

Phenotypic plasticity in adults of *Anticarsia gemmatalis* exposed to sub-doses of Bt-based bioinsecticide

Fabricio Oliveira Fernandes (✉ fabriciof9@gmail.com)

UNESP: Universidade Estadual Paulista Julio de Mesquita Filho <https://orcid.org/0000-0002-8776-8308>

Tamires Doroteo de Souza

UNESP: Universidade Estadual Paulista Julio de Mesquita Filho

Ariadne Costas Sanches

USP: Universidade de Sao Paulo

Ivan Ricardo Carvalho

UNIJUI: Universidade Regional do Noroeste do Estado do Rio Grande do Sul

Naymã Pinto Dias

UNESP: Universidade Estadual Paulista Julio de Mesquita Filho

Janete Aparecida Desiderio

UNESP: Universidade Estadual Paulista Julio de Mesquita Filho

Ricardo Antonio Polanczyk

UNESP: Universidade Estadual Paulista Julio de Mesquita Filho

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Abstract

Anticarsia gemmatalis Hünber, 1818 is one of the main defoliating species in the soybean crop. *Bacillus thuringiensis* Berliner, 1915, is a bacterium used in the biological control of this pest species. Resistant populations and their sublethal effects caused by the use of the bacteria have already been reported; however, there are no studies on phenotypic plasticity in adulthood exposed to Bt-based bioinsecticide sub-doses. This study aimed to evaluate the morphometry of *A. gemmatalis* adults under laboratory conditions submitted to the Bt-based bioinsecticide Dipel SC over the three generations. The body segments mensured were width, length, and area of the anterior and posterior wings, the weight of the integument, chest, abdomen, wings, and the whole adult of males and females. Among the treatments, LC₅ in the first generation and CL₁₀ in the second generation were those with lower thresholds in relation to the weight of the chest and abdomen, considering the proportions of the body smaller than the females. The female's weight adulthood was reduced by 10% about males, and, only in the first generation. Males have larger body size and more pronounced phenotypic plasticity than females. Here, we demonstrate the first study assessing the phenotypic plasticity of *A. gemmatalis* adults.

Introduction

Anticarsia gemmatalis Hünber, 1818 is one of the main defoliating species in the soybean crop in the American continent (Haase et al., 2015). However, as it is considered a polyphagous species, it migrates to other regions to colonize new areas with greater food availability (Ford et al., 1975; Pashley and Johnson, 1986; Fernandes et al., 2018). The permanence of the insect pest in tropical and subtropical environments is attributed to the continuous cultivation throughout the year, which provides the formation of green bridges (Oliveira et al., 2014; Fernandes et al., 2020).

Bacillus thuringiensis Berliner, 1915, is a bacterium used in the biological control of this pest species. In the form of Bt-based bioinsecticides or biotechnology with the insertion of the *Cry* genes in plants to provide resistance to insects, known as transgenic plants or Bt plants (Konecka et al., 2018).

Resistant populations and their sublethal effects caused by the use of the bacteria have already been reported (Sedaratian et al., 2013; Janmaat et al., 2014; Souza et al., 2019; Rabelo et al., 2020); however, there are no studies on the phenotypic plasticity in adulthood, according to the exposure of the sub-dose of Bt-based bioinsecticides.

Phenotypic plasticity is the ability of an organism to respond to environmental stresses with changes in shape, state, movement, or activity (Brisson, 2010; West-Eberhard, 2003). In addition to being considered an important escape tool for survival in unstable environments or disturbed by human action (Gotthard and Nylin, 1995).

Studies demonstrate that these behavioral, morphological and physiological adaptations allow organisms to better adapt to disturbed environments on short time scales without changes in the genotype (West-Eberhard, 2003; Hayes et al., 2019).

Evaluations carried out on *Nilaparvata lugens* Stål, 1854 exposed to sub-doses of insecticides, showed differences in the morphometric parameters of the wings. The imidacloprid and dinotefuran treatment morphometrically provided smaller wings for both sexes, unlike the fenvalerate treatment that contributed to larger wings (Bao et al., 2009).

