

Feeding, oviposition, and antibiosis of *Paraselenis flava* in sweet potato

Abstract – The objective of this work was to evaluate the feeding, oviposition, and antibiosis of *Paraselenis flava* on sweet potato. The experiment had two phases, as follows: the first one was a choice test by which the insect had access to all genotypes; the second one was a no choice test. In the first phase, 15 genotypes were evaluated and divided into three flesh color groups; and three commercial genotypes were used as control treatments. The six least-preferred genotypes by the insect were subjected to the no choice test. All tests were performed in a laboratory under controlled conditions. The percentage of defoliation, number of adults, and oviposition were evaluated. Chemical compounds and leaf trichomes were quantified 90 days after planting. The insect caused defoliation in all sweet potato genotypes. It did not laid eggs on the white flesh LI-17, C-12, and LI-04, or on the orange flesh C-14 and U2-05, or the purple flesh U2-10, F-21, U1-15, and U2-12, or on the 'Luiza' genotype. The phenolic compounds, total flavonoids, total sugars, and leaf trichomes varied among genotypes. Although the genotypes vary for chemical compounds and leaf trichomes, there is no correlation between these factors and the resistance to *P. flava*.

Index terms: *Ipomoea batatas*, *Paraselenis flava*, antixenosis, plant breeding, leaf-eating insects.

Alimentação, oviposição e antibiose de *Paraselenis flava* em batata-doce

Resumo – O objetivo deste trabalho foi investigar a alimentação, oviposição e antibiose de *Paraselenis flava* em batata-doce. O experimento teve duas fases, conforme a seguir: a primeira foi um teste de escolhas, quando o inseto tinha acesso a todos os genótipos; a segunda foi um teste sem escolhas. Na primeira fase, 15 genótipos de batata-doce foram avaliados e divididos em três grupos de coloração de polpa, e três genótipos comerciais foram usados como tratamento-controle. Os seis genótipos menos preferidos pelo inseto foram submetidos ao teste sem escolha. Todos os testes foram realizados em laboratório, em condições controladas. A percentagem de desfolha, o número de adultos e a oviposição foram avaliados. Os compostos químicos e os tricomas foliares foram quantificados aos 90 dias após o plantio. O inseto *P. flava* causou desfolha em todos os genótipos de batata-doce. O inseto não depositou ovos sobre os genótipos de polpa branca LI-17, C-12 e LI-04, nem sobre os de polpa laranja C-14 e U2-05, os de polpa roxa U2-10, F-21, U1-15 e U2-12, e nem sobre o genótipo 'Luiza'. Os compostos fenólicos, flavonoides totais, açúcares totais e tricomas foliares variaram entre os genótipos. Os genótipos variam quanto aos compostos químicos e tricomas foliares, mas não se encontra correlação entre esses fatores e a resistência ao inseto *P. flava*.

Termos para indexação: *Ipomoea batatas*, *Paraselenis flava*, antixenose, melhoramento de plantas, praga de parte aérea de plantas.

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Introduction

Sweet potatoes (*Ipomoea batatas*) are a great alternative for least developed regions and can be grown in low to medium fertility soils (Motsa et al., 2015; Kwak, 2019). Currently, even in relevant sweet potato production hubs, the average yield of the crop is low. In an important Brazilian sweet potato producing region, in the western São Paulo state, the average yield is 17 Mg ha⁻¹, which is below the genetic potential already verified in other producing countries (IBGE, 2021; Toroco et al., 2023). The average sweet potato yield in Brazil is 14.6 Mg ha⁻¹ (IBGE, 2021). The relatively low Brazilian yield is closely related to the use of obsolete genotypes that are susceptible to pests and pathogens, which respond poorly to improvements in the management of the crop (Otoboni et al., 2020; Zeist et al., 2022).

Sweet potato breeding efforts have focused mainly on traits related to yield, quality of tuberous roots, and resistance to soil pests. However, no reports could be found on selections regarding the resistance to pests that damage aboveground structures of the plant. Numerous insects can feed and reproduce on sweet potato leaf and vines, but few pests cause economic damage. Until now, soilborne pests were the only ones of economic importance in Brazilian sweet potato crops (Leal et al., 2021).

