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Cassava roots damaged by *Migdolus fryanus* produce poor quality starch

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ABSTRACT

The aim of this study was to evaluate the quality of starch produced by plants that suffer damage caused by *M. fryanus*, analyzed by colorimetric and morphological indices. Colorimetric data were composed of luminosity indices, “a” and “b” chroma coordinates and darkening index. For morphological data, the perimeter and area of starch granules extracted from cassava roots were analyzed. The variables were summarized in a principal component analysis and the axes scores retained for interpretation were correlated with the damage caused by the *M. fryanus*. From these analyses and observing the main effects, linear regression models for the variables were determined, according to the level of damage caused by the pest. The damage caused by *M. fryanus* in the roots strongly influenced the indices related to starch colorimetry, hence, under conditions of greater damage, the tendency of the chroma coordinates to vary toward red and yellow was observed and a decrease in starch luminosity occurred, inversely proportional to the increase in darkening. Similar to what was observed in the colorimetric data, a drop in the size of starch granules was noted in the roots, which suffered greater damage from the pest.

Keywords: Soil pests, Tuberous roots, Pests, Processing industry.

Raízes de mandioca danificadas por *Migdolus fryanus* produzem amido de baixa qualidade

RESUMO

O objetivo deste trabalho foi avaliar a qualidade do amido produzido por plantas que sofrem danos causados por *M. fryanus*, analisada por meio de índices colorimétricos e morfológicos. Os dados colorimétricos foram compostos por índices de luminosidade, coordenadas de croma “a” e “b” e índice de escurecimento. Para os dados morfológicos, foram analisados o perímetro e a área dos grânulos de amido extraídos das raízes da mandioca. As variáveis foram resumidas em uma análise de componentes principais e os escores dos eixos retidos para interpretação foram correlacionados com os danos causados por *M. fryanus*. A partir dessas análises e observando os principais efeitos, foram determinados modelos de regressão linear para as variáveis, de acordo com o nível de dano causado pela praga. O dano causado por *M. fryanus* nas raízes influenciou fortemente os índices relacionados à colorimetria do amido, portanto, em condições de maior dano, observou-se a tendência das coordenadas de croma de variar para vermelho e amarelo e ocorreu diminuição da luminosidade do amido, inversamente proporcional ao aumento do escurecimento. Semelhante ao observado nos dados colorimétricos, observou-se queda no tamanho dos grânulos de amido nas raízes, que sofreram maiores danos da praga

Palavras-chave: Pragas de solo, Raízes tuberosas, Pragas, Indústria de processamento.

1. Introduction

Cassava (*Manihot esculenta* Crants) is a perennial plant of the Euphorbiaceae family that has South America as its center of origin. It is one of the main sources of carbohydrates consumed in the world and has several locations, such as human and animal food, alcohol production, and pharmaceutical products, in addition to other uses of industrial interest (Reinhardt et al., 2013; Pinto-Zevallos et al., 2016). Cassava can be considered as one of the plants that positively influences food security in tropical and subtropical areas of the world, due to its ability to tolerate periods of drought, soils with low fertility, and limited use of resources for its management (Saengchan et al., 2015; Shigaki, 2015).

The storage capacity and ease of cassava starch extraction is significant and justifies its use, either for food or industrial purposes. The commercial destination and market value of cassava starch can be measured by its degree of purity, characteristics of its morphological structure, neutral taste, clarity, and bright color presented by its paste (Eke et al., 2009). The amount of starch produced in cassava roots, as well as its quality, is closely related to plant development. This reserve structure may have its formation and accumulation influenced by abiotic or biotic factors (Daiuto and Cereda, 2006 b).

Starch extracted from plants that have gone through suitable development conditions has a brilliant coloration, intact morphological structures, and significant criteria to define the quality of starch, which directly influence the properties of this product (Eke et al., 2009). However, based on the cassava, any negative change in starch quality can influence the properties of many marketed products (Zhu, 2015). Despite the important natural resistance of cassava to changes in abiotic and biotic factors considered to be adequate (Pinto-Zevallos et al., 2016), the crop can suffer significant damage due to the occurrence of pest insects. These organisms are among the biotic factors that can lead to decreased production and quality of starch (Bellotti et al., 1999).

The species *Migdolus fryanus* Westwood (Coleoptera: Vesperidae), popularly called sugarcane root borer, has emerged in the cassava planting areas and caused severe damage to tuberous roots, resulting in a decrease in starch production. *M. fryanus* is a species that lives part of the time in the soil, feeding on roots, causing a lot of damage ranging from small scrapings to large perforations (Guerreiro et al., 2019). As the *M. fryanus* larvae enter the soil to feed on cassava roots, they can cause deposition of soil in the feeding holes and allow the entry of opportunistic pathogens, which predisposes to rot and loss in the amount of starch produced by the roots that have been attacked (Bellotti et al., 2012; Guerreiro et al., 2019).

Although the symptoms of damage caused by *M. fryanus* on cassava as well as its direct influence on the decrease of starch production in damaged roots are known, little is known about the effect of the damage caused by this pest species on the final quality of starch extracted from the plants. With regard to such concerns, this study aims to evaluate the change in the quality of starch produced by plants that suffer damage caused by *M. fryanus* larvae, which is evaluated by the variation of colorimetric and morphological indices.

2. Material and Methods

The study was carried out in a commercial cassava planting area of approximately three hectares, located in the municipality of Perobal - PR, Brazil (23°51′ S, 53°20′ W, and 425 m altitude). The variety used to carry out the experiment was IAC 90, with 12 months of development. To identify the insects and relate the damage caused by the larvae to the cassava roots, pitfall traps containing the Migdo® pheromone were randomly installed at 78 points in the root collection area. The collected insects, *M. fryanus* males, were assembled and sent to the Laboratory of Chemical Ecology and Insect Behavior of the Department of Entomology and Acarology of the "Luiz de Queiroz" School of Agriculture - University of São Paulo.

