Characterization of sweet and bitter cassava (*Manihot esculenta* Crantz) genotypes through multivariate analysis

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Abstract

Cassava has importance as a source of human and animal food. With the objectives to select promising sweet and bitter cassava varieties for breeding programs, 27 genotypes were characterized in terms of their quantitative and qualitative properties. Roots were harvested from three plants per genotype, washed, peeled, sanitized. Regarding the yield, the storage root number (SRN), and the fresh storage root weight (FSRW), were determined, as well as the root fresh matter content (RFMC), and root dry matter content (RDMC), both expressed as a percentage. Among the cassava genotypes, the protein content ranged from 0.1-0.7%; lipids 0.3-2.1%; moisture 58.0-65.2%; 0.1-1.0% ash; fibers 0.9-1.9%; acidity 1,1-2,7%; pH 6.3-6.8; TSS between 0.8-1.2 ºBrix; glucose 0.1-0.8% and sucrose 0.5-1.0%, except for the fructose and starch contents, which did not vary significantly. The principal component analysis showed that the factors explain 84.2% of the total variability and through cluster analysis, evidencing cluster III for the highest starch yield and cluster I for the highest average of lipids and proteins.

Keywords: clustering, genotypes, starch, principal component analysis.

Introduction

Cassava (*Manihot esculenta* Crantz) has significant economic and food importance as it is a source of calories that serves as a staple food for millions of inhabitants, mainly in developing countries such as Africa, Asia, and Latin America (FAO, 2018; Uchechukwu- Agua et al., 2015).
Research indicates that many varieties of cassava were cultivated and preserved by native peoples of the Amazon approximately 8000 years ago. In this way, Brazil can be the probable center of origin and domestication of the culture because it presents a great diversity of genotypes cultivated in almost all regions. After all, the plant is resistant to several edaphoclimatic conditions (Piperno et al., 2000; Olsen, 2004; Nick et al., 2008). These genotypes are maintained in active germplasm banks (AGB) across the country.
Cassava varieties can be called biter or sweet according to hydrocyanic acid (HCN) content. Among these two groups, bitter cassava is characterized by its high content of HCN (above 100 mg of HCN per kilogram of fresh weight of roots) while sweet cassava with low cyanide content (below 100 mg of HCN per kilogram of fresh weight of roots) (Araújo et al., 2015).
The chemical composition of cassava roots according to the genotype can constitute approximately 80% of the fresh weight, where the plant stores carbohydrates (starch and sugars), fibers, ash, proteins, and other substances (Araújo et al., 2019).
Due to the high heterozygosity of intraspecific crosses that occur naturally over time, there are a large number of different genotypes (Lorenzi, 1994). Cassava genotypes can present peculiar qualitative and quantitative properties in the roots (Oyeyinka, et al., 2019), which need to be characterized so that they can be selected, identified, and introduced into breeding programs.
However, there are still few studies related to the characterization of these different genotypes to yield parameters and their physicochemical properties, with potential use in several sectors, such as the food industry, chemistry, and in the development of biomaterials. Thus, the present study aimed to comparatively evaluate the physicochemical characteristics of genotypes of sweet and bitter cassava belonging to the AGB of the Brazilian Agricultural Research Corporation (EMBRAPA) in the Eastern Amazon, with the aid of multivariate analyzes as a tool for selecting genotypes based on their associated attributes.

Results and Discussion

Root yield and extracted starch
The yields of root and starch extracted per plant in relation to the genotypes of sweet cassava (SC) and bitter cassava

(BC) in Tables 1 and 2, respectively, showed significant variations in the parameters evaluated, except for the fresh storage root weight (FSRW) and root dry matter content (RDMC).

Table 1 shows a variation in the number of SRN for sweet cassava from 2.7 to 7.7 units (SC08-SC09), respectively; 0.6 to 2.5 kg of FSRW (SC02- SC08); 72.6 to 88.6% of RFMC (SC10- SC03); 35.6 to 44.7% of RDMC (SC08- SC12); and 28.2 to 65.7% of starch yield in dry mass (SC01- SC05). Noteworthy is the SC12 genotype, which presented higher RDMC and SC05 higher starch yields in dry weight.

In Table 2, for bitter cassava, the SRN varied from 2.3 to 8.3 units (BC15-BC12) respectively; 1.2 to 3.2 kg of FSRW (BC09-BC03); 73.1 to 86.4% of RFMC (BC05-BC14) and 17.6 to 42% of RDMC (BC10-BC06); and 27.4 to 64.7% of starch yield in dry weight (BC12-BC04). The genotype, BC05 presented higher RDMC and BC04 higher starch yield in dry weight.

The general average of the samples was 5.3 units of SRN for sweet cassava and 4.4 for bitter cassava, while Chipeta et al. (2017) evaluating 16 cassava genotypes 12 months after planting obtained an average between 3.7 - 6.9 root units per plant.

The general average of RFMC found was relatively higher in the genotypes of bitter cassava (2.0 kg.plant-1) compared to the genotypes of sweet cassava (1.4 kg plant-1) as shown in Tables 1 and 2. However, Oliveira and Moraes, (2009) when evaluating a cultivar at different harvest times, obtained an average of 4.36 kg plant-1 in 12 months, higher than those found in this work.

The percentage of RDMC was 44.7% for SC12 genotype (Table 2) and 42.0% for BC05 genotype (Table 3). The cassava culture presents, on average, 30% of the root in dry mass, having already been found up to 45% (Borges et al., 2002; Mendonça et al., 2003), an important factor because the higher the dry mass content, the greater the yield of the final product per unit of cultivated area (Vidigal Filho et al., 2000).

The maximum values for SY were SC05 (65.7%) and BC04 (64.7%), as seen in Tables 1 and 2. Nuwamanya et al. (2010), in turn, found higher values of starch in dry weight, ranging from 70 to 90%.

