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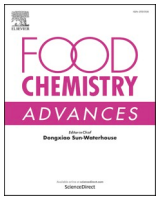

Cassava waste (stem and leaf) analysis for reuse

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ABSTRACT

Cassava is an important crop for developing countries. In addition to its roots, the leaves are rich in proteins and minerals that could substantially supplement diets if treated properly, as they contain some anti-nutritional factors. Plant residues can be used as raw material in the food industry and as bioenergy. This work aimed to characterize the profile of macronutrients and polysaccharides in leaves and stems of two cassava clones generated by the breeding program (classical genetic improvement/grafting) of Embrapa Mandioca & Fruticultura. Dehydrated and crushed leaves and stems evaluated for chemical composition, in addition to the determination of cyanogenic compounds and polysaccharides. Macronutrients were similar between the two studied clones in stem and leaves. Moisture values of approximately 8.5 %, a protein content of 20.44 % and small amounts of soluble sugars and starch, overlapping the fiber content that approaches 25 %, containing a low concentration of pectins but high levels of lignin, which gives the material potential for saccharification mainly in the trunk. The results also showed that these foods, used in animal feed, represent potential raw materials for the food and bioenergy industry with high added value.

Introduction

Reusing organic waste has gained prominence as a way to reduce waste, reduce environmental impact and increase productivity in agriculture. Efficient management of agro-industrial waste is critical for the industry to become increasingly sustainable (Serpa-Fajardo et al., 2022). Agricultural production must have as a premise the recycling and reuse of generated waste as a way to reduce waste, reduce environmental impact and increase productivity in agriculture, in addition to promoting an activity that is socially fair, economically viable, and environmentally correct (Cardoso & Vieira, 2019). When dealing with harvest residues, Brazil, being the fifth producer of cassava in the world, with 18.1 million tons in 2021 (FAO, 2023). Others biggest producers are Nigeria (63.0 million tons), the Democratic Republic of Congo (45.7 million tons), Thailand (30.1 million tonnes) and Ghana (22.7 million tonnes) (FAO, 2023). This root (*Manihot esculenta* Crantz*)* is one of the main energetic foods for more than 800 million people worldwide, mainly in developing countries (Tappiban et al., 2019). Therefore, cassava production generates a lot of agricultural waste, which can potentially be treated and its by-products used (Silva et al., 2022).

In Brazil, the largest producing regions are the north and northeast, which account for approximately 59 % of the national production. In the northeast region, the states of Ceará and Bahia stand out, having produced 760 and 700 thousand tons of cassava in 2022, respectively (IBGE, 2023).

Cassava varieties with good agronomic traits and good culinary quality have been released or recommended by Embrapa Cassava & Fruits for the northeast region. In this context, the following varieties stand out: Saracura (white pulp), a high-yielding variety recommended for the regions of Nossa Senhora das Dores and Lagarto - SE (Reis et al., 2021). Cassava varieties normally grown and marketed are deficient in micronutrients such as vitamin A, which limit their use as the only food in the diet considering the importance of these micronutrient (Oluranti et al., 2016). Due to cassava's importance in the diet of sub-Saharan

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Africa populations, which often present health problems caused by vitamin A deficiency, the consumption of cassava varieties with higher levels of β-carotene may be a sustainable way of preventing this deficiency (Oluranti et al., 2016; Ceballos et al., 2012). In this sense, the hybrid (2009 12–20) were generated by the breeding program by Embrapa Cassava & Fruits with higher levels of pro-vitamin A than commercial varieties of white pulp.

