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Nutritional and Functional Characteristics of Whole-grain Cookies with Added Gurguéia Nut (*Dipteryx Lacunifera* Ducke) and Soursop Residue (*Annona Muricata* L.)

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ABSTRACT

The objective was to determine the nutritional and functional characteristics of cookies with the addition of Gurguéia nut and soursop residue. Three whole-grain cookie formulations were developed from a standard (SC). Whole-grain cookies 2 (WC2) had greater acceptance and preference. Proximate composition, microbiological, and bioactive compound analyses were performed for WC2 and SC. WC2 exhibited higher levels of proteins, lipids, ash, total phenolics, condensed tannins, vitamin C, and antioxidant activity, with lower carbohydrate content and equivalent energy. In the microbiological analysis, the cookies were shown to be adequate for consumption. Cookies produced with regional raw materials proved to be a healthier option.

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Cerrado fruits; soursop residue; gurguéia nut; sensory analysis; whole-grain cookies

Introduction

The demand for products with easier and faster preparation is increasing, making the pulp market in Brazil promising. However, this process generates waste, and there are concerns about its form of disposal, since most of them are potential polluters presenting high organic, showing high organic value (Martins, Barros, Silva, Silva, & V, 2019; Souza et al., 2018).

Soursop (*Annona muricata* L.), belonging to the family Annonaceae, is an important fruit cultivated in northeast Brazil, mainly in Paraíba, Ceará, Pernambuco, and Bahia, and is considered one of the most commercially accepted tropical Brazilian fruits (Silva, 2019). Its residues can be used as a source of nutrients, mainly the phenolic compounds, since this fruit has numerous properties such as antioxidant activity (Silva, Guimarães, Costa, Cruz, & Meireles, 2019). Several compound classes have been isolated from soursop through chemical studies, such as acetogenins, alkaloids, terpenoids,

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carbohydrates, polyphenols, lipids, and amino acids, with some of these substances being associated with the sequestration of free radicals formed in degenerative processes (Coria-Téllez, Montalvo-Gonzalez, Yahia, & Obledo-Vázquez, 2018).

Gurguéia nuts (*Dipteryx lacunifera* Ducke) are also known as bat bean, donkey nut, and garampara. It is a plant of the family Leguminosae, native to the mid-north region of Brazil, mainly the southern and central-south Cerrado in Piauí and Maranhão (Cavalcante et al., 2015). They are rich in lipids, proteins, and ashes (Araújo et al., 2020), and present excellent properties for the nut market. Therefore, there is a possibility of using its flour to prepare and improve the nutritional value of cookies.

A cookie is defined as a product, fermented or unfermented, obtained by kneading and baking different types of starch or flour, mixed with other ingredients. It can also have different frosting, filling, shape, and texture (Brasil, Ministério da Saúde. Agência Nacional de Vigilância Sanitária, 2005).

Being a product that can have ingredients added or substituted, cookies are being increasingly consumed (Costa et al., 2019), since it can supply nutrients, fibers, and antioxidant compounds to people of any age. Thus, the objective of this study was to develop and analyze the nutritional and functional value and the microbiological quality of standard and whole-grain cookies incorporated with gurguéia nut and soursop residue flour.

Material and methods

Acquisition of raw materials

Gurguéia nut was supplied by Vão dos Negros farm, in the municipality of Landri Sales, Piauí, Brazil. They were stored at a temperature of −18°C until the flour was prepared. Soursop residue (bagasse) was supplied by Fruta Polpa company, Teresina, Piauí, Brazil.

The sanitized residues were obtained in two 5 kg polyethylene bags. The seeds adhered to the residues were manually removed, packed in 1 kg polyethylene bags and stored at −18°C until the flour was prepared. Free-range chicken eggs were purchased at São Carlos farm, located in the municipality of Teresina, and stored at 10°C until cookie preparation. The other raw materials were purchased from a retail store.

Flour production

To obtain the flour, soursop residue was dried in a ventilated oven 314D242 model, Quimis, São Paulo, Brazil for 3 hours at 60°C and ground in blender Problend 4 model, wallita, São Paulo, Brazil until desired granulometry, followed by sieving. For the production of gurguéia flour, the nuts were thawed for 24 hours under refrigeration, ground in a blender for 10 minutes, and then sieved in a sieve 0,5 mesh. The whole-grain cookies were prepared using the standard cookie (SC) formulation and three variations of gurguéia flour content were tested.

Obtaining the cookies

The cookies were prepared using a standard formulation adapted (Santos, Storck, & Fogaça, 2014) Three formulations WC1, WC2, and WC3 were developed with mixtures of refined and whole-wheat flour ranging from 10% to 40% and 25% to 65% respectively, soursop residue from 5.0% to 25% and walnut flour from 5.0% to 25% gurguéia with partial replacement of refined wheat flour, keeping the other ingredients constant crystal sugar 15% to 45%, honey 5% to 25%, free-range chicken egg 5% to 25%, butter 10% to 35%, and 1% to 10%. The raw materials used in the preparation of integral cookies 2 (WC2) and standard cookie (SC) are shown in Table 1. [Table 1 near here].