Physical and Chemical Properties of Roots

The physicochemical properties of the cassava roots (Tables 3) show that the cassava genotypes present significant differences for most of the physicochemical parameters (P <0.05) such as protein content that varied from 0, 4 to 1.3% (SC10-SC03); lipids 0.3 to 1.9% (SC10-SC02); moisture 55.3 to 64.4% (SC12-SC08); acidity 2.3 to 3.6% (SC07, SC08-SC03); and total soluble solids between 1.0 to 1.4 ºBrix (SC12-SC06), except for ash, fiber and pH contents, which did not vary significantly.

In the cassava genotypes (Table 4), significant statistical differences can be observed in all parameters analyzed (P <0.05). The protein content varied from 0.1 to 0.7% (BC02-BC08, BC11); lipids 0.3 to 2.1% (BC04, BC05, BC06-BC10); moisture 58.0 to 65.2% (BC05-BC10); ash from 0.1 to 1.0% (BC10-BC05); fibers 0.9 to 1.9% (BC09, BC15-BC05); acidity 1.1 to 2.7% (BC13-BC11); pH 6.3 to 6.8 (BC10-BC05, BC11); and ºBrix 0.8 to 1.2 (BC09-BC02, BC05, BC11).

Tables 5 and 6 show the results of the concentrations of soluble sugars, starch content present in the root pulp, and the total carbohydrates. The sweet cassava genotypes showed statistically significant differences (P <0.05) between the parameters, such as: glucose levels that varied from 0.2 to 1.0% (SC03, SC07, SC11, SC12 and SC08); fructose 0 to

0.3% (SC05, SC11, SC12, and SC08); sucrose 0.6 to 1.0% (SC01, SC02, SC03, SC04, SC09, and SC12 - SC06); starch 26 to 38.9% (SC12-SC02), with a total of 26.8 to 40% (SC12-SC02) (Table 5) . In Table 6, there were statistical differences (P <0.05) in the cassava genotypes for glucose levels that varied from 0.1 to 0.8% (BC11-BC14) and sucrose 0.5 to 1.0% (BC06- BC07), the other properties did not differ statistically. The levels of TSS (1.3 SC and 1.1 BC), moisture (60.1 SC and 63.2 BC), and ash (0.5 for SC and BC) were lower than those found by Araújo et al. (2019) which obtained TSS values of 3.55 for BC and 2.50 for SC, ashes of 2.0 for SC and BC, and moisture of 62.3 for SC and 65.5 for BC. The differences in TSS may have occurred due to the variation in moisture values in relation to the age of the plant.

Glucose, fructose and sucrose average levels among SC groups was 0.4%; 0.1%; 0.7%, and among the BC groups was 0.4%; 0.1%; 0.7% respectively. Lower results were obtained by Araújo, et al. (2015); Araújo et al. (2019).

Regarding the pH, the evaluated genotypes showed values close to neutrality for SC (6.7) and BC (6.6), similar to the results obtained by Araújo et al. (2015) between 6.50 and 5.68. Lower values could indicate deterioration caused by microorganisms in the samples.

The fiber data observed in this work showed averages of 0.9% SC and 1.2% BC, that is, a low source of fibers such as lignin and cellulose. According to Mattos and Martins (2000) foods are classified as having a very high fiber with content greater than or equal to 7%, moderate between 4.5 to 6.9%, and low with content less than 4.4%.

The levels of crude protein found in the roots were higher in SC with average levels of 0.8% compared to the results of Luna et al. (2013) with 0.43%, however equal to the average levels found for BC of 0.4%.

The lipid content obtained in this study (0.8% for SC and 0.4% for BC) was higher than the data from Ceni et al. (2009) who found values in the range of 0.2 to 0.7% when evaluating different cassava roots.

The average of starch content was 31% for SC and 22.5% for BC, while for williams et al. (2019) and araújo et al. (2015), the averages were between 25.67- 29.33%, which are relatively close to those found in this study.

Analysis of Principal Components for the Physicochemical Properties of the Roots

In the multivariate analysis of principal components (PCA) using Pearson's correlation, the loadings of the variables analyzed are shown in Figure 1A and the genotype scores in Figure 1B, using two factors (F1 and F2) that together explain 84.2% of the total variability observed between the genotypes of sweet and bitter cassava.

The hierarchical cluster analysis (HCA) was also performed using Euclidean distance (Table 7) to explore the variability of the dataset, as shown in the dendrogram (Figure 2), where cassava genotypes formed 4 clusters with 75% similarity. In addition, it can be seen that there was no interaction between the genotypes of SC and BC.

Table 8 shows that the variables Lipids, Proteins, and pH were responsible for cluster I, formed by SC01 and SC08 genotypes, Moisture and Fibers formed cluster II (BC10, BC12, BC14), Starch yield, Ashes, and acidity formed cluster III (SC02, SC03, SC04, SC05, SC06, SC07, SC09, SC10, SC11, SC12), and TSS concentration formed cluster IV (BC01, BC02, BC03, BC04, BC05, BC06, BC07, BC08, BC09, BCB11, BC13, BC15).

The use of multivariate statistical techniques of both principal components and cluster analyzes has been widely

Table 1. Yield of roots and starch extracted from 12 genotypes of sweet cassava (SC). (1) storage root number; (2) fresh storage root number; (3) root fresh matter content; (4) root dry matter content (5) starch yield - dry weight. Each value represents the mean ± standard deviation (n = 3). The values of the columns followed by different letters differ from each other by the Tukey test (P <0.05).