Sub-doses of insecticides can alter the development of the wings in insects and other body segments and, therefore, developmental, reproductive and dispersal capacities (Hayes et al., 2019). Thus, it is expected that population interactions and biotic relationships established in a community are influenced by changes in the individual's morphometry (Vaz et al., 2004; Gorür et al., 2005).

However, due to the lack of studies on the phenotypic plasticity of *A. gemmatalis* exposed to sub-doses of Bt-based bioinsecticides, this study aimed to evaluate the morphometry of *A. gemmatalis* adults submitted to the Bt-based bioinsecticide Dipel SC in laboratory over three generations.

Materials And Methods

Insect Rearing

The population of *A. gemmatalis* used in the bioassays was maintained on an artificial diet (Greene et al., 1976) at the Laboratory of Microbial Control of Arthropod Pests of the State University of São Paulo "Júlio de Mesquita Filho" (UNESP – Jaboticabal). The insects were maintained at 25 ± 1 °C, 70 ± 10% RH, and 12 h photophase.

Sublethal Concentrations

To evaluate the toxicity of this product, spore-crystal suspensions of the Bt-based bioinsecticides (Dipel®) were defined by plating on nutrient agar to determine the CFU evaluated after seven days (Sedaratian et al. 2013).

The curve response was estimated using the Six Error Problems analysis (Sas University 2013). 200 µL on the surface of the artificial diet (4.8 cm³) were previously distributed in polyethylene cups (3.5 cm Ø). To estimate a curve response, 100 insects were used for each treatment, distributed in 10 repetitions. Deionized water was applied in equal volume as a control (Santos et al. 2020). The bioassay evaluations were carried out after seven days.

Based on mortality data from bioassays, artificial diet preparations containing sub-lethal concentrations of Dipel® or controls (untreated diet) were prepared and used to study sublethal effects of Bt on *A. gemmatalis*. The sub-lethal concentrations LC₅, LC₁₀, LC₁₅ and LC₂₀ (0.20509, 0.38126, 0.57929 and 0.80776 µg Bt.mL diet⁻¹) were chosen by the estimate response curve (Sedaratian et al. 2013).

In these assays, a 200 µL aliquot of each sub-lethal concentration was applied to the surface of the artificial diet (4.8 cm³), previously distributed in polyethylene cups (3.5 cm Ø) (Sas University 2013).

The surviving caterpillars were fed an artificial diet containing their respective sub-lethal concentrations for three generations (F₁, F₂ and F₃). For each generation, 100 insects were used for each treatment, considering each caterpillar as a sampling unit. Deionized water was applied in an equivalent volume in the control. The evaluations were carried out daily.

Assessment of Sublethal Effects

The surviving caterpillars in each treatment/generation were evaluated daily and were sexed when they reached the pupal stage (Butt and Cantu, 1962). The newly emerged adults were separated into couples and placed in PVC cages (10 x 20 cm), lined with white A4 sulfite paper (used as an oviposition substrate). At the bottom, a Petri dish with filter paper was used and the top was sealed with voile fabric.

The adults were fed with a 10% honey solution moistened with cotton wool placed in a polyethylene petri dish (49 x 12 mm) at the cages (Fernandes et al., 2017). The papers used as a laying substrate were removed, exchanged daily, packed in plastic pots (14.0 cm Ø, 10 cm h), and with the hatching of the larvae, these were used to originate the subsequent generations (Kalvnadi et al., 2018).

Morphometry

The adults of *A. gemmatalis* exposed to the Bt-based bioinsecticide sub-doses were weighted within 24 hours. The different parts of the individuals were separated with the aid of fine-tipped surgical scissors and then weighed on an analytical balance (Belmark – 210A). The weighing was performed with the tegument, thorax, abdomen, wings and whole adult.

After mounting on the lamina, coverslip and sealed with a thin layer of colorless nail polish dried for two hours. The measurements of the length, width and area of the anterior and posterior wings were obtained with the aid of a stereoscope microscope with an attached camera (Leica S9 i), according to the technique described by Mare and Corseuil (2004).