The raw materials were weighed, mixed by hand, and homogenized. The dough was rolled out with a roller and molded using a star-shaped cutter. The raw cookies were distributed in rectangular baking pans greased with butter and refined wheat flour, and baked at 200°C for 15–20 minutes in a stove oven Atlas, Monaco model, Paraná, Brazil. After baking and cooling the cookies to room temperature, they were packed and stored in hermetically sealed glass containers until the time of analysis. [Figure 1 near here]

Sensory analysis

The sensory analysis was performed with 116 untrained participants of both sexes 18–50 years old, who signed an Informed Consent Form, according to current legislation. Sensory analysis tests were performed, as described by

Table 1. Percentages of raw materials used to make the standard cookie (SC) and cookie with added soursop and gurguéia flour (WC2).

	Types of cookies	
	SC	WC2
Raw materials	Quantity %	Quantity %
Gurguéia nut flour	-	5.0–25.0
Soursop residue flour	-	5.0–25.0
Whole wheat flour	-	25.0–65.0
Refined wheat flour	35.50	10.0–40.0
Crystal sugar	39.74	15.0–45.0
Honey	-	5.0–25.0
Free-range chicken egg	9.10	5.0–25.0
Butter	14.95	10.0–35.0
Chemical yeast	0.71	1.0–10.0

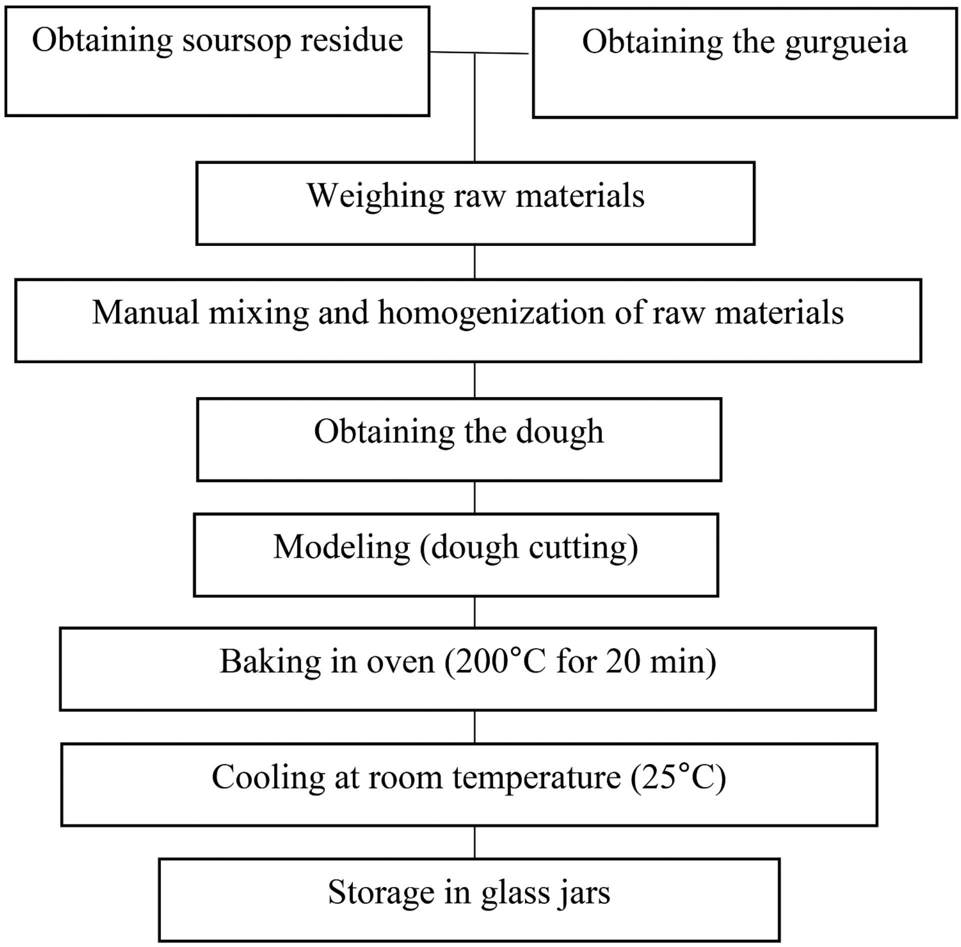

Figure 1. Flowchart of the process to obtain cookie with added soursop residue and gurgueia (WC2) nut flours.

(Dutcosky, 2013). The cookie samples were presented in a monadic way as complete balanced blocks coded with three-digit random numbers in two sessions with four repetitions.

To verify acceptance, a nine-point hedonic scale was used 1 = really disliked; 5 = indifferent; 9 = really liked. A scale test of five points was used to determine the intention to purchase the most widely accepted product, with the terms: 1 – would certainly not buy to 5 – would certainly buy it. After the hedonic scale test, the cookie with the highest acceptance was evaluated using Quantitative Descriptive Analysis (QDA) by a team composed of five trained participants of the Product Development and Sensory Analysis Laboratory – LASA/UFPI.

