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Genetic diversity of *Eugenia stipitate* based on the physical and physicochemical characteristics of the fruits in tropical conditions

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Abstract: The objective of this study was to evaluate the genetic diversity between genotypes based on the physical, chemical and physicochemical characteristics of araçá-boi in tropical conditions. The experiment was carried out at the Federal University of Roraima, in the municipality of Boa Vista, Roraima, Brazil. Genotypes were obtained in the F1 generation, through open pollination. Twelve plants were evaluated, with 10 fruits being collected per plant. The physical and physicalchemical characteristics of the fruits were evaluated: fresh mass, length and diameter of the fruits, pulp yield, firmness, mass of the pulp, peel and seeds, number of seeds, soluble solids, tritratable acidity, pH, ratio SS/AT and vitamin C from fruits. The araçá-boi genotypes showed medium to high genetic variability, obtaining averages for the physical and physicalchemical characteristics higher than those observed in the literature. G genotypes A9, A10 and A6 adopted fruits with larger sizes and produced a greater number of fruits genotypes A1 obtained fruits with higher organoleptic quality. Genotypes A3 produced fruits with higher levels of Vitamin C and fruits with greater firmness. Genotypes A3, A6, A9 and A10 were promising with high agronomic potential and could be selected for the genetic improvement program of the species.

Index terms: araçá-boi, physical-chemical quality, Myrtaceae, genetic variability.

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Diversidade genética de *Eugenia stipitate* Mcvaugh. baseada nas características físicas e físico-químicas dos frutos em condições tropicais

Resumo: O objetivo deste estudo foi avaliar a diversidade genética entre genótipos, com base nas características físicas, químicas e físico-químicas do araçá-boi em condições tropicais. O experimento foi realizado na Universidade Federal de Roraima, no município de Boa Vista, Roraima, Brasil. Os genótipos foram obtidos na geração F1, através de polinização aberta. Foram avaliadas 12 plantas, sendo coletados 10 frutos por planta. Foram avaliadas as características físicas e físico-químicas dos frutos: massa fresca, comprimento e diâmetro dos frutos, rendimento de polpa, firmeza, massa de polpa, casca e sementes, número de sementes, sólidos solúveis, acidez titulável, pH, relação SS/ AT e vitamina C de frutas. Os genótipos araçá-boi apresentaram variabilidade genética de média a alta, obtendo médias das características físicas e físico-químicas superiores às observadas na literatura. Os genótipos G A9, A10 e A6 adotaram frutos com tamanhos maiores e produziram maior número de frutos. Os genótipos A1 obtiveram frutos com maior qualidade organoléptica. Os genótipos A3 produziram frutos com maiores teores de Vitamina C e frutos com maior firmeza. Os genótipos A3, A6, A9 e A10 mostraram-se promissores, com alto potencial agronômico, e puderam ser selecionados para o programa de melhoramento genético da espécie.

Termos para indexação: araçá-boi, qualidade físico-química, Mirtáceas, variabilidade genética.

Introduction

The araçá-boi (*Eugenia stipitata* Mcvaugh) is a fruit species native to the Amazon region, and its geographic distribution extends between Brazil, Colombia, Ecuador and Peru. Belonging to the Myrtaceae family, it is adapted to tropical and subtropical climates and among the native fruits of the Amazon, it stands out as one of the most recent promising species. The araçá-boi has led to agro-industrial interest, which boosts its economic potential due to its high levels of nutraceuticals, antioxidants, drugs and biocompounds, vitamins and minerals. Therefore, due to its beneficial health properties, several by-products can be produced, since the fruits have high acidity for fresh consumption (NERI-NUMA et al., 2013; ARAÚJO et al., 2019, 2021; ARAUJO et al., 2021).

The specie has easy adaptation to tropical and subtropical climates, the genetic diversity found in the Amazon region is great due to the reproduction mode of the species. In addition, plants suffer several influences on their phenological behavior, on the physical, physicochemical and biochemical properties of the fruits due to the high genetic variability, which occur mainly due to genetic factors and edaphoclimatic conditions (CASTRO et al., 2020; MOURA et al., 2020; ARAÚJO et al., 2021; ITURRI et al., 2021; ANDRADE et al., 2022). In this way, the exploitation of the species is conditioned only in an extractive way, limiting the potential for economic exploitation of the species.