Cassava is originally from South America and is highly prevalent in Brazil, where the tuberous roots are highly consumed due to the large content of starch. At the same time, the leaves and stems are considered a by-product, are little used, and are often left as a residue in the soil. Serpa-Fajardo et al. (2022) show cassava residue is prevenient only on the bagasse generated from starch production, the leaves, stalks, and other raw materials are usually excluded. In African countries, the cassava plant is considered complete because it has a high concentration of carbohydrates in the roots, and the leaves are protein-rich (Achidi et al., 2005). Cassava leaves are a potential alternative source of protein for humans and animals (Pereira et al., 2018; Macedo, 2016; Oresegun et al., 2016; Latif & Müller, 2015). The leaf can be used in diets and as a complementary food source, being an important ingredient in the formulation of the multi mixture as an effort to combat nutrition (Câmara & Madrugada, 2001), and are also used as a source of livestock feed for animals (Lukuyu et al., 2014). Using waste as bulky economic viability is a way of reducing animal production costs. Moreover, cassava leaves are rich in starch and lignin Zhu et al. (2015), thus there is no perspective to replace the roots as food, being soil as an incorporated form of nutrient complements and fertilizer for the plants. If irregularly discarded or burned, it can result in environmental problems (Howeler, 2012). The nutritional quality of these residues depends on several factors, such as soil, plant age, variety, and others. Another factor is the proportion between leaves and stems, which is also an important nutritional factor since the greater proportion of leaves in the plant improves the nutritional quality of the product (De Almeida & Ferreira Filho, 2005). For example, the stem biomass of a plant at harvest age (from 12 months after planting) can reach 50 % of the root mass (Wei et al., 2015). De Souza et al. (2017) emphasize the importance of genetic studies to improve cassava quality, especially photosynthetic efficiency for better biomass accumulation, presenting results of photosynthetic improvement of 14 %, which improves the accumulation of organic matter in the leaves. The leaves, depending on the varieties, are rich in minerals, proteins, vitamins, carotenoids, and fibers. However, they also have some anti-nutritional compounds and toxic substances, including cyanogenic glycosides. These substances interfere with the digestibility and absorption of nutrients and may have toxic effects depending on the amount consumed (Wobeto et al., 2006). However, in the study by Oresegun et al. (2016) with 6 cassava varieties, most had low levels for most anti-nutrients and, consequently, the highest content for most nutritional value. Cassava leaves have two cyanogenic glycosides, linamarin (93–95 % of the total glycocyanide content) and lotaustralin (5 to 7 %), which can generate cyanide. Cyanogenic glycosides are present in all cassava plant tissues, and several factors affect the content of these compounds, such as variety, soil nitrogen content, climate, and plant age (Hidayat et al., 2002; Nambisan, 2011; Montagnac et al., 2009b). Processing aims to reduce the levels of cyanogenic compounds in cassava and some methods are better than others, so the final concentrations depend on the level of these substances before processing and the process carried out (FAO/WHO, 2012; Nambisan, 1994). When the cell structure of any part of the plant breaks down (by crushing, maceration, crushing, grinding), the enzyme linamarase, which is found in the cell wall, catalyzes the hydrolysis of cyanogenic glycosides (released from vacuoles), forming glucose and cyanohydrins correspondents. The cyanohydrins then decompose spontaneously at a pH greater than 4 or through the action of the α-hydroxynitrile lyase enzyme, releasing cyanide (Montagnac et al., 2009b; FAO/WHO, 2012). The cyanogenic glycosides, the corresponding cyanohydrins, and cyanide are soluble in water, and hydrocyanic acid is volatile at 25.6 °C (Sheikh et al, 2021). The lack of knowledge of the nutritional characteristics of the producers and the importance of their use in animal and human food has contributed to the low use of these residues worldwide. Therefore, studies that characterize the leaves and stems in nature of this culture are needed to contextualize its use in supplementary feeding and indicate applications in the food, cosmetic, pharmaceutical, and biofuel industries (Wei, 2015) as well as suggest alternatives for sustainable use of waste generated by the cassava crop and its by-products. Therefore, this work aimed to characterize the proximate composition and the polysaccharide profile of residues (stem and leaves) of cassava harvest to verify possible future applications for these residues.

Material and methods

Plant material

Residues from the production of cassava, stem, and leaf from a commercial variety (Saracura, white pulp) and a hybrid (2009, 12–20, yelow pulp) generated by the breeding program (classical genetic breeding/grafting) by Embrapa Cassava & Fruits were analyzed. The commercial variety and the hybrid were grown in the experimental field of Embrapa Cassava & Fruits, in Cruz das Almas, Bahia, Brazil (12° 67′ S and 39°15′ W, 199 m asl), from July 2016 to August 2018. Samples of the cassava stem and leaf (the upper third of five plants) were harvested, dried in a forced air oven at 65 °C, and processed in an ultra-centrifugal mill for testing and material analysis.

