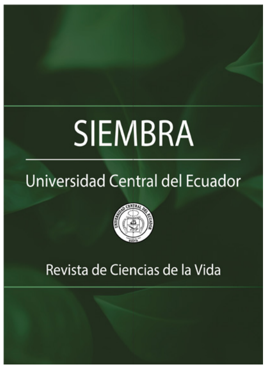
Effect of infestation levels of the rice stink bug (Oebalus insularis Stal.) on mechanical damage in different rice genotypes



Efecto de los niveles de infestación del chinche vaneador (Oebalus insularis Stal.) sobre el daño mecánico en diferentes genotipos de arroz

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Abstract

The stink bug (*O. insularis*) represents a significant threat to rice production in various regions of the world. This hemipteran causes considerable damage by feeding on developing grains, which facilitates the entry of pathogens that promote spotting, resulting in yield and quality losses of rice. We aimed to evaluate the effect of the infestation levels of the stink bug on mechanical damage in rice genotypes under controlled conditions. For this purpose, a rearing system was established to infest rice genotypes with young adults at levels of 0, 2, 4, and 6 insects per plant, incorporated during flowering. Four genotypes (Go-04209, Go-05070, INIAP FL-ÉLITE, and INIAP-20) were evaluated in a Completely Randomized Design [CRD]. The variables analyzed were healthy grains per spike, spotted grains per spike, missing grains per spike, and weight of spike per plant. The data were processed using the SPSS statistical program to perform the analysis of variance, the multiple comparisons tests (Tukey 0.05), and correlation analysis. We determined that the higher the infestation, the yield decreases 46%. Hereby, we propose an action threshold of 1 insect per plant to limit the economic loss to 17%. Genotype Go-04209 showed the least affectation compared to the others, standing out in quality and maturity parameters, constituting promising botanical material for genetic improvement and strengthening of productive development against this pest.

Keywords: pest, infestation, grains, Poaceae, action threshold.

Resumen

El chinche vaneador del arroz (*O. insularis*) representa una amenaza significativa para la producción arrocera en diversas regiones del mundo. Este hemíptero causa daños considerables al alimentarse de los granos en desarrollo, facilitando, además, el ingreso de patógenos promotores del manchado, derivando en pérdidas de rendimiento y calidad del arroz. Esta investigación se propuso evaluar el efecto de los niveles de infestación del chinche vaneador sobre el daño mecánico en genotipos de arroz bajo condiciones controladas. Para su efecto, se estableció una cría que permitió infestar con adultos jóvenes los genotipos de arroz, en niveles de 0, 2, 4, y 6 insectos planta^{-1}, incorporados durante la floración. Se evaluaron cuatro genotipos (Go-04209, Go-05070, INIAP FL-ÉLITE e INIAP-20) en un Diseño Completamente al Azar [DCA]. Las variables analizadas fueron: granos sanos por espiga, granos manchados por espiga, granos vanos por espiga y peso de espiga planta^{-1}. Los datos se procesaron mediante el programa estadístico SPSS, para establecer el análisis de varianza, las consiguientes, pruebas de comparaciones múltiples (Tukey 0,05); y, un análisis de correlación. Como resultado se determinó que a mayor infestación el rendimiento disminuye hasta un 46%; por lo que se propone un umbral de acción de 1 insecto planta^{-1} para limitar la pérdida económica al 17%. El genotipo Go-04209 presentó la menor afectación comparado con los demás, sobresaliendo en parámetros de calidad y cantidad, constituyendo un material botánico promisorio para el mejoramiento genético y el fortalecimiento del desarrollo productivo frente a esta plaga.

Palabras clave: plaga, infestación, granos, Poaceae, umbral de acción.

1. Introduction

Rice (*Oryza sativa* L.) is one of the world's most important crops, forming the dietary foundation for a large portion of the global population (Jiménez-Martínez, 2021). In 2023, the global harvested area of paddy rice reached 168,356,566.00 ha, with a production of 799,999,504.87 t, with India and China being the leading producers (Organización de las Naciones Unidas para la Alimentación y la Agricultura [FAO], 2025). In Ecuador, the national planted area in 2023 was 358,231.00 ha, of which 343,050.00 ha were harvested, resulting in a production of 1,636,349.00 t, mainly contributed by the provinces of Guayas, Los Ríos, Manabí, and El Oro (Instituto Nacional de Estadísticas y Censos [INEC], 2025).

The production of this cereal is consistently threatened by biotic and abiotic factors; among the former, insect pests stand out due to the significant damage they cause (Parrales et al., 2024; Escalona et al., 2023). In the agricultural context, pests are defined as any form of plant or animal life, or any pathogenic agent, that can be harmful to plants or plant products. Their impact varies in severity, generating yield losses that affect the economy and pose risks to food security (Arrua et al., 2022).

The rice stink bug, *O. insularis* Stal (Hemiptera: Pentatomidae), is an insect widely distributed throughout the American continent (Rodríguez-G. et al., 2006) and is present in many production systems such as maize (*Zea mays* L.), rice (*O. sativa* L.), and soybean (*Glycine max* L.), among others. Under certain conditions it can become a pest due to its feeding habits, as its piercing-sucking mouthparts allow it to feed on flowers and developing grains, affecting both yield quantity and quality through direct or indirect effects (Vivas & Astudillo, 2010; Zachrisson, 2010). Direct or mechanical damage results from the suction the pest exerts to extract photoassimilates necessary for feeding, which leads to partially filled or empty grains, giving rise to its common name stink bug (*"chinche vaneador"* in Spanish) (Bhavanam et al., 2021; Rodríguez-G. et al., 2006; VanWeelden et al., 2020; Vivas & Astudillo, 2010).

Indirect damage occurs as a consequence of feeding-related wounds, which create lesions that serve as entry points for various pathogens that compromise grain integrity. As a result, a loss of quality is observed alongside a reduction in yield (Rivero, 2008; Tumanyan et al., 2022, 2024; Weber et al., 2020). The economic losses associated with these damages can be substantial, reaching between 30% and 65% of the total production value (Pérez Iglesias & Rodríguez Delgado, 2019).

This insect can exploit a wide range of food resources, including the following alternative poaceous and fabaceous host plants: jungle rice (*Echinochloa colona* L.), barnyardgrass (*E. crus-galli* L.), large crabgrass (*Digitaria sanguinalis* L.), knotgrass (*Paspalum distichum* L.), Virginia paspalum (*Paspalum virgatum* L.), goosegrass (*Eleusine indica* L.), pangola grass (*Digitaria decumbens* S.), nutgrass (*Cyperus rotundus* L.), Indian fimbry (*Fimbristylis miliacea* L.), among others. Research has shown that these species are strongly inclined to complete their entire life cycle (Vivas & Astudillo, 2010; Zambrano Mero et al., 2024).

