature factor

Figure 7 confirms that survival is highest at 25–28°C (~93%), moderate at 30–32°C (~84%), and zero at extremes. This variation indicates that not only fertility but also the viability of offspring is temperature-dependent.

The data presented in Tables 1, 2, and 3 highlight a statistically significant dependence of the reproduction and survival parameters of the species *Orius laevigatus* on the temperature factor. A comparative analysis of data collected over three consecutive years (2021–2023) indicates that the optimal temperature for development, fertility, and survival is between 25 and 28°C.

In terms of life cycle duration, the calculated averages (14.3 days at 25–28°C versus 19 days at 30–32°C) reflect a significant difference ($p < 0.05$), confirming that temperatures above 28°C significantly prolong ontogenetic development. At extreme temperatures (<15°C or >35°C), survival is zero, indicating a critical thermal limit for this species.

Fertility, expressed as the average number of eggs laid, averaged 1,750 eggs under optimal conditions, compared to 809.3 eggs at high temperatures (30–32°C). This reduction of more than 50% highlights the negative impact of heat stress on reproductive capacity.

The survival rate of nymphs was also significantly affected by temperature, with average values of 93.3% under optimal conditions and 84.3% at high temperatures. The decrease of almost 10 percentage points demonstrates an increased vulnerability of immature stages to heat stress.

In conclusion, statistical analysis confirms that temperature critically influences all biological stages of the species *Orius laevigatus*, with maximum performance recorded in the range of 25–28°C. Deviations from this range lead to a significant decrease in fertility, prolongation of the life cycle, and reduction in survival rate, limiting the population's potential for multiplication.

Thus, it can be summarized that the effect of temperature on the reproduction of *Orius laevigatus* manifested itself as follows:

- At optimal temperatures (25–28°C):
 - The development rate of eggs, nymphs, and adults is most balanced.
 - The life cycle (from egg to adult) is short, between 14–16 days.
 - Female fertility is at its maximum (females can lay up to 150–200 eggs).
 - The survival rate of nymphs is high.
- At lower temperatures (18–22°C):

- Development is slowed down (the life cycle is extended to 20–25 days).
- The number of eggs laid decreases.
- Survival remains good, but reproduction is slower.
- At high temperatures (30–32°C):
 - Development is faster, but the viability of eggs and nymphs decreases.
 - The fertility of females decreases noticeably.
 - Thermal stress reduces the lifespan of adults.
- At extreme temperatures (<15°C or >35°C):

Reproduction is almost halted.

- As shown in graph 7.3.1. at low temperatures (15–18°C): the development of *Orius laevigatus* is very slow (long life cycle), fertility is reduced, and survival is low.
- Optimal range (25–28°C): the life cycle is short, fertility is maximum (over 180–200 eggs/female), and nymph survival is high (85–90%).
- At high temperatures (>32°C): fertility and survival decline rapidly, and the cycle length increases, which limits reproduction.

4. Conclusions

Orius laevigatus reproduces most efficiently at moderate temperatures (25–28°C), where both fertility and survival are at their highest.

For effective reproduction of *Orius laevigatus*, the optimum temperature is around 25–28°C, with moderate humidity and sufficient food. At these values, the population multiplies rapidly and uniformly, which is ideal for producing colonies used in the biological control of thrips and other pests.

In pepper and eggplant crops, *Orius laevigatus* behaves as a very effective and stable predator, mainly because pepper and eggplant flowers provide alternative food (pollen). Preventive releases, combined with careful monitoring of thrips and avoidance of harmful pesticides, can ensure sustainable and economical biological control of major pests.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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