Major components

Moisture, ash, lipids, protein, and total dietary fiber were measured using the methods described in AOAC (2006), Horwitz and Latimer (2007). Moisture determination of the samples was performed by the oven-drying method at 105 °C until constant weight (AOAC 925.45b). The ashes were determined by incineration at 550 °C (AOAC 923.03). Lipid content was extracted with ethyl ether using a Soxhlet extractor (AOAC 920.39). Protein content was estimated by total nitrogen using the micro-Kjeldahl technique, and the crude protein content was calculated by multiplying the total nitrogen content by the conversion factor 6.25 (AOAC 960.52). The total dietary fiber content (TDF) was determined from the dry and defatted sample according to the AOAC 991.43 method with modifications proposed by McCleary and Rossiter (2004) to avoid overlapping the resistant starch content (Horwitz & Latimer, 2007; McCleary & Rossiter, 2004).

Cyanogenic compounds

The determination of cyanogenic compounds (free cyanide, α-hydroxynitrile, and cyanogenic glycosides), were determined as proposed by Essers (1994). In 5 g of sample were added 40 mL of extraction medium. The cyanogenic compounds were extracted with 0.1 M phosphoric acid containing, 25 % (v/v) ethanol, and 2.5 % (m/v) sodium chloride. For analysis, in a test tube, 0.1 mL of the extract, 0.4 mL of 0.1 M phosphate buffer solution pH 7, and 0.1 mL of linamarase enzyme solution (3 U mL^{-1}) were heated in a water bath for 15 min at 30 °C. Incubation was followed by adding 0.6 mL sodium hydroxide 0.2 M for 5 min, 2.8 ml of 0.1 M phosphate buffer solution pH 6 and a subsequent reaction with 0.1 mL chloramine-T 1 % (w/v) and 0.6 mL isonicotinate 2.85 % (w/v)/1,3-dimethyl barbiturate 3.5 % (w/v) pH 7.5. subsequent reaction with chloramine-T and Isonicotinate/1,3-dimethyl barbiturate. The final reaction was read at 605 nm. Linamarase, used to hydrolyze cyanogenic glycosides, was extracted from the cassava's inner bark, according to Cooke (1978). The calibration curve was constructed from six points, with concentrations ranging from 0.05 to 0.30 μg HCN mL^{-1}.

Soluble carbohydrate extraction and analysis

Ten mg of pulverized biomass was extracted four times with 2 mL of ethanol 80 % (v/v) for 20 min each. The supernatants were recovered by centrifugation and vacuum concentrated (ThermoScientific® Savant SC 250 EXP) and resuspended in 1 mL of water. The recovered soluble sugars (glucose, fructose, and sucrose) were analyzed by High-Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD) in a Dionex® system (ICS 5000) using a CarboPac PA1 column and eluted with 150 μM sodium hydroxide in an isocratic run of 27 min. The Alcohol Insoluble Residue (AIR) was dried at 45 °C overnight for further analysis (Arenque et al., 2014).

Starch extraction and determination

The starch from AIR (alcohool insoluble residue) samples was digested with 120 U/mL of α-amylase (E.C. 3.2.1.1) of *Bacillus licheniformis* (Megazyme® Inc., Australia) diluted in 10 mM MOPS buffer pH 6.5 at 75 °C for 1 h and 30 U/mL of amyloglucosidase (E.C. 3.2.1.3) of *Aspergillus niger* (Megazyme® Inc., Australia) diluted in 100 mM sodium acetate pH 4.5 at 50 °C for 1 h. The supernatants containing the glucose from starch were quantified, and the residues were washed three times with 80 % ethanol and dried at 45 °C overnight for monosaccharides analysis. First, five μL of supernatant was diluted with 45 μL of deionized water, followed by the addition of 250 μL of a mixture containing glucose oxidase (1100 U/mL), peroxidase (700 U/mL), 4-aminoantipirin (290 μmol/L) and 50 mM of phenol at pH 7.5, were incubated at 30 °C for 15 min. and the absorbance was measured at 490 nm. The calibration curve was performed with commercial glucose (Sigma®) in the 0.02–0.2 mg/mL concentration range.