The selection of promising genotypes that make it possible to obtain stability in plant behavior, productivity and fruit quality is of great importance, as it makes it possible to develop cultivation techniques, and thus, to be able to exploit the species in commercial orchards. Currently, information regarding management at a commercial level to obtain fruits is scarce, making scientific research essential for the development of this crop.

Despite the great economic potential of the araçá-boi, the lack of information on agronomic management, production and fruit quality is undoubtedly one of the main obstacles to the establishment of commercial orchards and advances in the domestication of this fruit. Therefore, the objective of this study was to evaluate the genetic diversity between genotypes based on the physical, chemical and physicochemical characteristics of araçá-boi in tropical conditions.

Material and Methods

The araçá-boi (*Eugenia stipitate* Mcvaugh) genotypes were cultivated in the experimental orchard of the Federal University Roraima (UFRR), Boa Vista municipality, Roraima, Brazil, in partnership with Embrapa Roraima, whose geographic reference coordinates are 2º 52’ 14.9” N and 60º 42’ 46.8” W and 90 m high.

The climate of the region, according to the Köppen classification, is of the Awi type, with two well-defined seasons, one rainy (April to September) and the other dry (October to March) (ARAÚJO et al., 2024). The average annual precipitation is around 1700 mm, with a relative humidity of 70% and a daily temperature between 20.0 ºC and 38.0 ºC, with an annual average of around 27.4 ºC.

The soil in the experimental area is classified as a typical dystrocohesive Yellow Latosol (BENEDETTI et al., 2011). The ox guava plants have been implanted for 4 years at the site, planted in pits measuring 0.40 x 0.40 x 0.40 m mechanically dug. The genotypes were obtained in the F1 generation, through open pollination. Twelve plants were evaluated, each one divided into 4 quadrants and evaluated individually.

The fruits were harvested weekly, and their physical, chemical and physicochemical characteristics were evaluated. 10 fruits were harvested per plant in the 2022/2023 production cycle. The following were evaluated: yield - Prod (g plant^{-1}), number of fruits - NF, fruit diameter - FD (mm), fruit length - FL (mm), fruit mass - FM (g), seed mass - SM (g), peel mass - PeM (g) and number of seeds - SD (g). All the fruits were picked at the same stage of ripeness and were determined using a digital caliper (± 0.01 mm) and a precision analytical balance (± 0.01 g).

Fruit firmness (F) was determined using a manual Effegi penetrometer equipped with an 8 mm tip, and the results expressed in N.

Soluble solids (SS) were determined directly in the homogenized pulp juice in a digital refractometer (model PR – 100, Palette, Atago Co, LTD., Japan), with the results expressed in °Brix (AOAC, 2012). The titratable acidity (AT) was determined by volumetric titration, using 1 g of the pulp transferred to a 125 mL Erlenmeyer flask with the addition of 50 mL water, subsequently titration with 0.1 M NaOH solution until pH 8.1, previously standardized, expressing the results in g 100 g^{-1} citric acid pulp (AOAC, 2012).

The ratio SS/AT was determined by the ratio between the soluble solids values and the titratable acidity. The hydrogen ion potential (pH) was estimated using a potentiometer with automatic temperature adjustment (Model mPA-210 Tecnal®, Brazil), previously calibrated with pH 7.0 and pH 4.0 buffer solutions (AOAC, 2012). The measured data were expressed in actual pH values.

Vitamin C (Vit_C) was estimated by titrimetry with Tillmans solution (DFI - 2.6 dichloro-phenol indophenol at 0.02%), taking 1 g of the samples and diluted in a 100 mL volumetric flask with 0.5% oxalic acid and the results expressed in mg of ascorbic acid 100 g^{-1} pulp (STROHECKER; HENINING, 1967).

Data on the agronomic characteristics of each genotype were analyzed using descriptive statistics. Multivariate data analysis was also carried out, using principal components (PC). Genetic distances were calculated considering the 7 traits. Based on the genetic distance matrices, the genotypes were grouped by the "Ward. D" hierarchical grouping method (HAIR et al., 2005). The analyzes were performed in the R software (R CORE TEAM, 2020).

Result and Discussion

The evaluated genotypes had medium to high variability for the physical and physicochemical characteristics of the araçá-boi fruits Tables 1 and 2. The 12 genotypes showed variation for all studied traits. The production (Prod) and number of fruits (NF) of the araçá-boi plants had a high variation with an average of 57.83 (A12) to 673.34 g plant^{-1} (A9) and 2 (A1, 8 and 12) to 13 (A6), respectively.