However, with the emergence of important sweet potato production centers in Brazil, airborne pests have begun to cause direct and indirect injury to sweet potato crops (Oliveira et al., 2019). The airborne insect *Paraselenis flava* L. (Coleoptera: *Chrysomelidae*) causes injuries to leaves (Montes & Raga, 2010). The species *P. flava* has the popular name of tortoise beetle, which is due to its peculiar shape of the elytra that resembles a shell of a turtle (Chaboo, 2007). Adults of *P. flava* are characterized by cassidine shape, with a circular body and yellowish-green color.

The larvae and adults of *P. flava* feed on leaves, which reduces the photosynthetic area of the plant, delaying the vegetative development and compromising the accumulation of carbohydrates in roots. During the spring-summer cultivation of sweet potato, this pest has shown high levels of infestation, causing damage to large areas (Montes & Raga, 2010). At the same time, there are no insecticides registered to control this pest, thus making its control difficult at field level. An interesting alternative is the search for

genetic resistance. Screening genotypes for resistance to this insect is a strategy that may contribute to the production sustainability of sweet potato.

The objective of the present work was to evaluate the feeding, oviposition, and antibiosis of *P. flava* in sweet potato.

Materials and Methods

Fifteen sweet potato genotypes of three flesh color groups were tested. The white/cream flesh genotypes were L1-04, C-06, L1-17, K-39, and C-12. The orange flesh genotypes were C-14, U2-19, F-34, U2-05, and U1-25. The purple flesh genotypes were F-21, U2-12, U1-15, K-25, and U2-10. These genotypes were selected in previous trials because of their yield, tuberous root characteristics, precocity, and resistance to soil pests. Three commercial genotypes for each flesh color group were chosen as control treatments: 'Canadense' as white flesh; 'SCS370 Luiza' as purple flesh; and 'Beauregard' as orange flesh. Three separated experiments were conducted, one for each flesh color group.

Propagation material, vines, and leaves used in the experiment came from plants of a maintenance nursery, free of pathogens and pest arthropods, and without traces of phytosanitary products. The *P. flava* specimens used in the experiments were obtained from a stock of sweet potato harvested from an experimental area of the Universidade do Oeste Paulista (Unoeste), Campus II (22°07'S, 51°27'W, at 430 m altitude). The climate is Cwa, according to the Köppen-Geiger's classification, with 25°C average annual temperature, and 1,400 to 1,500 mm average annual precipitation. It is characterized by two distinct periods, a rainy one from October to March, and a low precipitation one from April to September.

The experiments were carried out in two phases; the first one was a choice test, when the insects were allowed to choose whatever genotype they wanted to pray and lay eggs. The genotypes that performed best (less praying and less eggs) were chosen for the second phase, when the experiment was a no choice test.

The choice test experiments were carried out to evaluate the preference of *P. flava* toward sweet potato genotypes for oviposition and feeding. Plants with five fully expanded leaves were kept in pots with 0.5 dm³ and placed on benches in an air-conditioned room, at

25±2°C, 70±10% relative humidity, and 12 hours of light.

The three experiments were conducted in a completely randomized experimental design, with nine replicates. Each of the experiments encompassed five genotypes, and their corresponding commercial genotypes of the same flesh color group as control. Each plot consisted of one plant (experimental unit). Twenty-four adults of *P. flava* were released in the center of each plot. The following evaluations were carried out at 24, 48, 72, and 96 hours after release: number of adults present on plants; number of eggs; and percentage of defoliation, using a 5-point scale (1, from 0% to 20% defoliation; 2, from 21% to 40% defoliation; 3, from 41% to 60% defoliation; 4, from 61% to 80% defoliation; and 5, from 81% to 100% defoliation).

The no choice test experiment was conducted in a completely randomized design, with nine replicates. There were nine treatments encompassing six genotypes that performed the best in previous phase, at least one belonging to each flesh color group, plus the three commercial controls.