Experimental Design and Data Analysis

The mortality data from virulence tests were submitted to Probit regression analysis and sublethal concentration values LC₅, LC₁₀, LC₁₅ and LC₂₀ were obtained using the SAS software (P > 95%) (Sas University, 2014). We used a completely randomized design (CRD) with ten repetitions per sex and the treatments arranged in a 3 x 5 x 22 factorial arrangement. There were three generations (F₁, F₂, F₃), five treatments (LC₅, LC₁₀, LC₁₅, LC₂₀ and control) and 22 variables being the weight of tegument, thorax, abdomen, wings, whole adult, and width, length and area of the anterior and posterior wings of both sexes.

Statistical analysis was performed on the GENES software (Cruz, 2001). The data were subjected to analysis of variance by F test and the stratified linear correlation was performed per generation and treatment. The Tukey test dismembered the variables that showed an interaction between treatment and the generation at 5% probability.

Results

Males Morphometry

In the weight of the tegument of the males, treatments LC₅, LC₁₀ and LC₁₅ differed in the first generation; the treatments LC₅, LC₁₀ and LC₂₀ in the second generation and only LC₁₅ in the third generation. The chest weight between all treatments obtained significance between the control in the first generation, with no significant differences being observed in the second and third generations. In the abdomen, the weight in the LC₂₀ in the first generation reached the highest average, differing significantly among all treatments. In subsequent generations, differences were observed only in the LC₁₀ treatment.

The weight of the wings showed significance only to the LC₁₀ treatment in the first generation. In the total weight of the males, treatments LC₂₀ in the first generation and LC₅ in the third generation reached higher averages, not differing significantly from the control. The length of the anterior wings in the LC₅ treatment reached lower averages over the three generations, with significance being observed for LC₁₀ in the first generation, LC₁₅ in the second generation and LC₁₀, LC₁₅ and LC₂₀ third generation. The same was not observed in the width of the anterior wings of the males. Therefore, the length of the posterior wings of the males obtained higher averages for the control, differing significantly between all treatments in the first generation and LC₅, LC₁₅ and LC₂₀ in the second generation. In the last generation, only LC₁₀ showed significance among all treatments. The same was not observed in the width of the posterior wings of the males.

The area of the anterior wing of the males obtained averages superior to the control in the three generations, differing significantly from the treatments LC₅, LC₁₀ and LC₁₅ in the first generation and LC₅, LC₁₀, LC₁₅ and LC₂₀ in the third generation. The same was not observed about the area of the anterior wing of the males (Table 1).

Table 1

Bodyweight, wing size and a wing area of *Anticarsia gemmatalis* submitted to sublethal doses of the bioinsecticide Dipel SC over three generations