Table 1. Physical characteristics of araçá-boi accessions harvested at the predominantly yellow stage.

Access	Prod	NF	FM	FL	FD	Rend	F	PuM	PeM	SM	SN
A1	109.71	2	54.86	41.8	49.91	43.9	10.0	33.41	4.91	10.96	9
A2	269.23	6	72.2	45.2	55.56	47.93	7.95	39.55	6.4	24.27	7
A3	162.32	3	67.72	39.47	53.48	44.01	20.7	28.09	13.84	23.72	8
A4	190.31	3	71.91	45.34	54.38	59.49	8.38	49.53	5.46	12.42	5
A5	256.41	7	46.7	38.54	45.18	43.4	6.88	35.92	9.54	3.31	4
A6	558.89	13	92.23	53.94	56.59	82.83	9.7	74.15	4.27	9.4	5
A7	134.29	3	57.08	41.6	49.65	49.98	10.78	37.0	7.19	7.1	10
A8	75.56	2	37.78	35.19	37.86	36.76	8.45	21.58	10.1	1.03	2
A9	673.34	9	112.96	55.73	60.94	89.25	8.53	71.22	11.63	23.71	10.5
A10	362.36	5	96.32	51.54	59.61	82.14	8.25	66.75	14.36	14.18	8
A11	190.6	4	54.25	40.77	51.4	40.88	11.65	33.24	6.62	13.37	6
A12	57.83	2	28.92	29.03	39.01	23.92	8.84	10.92	7.06	5.0	12
Average	**253.40**	**4.92**	**66.07**	**43.18**	**51.13**	**53.7**	**10.01**	**41.78**	**8.45**	**12.37**	**7.21**
CV%	**72.29**	**68.54**	**37.56**	**17.96**	**14.4**	**38.23**	**36.09**	**47.6**	**40.39**	**64.81**	**40.71**
SD	**183.20**	**3.37**	**24.82**	**7.76**	**7.36**	**20.53**	**3.61**	**19.89**	**3.41**	**8.02**	**2.93**

Prod - Production (g plant); NF - Number of fruits; FM - Fruit mass (g); FL - Fruit length (mm); FD - Fruit diameter (mm); Rend - Pulp Yield (%); F - Firmness (N); PuM - Pulp mass (g), PeM - Peel mass (g), SM - Seed mass (g) and SN - seed number; CV - Coefficient of variation (%); SD - Standard Deviation.

The variability of the genotypes in relation to the physical characteristics of the fruits was high, but with mean values similar to those observed by (SACRAMENTO; BARRETTO et al., 2008). Differences in physical characteristics in fruits of the genus Eugenia are probably due to climate variations, plant management and greater variability of the original germplasm (CASTRO et al., 2020; MOURA et al., 2020; ANDRADE et al., 2022).

The fruit mass (FM) ranged from 28.92 g (A12) to 112.96 g (A9), with an average of 66.07 g. The range of variation in fruit length (CF) ranged from 29.03 (A12) to 55.73 mm (A9), with an average of 43.18 mm. Fruit diameter (DF) ranged from (A8) 37.86 mm to 60.94 mm (A9), with an average of 51.13 mm. The fruits of araçá-boi had greater FM the greater the DF (Table 1).

The fruits of araçá-boi evaluated showed variation in pulp yield values (Rend), the lowest average was found in genotype 12 (23.92) and the highest average in genotype 9 (89.25%). Firmness (F) ranged from 6.88 (A5) to 11.65 N (A3). As for pulp mass (MP), peel mass (PeM), seed mass (SM) and seed number (SN), there was high variability between genotype, with means between 10.92 to 74.15 g, 4.27 to 14.36 g, 3.31 to 24.27 g and 2 to 12 seeds per fruit, respectively Table 1.

The soluble solids (SS) content had variations, genotype 2 had the lowest °Brix of 4.6, and genotype 7 had the highest value 8.2 °Brix, with an average of 6.48 °Brix. The titratable acidity (AT) ranged from 3.32 (genotype 2) to 5.83 (genotype 7) (g citric acid 100 g^{-1}) with an average of 4.41 Table 2. The SST/AT ratio had little variation between genotypes, from 1.27 (genotype 5) to 1.86 genotypes 11), with a general average of 1.48 Table 2.

Table 2. Physical characteristics of araçá-boi accessions harvested at the predominantly yellow stage.