There is limited information available on this topic. The damage caused is not restricted to crossed genotypes but also affects varieties that have not been genetically modified (native seeds). The lack of detailed knowledge hinders the development of adequate strategies to mitigate the impact of this insect.

The objective of this research was to evaluate the effect of rice stink bug infestation levels on mechanical damage in rice genotypes (promising lines obtained from crosses) under controlled conditions. This study is essential for identifying materials with strong agronomic performance, which could serve as a basis for rice breeding programs and, eventually, be distributed to farmers—contributing to improved productivity and profitability of the crop.

2. Materials and Methods

2.1. Geographic Location

The study was conducted in the greenhouse (screenhouse) of the Estación Experimental Litoral Sur [EELS] of the Instituto Nacional de Investigaciones Agropecuarias [INIAP], located in the Virgen de Fatima locality, Yaguachi canton, Guayas province, Ecuador (2° 15’ 26” S; 79° 38’ 43” W, y a 17 m a.s.l.).

2.2. Trial management

2.2.1. Establishment of the breeding colony of the rice stink bug

During the preliminary phase, infected rice fields with the pest were visited (Figure 1A), and the insects were randomly collected with an entomological net. The sampled insects were saved inside plastic bags. Then, the adults were separated and placed inside a clear plastic recipient of 18 x 10.5 cm, where rice spikes in the ‘milky’ stage and a moistened cotton pad were provided to them. The upper extreme of the recipient was sealed with tulle held with garters.

Figure 1. A) Collection of adult stink bugs (*O. insularis*) in rice fields. B) Rearing village.

The insects were transported to the greenhouse in a glass cage covered with tulle (50 × 50 × 50 cm) that had been previously disinfected. Inside the cage, a plastic container (9.5 × 6.0 cm) with water for hydration and 305 g of rice spikes (used as a vase) were placed, along with two Petri dishes containing cotton saturated with water and a third dish containing a 10% honey solution impregnated into the cotton.

The food was renewed every 48 hours, and the egg masses deposited on the leaf were removed and placed in Petri dishes with moistened cotton, remaining closed only until hatching (first generation).

In the greenhouse, the insects were kept under continuous monitoring (quarantine) for 15 days to verify the absence of infection by entomopathogenic agents; this procedure was necessary to prevent future contamination of the breeding colony.

For the establishment of the breeding colony, once the possibility of infection by entomopathogenic agents had been ruled out, the insects were placed in their respective glass cages in the rearing facility under controlled conditions (25 °C, 70% relative humidity, and a 12h photophase) as their permanent site (Figure 1B).

Each cage was positioned on four inverted containers placed inside plastic trays that served as the base and contained a water–detergent solution to prevent the entry of arthropods that could compromise the colony, such as ants. Inside each cage, three Petri dishes were placed: two with moistened cotton and one with a 10% honey solution saturated in the cotton.

Regarding feeding, a container with water and rice spikes at the milk-grain stage was placed in the adult cages. As an alternative food source, the use of weeds such as jungle rice (*E. colona* L.) and barnyardgrass (*E. crus-galli* L.) was considered, as well as bean pods (*Phaseolus vulgaris* L.) in case rice spikes were insufficient. However, it was not necessary to use these species due to the ready availability of rice spikes. Two Petri dishes were placed in this cage: one with moistened cotton and another with the previously mentioned energy solution.

In the nymph cages (Figure 2A), three pots with 15-day-old rice seedlings were incorporated to facilitate feeding (Figure 2B and Figure 2C), especially for the second instar, since their mouthparts are weak and they struggle to suck the starchy contents of the grain; otherwise, high mortality could occur. Additionally, rice spikes (Figure 2D) were placed in plastic containers with water, as nymphs of various instars coexisted within the same cage. The spikes were renewed every 48 hours, and the pots with rice seedlings every 3–4 weeks as the plant material deteriorated. The moisture of the pots with seedlings was regulated to avoid standing water and mortality due to drowning.

Figure 2. A) Nymph rearing cage. B) Built-in rice planters. C) Interaction and feeding of nymphs in rice seedlings. D) Contribution of spikes in a milky state.

Regarding the management of egg masses, nymphs, and adults, the egg masses were collected from the food provided every 48 hours and placed in Petri dishes (6.0 cm × 1.5 cm) with sufficient moisture to prevent dehydration. The hatching period was 4–5 days, after which the Petri dish was opened to allow the insects to emerge. Although the first instar is gregarious and does not feed on host plants, it survives thanks to symbionts (bacterial taxa) vertically transmitted by the mother; its developmental period lasts two days. Beginning with the second instar, the insects fed on the cellular contents of the host plants.

Adults were kept in cages separate from the nymphs (Figure 3A), because the coexistence of both stages limited colony growth. In this context, it was observed that adults pierced the egg chorion and fed on the internal contents, using the nutrients for their own development and preventing hatching (Figure 3B).

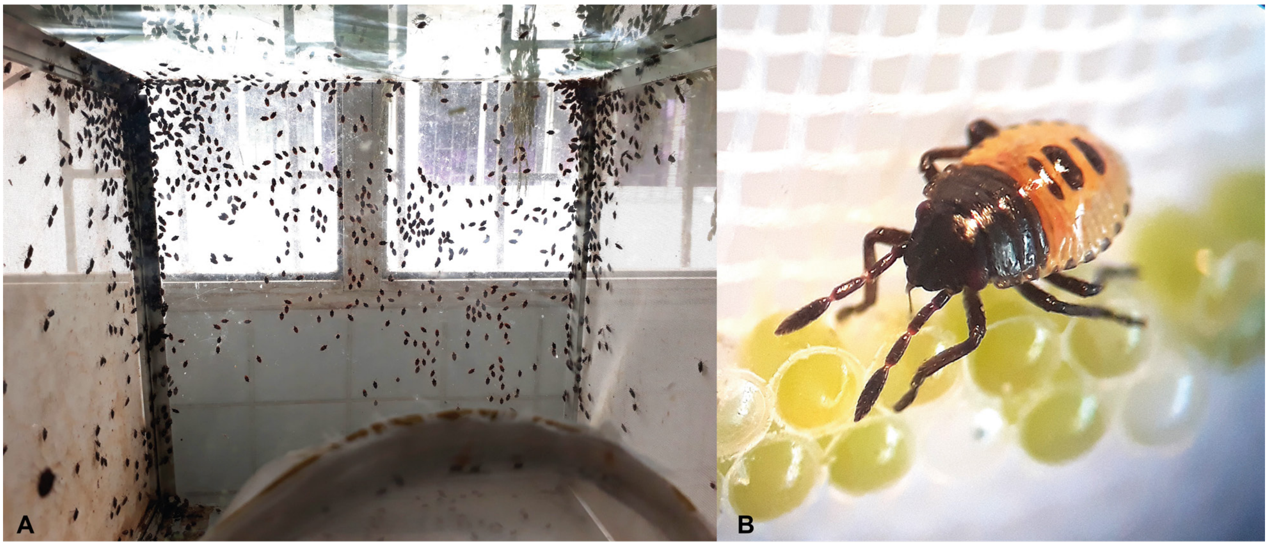

Figure 3. A) Confined adults. B) Nymph feeding on egg the chorion.