Males								
Treatments	Body Weight (mg)					Wing Sizes (mm)		
	Tegument	Thorax	Abdomen	Wing	Whole Adult	Anterior Length	Width	Posterior Length
1° Generation								
LC ₅	69.87±0.05cC ¹²	40.22±0.03cC	29.65±0.05cB	44.36±0.05aA	114.22±0.07bAB	17.36±0.39cA	9.52±0.29bB	12.12±0.
LC ₁₀	112.71±0.02bA	51.05±0.02bA	61.66±0.02bA	11.23±0.02bA	123.94±0.02bA	18.42±0.19bA	9.34±0.14cC	13.46±0.
LC ₁₅	64.96±0.02cB	20.59±0.02dB	44.37±0.03bcA	41.49±0.02aA	106.45±0.01bA	17.51±0.17cB	10.52±0.17aA	13.51±0.
LC ₂₀	126.22±0.04aA	49.16±0.01bA	87.06±0.04aA	40.25±0.04aA	176.47±0.05aA	17.93±0.33bcA	9.83±0.23bB	11.83±0.
Control	129.75±0.04aA	67.34±0.04aA	56.42±0.04bB	39.72±0.04aA	163.47±0.04aA	19.55±0.18aA	14.60±0.14aA	14.54±0.
2° Generation								
LC ₅	91.29±0.02cB	50.68±0.01aB	68.61±0.02aB	14.07±0.03aB	105.36±0.04bB	17.57±0.17bA	9.85±0.22bB	13.58±0.
LC ₁₀	93.39±0.01cB	53.32±0.01aA	40.06±0.01bB	13.14±0.01aA	106.53±0.01bA	17.93±0.19bA	10.13±0.21abB	14.40±0.
LC ₁₅	113.14±0.02abA	58.77±0.02aA	54.37±0.03abA	7.86±0.02aB	121.00±0.02bA	18.87±0.22aA	10.46±0.15aA	13.65±0.
LC ₂₀	106.02±0.03bcB	52.68±0.01aA	53.34±0.02abB	6.81±0.01aB	112.83±0.03bB	17.85±0.24bA	10.42±0.14aA	13.96±0.
Control	127.22±0.05aA	58.52±0.00aB	40.70±0.05bAB	21.02±0.04aB	148.24±0.05aAB	19.32±0.14aA	10.53±0.16aB	14.93±0.
3° Generation								
LC ₅	119.68±0.01aA	59.54±0.02aA	60.13±0.02abA	9.74±0.01aB	129.41±0.01abA	16.96±0.29cB	10.05±0.21bA	12.90±0.
LC ₁₀	106.20±0.02abAB	50.97±0.02aA	55.22±0.03bAB	9.35±0.02aA	115.55±0.02bA	19.35±0.34aA	10.90±0.27aA	15.01±0.
LC ₁₅	102.07±0.02bA	51.29±0.02aA	76.79±0.03aA	6.23±0.02aB	108.31±0.02bA	17.96±0.15bB	10.74±0.15aA	13.08±0.
LC ₂₀	107.38±0.02abB	49.08±0.02aA	58.30±0.03abB	6.81±0.02aB	114.19±0.02bB	17.58±0.17bcA	10.47±0.16abB	13.59±0.
Control	122.09±0.02aA	46.05±0.01aC	50.04±0.02bA	18.73±0.03aB	140.82±0.02aB	17.40±0.23bcB	9.46±0.20cC	13.93±0.

¹Mean ± standard error. ²Means followed by the same lower case letter between treatments and upper case letters between generations do not differ significantly.

Females Morphometry

In weight of the tegument of the females, the treatment LC₅ in the first generation, LC₁₀ in the second generation and LC₁₅ in the third generation reached higher averages. The weight of the chest of treatment LC₅ in the first generation, LC₁₀, LC₁₅, LC₂₀ and control in the second generation and LC₅, LC₁₅ and LC₂₀ in the third generation had the highest averages, not significantly different between both.

In the abdomen, LC₁₅ and LC₂₀ in the first generation, LC₅ and LC₂₀ in the second generation and LC₂₀ in the third generation reached lower averages among all other treatments. However, weight of the wings, did not show significance between treatments and generations. The total weight of the females revealed that the LC₅ in the first generation differed among all treatments. The same was not observed in the second and third generations.

In the length of the anterior wing, LC₁₀ and control in the first and second generation reached the highest averages, with significance with the treatments LC₅ and LC₂₀, respectively. In the third generation, there was no significance between treatments. In the width of the anterior wing, it was observed that only in the second generation, the treatments did not differ between both.

The length of the posterior wing showed higher averages at LC₁₀ and control over the first and second generation, with significance for the LC₅ treatment. In the third generation, there was no significant difference between treatments. Regarding the width of the posterior wing, LC₅ presented lower averages, differing significantly from the control treatment.

The area of the anterior wing in the LC₅ treatment in the first and second generation reached lower averages than the control treatment, differing significantly between both. In the third generation, there was no significant difference between treatments. The same was not observed in the area of the posterior wing of the females (Table 2).