Access	SS	AT	Ratio	Vit C
A1	6.0	4.15	1.53	700
A2	4.6	3.32	1.38	1000
A3	5.8	3.5	1.66	735.29
A4	5.95	4.0	1.49	879.63
A5	7.35	3.95	1.86	566.04
A6	7.65	5.63	1.36	424.53
A7	8.2	5.83	1.41	625
A8	6.0	3.96	1.52	462.96
A9	6.35	4.15	1.53	416.67
A10	6.5	5.12	1.27	432.69
A11	5.5	4.15	1.32	432.69
A12	7.55	5.15	1.47	480.77
Average	**6.48**	**4.41**	**1.48**	**596.36**
CV%	**15.93**	**18.54**	**10.78**	**32.82**
SD	**1.03**	**0.82**	**0.16**	**195.75**

SS - Soluble solids (°Brix); AT - Titratable acidity (mg $100g^{-1}$); *Ratio* SS/TA; Vit C – Vitamin C (mg $100g^{-1}$); CV - Coefficient of variation (%); SD - Standard Deviation.

The fruits showed averages of 596.36 mg of ascorbic acid per 100 g^{-1} of pulp. Genotype 9 had the lowest values of 416.67 mg of ascorbic acid 100 g^{-1} and genotype 2 had the highest value found 1000 mg of ascorbic acid 100 g^{-1} of pulp Table 2.

The total soluble solids content is composed of the ratio of the sugar content and the organic acid content present in the fruit, thus described as an important characteristic for products sold in nature, since the consumer prefers sweet fruits (BARRETT et al., 2010). Evaluations of araçá-boi plants identified values of 4.5 °Brix (CANUTO et al., 2010), 4.6 °Brix (GARZÓN et al., 2012) and 5.08 °Brix (BOHRY et al., 2019). The high genetic variability observed is inherent to the genus Eugenia, as observed for the species *Eugenia uniflora* L. (CASTRO et al., 2020; MOURA et al., 2020; ANDRADE et al., 2022), *Eugenia dysenterica* DC. (OLIVEIRA et al., 2014) and *Eugenia stipitate* L (BOHRY et al., 2019; GARZÓN et al., 2012). Presenting in this way, an enormous potential for selection during genetic improvement, since the fruits present countless possibilities of use, whether for fresh consumption, which requires fruits with greater sweetness and lower acidity, or for the agroindustry in which the acidity high levels provides advantages in microbial control.

The values of SS, AT, pH and ratio presented higher variations than the data observed in the literature, both for fruits evaluated in Brazil (ITURRI et al., 2021) and in Colombia (GARZÓN et al., 2012). All genotypes showed high levels of acidity. Directly influencing the low ratio values found. Ratio, is an indication of the degree of "flavor" of the fruit, obtained through the direct relationship between the sugar and acid content present in the fruit.

The dispersion of the characters studied by the principal component method (PCA), with two components, constituted 64.43% of the total variability of the data among the genotypes. The dispersion of PCA characteristics with two components PC 1 and PC 2 explained 44.17% and 20.26% of data variability, respectively Figure 1.

The vectors are distributed in two types dark and light (arrows) show how much weight each feature has on that main component. That is, the MF characters ($r=0.98$; $p<0.001$), Fruit length ($r=0.98$; $p<0.001$), Pulp Yield ($r=0.97$; $p<0.001$), pulp mass ($r=0.97$; $p<0.001$), production ($r=0.93$; $p<0.005$), fruit diameter ($r=0.90$; $p<0.005$) and number of fruits ($r=0.77$; $p<0.005$) are strongly correlated with PC 1. Vitamin C ($r=0.67$; $p<0.01$), titratable acidity (r=-0.85; p<0.001) and soluble solids (r=-0.86; p<0.001) were the main contributors to PC2. Genotypes A9, A6, A12, A8 and A10 were the ones that most influenced PC1. While only genotypes A3 and A2 presented the greatest contributions to PC2.