Asepsis of the cages consisted of removing dead insects with a brush, along with cleaning the interior walls and base using moistened cloth. The cotton pads containing water, as well as those containing the energy solution, were washed each time the food was renewed and permanently replaced when necessary (at the first signs of fungal growth). Daily observations were carried out to maintain control of the colony.

2.2.2. Controlled infestation of *O. insularis* populations in rice genotypes

In the trial, which occupied an area of 60 m², four rice genotypes were planted: Go-04209, Go-05070, INIAP FL-ÉLITE, and INIAP-20. For this purpose, polystyrene pots measuring 12 × 13 cm were used, each containing 350 g of loosened soil saturated with water to facilitate root development; the water layer was monitored periodically to ensure adequate saturation.

Transplanting was carried out at 18 days into containers with a capacity of 15 liters, filled with the previously mentioned substrate. Two plants of the same genotype were placed in each container to ensure the survival of the host plants during the experiment.

To preserve the integrity of the genotypes after transplanting, the plants were confined using tulle covers (1.35 m in height × 0.52 m in diameter), with each bucket supported by four wooden stakes (1.20 m) to provide stability. Each end of the tulle cover was secured with string to prevent insect escape. Finally, the cultivars were labeled with plastic tags (10 × 10 cm).

At the beginning of the flowering stage, one of the two plants was removed so that infestation could be carried out on the more robust plant. Subsequently, for the infestation, young adults were taken from the breeding colony (Figure 4A), carefully collected with a test tube, and introduced onto each of the host genotypes according to the infestation levels of 0, 2, 4, and 6 insects per plant (Figure 4B). The insects remained feeding for 15 days (Figure 5), after which the spikes were collected.

2.3. Experimental design

A Completely Randomized Factorial Design [CRFD] was used, consisting of 16 treatments (combinations of genotypes and infestation levels), four infestation levels (0, 2, 4, and 6 insects per plant), and three replicates for each combination, totaling 48 experimental units (Figure 6). A random number generator in the statistical software IBM® SPSS® version 21 was used to assign the genotype–infestation level combinations to the experimental units. The mathematical model described in equation [1] was applied.

$$Y_{ijk} = \mu + \tau_i + \gamma_j + (\tau\gamma)_{ij} + \epsilon_{ijk} \quad [1]$$

Where:

- Y_{ijk}: observation obtained at the i-th treatment of the study factor genotype, in the j-th treatment of the study factor level of infestation, and in the k-th replicate.
- μ: overall mean of the response variable across all treatment combinations.
- τ_i: main factorial effect produced by the i-th treatment of the genotype factor.
- $\gamma_{j:}$ main factorial effect produced by the j-th treatment of the study factor level of infestation.
- $(\tau\gamma)_{ij}$: interaction effect produced between the i-th treatment of the study factor genotype and the j-th treatment of the study factor level of infestation.
- εi_{jk}: experimental error associated to the i-th treatment of the study factor genotype to the j-th treatment of the study factor level of infestation, and k-th replicate.

Figure 4. A) Collection of young adults from the rearing cage. B) Incorporation of insects according to the level of infestation.

Figure 5. Stink bug feeding on rice genotypes.

2.4. Evaluation of the mechanic damage

The spikes from each plant and treatment were harvested individually, placed in paper bags (40 × 18 cm), sealed, identified, and transported to the laboratory. The variables measured were:

- Healthy grains per spikes. The filled grains with husk and a healthy appearance were counted in each spike, that is, those lacking complete or partial dark spots.

- Stained grains per spike. Grains showing dark coloration (filled, partially filled, and empty stained grains) with partial or total black spotting were recorded.
- Empty grains per spike. Empty grains without the presence of stains were separated.
- Spike weight (g). Each whole spike was weighed prior to grain removal and category separation.

This protocol was applied to record and summarize the data from each spike according to the studied variables, repetitions, infestation level and genotype.

INIAP-20	T1R1	0
	T1R2	0
	T1R3	0
	T2R1	2
	T2R2	2
	T2R3	2
	T3R1	4
	T3R2	4
	T3R3	4
	T4R1	6
	T4R2	6
	T4R3	6
Go-05070	T5R1	0
	T5R2	0
	T5R3	0
	T6R1	2
	T6R2	2
	T6R3	2
	T7R1	4
	T7R2	4
	T7R3	4
	T8R1	6
	T8R2	6
	T8R3	6
INIAP FL-ÉLITE	T9R1	0
	T9R2	0
	T9R3	0
	T10R1	2
	T10R2	2
	T10R3	2
	T11R1	4
	T11R2	4
	T11R3	4
	T12R1	6
	T12R2	6
	T12R3	6
Go-04209	T13R1	0
	T13R2	0
	T13R3	0
	T14R1	2
	T14R2	2
	T14R3	2
	T15R1	4
	T15R2	4
	T15R3	4
	T16R1	6
	T16R2	6
	T16R3	6