The experiment was carried out in laboratory under controlled conditions and plants kept in 2 dm³ pots. The test was conducted in an air-conditioned room at 25±2°C temperature, 70±10% relative humidity, and 12 hours of light. When plants had 5 fully expanded leaves, a plastic cage and *voile* fabric were attached to each pot. In each cage, 10 female adults of *P. flava* were released. After 48 hours, a single set of eggs and the corresponding offspring were kept. Females of *P. flava* oviposit in a single location in the abaxial surface of leaves, grouping their eggs and protecting them until adults emerge (Montes & Raga, 2010). Ten females were used in order to guarantee that at least one of them would oviposit within 48 hours. The offsprings were kept in a cage until adults emerged. The following evaluations were carried out: egg period, duration of the larval period, larval weight and size, pupal cycle, and egg-adult cycle.

Phenolic compounds, flavonoids, and total soluble sugars of the six sweet potato genotypes chosen for the second phase were quantified. Leaves were collected from plants 90 days after planting. In two leaves per replicate, leaf trichomes at the leaf base were quantified using a 5-point scale: 1, no trichomes present; 3, few trichomes present; and 5, trichomes

present in abundance. An area of 1.9 cm² was counted under a microscope at 1X magnification.

Leaf extracts were obtained according to methodology presented by José et al. (2015). The quantification of flavonoids were determined according to Yao et al. (2013). Total polyphenols were determined according to Stagos et al. (2012). Total sugars were determined according to the methodology of Yemm & Willis (1954).

Data were tested for normality of errors, using the Shapiro-Wilk's test; for the homogeneity of variances, using the Levene's test; and for the independence of errors, using the Durbin-Watson's test. The data were transformed according to the Box-Cox's methodology proposed by Hawkins & Weisberg (2017). The means were analyzed by two-way analysis of variance, and the results were compared using the F test at 1% probability. The multiple comparison of means were performed using the Scott-Knott's test at 5% probability. The analyses were carried out using the AgroEstat statistical program (Barbosa & Maldonado Júnior, 2015).

Results and Discussion

Defoliation and adults were observed in all white flesh genotypes and evaluations (Figure 1). Most genotypes showed defoliation class between 1 and 2, that is, up to 40% defoliation, except for L1-17 and C-12 in the last evaluation (96 hours). Genotypes L1-17, C-12 and L1-04 did not show eggs in any of the evaluations. The K-39 and 'Canadense' genotypes showed eggs within 48 hours after the beginning of the experiment, and the C-06 genotype showed eggs only in the last evaluation (96 hours).

There were adult insects and defoliation in all orange flesh genotypes, in all evaluations (Figure 2). Genotypes C-14 and U2-05 did not show eggs in any of the evaluations. Most of the damage to the leaf area (defoliation) seems to have occurred within the first 24 hours. Similarly, oviposition occurred in the first 24 hour after the beginning of the experiment. For the UBD F-34 genotype, there was a greater number of adults present in the first 24 hours than that of the last evaluation.

All purple flesh genotypes were defoliated, and adults were observed in all evaluations (Figure 3). Only the K-25 genotype showed eggs, which were

counted from the second evaluation (48 hours after the beginning of the experiment). Most genotypes showed defoliation class between 1 and 2, that is, up to 40% defoliation.