During the cassava harvesting process, a directed collection of roots was performed to carry out the analyses. The choice of material collected in the field was made according to the intensity of damage caused by *M. fryanus*, according to the damage scores established by Guerreiro et al. (2019), which have adopted the following standardization: Note 1, without apparent damage (scraping) at the root; Note 2, scraping of the root without exceeding the cortex; Note 3, root scraping beyond the cortex, but without any presence of galleries; Note 4, root with galleries; Note 5, root with galleries and presence of rot.

The starch extraction procedure from the roots with the respective damage scores was carried out at the Entomology Laboratory of the State University of Maringá, Campus Umuarama, PR. Ten samples (content 1 kg) of roots brought from the field were separated per each note of damage, in this case, the intensity of damage was used as a basis for the evaluation of the variables studied in this research. The processing of roots took place through the removal of the skin and the woody part of the end close to the stem of the plant, resulting in 1 kg of sample mass. The roots were cut into smaller pieces and crushed, using an industrial blender. According to the processing methodology proposed by Rajapaksha et al. (2017), for each mass of 1 kg of root, there was an increase of 1000 mL of distilled water, to assist in the starch extraction process.

The ground root mass was washed and filtered through a cotton cloth, until the extracted liquid no longer contained starch. The mixture obtained was left to rest in stoppered flasks for 24 hours, to decant the starch. According to the methodology of Guerreiro et al. (2019), the supernatant was rejected and the rest of the material subjected to the drying process, in a controlled environment, until it reached a constant mass.

To measure the effect of *M. fryanus* damage on starch coloration, a Croma Meter CR 400 colorimeter, calibrated on a porcelain surface, was used. The starch samples were fractionated into 50 grams standards, with 30 repetitions for each treatment, which were comprised of the damage scores caused by *M. fryanus*. Measurements were obtained in the L system, "a*" and "b*", with L being the luminosity ranging from 0 (black) to 100 (white) and "a*" and "b*" chroma coordinates that are variable (-a = green, +a = red; -b = blue and +b = yellow), both ranging from -60 to +60. The Darkening Index (DI) was calculated according to the methodology proposed by Palou et al. (1999):

$$DI = \frac{[100(\mathrm{x} - 0.31)]}{0.172}$$

and

$$\mathrm{x} = \frac{(\mathrm{a}^* + 1.75\mathrm{L})}{(5.645\mathrm{L} + \mathrm{a}^* - 3.02\,\mathrm{b}^*)}$$

For size and morphological analysis of the starch granules, it was necessary to standardize the samples, thus 10 mg of starch was diluted in 500 µL of distilled water and 500 µL of glycerin (50%). The glycerin solution was used to make water absorption difficult and to avoid changing the size of the granules and compromising the sampling after dispersion in water (Daiuto and Cereda, 2006a). To observe the granules, 50µL of the standardized sample solution was deposited on glass slides and covered with a cover slip. Immediately after this process, the material was observed under an optical microscope (Motic® brand) with a 40X objective, coupled to an image analysis system Camera Software Motic®Images Plus 2.0ML.

After obtaining the images through the analysis system, the starch granules were measured in terms of their perimeter and area. Five slides were mounted for each damage score and 30 measurements were taken per slide, totaling 150 determinations per treatment. First, the data were transformed into Log10 to linearize the relationships between the variables, and thus standardize the values in the database. The Shapiro-Wilk (W) normality test was used to verify whether the data set had a normal distribution (Shapiro e Wilk 1965), whether there were differences between the means, and whether the factors could influence the dependent variable, in addition to determining the possibility of using other analyses. The homogeneity of variances was assessed using the Levene test (Layard, 1973).

To identify the relationship between the variables in the data set and the visualization of trends in this set, the principal component analysis (PCA) was used, aiming to reduce or eliminate the number of variables and explain the changes suffered by cassava starch according to the damage caused by *M. fryanus.* To begin with, the set of all variables was analyzed in an abridged manner in two main components (PC1 and PC2).

The eigenvalues, associated with a component or factor in descending order versus the component or factor number, were displayed on a screen plot of the dataset, utilizing the Broken-Stick model (Colwell et al, 1988). The regression analysis was performed with the scores resulting from the PC1, PC2 axes of the model: Yi = β0 + β1xi + €i, for i = 1,...n. Pearson's correlation test was used, and a correlation matrix was generated to indicate the intensity and direction of the linear or non-linear relationship between the damage caused by *M. fryanus* and the results of the quality of starch produced.

The colorimetry and morphology data were submitted to regression analysis at 5% significance, in order to obtain model adjustments, to explain the changes in the dependent variables, according to the damage caused by *M. fryanus* on plant roots. For the analysis of the morphological characterization of starch granules, the observed frequency of the perimeter and area of granules obtained from the damaged roots, compared with the material emerging from the unharmed roots, was evaluated in relative terms. All analyses adopted in the experiment were performed using R software version 4.1.0.

3. Results and Discussion

The analysis of the principal components (PC) made it possible to observe its relationship with the response variable that could be evaluated with the PC1 and PC2 axes, which together modeled 88.60% of the total variance of the data on *M. fryanus* damage and starch quality produced by plants, in which PC1 was responsible for 63.72% and the second principal component, PC2, was responsible for 24.88% of the variance (Table 1).

Broken-Stick axes 1 and 2 explained most of the variability, with a steep decrease from axis 3 (Figure 1). Axes PC3, PC4, PC5, and PC6 modeled 8.27, 1.67, 1.39, and 0.067% of the variances of the variable data, respectively; values that could be considered low. The PCA with the first two components generated, retained all factors and had eigenvalues >1 (λi>1), demonstrating that all variables were available and represented in axes 1 and 2 and summarized the total sample variance and could be adopted for the study of the data set (Table 1 and Figure 1).

Table 1. Scores resulting from principal component analysis (PCA) containing eigenvalues, explained proportion and cumulative proportion by principal component of the variables.