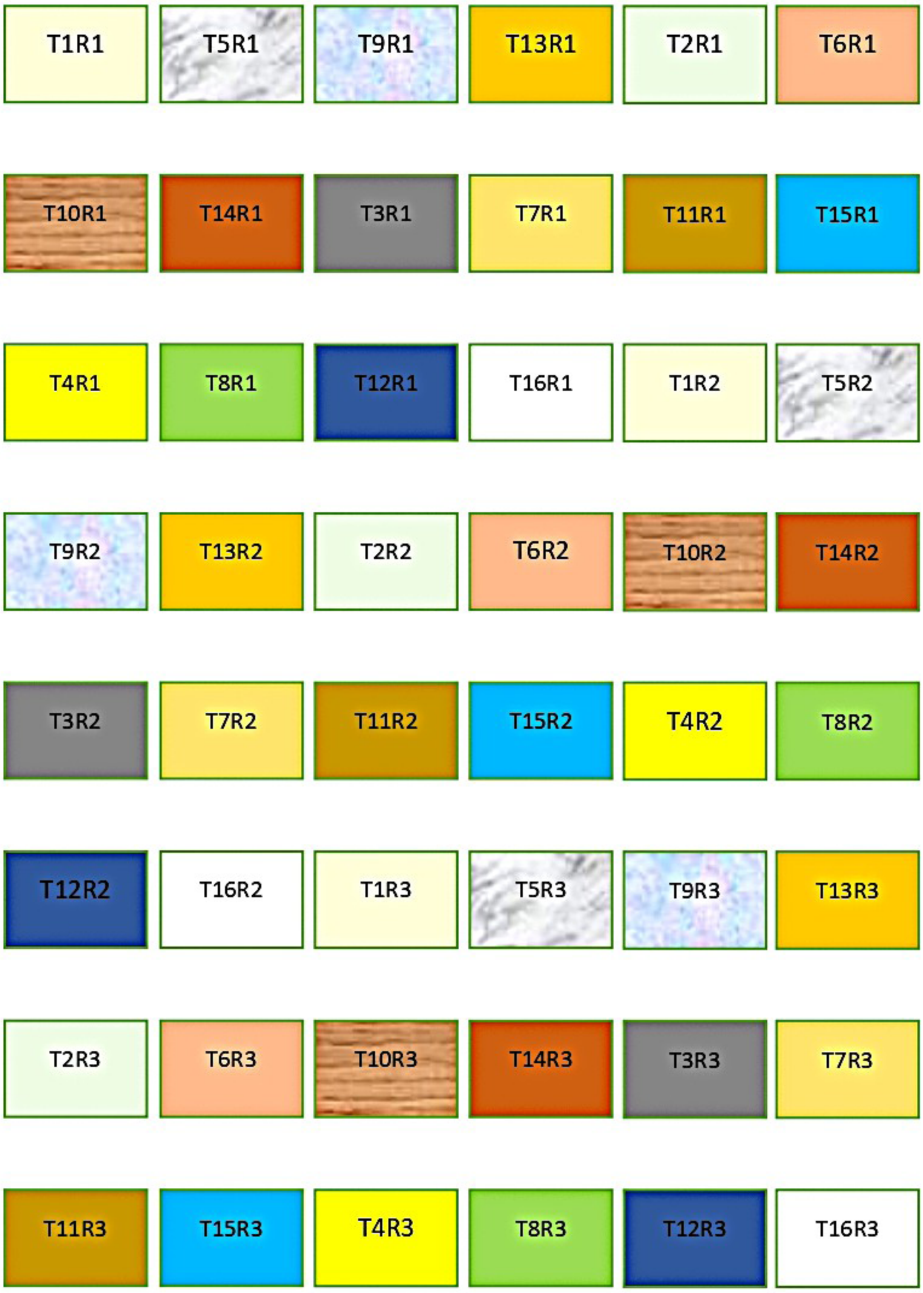

Figure 6. Sketch test.

2.5. Statistical procedure

A two-way analysis of variance [ANOVA] was performed to evaluate the effect of two independent variables (rice genotypes and infestation levels) on the dependent variables (healthy grains per spike, stained grains per spikes, empty grains per spike, and spike weight). This analysis is useful for assessing the main effects of each factor and detecting possible interactions. When the two-way ANOVA was significant, post-hoc tests (Tukey) were applied to conduct multiple mean comparisons and thus determine differences or similarities among the evaluated treatments.

A correlation analysis was carried out to determine the possible relationship between infestation level and the yield of the genotypes studied, for which an inferential hypothesis analysis was performed. In this context, the null hypothesis [H_0] established that there is no significant relationship between the infestation level of the stink bug (*O. insularis*) and the yield of the different rice genotypes; the alternative hypothesis [H_1] stated that a significant relationship exists between these variables. A significance level of 0.05 was used for all statistical tests. The data were processed using the statistical program IBM® SPSS® version 21, with 95% confidence.

Complementary to the parametric analyses, the percentage of increase and reduction (equations [2] and [3], respectively) was calculated, using the lowest and highest infestation levels as reference points, providing an additional criterion to evaluate the behavior of the genotypes under pest pressure.

$$\% \; of \; increase = \left(\frac{final \; value - initial \; value}{initial \; value}\right) \times 100 \qquad [2]$$

$$\% \; of \; reduction = \left(\frac{initial \; value - final \; value}{initial \; value}\right) \times 100 \qquad [3]$$

3. Results and Discussion

The results showed that the correlation analysis between infestation level and spike weight was significant ($p < 0.05$) for the genotypes Go-05070, INIAP-20, and Go-04209. This allows the null hypothesis to be rejected for these genotypes and confirms that there is a relationship between increased infestation and reduced yield. In contrast, the INIAP FL-ÉLITE genotype was not significant ($p = 0.383$), indicating that the null hypothesis could not be rejected in this case. This result suggests that this genotype may have greater tolerance to infestation, although additional studies are needed to confirm this possibility (Table 1).

Table 1. Correlation analysis between the variable infestation level and yield of each genotype.

Genotypes		
Go-05070	Pearson Correlation	-0.735**
	Sig. (bilateral)	0.006
INIAP-20	Pearson Correlation	-0.963**
	Sig. (bilateral)	0.000
Go-04209	Pearson Correlation	-0.807**
	Sig. (bilateral)	0.002
INIAP FL-ÉLITE	Pearson Correlation	-0.277
	Sig. (bilateral)	0.383

** Significant correlation at 0.01

3.1. Healthy grains per spike

The ANOVA revealed significant effects of both genotype ($F = 20.376$; $df = 3$; $p < 0.001$) and infestation level ($F = 14.937$; $df = 3$; $p < 0.001$) for this variable. Additionally, a significant interaction was observed between these factors ($F = 5.952$; $df = 9$; $p < 0.001$), indicating that the effect of infestation varies among genotypes. Overall, the genotype Go-04209 showed the highest number of healthy grains, with a mean and standard deviation of 60.83 ± 39.03 compared to the others (Table 2).

Regarding infestation level, a significant decrease in the number of healthy grains was observed as the number of insects per plant increased, with means ranging from 50.83 ± 38.46 healthy grains (no infestation) to 10.50 ± 8.56 healthy grains (6 insects per plant) (Table 2)

In Figure 7, the behavior of the variable is shown graphically, indicating that the genotypes respond differently to increasing infestation. In this context, the material Go-04209 maintains a relatively high number of healthy grains, remaining stable up to the fourth infestation level, compared with the highest level, where this variable decreases sharply; in fact, it responded differently from the others.