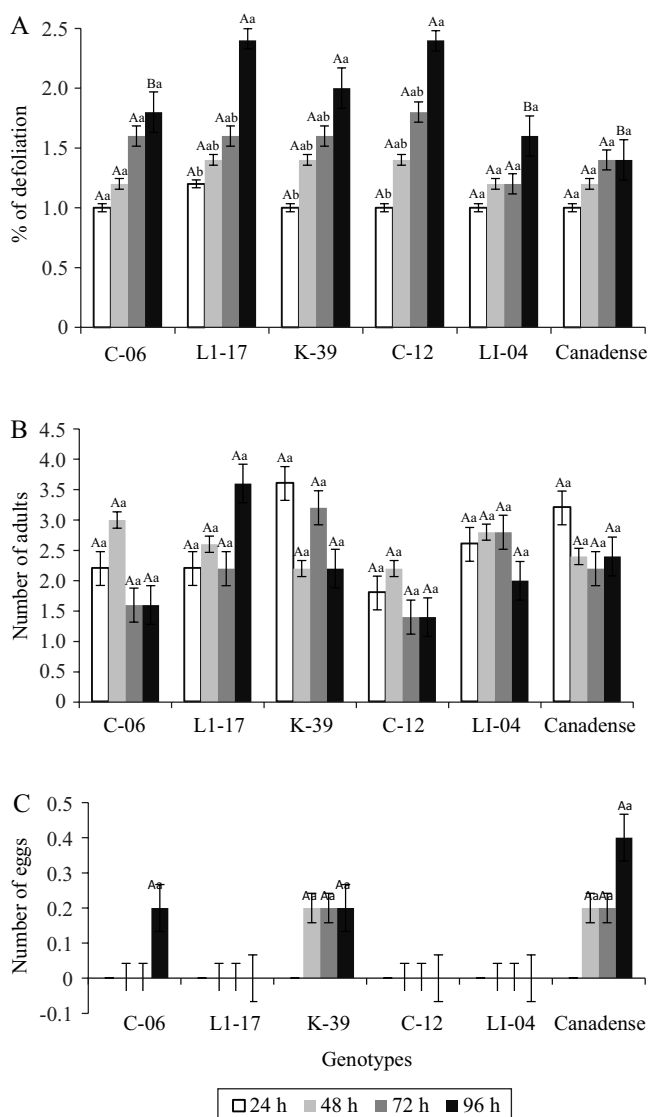


Figure 1. Mean and standard error of percentage of defoliation (A), number of adults of *Paraselenis flava* (B), and its number of eggs (C) in white flesh sweet potato (*Ipomoea batatas*) genotypes at different number of hours after the beginning of the experiment. Uppercase letters compare the genotypes in each evaluation, and lowercase letters compare the evaluations of each genotype, by the Scott-Knott's test, at 5% probability. The means presented in the graphs are those before transformation. The statistics are from transformed data using the Box-Cox's methodology proposed by Hawkins & Weisberg (2017).

The average number of insects present on plants decreased as the percentage of defoliation increased. In turn, defoliation increased throughout the evaluations. Damage from *P. flava* causes a decrease of the leaf area, reducing the photosynthesis and, consequently, negatively influencing the crop development, yield, and quality. There are no insecticides registered for control, which makes it difficult to handle the