Variations were observed in the wing area of males in all generations, with an increase in the area of the anterior and posterior wings according to the increase in the exposed sub-dose. The same fact was not observed in the LC₂₀ treatment in the second and third generations in males (Table 1). In females, the same

does not occur, but after the second generation, the area of the anterior wings became more stable, with no significant difference in the third generation. About the posterior wing, the fact occurred only in the second generation in females (Table 2).

Table 2
Bodyweight, wing size and a wing area of *Anticarsia gemmatalis* submitted to sublethal doses of the bioinsecticide Dipel SC over three generation

Females								
Treatments	Body Weight (mg)					Wing Sizes (mm)		
	Tegument	Thorax	Abdomen	Wing	Whole Adult	Anterior Length	Width	Poster Length
1° Generation								
LC ₅	131.81±0.03aA ¹²	68.98±0.03aA	62.82±0.02aA	18.62±0.03aA	150.42±0.03aA	16.89±0.22bA	9.37±0.29bA	11.97±
LC ₁₀	108.05±0.03bA	49.56±0.02bcA	58.49±0.02abA	16.38±0.03aA	124.43±0.03bA	18.37±0.27aA	10.34±0.16aA	14.32±
LC ₁₅	78.50±0.4cB	35.87±0.03dB	42.63±0.03cB	5.31±0.01aA	83.81±0.04cC	17.16±0.35bA	9.09±0.32bB	12.70±
LC ₂₀	86.96±0.03cA	42.93±0.02cdA	44.02±0.02cA	5.97±0.01aA	92.93±0.03cA	16.51±0.27bB	9.46±0.30bA	11.57±
Control	108.11±0.02bA	56.05±0.04bA	52.06±0.03bcB	14.87±0.02aA	122.98±0.02bA	19.03±0.33aA	10.77±0.38aA	14.22±
2° Generation								
LC ₅	87.79±0.00cA	44.28±0.01bB	43.51±0.03bB	7.81±0.01aA	95.60±0.03bC	17.50±0.16bA	9.83±0.23aA	12.57±
LC ₁₀	115.63±0.03abB	54.62±0.01aA	61.01±0.03aA	3.44±0.01aA	119.07±0.03aA	18.39±0.13aA	10.04±0.22aAB	13.47±
LC ₁₅	101.35±0.03bcA	49.89±0.02abA	51.46±0.02abA	3.06±0.01aA	104.41±0.03abB	17.73±0.23abA	10.46±0.17aA	12.72±
LC ₂₀	86.06±0.02cA	45.49±0.01abA	40.57±0.02bA	7.25±0.01aA	93.32±0.02bA	17.48±0.17bA	9.81±0.23aA	12.91±
Control	117.32±0.00aA	52.90±0.00abAB	64.42±0.02aA	3.58±0.01aA	120.90±0.02aA	18.44±0.16aA	10.54±0.23aA	13.92±
3° Generation								
LC ₅	98.18±0.03bA	47.14±0.03abB	51.04±0.02abB	16.20±0.03aA	114.38±0.04abB	17.48±0.28aA	9.54±0.25bA	13.10±
LC ₁₀	86.14±0.03bB	39.39±0.02bB	46.75±0.02bB	24.84±0.01aA	110.99±0.03abA	17.39±0.22aB	9.86±0.16abB	13.11±
LC ₁₅	114.24±0.03aA	53.80±0.02aA	60.44±0.02aA	13.91±0.01aA	128.15±0.03aA	16.85±0.16aA	9.49±0.16bA	12.62±
LC ₂₀	87.80±0.03bA	45.04±0.02abA	42.76±0.02bA	9.03±0.01aA	96.83±0.03bA	17.51±0.25aAB	10.40±0.24aA	13.01±
Control	92.25±0.02bB	46.11±0.01abC	46.14±0.02bB	38.32±0.03aA	130.57±0.03aA	17.19±0.18aB	9.95±0.17abB	12.57±

¹Mean ± standard error. ²Means followed by the same lower case letter between treatments and upper case letters between generations do not differ significantly.