	Principal component analysis					
	PC1	PC2	PC3	PC4	PC5	PC6
Eigenvalue	3.8229	1.4929	0.4963	0.1004	0.0835	0.0040
Proportion explained	0.6372	0.2488	0.0827	0.0167	0.0139	0.0007
accumulated proportion	0.6372	0.8860	0.9687	0.9854	0.9993	1.0000

In Figure 2, which represents the axes with the principal components, it is interesting to note the arrangement of variables along PC1, which modeled about 64% of the variance of the data matrix. The correlation between the variables and the PC1 axis can be considered high for the darkening index (r = 0.94), luminosity (r = -0.93), chroma_b (0.86), chroma_a (r = 0.81), and moderate for area (r = -0.60), and perimeter (r = -0.56). Note that the correlation of the luminosity variable is inversely proportional to the darkening index, chroma_a, and chroma_b.

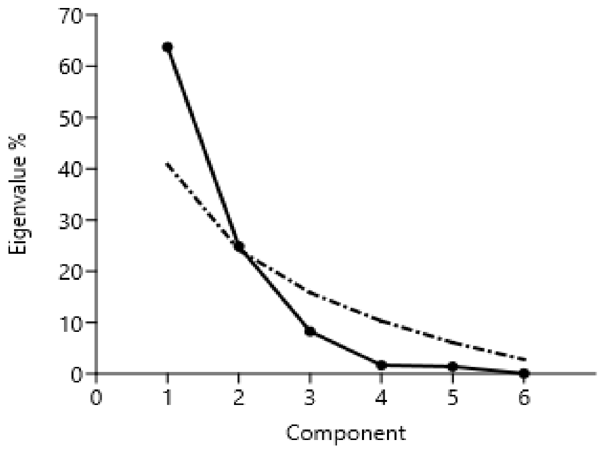


Figure 1. Broken-Stick model for the explanatory variables darkening index, luminosity, chroma_a, chroma_b, area and perimeter, from the principal component analysis (PCA).

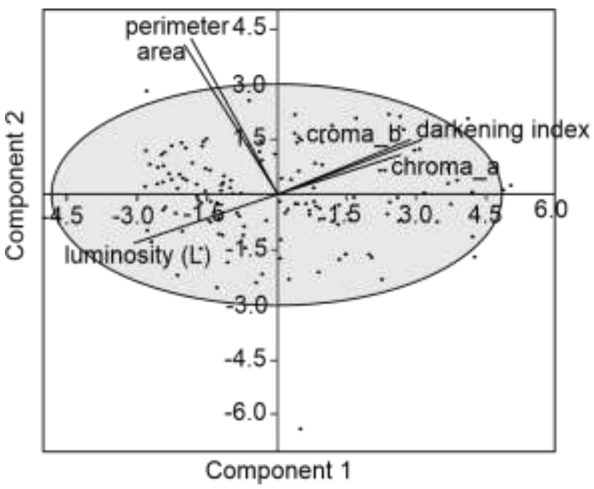


Figure 2. Principal component analysis (PCA) containing the explanatory variables darkening index (DI), luminosity, chroma_b, chroma_a, area and perimeter of the starch granule.

It is possible to observe that the second principal component (PC2), which models about 25% of the variance of the data matrix, is more related to the starch morphology data. In this case, it is possible to observe that the variable perimeters and area of the starch granules shift to positive values of the secondary component, with the correlation values considered high, being r = 0.80 and r = 0.77, respectively.

The coefficient of determination adjusted for damage caused by *M. fryanus* and chroma_a (R^2 adj. = 0.7708; n = 150) showed a fit of the regression analysis line with the data set. The relationship among the analyzed variable, data normality, and the F value (F = 84.538) for the *M. fryanus* damage and the chroma_a values can be considered high. These data have been used to determine the linear relationship between the response and explanatory variables (Table 2).

The development of a matrix that correlates the variable damage caused by the *M. fryanus* and the quality of starch extracted from the cassava roots, represented by colorimetry and the morphology data of the starch granules, demonstrates a high rate of positive correlation between the damage caused by the insect and the answer worth croma_a and the darkening index with a correction coefficient of 0.82 and 0.70, respectively. The value of the correlation coefficient between damage and chroma_b was 0.59.

A strong negative correlation was observed between damage and luminosity (r = -0.79), and a low correlation between the damage caused by *M. fryanus* and the variables related to the morphological characteristics, area and perimeter (r = -0.31 and -0.40) (Figure 3). The correlation matrix generated in response to damage caused by *M. fryanus* demonstrates a greater relationship with the starch color data, confirming the results of the data obtained using the PCA analysis. The effect of damage caused by the beetle *M. fryanus* on the colorimetric indices of starch can be represented by the models shown in Figure 4. A linear and significant relationship can be seen for all indices studied. The variable luminosity (L) showed a decrease proportional to the intensity of damage caused by the pest to the roots. In relative terms, starch extracted from the roots with a higher damage index lost about 7.7% of luminosity when compared to the undamaged roots.

Table 2. Adrien-Marie Legendre (1805) linear regression model based on principal component analysis (PCA) with PC1 and PC2 axes for the response variable chroma_a and PC1, PC2 for *M. fryanus* damage to cassava roots.

Response variable	N_starch	R^2 adj.	F	df1,df2	*p*- value
Chroma_a	150m função do	0.7708	84.538	5.13	1.53E-40