Monosaccharide analysis

The cell wall monosaccharide composition was determined by the de-starched AIR hydrolysis. The hydrolysis of pectin and hemicelluloses (non-celullolytic monosaccharides) was performed with two mg of de-starched AIR and 1 mL of 2 M trifluoroacetic acid (TFA), heated at 100 °C for 1 h, vacuum dried, resuspended in 1 mL of deionized water, and filtered through a 0.22 μm membrane filter. The monosaccharides were quantified by high-performance anion-exchange chromatography with pulsed amperometric detection (HPAEC-PAD) on a CarboPac SA10 column (ICS 5000 system, Dionex-Thermo®) eluted with 99.2 % of water and 0.8 % (v/v) sodium hydroxide (1 mL min^{-1}) and a post-column containing 500 mM sodium hydroxide (0.5 mL min^{-1}).

Lignin quantification

The biomass (30 mg) was treated for 15 min with 1 mL of water, ethanol, ethanol-chloroform (1:1 v/v), and acetone and incubated at 98 °C, 76 °C, 59 °C, and 54 °C, respectively with stirring of 750 rpm (Van Acker et. al., 2013). The precipitate was recovered by centrifugation for 5 min at 14,000 g and dried at 50 °C. The lignin content was determined using the acetyl bromide method (Fukushima & Kerley, 2011). To 10 mg of each treated dry sample was added 250 μL of 25 % acetyl bromide in acetic acid, heated for 2 h at 50 °C and 1 h with stirring of 1500 rpm, and cooled in an ice bath. Aliquots of 100 μL of acetyl bromide reaction were reacted with 400 μL of 2 M sodium hydroxide, 75 μL of 0.5 M hydroxylamine hydrochloride, and 1425 μL glacial acetic acid. The absorbance was read at 280 nm, and the lignin was stipulated by

Bouguer-Lambert-Beer law (Eq. (1)) and corrected by the cell wall amount used in the assay.

$$A = \varepsilon * c * l \quad (1)$$

where: A = absorbance; $\varepsilon = 23.35$; $c = g^{-1}\ cm^{-1}$; and $l = 0.1$ cm (Chang et al., 2008)

Table 1
Centesimal composition of leaf and stem cassava, hybrid 2009 12–20 and Saracura.

Variable	Plant part	Hybrid 2009 12–20	Saracura	*p value**
Moisture (%)	Leaf	8.60 ± 0.15	7.81 ± 0.28	***0.002***
	Stem	6.57 ± 0.31	5.20 ± 0.08	***0.000***
	p value	***0.000***	***0.000***	
Ashes (%)	Leaf	7.37 ± 0.04	7.77 ± 0.07	***0.000***
	Stem	5.33 ± 0.03	5.79 ± 0.01	***0.000***
	p value	***0.000***	***0.000***	
Lipids (%)	Leaf	7.35 ± 0.34	6.12 ± 0.38	***0.001***
	Stem	1.66 ± 0.05	1.30 ± 0.06	***0.000***
	p value	***0.000***	***0.000***	
Protein (%)	Leaf	20.24 ± 0.61	20.63 ± 1.71	*0.624*
	Stem	4.47 ± 0.30	4.75 ± 0.34	*0.185*
	p value	***0.000***	***0.000***	
TDF (%)	Leaf	21.77 ± 0.35	21.04 ± 0.85	*0.719*
	Stem	29.11 ± 0.37	26.81 ± 0.31	*0.315*
	p value	***0.023***	***0.000***	
Cyanogenic compounds (μg de HCN/g)	Leaf	112.15 ± 0.22	19.57 ± 0.11	***0.000***
	Stem	ND	ND	–

Data represent mean ± standard error in g/ 100 g dry mass *Statistically significant p-values are presented in italics according to the t-Student test ($p < 0.05$) ($n = 4$), significant differences between organs and varieties are indicated in bold italics. ND- not detected. TDF is total dietary fiber

Saccharification assay

The saccharification was performed as described by Gomez et al. (2010) with 10 mg of cassava de-starched AIR. First, the samples were pre-treated with 500 μL of 0.5 N NaOH at 90 °C for 30 min and washed three times with 1 mL sodium acetate (pH 4.5). The remaining particulate content was digested with 1.6 mL of Cellic Ctec2 (Novozymes®) enzymatic cocktail diluted 1:1000 in 25 mM sodium acetate buffer (pH 4.3) and heated at 50 °C for 16 h. The released sugars were quantified with 300 μL of the enzymatic digestion supernatants, 100 μL of 1 M NaOH, and 200 μL of MBTH reagent (3 mg L^{-1} MBTH and 1 mg L^{-1} DTT). After incubation of 20 min at 70 °C, the samples were added 400 μL of the oxidizing reagent (0.2 % $FeNH_4(SO_4)_2$, 0.2 % sulfamic acid, and 0.1 % HCl) for color development. The absorbance was read at 620 nm, and a standard curve with glucose concentrations of 50, 100, and 150 nmol mL^{-1} was used for measurements.