Linear Correlation Stratifies

In the first generation males, the parameters tegument + abdomen, thorax + healthy adult, and wing + healthy adult achieved moderate positive linear correlations to the LC₅ and control treatments. However, females in the parameters tegument + thorax, tegument + abdomen, intact adult + abdomen, inferior length + superior width, thorax + intact adult and intact adult + integument, reached, predominantly, moderate to strong positive linear correlations to the LC₁₅ and LC₂₀ treatments, respectively. The same was not observed in the second generation of males, with moderate linear correlations for LC₅ and LC₁₀ in the parameters tegument + abdomen, chest + abdomen and healthy adults + abdomen. In females, the treatments that presented strong linear correlations were LC₁₀ and LC₁₅ to the parameters integument + abdomen, integument + healthy adults and healthy adults + abdomen.

In the third generation, the males in the control treatment showed moderate positive linear correlations to the parameters wing + intact adult, integument + upper wing, wing + upper width and upper width + whole adult. Unlike females who obtained moderate negative linear correlations to the control treatment in the parameters thorax + upper size width, lower size length + thorax, integument + wing, integument + healthy adult, wing + abdomen, healthy adult + abdomen and thorax + abdomen. Therefore, a greater number of moderate positive correlations to treatment LC₅ with the parameters integument + abdomen, integument + intact adult, intact adult + abdomen, thorax + abdomen and integument + thorax, respectively (Figure 1).

Discussion

The sublethal effect of Dipel SC sub-doses on *A. gemmatalis* morphometry varied according to the concentration of the bioinsecticide. The sublethal effects observed in the bodyweight of adults may be associated with the differential susceptibility between the sexes exposed to sub-doses of the bioinsecticide

based on *B. thuringiensis*, in addition to the physiological changes that are reflected in the adult phase (Retnakaran et al., 1983; Alix et al., 2001; Desneux et al., 2007; Sedaratian et al., 2013).

The chest weight was higher than that of females in both generations. In field conditions, one should consider the higher energy expenditure of males to locate and court females and, therefore, the greater need for chest muscles to be developed (Srygley and Chai, 1990). The relative speed of flight in insects is correlated with the chest mass, and the sublethal effects caused by the bioinsecticide can interfere with the formation of muscles essential to flight. This region concentrates phasic muscles, which commonly work to move appendages in the exoskeleton (Howland, 1974). This arrangement of muscles within the insects' rib cage is directly related to weight, because the larger it is inferred that the male will have better physical conditioning (Srygley and Chai, 1990). Individuals who have these morphometric characteristics exhibit, for example, a higher frequency of copulations, better biological and even physical conditioning (Mare and Corseuil, 2004).

The abdomen is another fundamental structure for the proper functioning of all insect functions. This structure is responsible for energy reserves and the weight parameter is linked to the amount of this reserve. However, the balance between chest and abdomen must exist for the insect to perform the basic functions for survival (Srygley and Thomas, 2002). The hovering flight that insects present is a major component of the energy cost, requiring a greater energy reserve in the abdomen (Srygley and Chai, 1990). This type of flight has advantages because it allows the insect to escape from predators through high-speed flights (Marden and Chain, 1991).

Among the treatments, LC₅ in the first generation and CL₁₀ in the second generation were those with lower thresholds in relation to the weight of the chest and abdomen, considering the proportions of the body smaller than the females. Body size significantly affects most of the physiological characters linked to survival and reproduction, one of the most important quantitative characteristics subject to evolution (Darwin, 1859; Schmidt-Nielsen, 1984; Roff, 1992; Stearns, 1992).

Smaller individuals are potentially less likely to perpetuate their offspring, due to competitive disadvantages compared to other males and the lower acceptability of females (Stearns, 1976). The choice for the female, in this case, can occur, in such a way, that each female has its optimum male size to copulate. This fact, is closely linked to the hypothesis of the physiological capacity of insects to define patterns of allometric measurements (Borgia, 1979).

