

Greenbelly Stinkbug Biology in Different Temperatures

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Abstract

The green-belly stinkbug (*Dichelops melacanthus*) stands out due to its damage potential towards corn and wheat crops. The population distribution and size greatly influence the potential damage. The insect's reproductive capacity depends on different temperature conditions during various crop seasons and its lifecycle. The study aimed to evaluate the influence of different constant temperatures on the *D. melacanthus* biology. The study was performed twice, once in the year of 2017, and again in 2019. The biological features were observed in temperature-controlled climate chambers with the following temperatures: 11, 16, 21, 26, 31 and 36 °C, each with a fluctuation of ± 1 °C, using a RH of $65 \pm 15\%$ and photophase of 14 h. The evaluations were conducted from eggs to adults in terms of: nymphal hatch period, each instar duration, female fecundity and egg viability. There were no eggs hatching at 11 °C and it also reached 100% mortality during the second instar at 16 °C. The eggs-to-adult duration for the temperatures 21, 26, 31 and 36 °C in the trial of 2017 was 58.4, 30.1, 18.2 and 16.3 days, respectively. In the same temperatures, but during the 2019 trial, the eggs to adult duration was 58.1; 29.7; 21.3 and 19.1 days, respectively. The reproductive capacity in the temperatures of 21 and 36 °C impaired the female fecundity and egg viability. The temperatures 26 and 31 °C favored the development of *D. melacanthus*.

Keywords: *Dichelops melacanthus*, egg viability, fecundity

1. Introduction

Grain cultivation in Brazil is very competitive in the international market due to the possibility of more than one annual yield, using crop succession (CONAB, 2019). The adoption of the no-tillage system has brought several benefits to soil and crop development: dispersing the impact force of rainfall on the soil; preventing leaching; maintaining an organic cover over the soil; providing a degree of weed control (Silva et al., 2009). However, the reduction of soil movement and succession of susceptible monocultures has been favoring some arthropods, such as the green-belly stinkbug *Dichelops melacanthus* (Dallas, 1851) (Hemiptera: Pentatomidae), whose population in soybean, corn (first or second crop), and wheat crops has been constant (Carvalho, 2007).

The presence of the green-belly stinkbug in crop succession areas is favored due to the availability of plant hosts and shelter provided by the no-tillage resulting straw (Carvalho, 2007). Its feeding in the vegetal tissues occurs through the insertion of its sucking apparatus, constituted by juxtaposed stylets, forming two channels: salivary and alimentary. It can penetrate various plant structures, such as the stem, pod or seed, releasing its saliva with digestive enzymes, followed by the absorption of the liquefied cellular material (Chocorosqui, 2001). The extraoral digestion of the saliva changes the internal cellular constitution of tissues, leading to plant disturbances such as poor plant development; seed abortion or improper grain filling; tillering, and tissue damage, which serve as a gateway for pathogens. The penetration and consumption of grain reserves leads to a seed with a shriveled appearance and spotted pods (Roza-Gomes et al., 2011), thus dwindling the seed value.

D. melacanthus has a hemi-metabolic development, consisting of the egg phase, followed by five nymphal instars, whose duration depends on the accumulation of degree-days, in direct relation with the exposed daily temperature (Pereira et al., 2007). Since it does not have diapause period, the study of its development under different climatic conditions throughout the year, allows for the estimation of its population growth and planning of actions for its management. The study aimed to evaluate the influence of six constant temperatures on the development time, female fecundity and egg viability of *D. melacanthus* with a controlled diet and photophase.

2. Method

2.1 Location and Egg Origin

The study was carried out at the Ecologic Pest Management Laboratory from the department of entomology in the Instituto Agronômico do Paraná (IAPAR), in Londrina, State of Paraná. Two biological trials of *D. melacanthus* were performed, one was in 2017 and the other study was in 2019. The eggs used came from the IAPAR rearing of *D. melacanthus* and their collection was on the first day of each evaluation.

2.2 Temperature and Rearing Conditions

The green-belly stinkbug biology was studied in temperature-controlled climate chambers, using the temperatures of 11, 16, 21, 26, 31 and 36 °C, each with a fluctuation of ± 1 °C, with a RH of $65 \pm 15\%$ and 14 h photophase. Temperature and relative humidity readings were recorded with an AZ® Datalogger, model 8829, one placed in each climate chamber. Within each chamber were placed 10 containers, brand Gerbox® (11 × 11 × 3.5 cm), totaling 60 Gerbox®, and in each one, placed 10 eggs, giving a total of 600 *D. melacanthus* eggs.