Similar findings were reported by Vivas and Notz (2010), who evaluated in Venezuela the damage caused by six population densities of the rice stink bug (*O. insularis*) on the yield of the rice cultivar "*Cimarrón*" under field conditions. They found that increases in pest density were associated with significant reductions in the weight of normal (healthy) grains and in stained grains. In that study, for the first case, densities of 2, 3, 4, 6, and 8 pairs of insects were different from densities of 0 and 1 pair of insects.

In the United States, field studies confirmed the impacts of the rice stink bug (*O. pugnax* F.) during the flowering, milk, and soft-dough stages, with the greatest losses recorded during flowering, suggesting that the economic threshold is five insects per ten sweeps (Awuni et al., 2015).

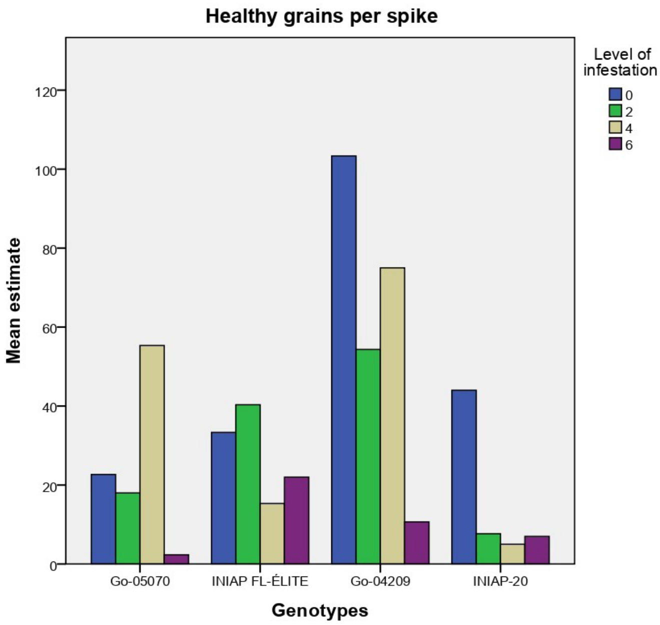

Figure 7. Influence of genotypes on the amount of healthy grains in the face of infestation.

3.2. Stained grains per spike

The results of the study showed that the incidence of stained grains in rice is influenced by genetic and environmental factors. The ANOVA revealed highly significant differences among the rice genotypes (F = 5.646; df = 3; p = 0.003) and the infestation levels (F = 10.653; df = 3; p < 0.001); although the interaction between both factors was not significant (F = 1.884; df = 9; p = 0.091). Tukey's mean comparison test determined that the genotypes Go-04209, INIAP FL-ÉLITE, and Go-05070 exhibited better performance for this variable (Table 2).

Regarding the infestation level, the same pattern was repeated: the higher the number of insects, the greater the number of stained grains. The mean for the highest infestation level was 49.42 ± 17.63 affected grains, while the control registered 19.42 ± 13.35 (Table 2). It is worth noting that the stained grains in the control (without infestation) can be attributed to biotic factors, such as the presence of diseases caused by propagule structures in the environment from fungal and bacterial pathogens that compromise grain quality (*Alternaria alternata* F., *Alternaria padwickii* G., *Aspergillus flavus* L., *Aspergillus niger* T., *Bipolaris oryzae* B., *Curvularia lunata* W., *Fusarium moniliforme* S., *Fusarium oxysporum* S., *Fusarium solani* M., *Xanthomonas oryzae* pv. *Oryzae* X., *Acidovorax avenae* M., *Pantoea agglomerans* E., *Burkholderia glumae* K., among others) (Bedoya Castañeda, 2022; Escalona et al., 2023; Lamsal et al., 2024; Sandoval-Martínez et al., 2022; Shamshad et al., 2024).

In Figure 8, an increment in the stained rice grains was observed as the exposition to the pest increased. In this sense, the genotype INIAP-20 had a more susceptible behavior expressing higher number of stained grains in comparison to the rest of materials. Conversely, the Go-05070 genotype showed fewer stained grains when exposed to maximum infestation, suggesting a potential tolerance response that warrants further investigation.

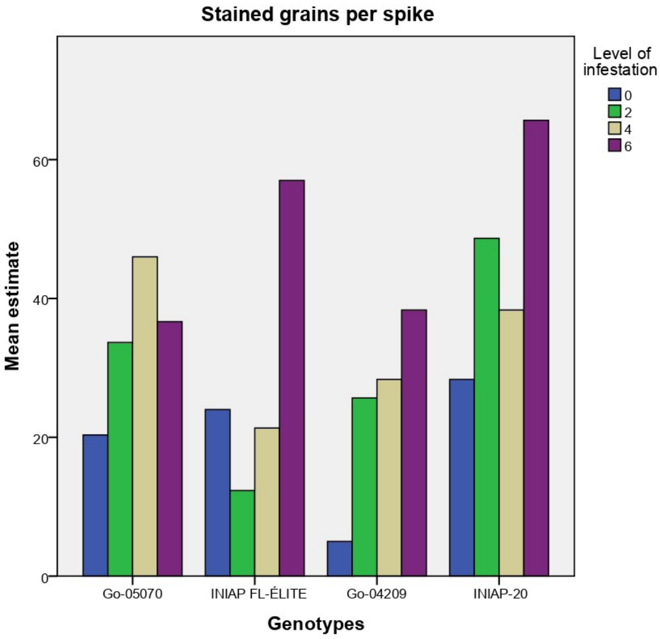

Figure 8. Influence of genotypes on the number of stained grains.

This information is important for selecting tolerant and/or resistant varieties in breeding programs and for strengthening integrated pest management (Smith, 2021). Some findings related to tolerance and/or resistance of rice cultivars in interaction with other pests are cited below.

In Brazil, the tolerance of the rice genotypes “Xingu,” “Canela de Ferro,” and “Primavera” to the brown stink bug *Tibraca limbativentris* S. (Hemiptera: Pentatomidae) was evaluated under greenhouse conditions, where physiological responses were analyzed (gas exchange rate, chlorophyll content in the leaves, and detoxification of reactive oxygen species) (de Sousa Almeida et al., 2021). The genotype “Primavera” showed greater tolerance to damage caused by the pest, exhibiting a smaller reduction in photosynthetic activity (41%) at 96 hours after infestation, compared with the genotypes “Xingu” (56%) and “Canela de Ferro” (65%) at 24 and 48 hours after infestation, respectively.