| Genotypes	[1]SNR	[2]FSRW (kg)	[3]RFMC (%)	[4]RDMC (%)	[5]SY (%)
SC01	ab 4.0 ± 1.7	ab 1.0 ± 0.6	c 72.8 ± 0.2	ab 43.1 ± 0.9	d 28.2 ± 3.7
SC02	ab 6.3 ± 1.2	b 0.6 ± 0.1	c 74.1 ± 0.1	ab 42.9 ± 2.4	ab 60.0 ± 3.0
SC03	ab 4.3 ± 0.6	ab 1.0 ± 0.2	a 88.6 ± 8.7	de 36.4 ± 0.7	bc 48.2 ± 6.3
SC04	ab 5.0 ± 0.0	ab 1.0 ± 0.6	a 82.6 ± 2.4	bcde 38.7 ± 1.9	cd 42.0 ± 6.2
SC05	ab 4.7 ± 0.5	ab 1.4 ± 0.4	bc 75.4 ± 1.0	abc 42.2 ± 1.5	a 65.7 ± 1.6
SC06	ab 3.7 ± 2.9	ab 0.8 ± 0.6	d 58.8 ± 0.3	de 35.8 ± 0.7	abc 55.6 ± 3.0
SC07	ab 6.0 ± 0.0	ab 1.8 ± 0.2	a 83.6 ± 1.6	bcde 39.5 ± 2.2	abc 56.2 ± 5.1
SC08	b 2.7 ± 1.5	a 2.5 ± 1.3	bc 76.1 ± 2.6	e 35.6 ± 0.5	d 29.1 ± 4.0
SC09	a 7.7 ± 3.1	ab 1.8 ± 0.1	bc 75.5 ± 0.3	cde 38.0 ± 1.1	ab 58.5 ± 3.7
SC10	ab 7.0 ± 1.7	ab 1.3 ± 0.4	c 72.6 ± 0.2	abc 41.5 ± 1.0	bc 51.3 ± 3.5
SC11	ab 5.7 ± 2.1	ab 1.7 ± 0.6	bc 75.0 ± 3.8	abcd 40.5 ± 2.8	ab 58.9 ± 3.4
SC12	ab 6.7 ± 0.6	ab 2.2 ± 0.9	ab 80.7 ± 0.7	a 44.7 ± 1.7	ab 57.5 ± 4.3
Overall mean	5.3	1.4	76.0	39.9	50.4
CV (%)	31.1	41.9	2.7	4.0	8.2

Source: Elaborated by the authors (2020).
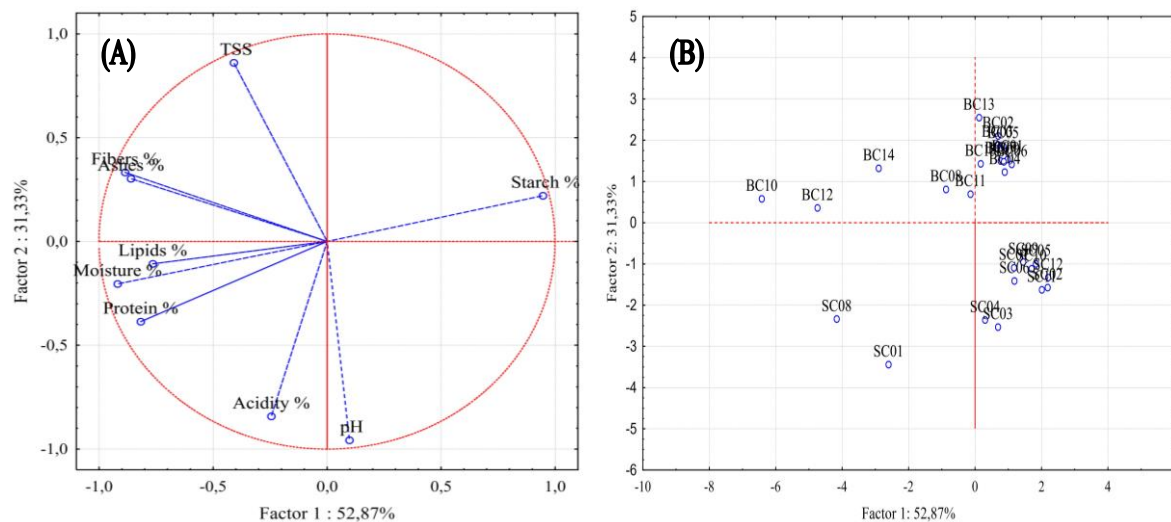
Figure 1. Principal components analysis (PCA) of starch, protein, lipid, moisture, ash, fiber, acidity, pH, and root TSS, among the 12 genotypes of sweet cassava (SC) and 15 genotypes of bitter cassava (BC). In (A) are shown the loadings of the physicochemical variables. (B) shows the scores of the genotypes evaluated according to the variables.
Source: Elaborated by the authors (2020).

Table 2. Yield of roots and starch extracted from 15 genotypes of bitter cassava (SC). (1) storage root number; (2) fresh storage root number; (3) root fresh matter content; (4) root dry matter content (5) starch yield - dry weight. Each value represents the mean ± standard deviation (n = 3). The values of the columns followed by different letters differ from each other by the Tukey test (P <0.05).