2.2.1 Rearing Container Preparation

For the Gerbox® asepsis they were washed with distilled water and subsequently cleaned with 70% alcohol. At the bottom of the container, a cut filter paper was placed and wet using 3 mL of autoclaved distilled water. To ensure adequate relative humidity for the stinkbug development, within each Gerbox was placed an Eppendorf® (1.5 mL) sealed tube with a moistened dental-cotton containing 2.5 mL of autoclaved distilled water. Occurring a daily wetting of the filter paper with 3 mL of water and replacing each Eppendorf® with 2.5 mL of water.

2.3 Diet

The *D. melacanthus* diet was composed of two green pods (*Phaseolus vulgaris*), previously washed with detergent in running water, three soybean seeds (*Glycine max*) variety BMX Potência RR and two peanut seeds (*Arachis hypogea*), all food underwent ultraviolet sterilization for an hour on each face.

2.4 Research Design and Statistical Analysis

The experiment used a completely randomized design, in which each Gerbox® containing the ten eggs were randomly arranged within the chambers, and their positions were alternated daily, resulting in ten replications per temperature. The study parameters were recorded daily, in which the hatching, development and adult maturation time was evaluated, followed by the number and viability of the eggs per reared adults. The statistical analysis was comprised of ANOVA, Skott-Knott (parametric analysis), Kruskal-Wallis (non-parametric analysis) and Shapiro-Wilk, using the SISVAR® and BioStat® software.

3. Results and Discussion

The egg-to-adult development time of the *D. melacanthus* varied at different temperatures, the temperatures 21, 26, 31 and 36 °C in the 2017 trial had a development time of 58.4; 30.1; 18.2 and 16.3 days, respectively. During the test with the same temperatures in 2019, the time was 58.1; 29.7; 21.3 and 19.1 days, respectively (Table 1 and 2).

At 11 °C, no eggs hatched in either of the trials, at 16 °C in the first trial, there were hatchings, but every individual died during the second instar, during the second trial, there was no hatch at all. Thus, low temperatures demonstrate a lethal effect on developing embryos. Corresponding to the results gathered during the rearing of this arthropod in both trials, Chocorosqui et al. (2008) found that green-belly stinkbug eggs reared at 15 and 20 °C resulted in total egg mortality, also, when first instar nymphs, reared at 25 °C, were placed at these two temperatures, they died before reaching the second instar.

Rodrigues et al. (2014) found that *D. melacanthus* reared at a temperature of 31 ± 2 °C had a much shorter development time, coinciding with the present study, as the development time of each instar in the temperatures 31 and 36 °C, in both trials, demonstrated the shortest periods in days (Tables 1 and 2).

Table 1. Average time (n = 10), in days, to reach the different development stages of the *D. melacanthus* stinkbug, reared in five constant temperatures, first trial. IAPAR, Londrina, State of Paraná, 2017

Temperature (°C)	Nymphal stages						
	Hatch	1°	2°	3°	4°	5°	Egg-to-Adult
16	14.2±2.2 a ¹	9.0±1.1 a	D ²	D	D	D	D
21	7.9±1.3 b	5.5±0.7 b	9.6±1.0 a	9.4±1.8 a	11.4±2.3 a	14.6±2.5 a	58.4±5.5 a
26	4.8±0.6 c	3.2±0.3 c	5.5±0.6 b	4.6±1.0 b	4.8±0.9 b	7.1±1.1 b	30.1±1.7 b
31	3.1±0.1 d	1.9±0.3 d	3.8±0.5 c	2.5±0.6 c	2.7±0.5 c	4.2±0.4 c	18.2±0.9 c
36	2.7±0.5 d	1.7±0.4 d	3.3±0.5 c	2.3±0.4 c	2.3±0.4 c	3.7±0.4 c	16.3±0.7 c
C.V. (%)	17.3	14.6	12.9	24.3	26.6	17.9	9.43

Note. ¹ Averages followed by the same letter in the columns do not differ, using Scott-Knott test ($\alpha = 5\%$).

² Dead stinkbugs after the second instar (D), were not statistically evaluated.