In the United States, the performance of eight inbred rice cultivars under combined infestations of the rice water weevil *Lissorhoptrus oryzophilus* K. (Coleoptera: Curculionidae) and the stalk borer *Eoreuma loftini* D. (Lepidoptera: Crambidae) was evaluated using insecticide seed treatments under field conditions (Villegas et al., 2021). It was found that the genotype “Jupiter” harbored the highest number of immature water weevils, while densities in the other cultivars were intermediate. In the case of the borers, low damage levels were observed in the genotypes “Cheniere” and “Jazzman-2,” suggesting that these cultivars express some resistance to this pest, with yield losses among cultivars being minimal (18%).

In Colombia, the resistance mechanisms of six rice lines in interaction with the planthopper *Tagosodes orizicolus* M. (Hemiptera: Delphacidae) were studied through forced-feeding tests (15, 30, and 45 individuals per plant) and free-feeding tests (10, 20, 30, and 40 insects per plant), using fifth-instar nymphs and adults under controlled conditions. Antixenotic, antibiotic, and tolerant genotypes were identified (Chávez Sosa, 2022).

3.3. Empty grains per spike

The ANOVA revealed that there were no significant differences among the genotypes ($F = 2.437$; $df = 3$; $p = 0.083$); however, highly significant differences were found among the infestation levels ($F = 6.617$; $df = 3$; $p = 0.001$). In addition, a highly significant interaction between genotypes and infestation levels was detected ($F = 3.021$; $df = 9$; $p = 0.010$). Despite this, in the mean comparison, all genotypes were statistically similar

(Table 2).

It is worth noting that the non-infested genotypes showed a mean of 44.67±24.39 empty grains (Table 2), which could be attributed to factors such as the presence of mites, phytopathogens (fungal and bacterial), genetic characteristics of the cultivar itself, and environmental conditions (Awuni et al., 2024; Kayal et al., 2024; Méndez, 2011; Meneses, 2008; Weber et al., 2020).

In INIAP FL-ÉLITE (Figure 9), the number of empty grains did not increase directly with the infestation by the rice stink bug. For example, under non-infested conditions it showed an average of 55 empty grains, possibly attributable to the previously mentioned factors (genetic, environmental, phytopathogenic, and others). With an infestation of 2 insects per plant, a reduction in empty grains (38 on average) was observed, possibly as a compensatory response by the plant that optimized grain filling. However, with 4 insects per plant, the damage was more severe, significantly increasing the number of empty grains (108 on average), indicating that the plant's compensatory capacity had been exceeded. Meanwhile, with 6 insects per plant, the number of empty grains decreased again to 53, which could be explained by a defensive response of the plant and an apparent tolerance that would need further studies to confirm.

In the Dominican Republic, it was determined under field conditions that high stink bug populations significantly affect grain emptying, with rates of 23.5% and 32.0% for 20 and 40 specimens of *O. ornatus* S. per m², respectively (Núñez et al., 2015).

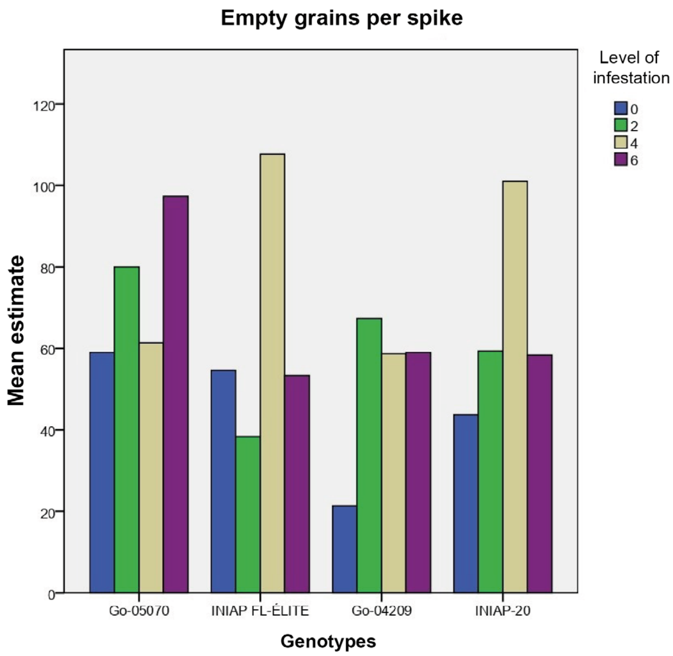

Figure 9. Influence of genotypes on the amount of empty kernels in the face of infestation.

3.4. Weight of the spikes (g)

The genotypes showed significant effects ($F = 14.009$, $df = 3$; $p < 0.001$), as did the infestation levels ($F = 12.241$; $df = 3$; $p < 0.001$). The interaction between genotypes and infestation level was not statistically significant ($F = 1.307$; $df = 9$; $p = 0.272$). The mean comparison revealed that the material with the highest spike weight was Go-04209 (2.69±0.81), unlike the others, which were statistically similar to each other (Table 2). When analyzing the infestation levels in general, it was confirmed that when the genotypes were exposed to the highest infestation level, spike weight decreased by 46%.

Figure 10 shows that the genotype Go-04209 exhibited optimal performance under non-infested conditions. When exposed to 2 and 4 insects per plant, it showed relative stability between these levels. In contrast, spike weight was affected under higher exposure.

In Colombia, significant reductions in rice yield were found, ranging from 27% to 65%, when spikes were infested with populations of 0.3 to 1.1 individuals of *Oebalus* sp., compared with the control. Additionally, it was confirmed that seed germination capacity decreased between 9% and 14% due to the damage caused by this pest (Meneses Carbonell et al., 2001).

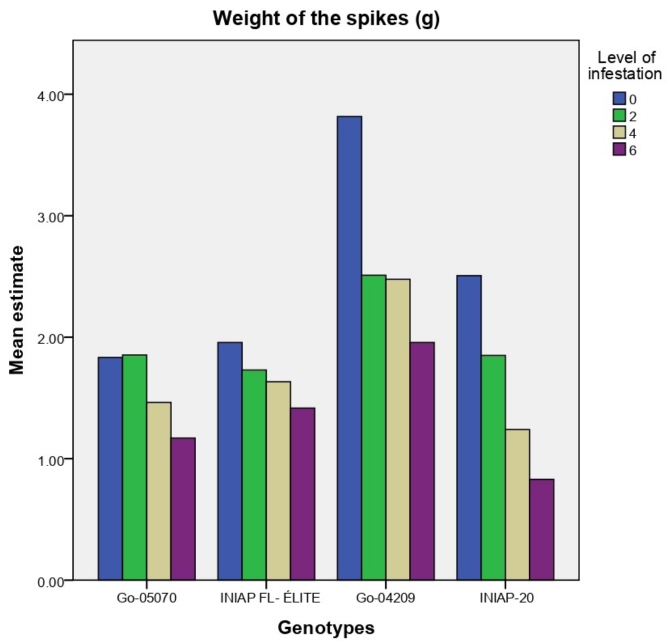

Figure 10. Influence of genotypes on spike weight in the face of infestation.