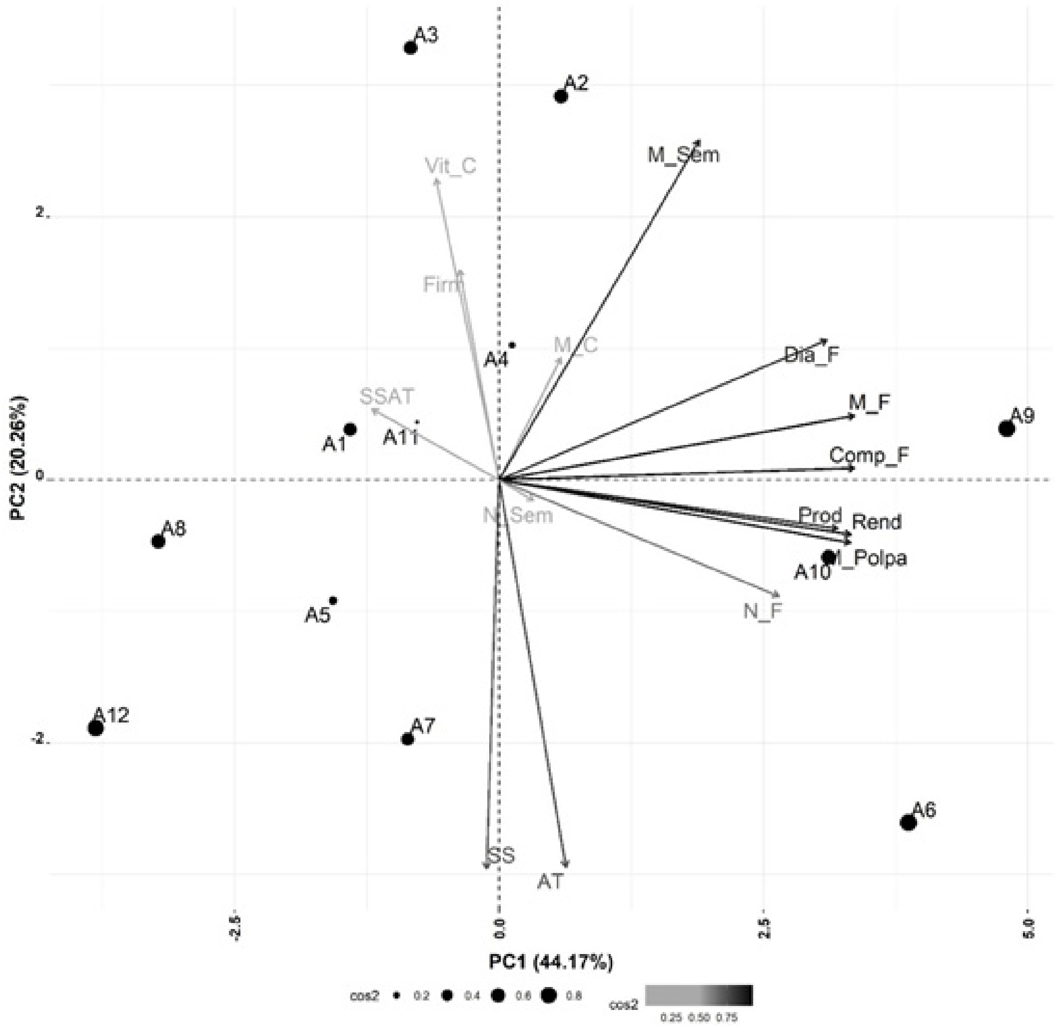

Figure 1. Principal component analysis (PCA) performed on genotypes of araçá-boi (*Eugenia stipitata* Mcvaugh) in relation to the physical and physicochemical characteristics of the fruits. Prod - Production, N_F - Fruit number, Dia_F - Fruit diameter, Comp_F - Fruit length, M_F - Fruit mass, N_Sem - Seed number, M_C - Peel mass, Firm - Fruit firmness, Vit_C - Vitamin C, M_pulp - Pulp mass, Yield - Yield, SS/AT - Soluble solids and titratable acidity ratio, SS - Soluble solids, AT - Titratable acidity.

The great variation of these characteristics allows for genetic variability between genotypes and provides subsidies for the selection of accessions with a high content of total soluble solids and, consequently, improvement in the flavor ratio of the fruits (BOHRY et al., 2019).

The araçá-boi genotypes tested in this study showed ascorbic acid values higher than those found in the literature. The content of this compound may vary due to several factors. Which may involve the high genetic variability of plants of the genus Eugenia, edaphoclimatic conditions, maturation stage, in addition to pulp processing, which can greatly affect the concentration of ascorbic acid (CASTRO et al., 2020; MOURA et al., 2020; ANDRADE et al., 2022; ARAÚJO et al., 2021).

Genotypes A9, A10 and A6 had larger-sized guava fruits, and produced a greater number of fruits, which resulted in greater yields Figure 1. Opposed to this, accession A1 obtained fruits with a higher quality index, however, the fruits and the production were not satisfactory. Genotypes A3 were the ones that produced fruits with higher levels of Vitamin C and fruits with greater firmness Figure 1.