Table 2. Average time (n = 10), in days, to reach the different development stages of the *D. melacanthus* stinkbug, reared in five constant temperatures, second trial. IAPAR, Londrina, State of Paraná, 2019

Temperature (°C)	Nymphal stages						
	Hatch	1°	2°	3°	4°	5°	Egg-to-Adult
21	11.2±0.8 a ¹	4.8±0.8 a	10.5±2.6 a	7.8±1.7 a	12.4±3.8 a	13.0±5.1 a	59.3±6.1 a
26	8.0±0.0 b	2.5±0.4 b	5.4±0.8 b	3.5±1.1 b	4.1±1.0 b	6.0±1.8 b	29.6±2.2 b
31	7.1±0.3 c	1.1±0.2 c	3.7±0.6 c	2.5±0.4 b	2.5±0.9 b	4.1±0.7 b	21.2±1.2 c
36	6.0±0.0 d	1.4±0.3 c	4.0±1.5 c	2.2±0.9 b	2.9±0.3 b	3.4±0.8 b	19.1±1.0 c
C.V. (%)	5.0	18.7	26.1	19.6	34.8	23.9	10.0

Note. ¹ Averages followed by the same letter in the columns do not differ, using Scott-Knott test ($\alpha = 5\%$).

The reared female fecundity and egg viability evaluation revealed that the utmost temperatures of 21 and 36 °C negatively affected the oviposition, producing, respectively, 6.7 and 136 eggs in average, with no hatchings, in the first trial. However, in the second trial there was no oviposition at all in these temperatures (Tables 3 and 4). The low oviposition as well as an unviability observed in the 21 °C, was similar to results found by Bortolotto et al. (2016), whom observed that *D. melacanthus* reared in a temperature of 19 °C, had no fecundity nor egg viability.

Egg viability and female fecundity varied between the two trials for the temperatures of 26 and 31 °C. During the first trial, the egg viability was 87.9 and 75.5%, and female fecundity was 205.2 and 337.4 eggs in average, respectively. During the second trial, an egg viability of 63.2 and 64.6%, and female fecundity of 120.1 and 125.1, respectively, was observed. Bortolotto et al. (2016) found that *D. melacanthus* reared at temperatures of 25 and 31 °C presented an egg viability of 79.7 and 79.4%, and female fecundity of 70.1 and 68.8 eggs, at average, respectively. Silva et al. (2011) reared 50 females of *D. melacanthus* in a constant 26 °C, and produced averagely 136.2 eggs each. In the present study, in the same temperature, with 10 females, an average of 205.2 eggs in the first trial, and 120.1 eggs in the second trial was observed. Rodrigues et al. (2014) while rearing the green-belly stinkbug at a temperature of 25 and 31 °C, found that the female fecundity was 86.3 and 104.63 eggs, in average, and an egg viability of 60.2 and 65.2, respectively. Similar egg viabilities, however, a higher egg production in the present study was probably due to the different diet the females were fed during the rearing.

Table 3. Average fecundity and viable eggs of the *D. melacanthus* stinkbug, reared in four constant temperatures. IAPAR, Londrina, State of Paraná, 2017

Temperature (°C)	N° of females	Fecundity (eggs/female)	(%) of viable eggs
21	19	6.8±3.1 b ¹	0.0±0.0 c
26	10	205.2 ±133.5 a	86.1±6.8 a
31	13	337.4±207.5 a	73.0±13.8 a
36	10	136±80.6 ab	0.2±0.3 b
p-value			< 0.01

Note. ¹ Averages followed by the same letter in the columns do not differ, using Kruskal-Wallis test ($\alpha = 5\%$).

Table 4. Average fecundity and viable eggs of the *D. melacanthus* stinkbug, reared in four constant temperatures. IAPAR, Londrina, State of Paraná, 2019

Temperature (°C)	N° of females	Fecundity (eggs/female)	(%) of viable eggs
21	10	0.0±0.0 b ¹	-
26	10	120.1±59.0 a	53.1±31.6 a ¹
31	10	125.1±103.0 a	49.8±34.4 a
36	10	4.3±7.5 b	0.0±0.0 b
p-value		< 0.01	0,04

Note. ¹ Averages followed by the same letter in the columns do not differ, using Kruskal-Wallis test ($\alpha = 5\%$).

Thus, the development of *D. melacanthus* is impaired in the utmost temperatures, above 31 °C and below 26 °C. However, temperatures between 26 and 31 °C favor its development, female fecundity and egg viability, thus demonstrating the adaptability of this pentatomidae to higher temperatures.

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