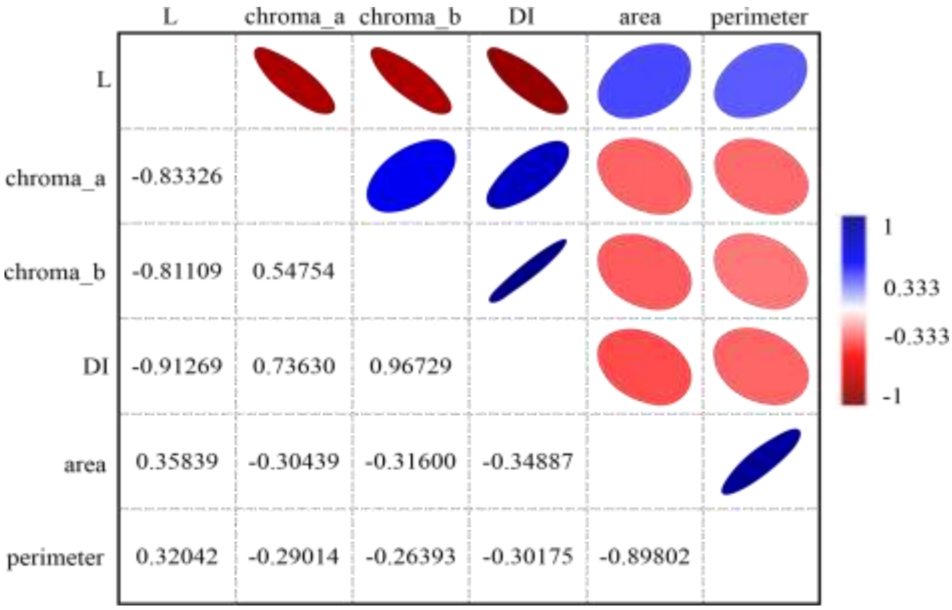


Figure 3. Linear correlation matrix between the response variables darkening index (DI), luminosity, chroma_b, chroma_a, area and perimeter and the explanatory variable *Migdolus fryanus* damage.

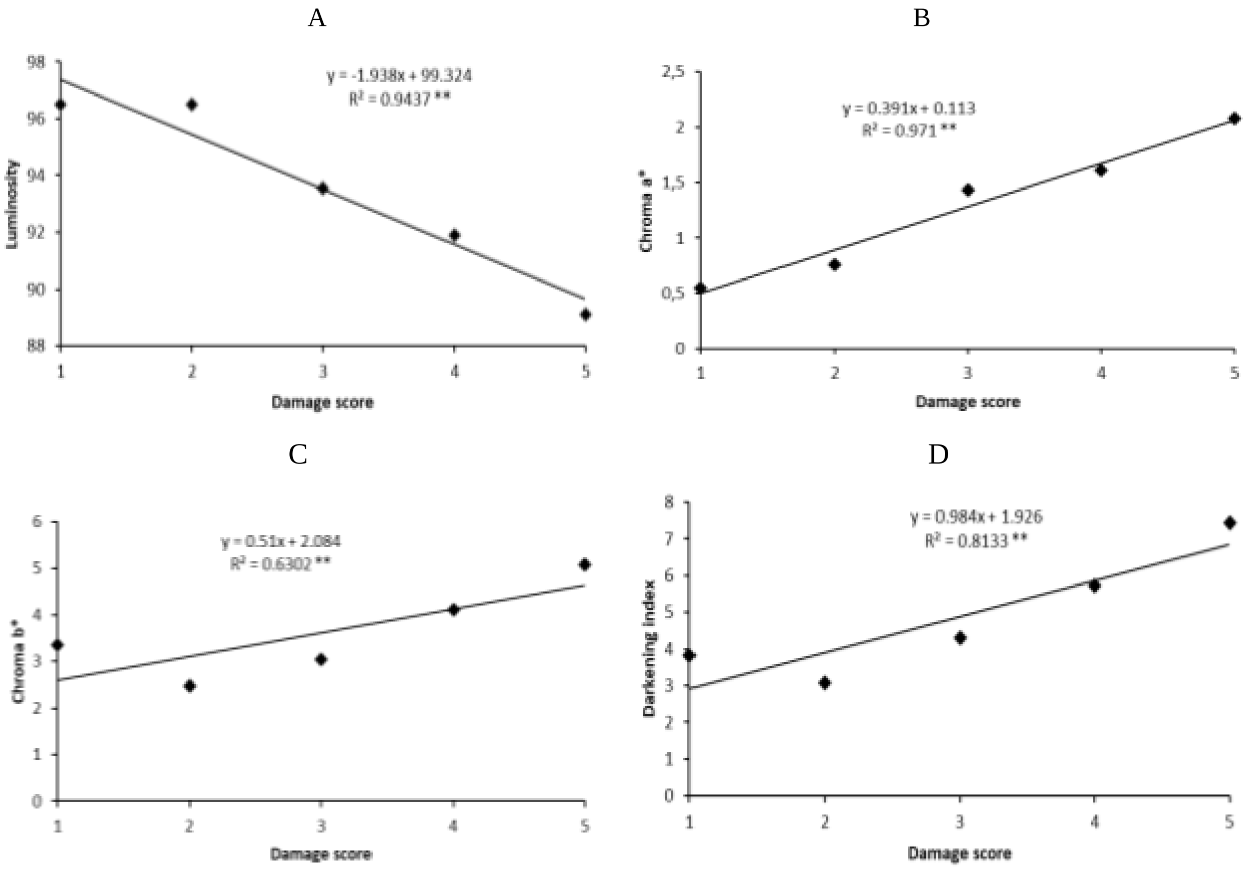


Figure 4. Mean values of luminosity (A), chroma_a (B), chroma_b (C) and darkening index (D) in cassava starch samples, the values were obtained as a function of damage scores of the beetle *Migdolus fryanus*.

The models developed for all other colorimetric variables showed increasing values according to the increase in the level of damage caused by the pest in the roots. This linear behavior can be confirmed, with emphasis on the variation obtained for chroma_a, which tended to be a reddish color (+60), and for values of the starch darkening index (DI), which varied for darker tones, as there was a greater intensity of damage (Figure 4). The data provided in Figure 5 allowed us to observe variations that occurred between measurements of the perimeter and area of starch granules extracted from the tuberous roots that did not suffer damage from the *M. fryanus* beetle (damage score 1), compared to starch extracted from roots that suffered intermediate to severe damage. Note that for roots that presented damage scores 2 and 3, considered intermediate, the frequency of occurrence of the highest values of starch granule size were closer to those presented by the undamaged roots.