Genotypes	[1]SNR	[2]FSRW (kg)	[3]RFMC (%)	[4]RDMC (%)	[5]SY (%)
BC01	[b] 3.3± 1.5	[a] 1.5 ± 1.0	[abc] 79.2 ± 3.1	[a] 38.9 ± 3,2	[ab] 57.9 ± 5.2
BC02	[ab] 4.0±0.0	[a] 1.5 ± 0.5	[ab] 84.0 ± 1.9	[a] 35.2 ± 8,4	[ab] 61.7 ± 9.4
BC03	[b] 3.7±1.2	[a] 3.2 ± 0.7	[ab] 83.6 ± 3.8	[a] 41.7 ± 2,2	[abcde] 51.5 ± 3.6
BC04	[ab] 5.3±3.1	[a] 2.8 ± 2.6	[abc] 79.7 ± 4.6	[a] 35.2 ± 6,4	[a] 64.7 ± 4.8
BC05	[b] 3.0±1.0	[a] 1.3 ± 0.8	[c] 73.1 ± 4.6	[a] 42.0 ± 3,0	[abc] 55.4 ± 8.5
BC06	[ab] 4.3±0.6	[a] 1.4 ± 0.2	[abc] 80.3 ± 2.5	[a] 39.8 ± 0,4	[ab] 60.9 ± 1.0
BC07	[ab] 5.7±1.2	[a] 3.1 ± 0.7	[abc] 79.2 ± 0.5	[a] 40.0 ± 3,7	[abcd] 55.2 ± 4.3
BC08	[ab] 4.7±0.6	[a] 1.7 ± 0.7	[abc] 79.0 ± 1.3	[a] 38.6 ± 1,0	[bcdefg] 42.0 ± 6.3
BC09	[b] 3.7±2.1	[a] 1.2 ± 0.5	[abc] 78.0 ± 4.2	[a] 36.9 ± 2,0	[ab] 61.2 ± 1.6
BC10	[ab] 5.3±2.5	[a] 1.9 ± 0.9	[bc] 76.7 ± 2.8	[b] 17.6 ± 5,0	[cefg] 34.7 ± 11.3
BC11	[ab] 5.0±1.0	[a] 1.7 ± 0.4	[bc] 76.3 ± 2.7	[a] 37.6 ± 1,9	[abcdef] 46.0 ± 4.2
BC12	[a] 8.3 ± 2.1	[a] 2.7 ± 0.6	[abc] 80.8 ± 2.0	[a] 38.3 ± 2,9	[g] 27.4 ± 2.6
BC13	[ab] 4.0±0.0	[a] 2.5 ± 1.7	[c] 73.4 ± 1.5	[a] 34.8 ± 7,6	[ab] 58.0 ± 7.2
BC14	[b] 3.3±1.5	[a] 2.4 ± 1.8	[a] 86.4 ± 1.2	[a] 39.9 ± 1,1	[fg] 32.0 ± 1.9
BC15	[b] 2.3±0.6	[a] 1.3 ± 0.6	[abc] 80.1 ± 2.7	[a] 34.9 ± 8,0	[abcde] 53.2 ± 10.4
Overall mean	4.4	2.0	79.3	36.8	50.9
CV (%)	34.6	54.3	3.6	12.5	11.3

Source: Elaborated by the authors (2020).
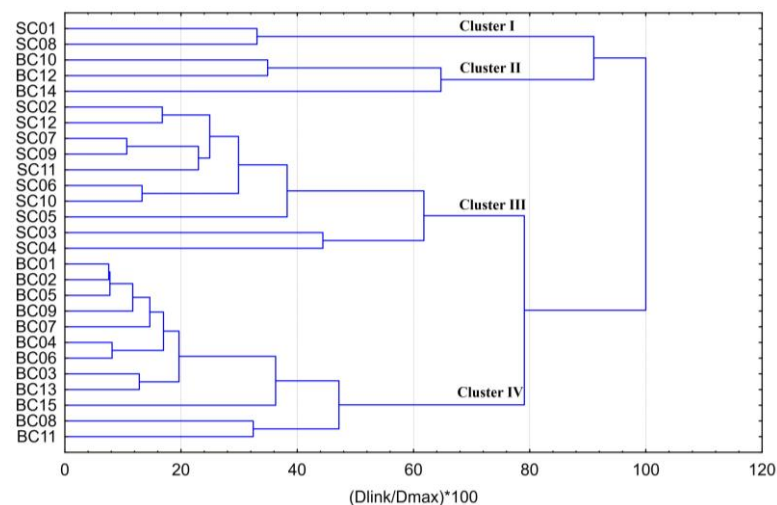
Figure 2. Dendrogram obtained from the variables characterizing the yield of starch, proteins, lipids, moisture, ash, fibers, acidity, pH, TSS, glucose, fructose, sucrose, and root starch, among the groups of 12 sweet cassava genotypes (SC) and 15 of bitter cassava (BC). Source: Elaborated by the authors (2020).

Table 3. Physicochemical characterization of peeled root of 12 sweet cassava (SC) genotypes, based on fresh weight.

Genotypes	Content in %						pH	**TSS
	Protein	Lipids	Moisture	Ash	Fiber	Acidity*		
SM01	[abc] 1.1 ± 0.3	[ab] 1.1 ± 0.2	[de] 56.9 ± 0.9	[a] 0.4 ± 0.1	[a] 1.1 ± 0.1	[cde] 3.0 ± 0.2	[a] 6.8 ± 0.2	[abc] 1.2 ± 0.1
SM02	[bcde] 0.7 ± 0.1	[a] 1.9 ± 0.3	[de] 57.1 ± 2.4	[a] 0.4 ± 0.1	[a] 1.2 ± 0.1	[ab] 3.5 ± 0.3	[a] 6.8 ± 0.3	[abc] 1.3 ± 0.2
SM03	[a] 1.3 ± 0.1	[b] 0.5 ± 0.2	[ab] 63.6 ± 0.7	[a] 0.4 ± 0.1	[a] 0.9 ± 0.2	[a] 3.6 ± 0.1	[a] 6.9 ± 0.1	[ab] 1.4 ± 0.2
SM04	[f] 0.4 ± 0.1	[b] 0.7 ± 0.5	[abcd] 61.3 ± 1.9	[a] 0.4 ± 0.0	[a] 0.7 ± 0.2	[abcd] 3.3 ± 0.1	[a] 6.8 ± 0.3	[abc] 1.1 ± 0.1
SM05	[ab] 1.2 ± 0.1	[ab] 1.1 ± 0.4	[cde] 57.8 ± 1.5	[a] 0.5 ± 0.1	[a] 1.1 ± 0.2	[ef] 2.7 ± 0.2	[a] 7.0 ± 0.2	[abc] 1.3 ± 0.3
SM06	[abcd] 0.9 ± 0.1	[ab] 1.0 ± 0.4	[ab] 64.2 ± 0.7	[a] 0.4 ± 0.1	[a] 1.1 ± 0.1	[bcde] 3.0 ± 0.0	[a] 6.7 ± 0.3	[a] 1.5 ± 0.1
SM07	[abcd] 1.0 ± 0.2	[ab] 1.1 ± 0.3	[abcd] 60.5 ± 2.2	[a] 0.5 ± 0.0	[a] 0.8 ± 0.1	[f] 2.3 ± 0.1	[a] 6.6 ± 0.1	[bc] 1.0 ± 0.2
SM08	[def] 0.7 ± 0.2	[b] 1.0 ± 0.4	[a] 64.4 ± 0.5	[a] 0.4 ± 0.1	[a] 1.2 ± 0.9	[f] 2.3 ± 0.2	[a] 6.6 ± 0.2	[abc] 1.2 ± 0.2
SM09	[def] 0.7 ± 0.1	[b] 0.5 ± 0.3	[abc] 62.0 ± 1.1	[a] 0.5 ± 0.1	[a] 0.8 ± 0.2	[ef] 2.6 ± 0.0	[a] 6.7 ± 0.1	[abc] 1.3 ± 0.1
SM10	[f] 0.4 ± 0.1	[b] 0.3 ± 0.1	[cde] 58.5 ± 1.0	[a] 0.6 ± 0.1	[a] 0.8 ± 0.2	[ef] 2.7 ± 0.4	[a] 6.8 ± 0.1	[ab] 1.4 ± 0.1
SM11	[cdef] 0.8 ± 0.1	[b] 0.5 ± 0.2	[bcde] 59.5 ± 2.8	[a] 0.5 ± 0.1	[a] 0.7 ± 0.1	[abc] 3.4 ± 0.1	[a] 6.6 ± 0.2	[ab] 1.4 ± 0.0
SM12	[ef] 0.5 ± 0.1	[b] 0.5 ± 0.2	[e] 55.3 ± 1.7	[a] 0.6 ± 0.1	[a] 0.4 ±0.1	[def] 2.8 ± 0.3	[a] 6.9 ± 0.3	[c] 1.0 ± 0.1
Overall mean	0.8	0.8	60.1	0.5	0.9	2.9	6.8	1.3
CV (%)	16.3	37.2	2.7	16.9	30.8	6	3.1	11.4