This optimal size would be the result of a trade-off between the negative influences that the female has with large males on fertility and the advantages of large males for the biological conditioning of the offspring (Clutton-Brock and Parker, 1992; Andersson, 1994). However, even individuals who presented smaller sizes such as LC₅ and LC₁₅ in the first generation, LC₅ and LC₁₀ in the second generation and LC₁₅ and LC₂₀ in the third generation may not perpetuate their offspring, considering that the larger body size generally increases the pairing success due to intraspecific competition or female choice (Clutton-Brock and Parker, 1992; Andersson, 1994).

Wing proportions are influenced, according to the size of the rib cage, as individuals with larger wings have more developed muscles (Marden and Chain, 1991). Morphometry studies confirm that the insects' anterior wings have an important allometric measurement in determining size and shape (Mare and Corseuil, 2004; Sane, 2003). This fact is called phenotypic plasticity (Gotthard and Nylin, 1995; Loh et al., 2008), and does not justify the great variation only in the wings, but in all the dimensioned segments of this study.

Anterior wings perform the aerodynamic capacity and are closely related to the flight speed. However, the posterior wings function as an airfoil that regulates the direction and maneuverability of the flight (Mare and Corseuil, 2004; Dudley, 2000). *A. gemmatalis* lives in open agroecosystems and travels over long distances, thus requiring a relatively larger wing area (Mare and Corseuil, 2004). Studies monitoring populations of *A. gemmatalis* have shown that these adults can migrate great distances, even crossing entire states in the USA (Buschman et al., 1977). The species is known to be unable to survive the winter in the continental USA. On many occasions, insect pest populations fly dozens of kilometers in search of favorable conditions for development (Buschman et al., 1977; Sosa-Gómez, 2004).

Studies evaluating the morphometry of adults in Pieridae, Nymphalidae, and Papilionidae families have shown positive correlations between the flight speed and chest weight, but negatively for the abdomen weight that has the function of storing energy and the reproductive organs (Srygley and Chai, 1990). Thus, the influence of weight distribution between the chest and abdomen may interfere with the allometric measurements of *A. gemmatalis* due to exposure to the bioinsecticide sub-dose based on *B. thuringiensis* (Sih, 1987; Srygley and Chai, 1990).

The parameters abdomen + intact adults and abdomen + integument in females had a predominance of positive correlations. Biologically, males aim to develop and fertilize females; in turn, females have the function of producing eggs, storing male sperm until the eggs are ready to be fertilized, generating offspring and perpetuating the species (Milano et al., 2008). The region where the female reproductive system is located is in the abdomen and requires that all basic functions communicate and have a good functioning to generate viable offspring, also, the minimum size is of great relevance for the perpetuation of the species (Milano et al., 2010).

In the integument + healthy adult parameters in both sexes, they reinforce the strong correlation between the balance of the segments, between weight and adequate wing size. The morphology of insect wings has a direct effect on a flight and, therefore, on the ability of flying species to explore their environment efficiently. The need to maneuver, hover, accelerate and fly at a low energy cost should affect the shape of the wing and lead to the diversification of wing morphometry, according to the stress exposed to the host (Meresman et al., 2020).

In insects, the variation in wing morphometry suggests that different selective pressures, such as bioinsecticides, act non-uniformly in different regions of the wings, probably due to differences associated with body size (Bai et al., 2012; Tocco et al., 2019; Le-Roy et al., 2019). Therefore, this can influence the ecology and physiology of the population and even the organization of the community. Additional effects can also occur in the type of defense used to prevent predators, parasitoids, entomopathogens and in the development and fertility rates of the insect pest (Srygley and Chai, 1990).

Conclusion

Here, we demonstrate the phenotypic plasticity of *A. gemmatalis* adults submitted to sub-doses of the bioinsecticide based on *B. thuringiensis* in tree generations. Due to the possible difference in susceptibility between the sexes, males have larger body size and more pronounced phenotypic plasticity than females. Phenotypic plasticity is very relevant in issues involving the interface between ecology and evolution, as it is the ability of a single genotype to express alternative forms of morphology, physiological state and/or behavior in response to environmental conditions. Thus, the study of this topic becomes increasingly important as it can predict population resistance, outbreaks and pest resurgence.