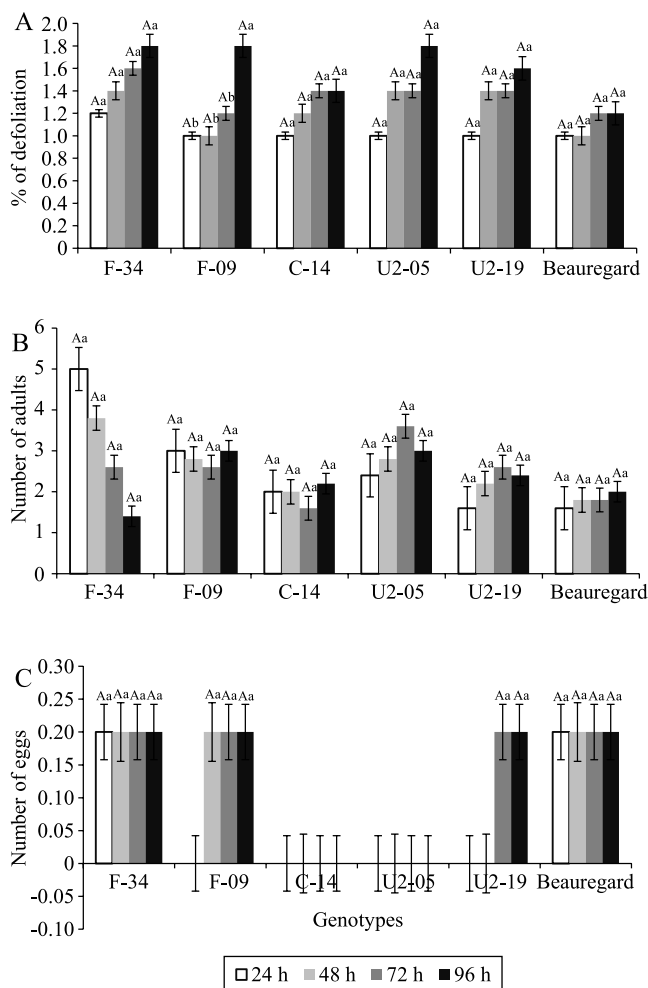


Figure 2. Mean and standard error of percentage of defoliation (A), number of adults of *Paraselenis flava* (B), and its number of eggs (C) on orange flesh sweet potato (*Ipomoea batatas*) genotypes at different number of hours after the beginning of the experiment. Uppercase letters compare the genotypes in each evaluation, and lowercase letters compare the evaluations of each genotype, by the Scott-Knott's test, at 5% probability. The means presented in the graphs are those before transformation. The statistics are from transformed data using the Box-Cox's methodology proposed by Hawkins & Weisberg (2017).

insects and requires that producers adopt alternative solutions (Oliveira et al., 2019). The identification of a resistant genotype can be an alternative for sustainable management of this insect.

Adults were detected for almost all genotypes, including commercial controls. However, some

genotypes were not oviposited by *P. flava*, which may present some level of resistance.

The genotypes subjected to the no choice test had similar and nonsignificant results for egg cycle, larval cycle, pupal cycle, and egg-adult cycle. However, the evaluated genotypes varied for larval weight, larval size, and number of adults obtained at the end of the egg-adult cycle (Table 1).

L1-04 increased the pupal and egg-adult cycle of *P. flava* (Table 1). The increase of the time required to complete part or all the insect cycle may indicate some level of resistance and constitute an important aspect in integrated pest management. The adoption of complementary pest management practices can be more effective by extending the development period.

A lower number of adults was found on the U2-12 genotype, although the period of pupal and total development was not changed (Table 1). This result may represent some intrinsic mechanism to this genotype, regarding the viability of completing the egg-adult cycle by the insect.

Plants have defense mechanisms that prevent or reduce pest attacks through properties that hinder the movement, feeding, or reproduction of insect pests (Yactayo-Chang et al., 2020). Defense mechanisms can be physical barriers, the presence of a sticky substance on the leaf or other part of the plant, or the production of specialized enzymes or metabolites (Kortbeek et al., 2021).

During the choice test, the content of phenolic compounds, total flavonoids, and total sugars were quantified in the six selected genotypes that showed a greater potential for sweet potato resistance to *P. flava*. The L1-04 genotype showed the highest value of phenolic compounds, but did not differ from 'Beauregard' and C-14 (Table 2). 'Canadense' stood out for total flavonoid content. 'Luiza' showed the worst performance for phenolic compounds and total flavonoids, but exhibited the highest average for total sugars, although it did not differ from 'Beauregard', C-14, and L1-04.

It is important to consider that L1-04 stood out for phenolic compounds and extended the pupal and total cycle (Tables 1 and 2). Thus, the relationship between the content of phenolic compounds in sweet potato leaves and *P. flava* antibiosis should be studied more closely in the future.