*mL (0.01M NaOH 100 g^{-1}); ** TSS- Total soluble solids. Source: Elaborated by the authors (2020).

Table 4. Physicochemical characterization of peeled root of 15 bitter cassava (BC) genotypes, based on fresh weight.

Genotypes	Content in %						pH	**TSS
	Protein	Lipids	Moisture	Ash	Fiber	Acidity*		
BC01	[bcd] 0.3 ± 0.1	[b] 0.7 ± 0.2	[b] 61.1 ± 3.2	[bc] 0.4 ± 0.0	[b] 1.0 ± 0.2	[abcd] 2.1 ± 0.2	[ab] 6.5 ± 0.1	[ab] 1.1 ± 0.1
BC02	[e] 0.1 ± 0.0	[b] 0.5 ± 0.4	[b] 64.8 ± 8.4	[bc] 0.4 ± 0.2	[ab] 1.2 ± 0.3	[cde] 1.5 ± 0.1	[ab] 6.6 ± 0.1	[a] 1.2 ± 0.1
BC03	[ab] 0.4 ± 0.0	[b] 0.4 ± 0.2	[b] 58.3 ± 2.2	[bc] 0.4 ± 0.1	[ab] 1.3 ± 0.2	[bcde] 1.7 ± 0.1	[ab] 6.6 ± 0.1	[ab] 1.1 ± 02
BC04	[bc] 0.4 ± 0.1	[b] 0.3 ± 0.0	[b] 64.8 ± 6.4	[bc] 0.4 ± 0.1	[ab] 1.1 ± 0.3	[ab] 2.5 ± 0.4	[ab] 6.4 ± 0.2	[ab] 1.0 ± 0.2
BC05	[a] 0.6 ± 0.1	[b] 0.3 ± 0.1	[b] 58.0 ± 3.0	[a] 1.0 ± 0.4	[ab] 1.2 ± 0.2	[abc] 2.3 ± 0.0	[a] 6.8 ± 0.2	[a] 1.2 ± 0.2
BC06	[ab] 0.5 ± 0.1	[b] 0.3 ± 0.0	[b] 60.2 ± 0.4	[b] 0.6 ± 0.1	[ab] 1.1 ± 0.2	[ab] 2.5 ± 0.3	[ab] 6.6 ± 0.2	[ab] 1.1 ± 0.1
BC07	[a] 0.6 ± 0.1	[b] 0.4 ± 0.0	[b] 60.0 ± 3.7	[bc] 0.5 ± 0.1	[b] 1.1 ± 0.0	[abcde] 2.1 ± 0.2	[ab] 6.6 ± 0.1	[ab] 1.1 ± 0.2
BC08	[a] 0.7 ± 0.1	[b] 0.6 ± 0.6	[b] 61.4 ± 1.0	[bc] 0.5 ± 0.1	[b] 1.0 ± 0.3	[abc] 2.4 ± 0.5	[ab] 6.7 ± 0.1	[ab] 1.0 ± 0.1
BC09	[cde] 0.2 ± 0.1	[b] 0.5 ± 0.2	[b] 63.1 ± 2.0	[bc] 0.4 ± 0.0	[b] 0.9 ± 0.2	[abc] 2.3 ± 0.5	[ab] 6.6 ± 0.1	[b] 0.8 ± 0.2
BC10	[cde] 0.2 ± 0.1	[a] 2.1 ± 0.5	[a] 82.4 ± 5.0	[c] 0.1 ± 0.0	[ab] 1.5 ± 0.4	[de] 1.2 ± 0.5	[b] 6.3 ± 0.1	[ab] 1.0 ± 0.1
BC11	[a] 0.7 ± 0.1	[b] 0.4 ± 0.1	[b] 62.4 ± 1.9	[bc] 0.5 ± 0.1	[b] 1.0 ± 0.0	[a] 2.7 ± 0.1	[a] 6.8 ± 0.1	[a] 1.2 ± 0.1
BC12	[a] 0.6 ± 0.1	[b] 0.4 ± 0.1	[b] 61.7 ± 2.9	[bc] 0.4 ± 0.0	[ab] 1.2 ± 0.3	[abcde] 2.0 ± 0.4	[ab] 6.7 ± 0.1	[ab] 1.1 ± 0.2
BC13	[de] 0.2 ± 0.1	[b] 0.4 ± 0.1	[b] 65.2 ± 7.6	[bc] 0.4 ± 0.0	[a] 1.9 ± 0.2	[e] 1.1 ± 0.1	[ab] 6.5 ± 0.2	[ab] 1.1 ± 0.1
BC14	[cde] 0.2 ± 0.0	[b] 0.4 ± 0.1	[b] 60.1 ± 1.1	[bc] 0.4 ± 0.1	[ab] 1.1 ± 0.4	[cde] 1.5 ± 0.3	[ab] 6.6 ± 0.2	[ab] 1.1 ± 0.1
BC15	[bcde] 0.3 ± 0.1	[b] 0.9 ± 0.5	[b] 65.1 ± 8.0	[bc] 0.3 ± 0.1	[b] 0.9 ± 0.1	[abcde] 1.9 ± 0.3	[ab] 6.6 ± 0.2	[ab] 1.1 ± 0.1
Overall mean	0.4	0.6	63.2	0.5	1.2	2.0	6.6	1.1
CV(%)	17.9	48.5	7.2	30.3	22.9	16	2.3	11.6