It is possible to confirm, by mathematical models, that there was a decreasing and significant linear relationship between the size of the starch granules extracted from the cassava roots and the damage caused by *M. fryanus*. The propensity of decreasing starch size was related to the intensity of damage suffered by the root (Figure 6). The drop in starch perimeter ranged from 4.98% to 19.9%, according to the intensity of pest damage. As for the area data, a higher and variable drop of 8.16% to 32.60% was noted with regard to the starch samples from damaged roots, with grades 4 and 5, respectively, compared with starch samples taken from the undamaged roots (Figure 6).

On the other hand, when the size of the starch granules extracted from the roots that were severely damaged by *M. fryanus* was evaluated, a decrease in this reserve structure was noted. The highest frequencies observed in the perimeter and area of granules stood out with regard to data obtained from the undamaged roots, with peaks of observations in classes of lower values, indicating that the root was negatively affected by severe injuries caused by the *M. fryanus*, which was possibly used as a strategy for the diversion of nutrients to other parts of the plant, causing a change in the size of the starch granule.

For roots that had no apparent damage, it was observed that there was formation of larger starch granules with average values of 63.54 µm and 211.46 μm^2 of the perimeter and area, respectively. In the evaluation of starches from roots that suffered the highest damage scores (notes 4 and 5), which represented the largest lesions caused by the *M. fryanus* larvae, the lowest average values of the starch granule size were observed, with an average of 56.38 and 49.25 µm and 172.49 and 134.78 μm^2, for perimeter and area, respectively (Figure 5).

Although changes in starch granule size were observed, no changes were observed in the morphological characteristics of this reserve material. Even in conditions of severe damage to the beetle, *M. fryanus*, the granules had circular, oval, and truncated aspects, some with formation of a concave–convex shape.

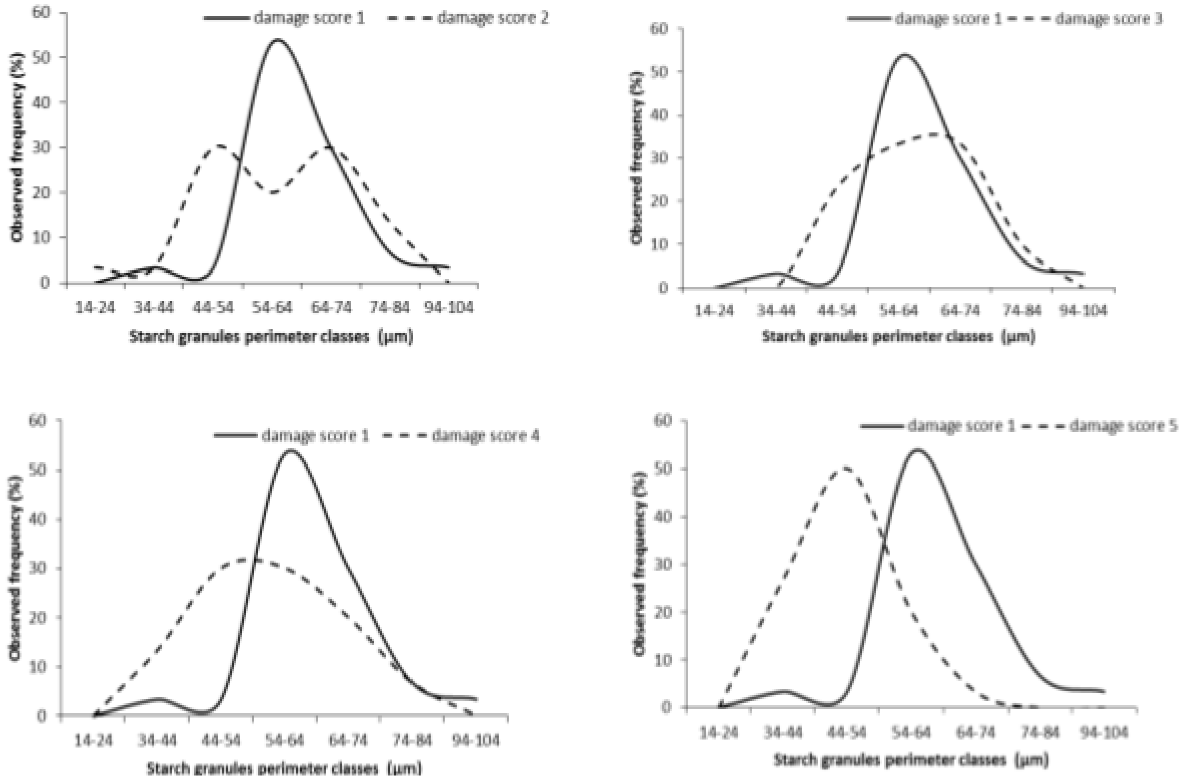
The chemical and structural composition of cassava roots depends on the variety, harvest time, weather conditions during cultivation, and other environmental factors. It is common for starch colorimetric values to be influenced by genetic factors, starch extraction, and processing, in addition to the soil where the planting is conducted, according to (Onitilo et al., 2007) and (Saengchan et al., 2015). However, as observed in this study, the damage caused by insect pests should also be considered as potential reasons for the occurrence of changes in the indices that measure the quality of the starch extracted from the cassava roots.

The beetle, *M. fryanus*, recently described as a potential cause of damage in cassava crops, is seen to be living most of its life in the soil, and its larvae feed on the plant roots in the drier times of the year. It is a pest that can be considered polyphagous, and its occurrence is more pronounced in regions with more sandy soils (Pietrowski et al., 2010). In the major producing regions of Paraná State cassava, has all the optimal conditions for the development of *M. fryanus*, we have seen a gradual and continuous expansion of this pest by consuming roots and causing production losses.