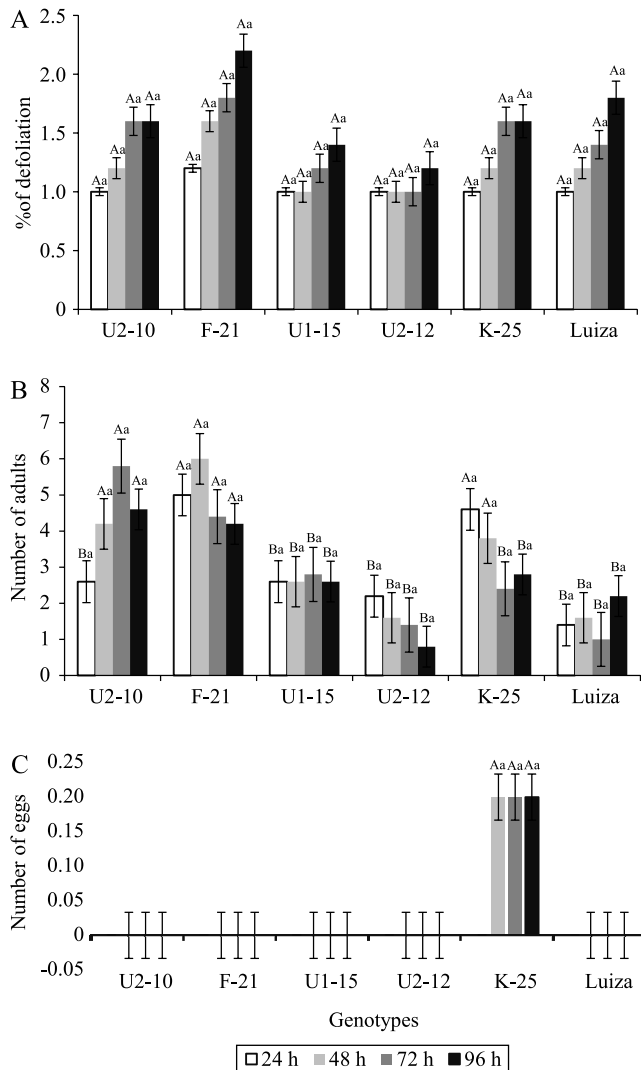


Figure 3. Mean and standard error of percentage of defoliation (A), number of adults of *Paraselenis flava* (B), and its number of eggs (C) on purple flesh sweet potato (*Ipomoea batatas*) genotypes, at different number of hours, after the beginning of the experiment. Uppercase letters compare the genotypes in each evaluation, and lowercase letters compare the evaluations of each genotype, by the Scott-Knott's test, at 5% probability. The means presented in the graphs are those before transformation. The statistics are from transformed data using the Box-Cox's methodology proposed by Hawkins & Weisberg (2017).

The genotypes differed for the presence of trichomes at the base of the leaf blade. 'Canadense', L1-04, C-14, and U2-12 did not showed trichomes on the abaxial and adaxial sides. 'Beauregard' and 'Luiza' had trichomes abundance on both sides. U2-12 showed some trichomes on the adaxial side, but none on the abaxial side. These results indicate that the presence of trichomes has little or no effect on the occurrence of *P. flava* on sweet potato.

Insect resistance on sweet potato has been poorly studied. The few available studies focus on soilborne pests, given the greater economic importance that these species can show, as they attack the commercial part, the tuberous roots (Leal et al., 2021; Tedesco et al., 2023; Yang et al., 2023). It is important to highlight that although *P. flava* is considered a secondary pest, it has become increasingly frequent in important sweet potato producing areas, especially in the summer, when sweet potato is most often cultivated.

On sweet potato, *P. flava* can occur since planting, if the propagation material is not properly selected (Montes & Raga, 2010). This aspect is relevant because sweet potato is vegetatively propagated, generally from crops in development within 60 to 90 days of planting. Thus, the occurrence of *P. flava* in the area of mother plants, from which vines will be obtained, may compromise the crop that will be implemented.

The egg-adult cycle of this pest is 30 days. So, it would be possible to have four to five cycles in a single sweet potato crop, if the pest occurs from the

beginning. This aspect is relevant, as the appropriate climatic conditions for the development of sweet potato are similar to those of *P. flava*, which could be a risk for a significant increase in the insect population. However, in the first month of development, the plant prioritizes the development of fibrous and tuberous roots in detriment of the leaf area. Therefore, if pest occurs in the first month, there is a high probability of impacting negatively rooting and yield, as the photosynthetic active leaf area would be compromised, which is not yet abundant.