*mL (0.01M NaOH 100 g^{-1}); ** TSS- Total soluble solids. Source: Elaborated by the authors (2020).

Table 5. Soluble sugars concentration, starch, and total carbohydrates from 12 sweet cassava (SC) genotypes. Each value represents the mean ± standard deviation (n = 3). The column values followed by different letters differ from each other by Tukey's test (P <0.05).

Genotypes	Content in %				
	Glucose	Fructose	Sucrose	Starch	Total
SC01	[b] 0.4 ± 0.0	[ab] 0.2 ± 0.1	[b] 0.6 ± 0.1	[bc] 26.9 ± 3.6	[bc] 28.0 ± 3.6
SC02	[b] 0.3 ± 0.0	[b] 0.1 ± 0.0	[b] 0.6 ± 0.1	[a] 38.9 ± 1.3	[a] 40.0 ± 1.2
SC03	[b] 0.2 ± 0.1	[b] 0.1 ± 0.0	[b] 0.6 ± 0.2	[ab] 37.6 ± 0.7	[ab] 38.7 ± 0.8
SC04	[b] 0.4 ± 0.2	[ab] 0.2 ± 0.1	[b] 0.6 ± 0.1	[abc] 32.7 ± 1.4	[abc] 33.8 ± 1.0
SC05	[b] 0.3 ± 0.0	[b] 0.0 ± 0.0	[ab] 0.8 ± 0.1	[abc] 35.3 ± 2.7	[abc] 36.5 ± 2.7
SC06	[b] 0.4 ± 0.1	[ab] 0.1 ± 0.0	[a] 1.0 ± 0.0	[abc] 30.8 ± 6.8	[abc] 32.3 ± 6.7
SC07	[b] 0.2 ± 0.0	[b] 0.1 ± 0.0	[b] 0.5 ± 0.1	[abc] 30.6 ± 3.2	[abc] 31.4 ± 3.3
SC08	[a] 1.0 ± 0.3	[a] 0.3 ± 0.1	[ab] 0.8 ± 0.2	[bc] 26.5 ± 1.2	[bc] 28.5 ± 1.5
SC09	[b] 0.3 ± 0.0	[b] 0.1 ± 0.0	[b] 0.6 ± 0.1	[abc] 32.8 ± 2.8	[abc] 33.7 ± 2.8
SC10	[b] 0.5 ± 0.1	[ab] 0.2 ± 0.0	[b] 0.7 ± 0.2	[abc] 29.1 ± 4.6	[abc] 30.4 ± 4.4
SC11	[b] 0.2 ± 0.1	[b] 0.0 ± 0.0	[ab] 0.7 ± 0.1	[abc] 29.8 ± 1.2	[abc] 30.8 ± 1.1
SC12	[b] 0.2 ± 0.1	[b] 0.0 ± 0.0	[b] 0.6 ± 0.0	[c] 26.0 ± 4.4	[c] 26.8 ± 4.3
Overall mean	0.4	0.1	0.7	31	32.2
CV(%)	30.5	43.8	17.5	11.13	10.6

Source: Elaborated by the authors (2020).

Table 6. Soluble sugars concentration, starch, and total carbohydrates from 15 bitter cassava (BC) genotypes. Each value represents the mean ± standard deviation (n = 3). The column values followed by different letters differ from each other by Tukey's test (P <0.05).

Genotypes	Content in %				
	Glucose	Fructose	Sucrose	Starch	Total
BC01	[ab] 0.5 ± 0.0	[a] 0.1 ± 0.0	[b] 0.7 ± 0.1	[a] 25.1 ± 1.2	[a] 26.3 ± 1.2
BC02	[ab] 0.5 ± 0.4	[a] 0.2 ± 0.2	[b] 0.6 ± 0.1	[a] 24.7 ± 4.3	[a] 26.0 ± 3.7
BC03	[ab] 0.4 ± 0.1	[a] 0.1 ± 0.0	[ab] 0.8 ± 0.2	[a] 24.1 ± 1.5	[a] 25.3 ± 1.4
BC04	[ab] 0.6 ± 0.0	[a] 0.2 ± 0.1	[b] 0.7 ± 0.1	[a] 18.7 ± 4.2	[a] 20.2 ± 4.2
BC05	[ab] 0.3 ± 0.1	[a] 0.1 ± 0.0	[ab] 0.7 ± 0.1	[a] 22.8 ± 2.7	[a] 23.9 ± 2.8
BC06	[ab] 0.2 ± 0.1	[a] 0.1 ± 0.0	[b] 0.5 ± 0.2	[a] 21.9 ± 2.1	[a] 22.8 ± 1.9
BC07	[ab] 0.6 ± 0.2	[a] 0.1 ± 0.1	[a] 1.0 ± 0.0	[a] 22.0 ± 2.9	[a] 23.7 ± 2.8
BC08	[ab] 0.3 ± 0.1	[a] 0.1 ± 0.0	[b] 0.6 ± 0.2	[a] 19.2 ± 1.5	[a] 20.3 ± 1.5
BC09	* NA	NA	NA	NA	NA
BC10	NA	NA	NA	NA	NA
BC11	[b] 0.1 ± 0.0	[a] 0.0 ± 0.0	[ab] 0.7 ± 0.1	[a] 23.3 ± 1.0	[a] 16.4 ± 13
BC12	NA	NA	NA	NA	NA
BC13	[ab] 0.5 ± 0.1	[a] 0.2 ± 0.1	[ab] 0.7 ± 0.30	[a] 19.6 ± 2.9	[a] 20.8 ± 3.1
BC14	[a] 0.8 ± 0.3	[a] 0.3 ± 0.2	[ab] 0.9 ± 0.10	[a] 23.6 ± 7.1	[a] 26.1 ± 6.1
BC15	[ab] 0.4 ± 0.1	[a] 0.1 ± 0.1	[b] 0.6 ± 0.10	[a] 25.5 ± 3.1	[a] 26.7 ± 3.3
Overall mean	0.4	0.1	0.7	22.5	23.2
CV (%)	41.8	69.0	17.3	13.8	14.5