Some studies have reported that the damage caused by *M. fryanus* in cassava can range from simple scraping of the root cortex to progress into open galleries, implying the possibility of penetration of opportunistic microorganisms, according to Bellotti et al. (1999) and Pietrowski et al. (2010). Furthermore, it was possible to relate the possibility of a decrease in the starch content produced in the roots damaged by *M. fryanus*, according to Guerreiro et al. (2019), however, the influence of damage caused by soil pests on the quality of starch produced by cassava roots was still not common knowledge, hence the emphasis on the results presented in this research

We noted that as a result of feeding and damage caused by *M. fryanus* there was a strong relationship in the alteration of starch quality, and it was found that the infestation by the coleopteran larvae of the cassava roots could directly interfere with the color of the extracted starch, with a tendency to darken it, as the intensity of pest damage to the roots increased. The damage caused by *M. fryanus* and observed in this study indicate a loss in the excellence of the quality of the cassava starch, which according to (Abraham, 1993) must be related to the white color of this material.

Granules Perimeter (µm)



Granules Area (µm^2)

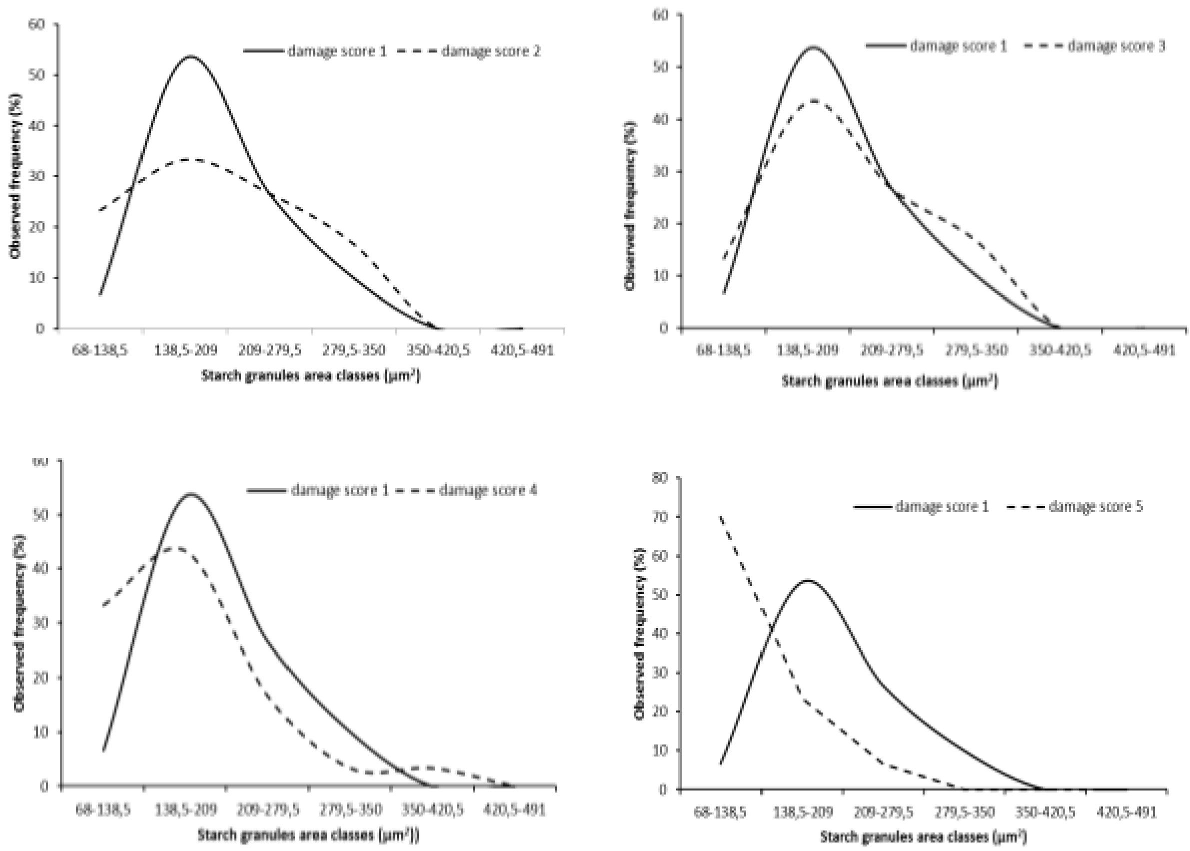


Figure 5. Perimeter (µm) and area (µm^2) measurements of starch granules according to damage caused by *Migdolus fryanus* in cassava roots.

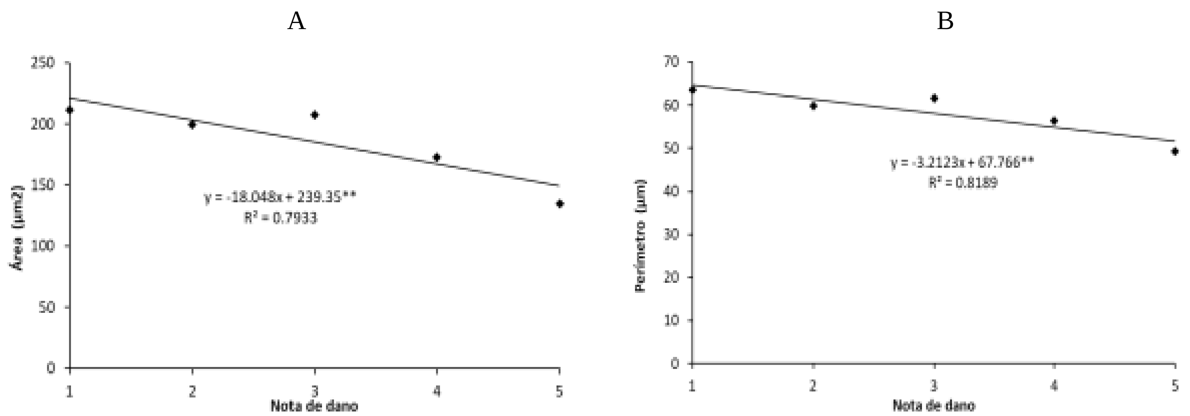


Figure 6. Regression representing the effect of damage caused by *Migdolus fryanus* on the perimeter (A) and area of starch granules (B)

In this sense, for a high-quality material it is important that the combination of the luminosity parameter is close to 100, with the darkening index parameters of chroma_a and b being close to zero. These patterns may indicate starches with a more intense white color, which are the more common characteristics for roots from plants that have had optimal development conditions. It was observed from the data shown in Figure 4 that when performing damage to the roots during consumption, the larvae of *M. fryanus* caused losses in the quality of the starch, resulting in lower values, lower than 90 and browning indices, represented by chroma_a and chroma_b being above zero, characterizing greater browning of the starch and configuring color patterns considered different from those described as ideal for cassava starch.