Data analysis

Statistical analysis of the evaluated sugars was performed for homogeneity analysis and subsequent applied Test t-Student (JMP® Statistical Discovery Software, version 5.0.1, SAS Inc., Cary, NC, USA) with significance $p \leq 0.05$ to compare cassava cultivars and organs (stem and leaves). All the analyses were conducted with four replicates ($n = 4$). When relevant, the data were analyzed by ANOVA one-way ($p \leq 0.05$) with a post-hoc by Tukey's test ($p \leq 0.05$) at R version 3.6.1.

Results and discussion

The results obtained in this work present chemical compositions and saccharification potential, which are important data for possible applications of residues, such as leaves and stems from leftover cassava

Table 2
Non-structural carbohydrates of leaf and stem cassava of hybrid 2009 12–20 and Saracura.

Variable	Plant part	Hybrid 2009 12–20	Saracura	*p* value*
Starch (μg/ g)	Leaf	8.10 ± 1.74	10.22 ± 3.65	*0.106*
	Stem	75.21 ± 7.42	86.44 ± 8.88	***0.000***
	p value	***0.000***	***0.000***	
Glucose (μg/ g)	Leaf	12.30 ± 2.30	10.80 ± 1.60	*0.082*
	Stem	14.90 ± 0.70	27.80 ± 3.00	***0.001***
	p value	***0.002***	***0.000***	
Fructose (μg/ g)	Leaf	8.20 ± 1.20	9.90 ± 1.80	*0.096*
	Stem	11.00 ± 0.60	21.20 ± 2.40	***0.001***
	p value	***0.001***	***0.001***	
Sucrose (μg/ g)	Leaf	14.40 ± 0.50	11.30 ± 5.90	*0.100*
	Stem	12.50 ± 0.80	22.40 ± 1.70	***0.000***
	p value	***0.023***	***0.004***	

Data represent mean ± standard error (μg/ mg dry mass).*Statistically significant *p*-values are presented in italics according to the t-Student test ($p < 0.05$) ($n = 4$), significant differences between organs and varieties are indicated in bold italics.

harvest. The Table 1 presents the results of the two studied clones' composition of leaves and stems. Differences are statistically evidenced between leaves and stems, differing among themselves and among clones. Moisture analysis was performed on the dehydrated samples, resulting in values of 7.81 and 8.60 % for leaves of the Saracura and hybrid 2009,12–20 varieties, respectively. Other works that analyzed the dry matter obtained a similar value in the dry cassava leaf, with a moisture of 7.15 % in the Branca de Santa Catarina variety (Ortega--Flores, 2003) and of 7.70 and 8.32 % in seven different clones. Fresh cassava leaves have 79–90 % moisture (Oni et al., 2011). It is worth mentioning that for agroindustry one of the biggest challenges is to remove moisture from large volumes of cassava residue, which ends up fermenting in an uncontrolled way and producing unpleasant odors. This waste has become an issue of environmental pollution (Grasso, 2020).

The ash values found in the dry leaves of the studied clones (7.37 and 7.77 %) were higher than in the varieties studied by Achidi et al. (2008), which on average were 6.5 % in the study mentioned above. The ash content in the stems was similar to values described in the literature for other varieties (2.7–5.5 %) (Martín et al., 2017). The lipid content observed in the leaves of the hybrid 2009 12–20 (7.35 %) and Saracura (6.12 %, corroborated the described by Achidi et al. (2008), which describe an average of 6.3 % in their studies. A significant nutritional factor is the protein content, which is 5 times higher in the leaf compared to the stem. However, among the leaves of the clones, the values are statistically similar (approximately 20.44 % protein), corresponding with the studies by Leguizamón et al. (2021) and Oni et al. (2011)