*Not analysed, Source: Elaborated by the authors (2020).

Table 7. Pearson correlation matrix of the variables proteins (%), lipids (%), moisture (%), ash (%), fibers (%), acidity (%), pH, Total Soluble Solids (TSS). Starch (%), evaluated from the sweet and bitter cassava genotypes.

	Protein	Lipids	Moisture	Ashes	Fibers	Acidity	TSS	pH
Lipids	* 0.641							
Moisture	* 0.767	* 0.599						
Ashes	* 0.561	* 0.589	* 0.691					
Fibers	* 0.603	* 0.673	* 0.709	* 0.844				
Acidity	* 0.481	0.15	* 0.448	- 0.0507	- 0.136			
TSS	0.000511	0.113	0.255	* 0.577	* 0.591	**- 0.5		
pH	0.276	0.0569	0.0828	- 0.326	- 0.355	* 0.676	**- 0.932	
Starch	**- 0.813	** -0.653	** -0.993	**- 0.726	**- 0.74	**- 0.46	- 0.24	- 0.104

* Pairs of variables with positive correlation coefficients and P-values <0.05 tend to increase together; ** Pairs of variables with negative correlation coefficients and P-values <0.05 tend to decrease while the others increase. There is no significant relationship between the other pairs of variables.
Source: Elaborated by the authors (2020).

Table 8. Comparison between means of physicochemical properties according to the cluster formed between 12 genotypes of sweet cassava (SC) and 15 genotypes of bitter cassava (BC).

Cluster Groups				
	I	II	III	IV
Genotypes	SC01, SC08	BC10, BC12, BC14	SC02, SC03, SC04, SC05, SC06, SC07, SC09, SC10, SC11, SC12	BC01, BC02, BC03, BC04, BC05, BC06, BC07, BC08, BC09, BC11, BC13, BC15
Properties	Means			
Lipids (%)	*1.01	0.97	0.78	0.48
Moisture (%)	60.65	*68.07	59.98	62.03
Ash (%)	0.42	0.31	0.47	*0.56
Protein (%)	*0.90	0.33	0.79	0.42
Fiber(%)	1.15	*1.27	0.85	1.14
Acidity (%)	2.65	1.57	*2.99	2.17
TSS	1.20	1.07	*1.27	1.08
pH	* 6.70	6.53	5.45	6.61
SY(%)*	28.65	31.37	*55.39	53.09

* Highest mean values , Source: Elaborated by the authors (2020).

Table 9. Identification of 12 genotypes of sweet cassava (SC) and 15 genotypes of bitter cassava cultivated in the active bank of germplasm (AGB) of EMBRAPA Eastern Amazon.

Genotype	Code	Local name/city	State	Country/Origin	Acquisition date
SC01	CPATU 390	Acará	PA	Brazil	06/07/2004
SC02	CPATU 034	Marabá	PA	Brazil	01/02/1960
SC03	CPATU 177	Ferreira Gomes	AP	Brazil	26/11/1996
SC04	CPATU 239	Santa Luzia	PA	Brazil	01/02/2000
SC05	CPATU 340	Santarém	PA	Brazil	27/06/2001
SC06	CPATU 162	Unknown	BA	Brazil	01/11/1995
SC07	CPATU 268	Rondon do Pará	PA	Brazil	14/09/2000
SC08	CPATU 103	Machadinho D'Oeste	RO	Brazil	26/08/1992
SC09	CPATU 216	Igarapé-Açu	PA	Brazil	18/06/1998
SC10	CPATU 339	Santarém	PA	Brazil	27/06/2001
SC11	CPATU 387	Belém	PA	Brazil	10/03/2004
SC12	CPATU 307	Santarém	PA	Brazil	27/06/2001
BC01	CPATU 018	Santarém	PA	Brazil	01/02/1950
BC02	CPATU 403	*CNPMF	BA	Brazil	29/09/2005
BC03	CPATU 125	Oiapoque	AP	Brazil	06/05/1993
BC04	CPATU 267	Rondon do Pará	PA	Brazil	14/09/2000
BC05	CPATU 022	Bragança	PA	Brazil	01/02/1950
BC06	CPATU 229	Nova TiBCoteua	PA	Brazil	16/12/1998
BC07	CPATU 033	Santarém	PA	Brazil	01/02/1960
BC08	CPATU 086	Nossa senhora das Dores	SE	Brazil	-
BC09	CPATU 438	Belém	PA	Brazil	01/11/2007
BC10	CPATU 352	Paragominas	PA	Brazil	12.08.2010
BC11	CPATU 437	Belém	PA	Brazil	01/11/2007
BC12	CPATU 489	Paragominas	PA	Brazil	12/08/2010
BC13	CPATU 488	Santarém	PA	Brazil	27/06/2001
BC14	CPATU 357	Castanhal	PA	Brazil	09/10/2001
BC15	CPATU 348	Santarém	PA	Brazil	27/06/2001

* CNPMF-EMBRAPA Cassava and Tropical Fruit / Cruz das Almas- BA; - No registry. Source: Elaborated by the authors (2020).

used to quantify the genetic divergence in cassava (Agre et al., 2015; Karim et al., 2019) when concisely viewing the results of the variables of the analyzed genotypes the principal component and grouping technique allows to reduce a wide number of original variables from the linear combinations of the characters, representing the original variation maximum of the individuals, which makes possible to use the genotypes according to their real potential, directly or in breeding programs.