Cluster analysis performed using the "Ward.D" method allowed the joining of accessions that presented similar characteristics. Therefore, we had the formation of seven groups. Three groups had only one access each. Three groups were constituted by two accessions. And only one group was formed by three genotypes Figure 2.

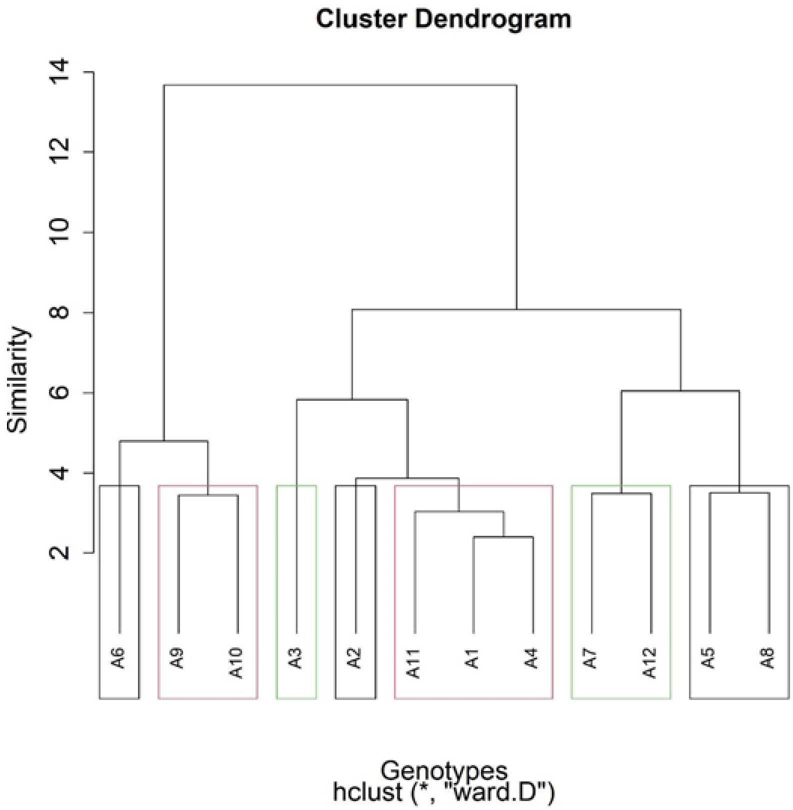

Figure 2. Cluster analysis by the Ward.D method using Euclidean distance organized the 12 genotypes of araçá-boi (*Eugenia stipitata* Mcvaugh) into seven groups.

Genotype A6 was isolated individually in group 1, as it produced a greater number of fruits, good levels of titratable acidity and soluble solids, however, low SS/AT quality index Figure 2. Genotypes A9 and A10 were similar due to their production and physical quality of the fruits produced, forming group 2. Confirming the PCA, genotype A3 was isolated individually, forming group 3, due to the excellent levels of vitamin C and firmness. Genotype A2 produced less acidic and sweet fruits with greater seed mass, being isolated individually in group 4. Group 5 grouped genotypes A1, A11 and A4, which were similar in terms of good SS/TA quality indices. genotypes A5, A7, A8 and A12 showed to be plants with low production, with smaller fruits and with low levels of vitamin C Figure 1. These genotypes were similar and isolated in pairs, forming groups 6 and 7 in the cluster analysis Figure 2.

The formation of groups with only one genotypes, or the formation of groups with up to two genotypes characterizes a good level of genetic divergence between genotypes (BOHRY et al., 2019). Similar results were observed 30 genotypes of cagaiteira (*Eugenia dysenterica* DC.), which belongs to the same genus as araçá-boi, found similar results with the formation of 9 groups, where four groups consisted of only one accession (OLIVEIRA et al., 2014).

Multivariate analysis allowed the selection of the best genotypes according to physical and physicochemical parameters. Variations are mainly influenced by environmental conditions and the behavior of each genotype in the environment (MOURA et al., 2020).

Conclusion

Genotypes A6, A9 and A10 showed better averages for the evaluated variables, showing high agronomic potential. Preliminary results make it possible to select genotypes for the genetic improvement program of the species, however, more evaluations seeking more biochemical and nutraceutical parameters are desirable for selection of superior accessions.

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