Starch obtained from the roots that suffered more severe damage from *M. fryanus* could cause direct problems for the industry due to the need for standardization of the materials. It could increase the possibility of occurrence of chemical or enzymatic degradation of the material, because the darker an object became, the greater the tendency it would have to absorb visible light. With the root damaged by *M. fryanus*, and the tendency to produce darker starch, the possibility of starch oxidation reactions, which might be responsible for changes in starch gelatinization, increase.

Thus, the starch obtained under conditions observed in this study, and coming from roots that suffered more severe damage by *M. fryanus*, can cause changes in the viscosity of the starch suspension and of the gel that is formed, resulting in the restriction of its use, mainly in conditions in which a product with a high degree of transparency is needed, according to (Song et al., 2010). The study performed by Guerreiro et al. (2019) in addition to demonstrating a drop in starch productivity, describes the external and internal damage of the attacked roots, and determines that the roots with the greatest losses, in terms of production, are those that suffer from perforation and penetration of larvae in their internal part.

The darker color of starch in roots most damaged by *M. fryanus*, as observed in this study, is related to the direct and indirect damage behavior of the larva, such as, when penetrating the root, it can carry away soil particles and cause damage that is considered direct, on the other hand, by providing root rot due to the entry and action of opportunistic soil fungi, the insect can cause indirect damage. The properties and functionalities of cassava tuberous roots, demonstrated through their ability to store starch, can be managed by environmental and physiological factors according to Santisopasri et al. (2001) and Souza et al. (2019).

Among the biotic environmental factors, the occurrence and damage of pests to the cassava crop directly influences the productivity administered by environmental and physiological factors as (Bellotti et al., 1999). However, until the development of this research, the influence of damage on the plant was not known, specifically on tuberous roots, and the modifications caused in the processes of formation of starch granules. It is possible to say that the process of formation of starch granules, demonstrated by its perimeter and area, is directly related to the root damage caused by *M. fryanus*.

It was possible to notice that the largest lesions caused on the roots by beetles of the species *M. fryanus*, configured by holes and rotting parts of the tuberous roots, resulted in the formation of smaller starch granules, when compared to the starch granules formed in roots classified as grade 1, no apparent damage (scraping). The decrease in the size of the starch granules produced, influenced by the consumption by *M. fryanus*, can negatively affect the total productivity of root starch.

This behavior was observed by Guerreiro et al. (2019), who noted a drop in yield of close to 30%, when there was a severe attack by the insect *M. fryanus* on the cassava roots. Based on this observation, prior to this study, it is possible to say that the damage caused by *M. fryanus* activates possible plant responses which lead to less starch storage, thus inferring the occurrence of smaller structures, compared to those found in roots not damaged by the insect. On the other hand, when studying the morphological attributes, it was observed that the granular structure of starch has a variable shape according to its origin, compared to those found in roots undamaged by the insect. According to (Ellis et al., 1998), the characteristic of the starch granule shape is more related to the genus or species of the producing plant, than to the differences in the conditions of occurrence of biotic or abiotic factors.

Under the conditions of this study, no occurrences of morphological alteration of starch granules were observed, in the roots attacked by *M. fryanus*, thus it is possible to argue that the stress caused by the pest did not infer morphological changes in these reserve organs, corroborating the results of Souza et al. (2019) and (Gu et al., 2013), who did not find changes in the morphological characteristics of starch granules stored in cassava roots, from plants with different stages of development and subjected to different climatic conditions and environmental stress.

The changes observed in the size of starch granules caused in this study by damage due *M. fryanus* may, according to Souza et al. (2019), affect the physicochemical properties of starch and cause difficulties in its use in the industrial phase, because according to (Ellis et al., 1998) every purpose of using starch requires that the starch has it common characteristics.

4. Conclusions

Feeding damage caused by *M. fryanus* larvae on cassava roots causes negative changes in the starch colorimetric indices and starch granule size, decreasing the quality of starch. The morphology of starch granules extracted from cassava roots is not altered, even under the stress caused by *M. fryanus*.

Authors' Contribution

Julio César Guerreiro, Ana Paula Azevedo, Rudiney Ringenberg planned, designed, executed experimental work; Evandro Pereira Prado, Pedro José Ferreira-Filho e Thais Carneiro Giotto conducted data analyses; Julio César Guerreiro, Vanda Pietrowski, Cristiane Mengue Feniman Moritz; wrote the manuscript. Thaíse Milena Pascutti, Ana Paula Azevedo helped with data analyses. All authors read and approved the manuscript.

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Bibliographic References

Abraham, T.E. 1993. Stabilization of Paste Viscosity of Cassava Starch by Heat Moisture Treatment. Starch/Stärke, 45(4), 131–135. DOI: https://doi.org/10.1002/star.19930450404

Bellotti, A.C., Smith, L. Lapointe, S.L. 1999. Recent Advances in Cassava Pest Management, Annual Review of Entomology, 44(1), 343–370. https://doi.org/10.1146/annurev.ento.44.1.343.

Bellotti, A., Campo, B.V.H. and Hyman, G. 2012. Cassava Production and Pest Management: Present and Potential Threats in a Changing Environment. Tropical Plant Biology. 5, 39-72. DOI: https://doi.org/10.1007/s12042-011-9091-4.

Colwell, J.D., Suhet, A.R. Van Raid, B. 1988. Statistical procedures for developing general soil fertility models for variable regions. CSIRO, Australia:, 68p. (CSIRO. Division of Soils. Divis. Report, 93).