As observed in Figures 2A and 2B and Table 5, the quantitative and qualitative results can characterize genotypes with properties that may be of potential commercial and biotechnological interest for the industry. Among the genotypes, we highlight those from group III, characterized by the highest concentration of starch.

Materials and methods

Plant Material and Growth Conditions
The experiment was carried out under field conditions, with 1-year-old cassava plants, grown at 1x1 m spacing, under the same management conditions and cultural treatments collected at the EMBRAPA Amazônia Oriental active germplasm bank (AGB), located in Belém do Pará with an average temperature of 27.2 °C and an average of 3418.7 mm, from February 2012 to February 2013 according to the agrometeorological station (Lat. 01º 28'S, Longitude: 48º 27 'WGr and Altitude: 12.8 m). For the development of this study, 27 genotypes of sweet cassava (SC) and bitter cassava (BC) (Table 9) were selected based on the uniformity of the height of the genotypes.

Determinations of Root and Starch Yield
The experiment was conducted at the EMBRAPA Agroindustry Laboratory. Roots were harvested from three plants per genotype, washed, peeled, sanitized (0.02% NaClO), and processed in an industrial crusher to disintegrate the cells and release the starch granules. Then, the resulted product was filtered on synthetic voile-type fabric. Regarding the yield, the storage root number (SRN) and the fresh storage root weight (FSRW) (kg plant^{-1}) were determined, as well as the root fresh matter content (RFMC) and root dry matter content (RDMC), both expressed as a percentage. The RDMC was determined through the relationship between the fresh root mass (Kg) and the dry root mass (Kg) multiplied by 100. The samples were weighted to determine the percentage of starch yield (SY) in dry weight, and physicochemical analyzes were done.

Physical and Chemical Properties for Roots and Extracted Starch
• **Moisture and Root Ash**
The determination of moisture and ash content was carried out gravimetrically, following methodology No. 920,151 and methodology No. 930.05 (AOAC, 1997).
• **Lipids**
The percentage of lipids was determined by the Bligh-Dyer method (1959).
• **Crude Protein**
Protein determination was performed using the Micro-Kjeldahl method, according to methodology No. 950.48 (AOAC, 1997).

1276

• Total Fiber

The determination of fibers was carried out by acid detergent by the method Van Soest et al., (1963) method nº 973.18 AOAC (1997).

• PH and Total Titratable Acidity (TTA)

For pH determination 0.5 g of unpeeled root was macerated in 25 mL of distilled water through direct reading in a portable digital pH meter, according to method No. 981.12 (AOAC, 1997). Then the total titratable acidity content was determined by volumetry with the phenolphthalein indicator 1%, No. 942.15 A (AOAC, 1997).

• Total Soluble Solids

The determination of the total soluble solids consists in the measurement of the refractive index of the solutions, the results were expressed in Brix degree, through the use of a digital refractometer HISEG RTD-45 (0-32 ºBrix), according to AOAC method No. 932.12 (1997).

• Glucose, Fructose, Sucrose, and Root Starch

The determination of glucose, fructose, and sucrose (soluble sugars) were carried out according to the methodology of Stitt et al. (1989). The selected roots were sectioned (100 mg of the root in fresh weight), macerated, and homogenized in 80% ethanol. Then, the resulting product was centrifuged for 10 minutes at 13 000 rpm and the ethanolic extract separated from the precipitate. An aliquot of the soluble fraction of each sample was added to the reaction buffer in an Elisa plate. Readings were performed at 340 nm, using the following steps: Hexokinase enzyme is first added for glucose determination. Then, after glucose levels stabilization, phosphoglucoisomerase is added, and finally, β-fructosidase for fructose and sucrose determination, respectively.

The methodology of Trethewey et al. (1998) was used to determine starch. The ethanolic extract precipitate was washed three times in 80% ethanol for the elimination of impurities. Subsequently, 1 mL of 0.2 M KOH was added, homogenized, and immersed in water bath at 95 ºC for 2 hours, pH was corrected to 5.5, and the resultant solution was centrifuged for 10 minutes at 13 000 rpm. The supernatant was removed and incubated with citrate buffer0.3 M at pH 4.6. Then, α-amylase and amyloglucosidase enzymes were added, supplemented with milli-Q water, and taken back to the water bath at 60 ºC for 60 minutes. Finally, glucose determination was performed in the same way.

• Statistical Analysis

The STATISTICAL method used in this study was a completely randomized design (CRD) with three replicates per genotype. Statistical analysis of the data was performed in ASSISTAT software version 7.7 Beta, covering the descriptive analysis of the data and comparison of means by the analysis of variance (ANOVA) using the Tukey's test (P <0.05). Principal components analysis (ACP) and hierarchical cluster analysis (HCA) were also performed using Statistica 7.0 software (StatSoft®, USA).

Conclusion

The evaluation of cassava physicochemical properties and root yields allow us to highlight among the genotypes of sweet and bitter cassava those that have the most relevant characteristics in root yield (SC12 and BC05) and starch (SC05 and BC04). Through multivariate analysis, we can identify cluster I (with higher averages of lipids and proteins) and cluster III (with a higher average of starch) as groups containing genotypes of biotechnological interest in the development of new studies and products.

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