The composition of total dietary fiber (TDF) in both organs of the clones was estimated at approximately 25 % of the total weight, data consistent with those found in the work of Achidi et al. (2008). Ortega-Flores et al. (2003) found fiber concentration in other varieties of 48 % of the total weight for the same organs. In contrast, Lambebo and Deme (2022) found much lower values for fiber content (4.86–6.63 %). Thus, cassava varieties in terms of leaves and stems have significantly different chemical compositions, which may also be related to the planting location, soil and climate, as is expected in analyzes of the plant's roots. All parts of the cassava plant contain cyanogenic glycosides (linamarin and lotaustralin), compounds that, enzymatically, can release hydrocyanic acid in vivo, a toxic compound (Hidayat et al., 2002; Nambisan, 2011; Nambisan, 2011; Montagnac et al., 2009a).The leaves of the hybrid contain more cyanogenic glycosides than the leaves of the Saracura variety, with a content five times higher (Table 1). In the stems, such compounds were not detected due to their low concentration (values below 1 μg of HCN per g of the stem). Quantification was performed on dehydrated samples.

Table 3
Non-celullolytic monosaccharides (μg/ mg dry mass) and lignin (% related Cell Wall) content from the leaf and stem of the cell wall of two cassava clones.

Variable	Plant part	Hybrid 2009 12–20	Saracura	*p* value*
Fucose	Leaf	2.52 ± 0.21	2.36 ± 0.20	*0.623*
	Stem	1.40 ± 0.07	1.27 ± 0.44	*0.857*
	p value	***0.005***	***0.049***	
Arabinose	Leaf	25.85 ± 1.63	25.32 ± 2.18	*0.928*
	Stem	8.34 ± 0.17	8.94 ± 3.27	*0.946*
	p value	***0.001***	***0.008***	
Galactose	Leaf	24.43 ± 2.20	23.40 ± 1.94	*0.787*
	Stem	14.48 ± 0.58	13.68 ± 4.97	*0.979*
	p value	***0.011***	*0.076*	
Rhamnose	Leaf	3.94 ± 0.33	3.76 ± 0.19	*0.631*
	Stem	2.80 ± 0.38	2.48 ± 0.87	*0.924*
	p value	***0.003***	*0.158*	
Glucose	Leaf	8.34 ± 0.67	15.03 ± 1.55	***0.012***
	Stem	7.96 ± 0.57	5.32 ± 1.64	*0.238*
	p value	*0.329*	***0.005***	
Xylose	Leaf	14.86 ± 1.16	12.28 ± 1.00	*0.157*
	Stem	67.43 ± 2.66	47.41 ± 13.61	*0.247*
	p value	***0.000***	*0.027*	
Mannose	Leaf	*5.16 ± 0.29*	*5.85 ± 0.47*	*0.147*
	Stem	*7.16 ± 0.58*	*4.41 ± 1.53*	*0.129*
	p value	***0.037***	*0.328*	
Lignin	Leaf	6.37 ± 0.77	5.86 ± 0.61	*0.537*
	Stem	13.23 ± 1.12	12.25±1.66	*0.648*
	p value	***0.000***	***0.007***	

Data represents mean ± standard error. *Statistically significant p-values are presented in italics according to the t-Student test ($p < 0.05$) ($n = 4$), significant differences between organs and varieties are indicated in bold italics.

The data found in our study (Table 2) differ from values described in other varieties, as observed by Achidi et al. (2008) report between 20 % of starch in leaves, and Martín et al. (2017) found variations from 18.5 % to 42.4 % of starch in stems. Although the starch concentration, in our study for both varieties, is lower than 8 % in stem and around 1% in leaf (Table 2). The concentration of soluble sugars and starch analyzed showed different concentrations in different organs of the same variety, which does not occur when comparing leaves of different varieties. Among the soluble sugars evaluated, the highest concentration is found in the Saracura stem (> 20 μg/g), corroborating what was described by Martín et al. (2017). It is important to highlight that the starch concentration was carried out using the enzymatic methodology (Amaral et al., 2007), and it cannot be compared with the data from the other methodologies.