Daiuto, E.R., Cereda, M.P., 2006a. Influência da granulometria de grânulos de amido sobre a densidade aparente de extratos atomizados. Revista de Ciências Farmacêuticas Básica e Aplicada, 27(1), 51–56. DOI:http://hdl.handle.net/11449/69329 (Accessed May 03, 2021).

Daiuto, E.R., Cereda, M.P., 2006b. Physical characteristics and energetic evaluation of tuber and root starch gels under sterilization stress, Energia na Agricultura. 21(2), 45–60.

Eke, J., Sanni, L.O., Oyewole, O.B., Maziya-Dixon, B. 2009. Pasting, color, and granular properties of starches from local and improved cassava varieties in high rainfall region of Nigeria. International Journal of Food Properties, 12(2), 438–449. https://doi.org/10.1080/10942910701867665.

Ellis, R.P., Cochrane, M.P., Dale, M.F.B., Duffus, C.M., Lynn, A., Morrison, I.M., Prentice, R.D.M., Swanston, J.S., Tiller, S.A. 1998. 'Starch production and industrial use', Journal of the Science of Food and Agriculture, 77(3), 289-311. DOI: https://doi.org/10.1002/(SICI)1097-0010(199807)77:3%3C289::AID-JSFA38%3E3.0.CO;2-D.

Fernandes, D.S., Santos, T.P.R., Fernandes, A.M., Leonel, M., 2019. Harvest time optimization leads to the production of native cassava starches with different properties. International Journal of Biological Macromolecules, 132, 710–721. https://doi.org/10.1016/j.ijbiomac.2019.03.245.

Gu, B., Yao, Q., Li, K., Chen, S. 2013. Change in physicochemical traits of cassava roots and starches associated with genotypes and environmental factors. Starch/Staerke, 65(3–4), 253–263. DOI: https://doi.org/10.1002/star.201200028.

Guerreiro, J.C., Azevedo, A.P., Espessato, R.R., Pietrowski, V., Ringenberg, R., Ferreira Filho, P.J., Hora, R.C., Padro, P.E., Pascutti, T. M. 2019. *Migdolus fryanus* Damage Causes Decrease in the Starch Content in *Manihot esculenta*. Journal of Agricultural Science, 11(15), 1–9. DOI: https://doi.org/10.5539/jas.v11n15p97.

Layard, M.N.J. 1973. Robust large-sample tests for homogeneity of variances. Journal of the American Statistical Association, 68 (341), 195-198.

Onitilo, M.O. Sanni, L.O., Oyewole, O.B., Maziya-Dixon, B. 2007. Physicochemical and functional properties of native starches from cassava varieties in Southwest Nigeria. Journal of Food, Agriculture and Environment, 5(3–4), 108–114. DOI: https://hdl.handle.net/10568/92196 (Accessed January 20, 2021)

Palou, E., Malo-Lópes, A., Barbosa-Cánovas, G.V., Welti-Chanes, J., Swanson, B.G. 1999. Polyphenoloxidase Activity and Color of Blanched and High Hydrostatic Pressure Treated Banana Puree. Journal of Food Science, 64(1), 42–45. DOI: https://doi.org/10.1111/j.1365-2621.1999.tb09857.x.

Pietrowski V., Ringenberg R., Rheinheimer A.R., Bellon P.P., Gazola D., Miranda A.M. 2010. Insetos-praga da cultura da mandioca na região Centro-Sul do Brasil. Unioeste, Marechal Cândido Rondon.

Pinto-Zevallos, D.M., Pareja, M., Ambrogi, B.G. 2016. Current knowledge and future research perspectives on cassava (Manihot esculenta Crantz) chemical defenses: An agroecological view. Phytochemistry, 130, 10–21. DOI: https://doi.org/10.1016/j.phytochem.2016.05.013

Rajapaksha, K.D.S.C.N., Somendrika, M.A.D., Wickramasinghe, I. 2017. Nutritional and toxicological composition analysis of selected cassava processed products, Potravinarstvo Slovak Journal of Food Sciences, 11(1), 35–42. DOI: https://doi.org/10.5219/689.

Reinhardt, H., NeBambi, L., Graeme, T. 2013. Save and Grow Cassava: A Guide to Sustainable Production Intensification, first ed. Food and Agriculture Organization of the United Nations (FAO). http://www.fao.org/docrep/018/i3278e/i3278e.pdf. (Accessed December 10, 2020)

Saengchana, K., Nopharatana, M., Lerdlattaporn, R., Songkasiri, W. 2015. Enhancement of starch-pulp separation in centrifugal-filtration process: Effects of particle size and variety of cassava root on free starch granule separation. Food and Bioproducts Processing, 95, 208–217. DOI: https://doi.org/10.1016/j.fbp.2015.05.008.

Santisopasri, C., Kurotjanawong, K., Chotineeranat, S., Piyachomkwan, K., Sriroth, K., Oates, C.G. 2001. Impact of water stress on yield and quality of cassava starch. Industrial Crops and Products, 13(2), 115–129. DOI: https://doi.org/10.1016/S0926-6690(00)00058-3.

Shapiro, S.S., Wilk, M.B., 1965. An Analysis of Variance Test for Normality (Complete Samples). Biometrika, 52, 591-611. DOI: https://doi.org/10.1093/biomet/52.3-4.591.

Shigaki, T., 2015 Cassava: The Nature and Uses. In: Caballero, B., Finglas, P.M., Toldrá, F. (Ed) Encyclopedia of Food and Health, National Agricultural Research Institute, Lae, Elsevier Ltd, Papua New Guinea, 687-693. DOI: https://doi.org/10.1016/B978-0-12-384947-2.00124-0.

Song, X., Zhu, W., Li, Z., Zhu, J. 2010. Characteristics and application of octenyl succinic anhydride modified waxy corn starch in sausage. Starch/Staerke, 62(12), 629–636. DOI: https://doi.org/10.1002/star.201000043.

Zhu, F. 2015. Composition, structure, physicochemical properties, and modifications of cassava starch, Carbohydrate Polymers, 122, 456–480. DOI: https://doi.org/10.1016/j.carbpol.2014.10.063.