Furthermore, the feeding frequency of *O. insularis* and *O. ornatus* on rice spikes at the milk stage has been compared under field and laboratory conditions. Greater voracity was observed in the first species, causing lower spike weight (Pantoja et al., 1993). In the same study, higher feeding activity in females of both species was recorded compared with males. This is attributed to females requiring greater quantities of nutrients for oocyte maturation, oviposition, and their own maintenance (Weber et al., 2020).

In crops such as sorghum, field evaluations have quantified the economic damage caused by *O. insularis*, with yield losses ranging from 10% to 40% under infestation levels of 2, 4, 6, 8, and 10 insects per spike, in Tolima, Colombia (Osorio Rivera et al., 1998).

Table 2: Impact of *O. insularis* infestation and performance of rice genotypes on grain quality and quantity.

Infestation	Healthy grains per spike	Stained grains per spike	Empty grains per spike	Weight of the spike (g)
0	50.83±38.46 a	19.42±13.35 b	44.67±24.39 b	2.52±1.02 a (0%)
2	30.08±22.16 b	30.08±17.85 b	61.25±28.02 ab	1.98±0.57ab (21%)
4	37.67±31.63 ab	33.50±15.29 b	82.17±27.12 a	1.70±0.57 bc (32%)
6	10.50±8.56 c	49.42±17.63 a	67.00±22.71 ab	1.34±0.49 c (46%)
Genotype				
INIAP-20	15.92±17.73 b	45.25±19.75 a	65.58±25.86 a	1.60±0.68 b
Go-05070	24.58±20.38 b	34.17±12.26 ab	74.42±24.37 a	1.58±0.38 b
INIAP FL-ÉLITE	27.75±21.28 b	28.67±21.73 b	63.50±32.98 a	1.68±0.72 b
Go-04209	60.83±39.03 a	24.33±16.28 b	51.58±27.88 a	2.69±0.81 a

3.5. Effect of the interaction between genotypes and infestation in the four studied variables.

In Table 3, for the genotype Go-05070, it was observed that the control showed an average of 22.67±2.52 healthy grains, and at infestation level 4, an unexpected 55.33±3.06 healthy grains were recorded. This behavior could be attributed to an adaptive physiological response, in which the moderate presence of four bugs may have activated defense mechanisms such as increased translocation of resources toward grain filling, activation of metabolic defense pathways that enhance grain development, and the production of defensive compounds that in turn promote development (Chen et al., 2012; Velásquez Salazar, 2012).

Another possible explanation would be a specific hormetic effect (Erofeeva, 2023; Jalal et al., 2021), in which the level of four insects may represent an optimal “moderate stress” that triggers positive responses. This effect would explain the reason for:

- With 0 insects there is no stimuli to activate defenses (22.67±2.52 healthy grains).
- With 4 insects there is an optimal response (55.33± 3.06 healthy grains).
- With 6 insects the damage surpasses the capacity of response (2.33±0.58 healthy grains), because the plant cannot defend itself against this level of infestation.

When analyzing the infestation levels, they showed a drastic reduction pattern of 90% (22.67±2.52 to 2.33±0.58) when the genotype was exposed to the maximum number of insects, presenting statistical differences. This highlights the need to implement control measures before reaching the critical level.

Stained grains exhibited a considerable increase at intermediate infestation levels, reaching 46±3.61 with 4 insects, equivalent to a 126% rise. Regarding the variable “empty grains,” no statistical differences were found; however, a proportional increase was observed from 59.00±23.26 to 97.33±8.51, equivalent to 64%.

Spike weight showed a gradual decrease (1.85±0.13 to 1.17±0.22 g spike^{-1}), equivalent to 36% (under maximum infestation). Therefore, an action threshold of 2–4 insects per plant could be established, since yield losses of up to 20% are observed at this level, indicating the need to apply control measures before the damage increases.

Vivas and Notz (2010) established economic thresholds of 2–4 insects per plant for the rice variety Cimarron in interaction with *O. insularis*.

In Table 3, for the genotype Go-04209, a 90% reduction in healthy grains was observed (Figure 11A) when exposed to the highest infestation level (from 103.33±38.37 down to 10.67±5.03). For the variable stained grains (Figure 11B), there was a 666% increase (from 5.00±4.58 to 38.33±5.67) when subjected to 6 insects per plant, while at the 2-insect level the increase was 413%. No statistical differences were found in the number of empty grains (Figure 11C).

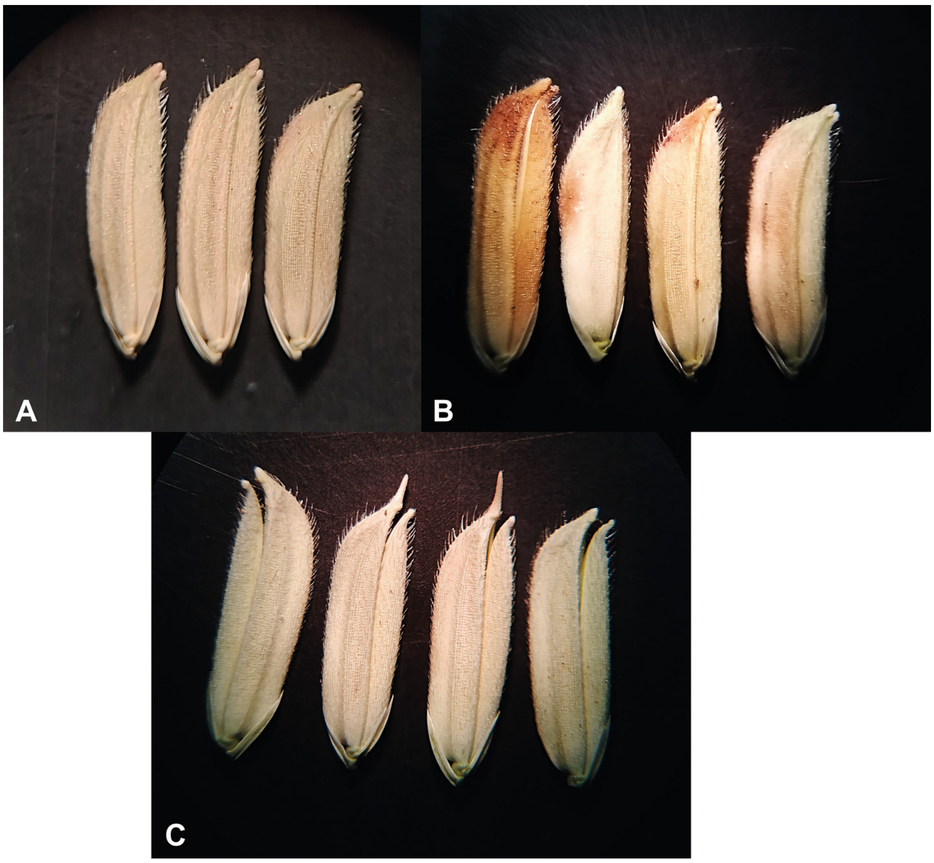

Figure 11. Genotype Go-04209 with an infestation level of 4 individuals plant^{-1}. A) Healthy grains. B) Stained grains. C) Empty grains.