After the tuberous roots formation, the energy source-sink relationship in the plant changes. The plant starts to produce more vines and leaves, in order to synthetize photoassimilates to fill the tuberous roots. At this stage, depending on the genotype and cultivation conditions, there is a large production of branches and leaves, and the attack by *P. flava* would be less significant. However, if a severe attack occurs, and the weather conditions are favorable, a more relevant impact is expected.

Based on the no choice test experiment, there is a preference for feeding and oviposition of *P. flava* specimens for certain genotypes, indicating that there is a possibility of obtaining a resistant cultivar. Furthermore, L1-04 was observed as influencing the number of adults and the period of development of the insect.

The adoption of cultivars with some level of resistance to *P. flava* can be part of an integrated pest management in sweet potato cultivation, as well

Table 1. Mean and standard error of egg cycle (EC), larval cycle (LC), larval weight (LW), larval size (LS), pupal cycle (PC), egg-adult cycle (EAC), and number of adults (NA) of *Paraselenis flava* on sweet potato (*Ipomoea batatas*) genotypes subjected to a no choice test experiment.

Genotype	EC (days)	LC (days)	LW (g)	LS (cm)	PC (days)	EAC (days)	NA
Luiza	7.0±0.00	14.0±0.00	0.009±0.0007	0.80±0.10	9.0±0.00b	30.0±0.00b	26.6±1.53a
Beauregard	6.7±0.58	14.3±0.58	0.0083±0.0009	0.70±0.17	9.3±0.58b	30.3±0.58b	263±2.08a
Canadense	7.0±0.00	14.0±0.00	0.0093±0.0009	0.76±0.15	9.0±0.00b	30.0±0.00b	27.0±1.73a
U2-12	7.0±0.00	14.0±0.00	0.0091±0.0006	0.80±0.10	9.0±0.00b	30.0±0.00b	20.6±1.53b
C-14	7.0±0.00	14.0±0.00	0.0086±0.0001	0.70±0.06	9.0±0.00b	30.0±0.00b	26.0±1.00a
L1-04	7.0±0.00	14.0±0.00	0.0085±0.0016	0.63±0.15	10.0±0.00a	31.0±0.00a	25.6±1.15a
CV (%)	3.39	1.68	10.71	17.31	2.56	0.78	6.10
F test	1.00 ^{ns}	1.00 ^{ns}	0.53 ^{ns}	0.75 ^{ns}	8.80**	8.80**	7.81**
p-value	0.46	0.46	0.75	0.60	0.001	0.001	0.0018

⁰¹Means followed by different letters indicate statistical differences, by the Scott-Knott's test, at 5% probability. For the F test: **Significant at 1% probability. ^{ns}Nonsignificant.

Table 2. Mean and standard error of the contents of phenolic compounds, total flavonoids, and total sugars in leaves of sweet potato (*Ipomoea batatas*) genotypes of the no choice test experiment⁽¹⁾.

Genotype	Content (mg g ⁻¹)		
	Phenolic compounds	Total flavonoids	Total sugars
Canadense	4.50±0.99c	29.33±4.14a	9.10±2.90b
Beauregard	6.52±0.36b	21.75±1.56b	13.48±2.15a
Luiza	5.16±1.50c	13.91±2.55c	16.39±2.28a
L1-04	8.38±1.12a	19.54±1.99b	10.66±2.30b
C-14	5.74±0.24c	17.78±1.81b	13.38±1.78a
U2-12	4.82±1.05c	20.83±1.49b	10.40±0.97b

⁽¹⁾Means followed by equal letters do not differ, by the Scott-Knott's test, at 5% probability.

as representing a sustainable practice that would contribute to avoid the use of insecticides.

Conclusions

1. *Paraselenis flava* causes defoliation in all evaluated sweet potato (*Ipomoea batatas*) genotypes, but it does not lay eggs in several genotypes.

2. Sweet potato genotypes vary for chemical compounds and leaf trichomes, but there is no correlation between these factors and the resistance to *P. flava*.

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