The composition of non-celullolityc monosaccharides from cell wall is similar between the organs of the hybrid and Saracura, except for the glucose content in the leaves (Table 3). The leaves showed a higher concentration of arabinose (25 μg/ mg dry mass) and galactose (24 μg/ mg dry mass) in both clones, followed by xylose (13.6 μg/ mg dry mass). The lower concentration of monosaccharides in leaves and stems is fucose (2.4 μg/ mg dry mass). In stems, there is a higher proportion of xylose (67.4 and 47.4 μg/ mg dry mass), followed by galactose (14.5 and 13.7 μg/ mg dry mass). The results suggest a low content of pectins (rhamnose and fucose) and, among the hemicelluloses, it is possible to indicate a low content of mannose and a high content, mainly of xyloglucans (xylose and glucose), and arabinoxylan (arabinose and xylose) (Table 3). The phenolic compound found in the cell wall (lignin), which is responsible for the recalcitrance of the wall and plant support, was found in greater proportions in the stem (13.23 % and 12.25 %) compared to the leaves (6.37 % and 5.86 %) in the hybrid and Saracura (Table 3), corroborating what was described by Leguizamón et al. (2021) and Martín et al. (2017). The results of the concentration of non-celullolytic monosaccharides found in cassava stem and leaf are slightly lower than the values described in products that are currently used in animal feed, namely: soybean meal, ground corn, and star grass (Dos Reis et al., 2015; Viroli et al., 2022), foods that are produced directly for animal nutrition.

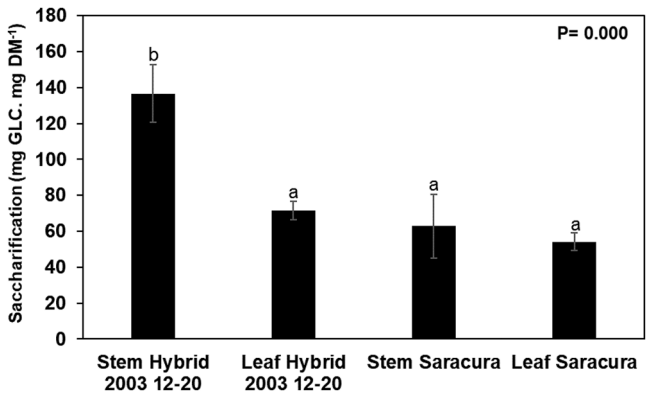

Fig. 1. The saccharification from the leaf and stem of the cell wall of two cassava clones. Data represent mean ± standard error. Different letters are statistically significant according to ANOVA one-way followed by Tukey's test ($p < 0.05$) ($n = 4$).

The saccharification capacity was higher in the stem for hybrid and Saracura (136.6 mg GLC.mg DW-1 and 62.9 mg GLC.mg DW-1) when comparing the plant parts (71.4 mg GLC.mg DW-1 and 54.0 mg GLC.mg DW-1) (Fig. 1). The saccharification capacity of the leaves was statistically distinct between hybrid and Sacarura (p-value 0.007 for t-test), being more elevated to a hybrid than for Saracura by 1.3 times. Comparing the saccharification capacity of hybrid it a distinction was found among the plant organs tested (Fig. 1). As presented by Sivamani et al. (2018), the potential of cassava residues as a raw material for the production of biofuels and the optimization of pre-treatment can yield up to 70 % in saccharification in cassava stem.

Conclusion

The characterization of the cassava leaves and stems of the Saracura variety and the 12–20 hybrid from 2009 revealed that although the residues are not used for human consumption, the by-products do have potential for use. They are by-products with a composition rich in starch, total soluble sugars, as the consumption of starch increases satiety, providing the characteristic of resistance to the degradation of the long chain of carbohydrates, making it possible to use them to increase animal feed. Another result to be highlighted is the high rate of saccharification, which is a residue with high saccharification performance and can be used for the production of biofuel. However, they are considered waste and have the potential for sustainable use.

CRediT authorship contribution statement

Isabela Simões Soares: Writing – original draft, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Fabiana Perrechil:** Writing – original draft, Methodology, Formal analysis. **Adriana Grandis:** Writing – original draft, Methodology, Formal analysis, Conceptualization. **Débora Pagliuso:** Writing – original draft, Methodology, Formal analysis, Conceptualization. **Eduardo Purgatto:** Writing – original draft, Methodology, Data curation. **Luciana Alves de Oliveira:** . **Aline Andreia Cavalari:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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