Under maximum infestation, spike weight was reduced by 49% (from 3.81±0.39 to 1.95±0.51 g spike^{-1}). An action threshold of 1 insect per plant is suggested, as yield reduction already reaches 17%, and with 2 insects losses rise to 34%.

In Brazil, the performance of the cultivar IRGA-424 IR in interaction with *O. poecilus* D. was evaluated, determining quantitative damage with 2 bugs per spike, resulting in a 32.7% reduction. However, even 1 bug per spike caused important quality losses (Weber et al., 2020).

For the genotype INIAP-20, statistical differences were observed between the control and the infestation levels, with an 84% reduction in healthy grains (from 44.00±8.55 to 5.00±5.00) (Table 3). For the variable stained grains, the values were statistically the same.

On the other hand, when exposed to 4 insects per plant, empty grains increased by 131% (from 43.67±18.56 to 101.00±20.88). Regarding spike weight, under high infestation it was reduced by 66% (from 2.50±0.06 to 0.83±0.04 g spike^{-1}). The action threshold for this genotype could be set at 2 insects per plant, as this level has been observed to reduce spike weight by 26%.

Based on the interaction observed, it is possible that this genotype has a lower silica content in the glumes, making it more acceptable to the pest. In contrast, studies show that high silica concentrations benefit the plant by hindering stylet penetration, resulting in low feeding preference for insects; in this regard, reductions in damage of up to 20% have been observed in pests such as borers (Jinger et al., 2017).

For the genotype INIAP FL-ÉLITE, no statistical differences were observed for the variable healthy grains, indicating that the infestation levels did not significantly affect the quantity (Table 3). Regarding stained grains, there was a 362% increase under high infestation compared with level 2 (from 12.33±6.43 to 57.00±25.36) and a 137% increase compared with the control (from 24.00±14.00 to 57.00±25.36). Meanwhile, for empty grains, infestation levels 2 and 4 produced means with a 180% increase (from 38.33±19.50 to 107.67±15.28); and at maximum infestation levels, a relative tolerance was observed, suggesting the need for further research.

A 38% reduction in spike weight was recorded (from 1.95±1.28 to 1.41±0.23 g spike^{-1}). In this regard, the action threshold could be set at around 2–4 insects per plant, since with 4 insects the loss for this variable would reach about 20%.

It is important to mention that the thresholds proposed for each genotype are estimates based on the data provided; however, to obtain more precise values, additional information would be needed on production costs, the cost of biocidal treatments, and the market value of the grain.

Table 3: Effect of interaction between genotypes and infestation in four study variables.

Genotype	Infestation	Healthy grains per spike	Stained grains per spike	Empty grains per spike	Weight of the spike (g)
Go-05070	0	22.67±2.52 b	20.33±11.02 b	59.00±23.26 a	1.83±0.43 a
	2	18.00±6.00 b	33.67±13.58 ab	80.00±32.61 a	1.85±0.13 a
	4	55.33±3.06 a	46.00±3.61 a	61.33±11.93 a	1.46±0.25 a
	6	2.33±0.58 c	36.67±1.53 ab	97.33±8.51 a	1.17±0.22 a
Go-04209	0	103.33±38.37 a	5.00±4.58 b	21.33±15.04 a	3.81±0.39 a
	2	54.33±1.53 ab	25.67±4.73 ab	67.33±38.69 a	2.51±0.45 b
	4	75.00±5.29 a	28.33±22.37 ab	58.67±15.69 a	2.47±0.44 b
	6	10.67±5.03 b	38.33±5.67 a	59.00±20.08 a	1.95±0.51 b
INIAP-20	0	44.00±8.55 a	28.33±13.32 a	43.67±18.56 b	2.50±0.06 a
	2	7.67±3.06 b	48.67±21.55 a	59.33±1.53 b	1.85±0.22 b
	4	5.00±5.00 b	38.33±15.50 a	101.00±20.88 a	1.24±0.33 c
	6	7.00±6.25 b	65.67±10.97 a	58.33±12.50 b	0.83±0.04 c
INIAP FL-ÉLITE	0	33.33±27.03 a	24.00±14.00 ab	54.67±29.74 ab	1.95±1.28 a
	2	40.33±25.33 a	12.33±6.43 b	38.33±19.50 b	1.73±0.94 a
	4	15.33±23.09 a	21.33±0.58 ab	107.67±15.28 a	1.63±0.32 a
	6	22.00±4.58 a	57.00±25.36 a	53.33±18.34 ab	1.41±0.23 a

4. Conclusions

The genotype Go-04209 presented better behavior, both in terms of quality and quantity. This means that it exhibited a higher average of healthy grains per spike, less than stained grains per spike, but higher spike weight in relationship to the rest, which constitutes a promissory botanic material. The threshold decision is established at 1 insect per plant for this genotype, which could cause economic losses of 17%.

The search for cultivars that could tolerate the damage by insects, prompt monitoring, and the implementation of more environmentally friendly control strategies, are important measures in the integrated management. This set of practices favors the sustainable agricultural production, so it is essential to generate research in this direction, which could serve as the basis for future studies that could significantly support the strengthening of the productive matrix.

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Contributor roles

- Jessica Daniela Zambrano Mero: conceptualization, investigation, methodology, data curation, writing – original draft.
- Alex Gabriel Delgado Párraga: data curation, formal analysis.
- Roberto Evaristo Celi Herán: supervision, resources, project administration.
- Carmen Isabel Castillo Carrillo: writing – review & editing.
- Héctor Antonio Reyes Villón: supervision, validation.
- Daniel Fernando Navia Santillán: supervision, writing – review & editing (support).

Ethical Implications

Ethics approval not applicable.

Conflicts of Interest

The authors declare that they have no affiliation with any organization with a direct or indirect financial interest that could have appeared to influence the work reported.

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