Declarations

Ethics approval and consent to participate

This article does not contain any studies with animal. Informed consent was obtained from all individual participants included in the study.

Consent for publication

Informed consent was obtained from all individual participants included in the study for the publication this work.

Availability of data and material

All data generated and analyzed for the current study are presented in this manuscript, and the corresponding author has no objection to the availability of data and materials.

Competing interests

The authors declare no competing interests.

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Authors' contributions

FOF, JAD and RAP conceived and designed the research. FOF, TDS and ACS conducted experiments. FOF, IRC and TDS analyzed the data. FOF wrote the first draft. NPD and RAP revised and edited the manuscript. All authors read and approved the manuscript.

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Figures

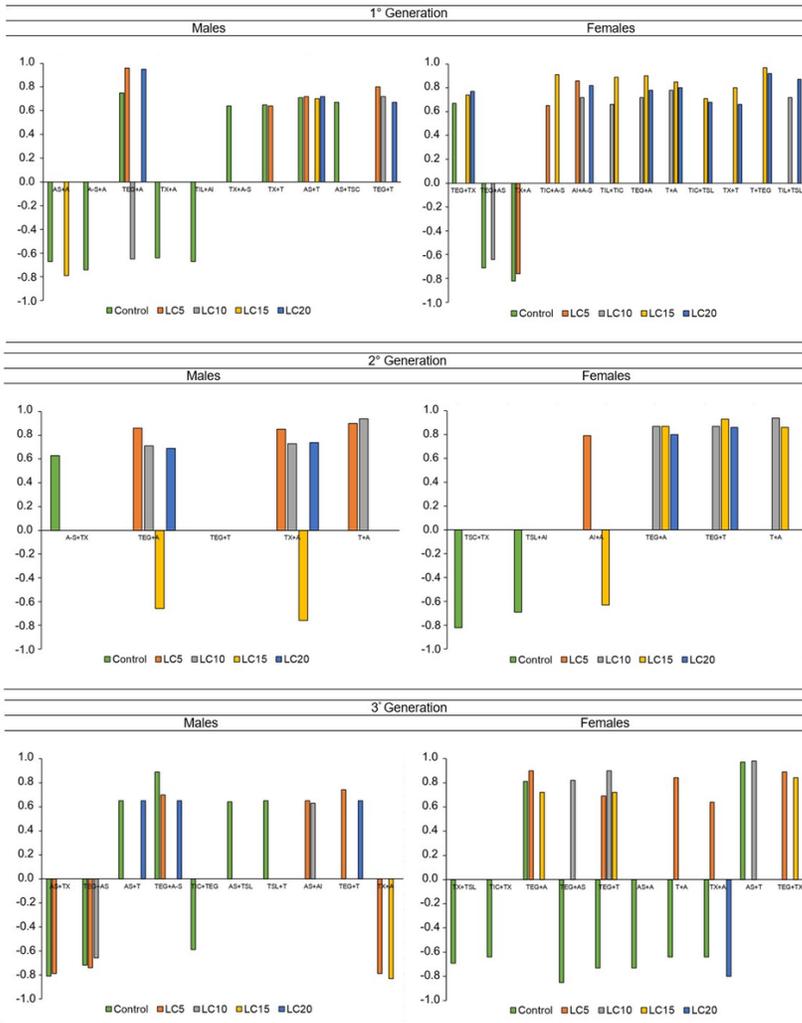


Figure 1
 Linear stratified morphometric correlation of *Anticarsia gemmatalis* submitted to sublethal doses of the bioinsecticide Dipel SC over three generations at $25 \pm 1^\circ\text{C}$, $70 \pm 10\%$ RH and photoperiod L12: D12 h. Tegument (TEG), thorax (TX), abdomen (A), wing (AS), upper wing (A-S), whole adult (T), upper wing length (TSC), lower wing length (TIC), upper wing width (TSL), bottom wing width (TIL), lower wing area (A).