From the results of the sensory analysis, the other analyses were performed only on the standard cookies (SC) and on the biscuit that obtained the highest percentage of acceptance (WC2).

Microbiological analyses

Microbiological analyses were performed to evaluate product quality at the Study, Research, and Food Processing Center (NUEPPA) of the UFPI Agricultural Sciences Center. Total and tolerant coliforms and coagulase-positive *Staphylococci* and *Salmonella spp* were determined. The analyses were obtained according to the methodology described in the manual of microbiological food analysis methods of (Silva, Junqueira, & Silveira, 2007), and the results were analyzed according to the current legislation on Microbiological Standards for Food (Brasil, Ministério da Saúde. Agência Nacional de Vigilância Sanitária, 2001).

Centesimal composition and total energy value (TEV)

The experiments were conducted at the Product Development Laboratory (LASA) and the analyses at the Bromatology and Biochemistry Laboratory (LABROM) of the Nutrition Department of the Federal University of Piauí, Brazil. Centesimal analysis was performed in triplicate, and the parameters of humidity, ashes, proteins, lipids, and carbohydrates, which were determined using the methods described in the analytical standards of the AOAC, 2005.

TEV was estimated according to the Atwater conversion values, which are based on macronutrient content (proteins, lipids, and carbohydrates) and multiplied by factors 4, 9, and 4 in kcal. g^{-1}, respectively, to obtain TEV (Watt & Merrill, 1963).

Extract preparation

Cookie extracts were prepared to analyze total phenolic, total flavonoid, and condensed tannin levels according to the adapted methodology by (Rufino et al., 2010). Antioxidant compounds were extracted with a solvent mixture containing methanol (50%), acetone (70%), and distilled water in a 2:2:1 ratio. One gram of cookie was mixed with 10 mL of methanol (50%) in a falcon centrifuge tube and placed in an ultrasonic (UltraCleaner 1600A, Indaiatuba, São Paulo) for 30 min at 24°C Next, it was transferred to Centrifuge 5702, Hamburg, Germany, at 1,207 x *g* for 15 min. Subsequently, the supernatant was collected, acetone 70% was added, and the ultrasound and centrifuge processes were repeated. The supernatant was again collected, added with 5 mL of distilled water, filtered, and stored at −6°C in the absence of light.

Determination of bioactive compounds

Phenolic compound content was determined by the spectrophotometric method using the Folin-Ciocalteau reagent (Singleton & Ross, 1965). The standard curve was obtained with gallic acid, and the results were expressed in mg of gallic acid per 100 g of sample. Flavonoids were determined by using spectrophotometry according to the procedure of (Kim, Jeong, & Lee, 2003) and modified by (Blasa et al., 2006). Total condensed tannin content was obtained by the spectrophotometric method of (Price, Scoyoc, & Butler, 1978) with the vanillin reagent. Vitamina C determined by the Tillmans method, modified by (Benassi & Antunes, 1988), which is based on the reduction of the 2,6-dichlorophenolindophenol sodium salt dye by an acidic solution of vitamin C.

Antioxidant activity was performed by the DPPH (2,2-diphenyl-1-picrylhydrazyl) free radical capture method using spectrophotometry, and the results were expressed in Trolox μmol. g^{-1}.

Statistical analysis

A database was created in the *Statistical Package for the Social Sciences*, version 21.0 (2016). Tukey's and Student's t-test were used to check differences between means, with a 5% significance level $p \leq .05$ and 95% confidence interval, respectively.

Ethical aspects

The research is part of the project entitled "Elaboration of products using regional raw materials," approved by the Research Ethics Committee of the Federal University of Piauí, under opinion No. 750,942. The study participants voluntarily signed an Informed Consent Form, according to Resolution No. 466 of the National Health Council (Conselho Nacional de Saúde, Brasil. 2012). This study is also part of a PhD dissertation for the Graduate Program in Food and Nutrition-PPGAN/UFPI.

Results

Table 2 shows the notes obtained by the hedonic scale test of SC and WC2 cookies [Table 2 near here].

The mean scores in the acceptance test are presented in Table 3, indicating that there was a significant difference $p \leq .05$ between SC and WC2, showing that WC2 acceptance was higher. [Table 3 near here]

As for the intent to buy, most participants stated that they would buy WC2 83.2% and SC 70.8%, and the Kruskal-Wallis$\chi 2$ (chi-square) test showed a significant difference $p = .032$ between SC and WC2.

Table 2. Acceptance of the standard cookie (SC) and the whole-grain cookie with added soursop residue and gurgueia nut flours (WC2).

Grade	Concept	SC	WC2
1	I really disliked it	4	0
2	I disliked it a lot	1	0
3	I moderately disliked it	2	1
4	I slightly disliked it	6	4
5	I did not like it; I did not dislike it	3	3
6	I slightly liked it	14	13
7	I moderately liked it	17	30
8	I liked it a lot	36	43
9	I really liked it	33	22
TOTAL		116	116

Table 3. Mean grades given in the acceptance test for the standard cookie (SC) and the cookie with added soursop residue and gurgueia nut flours (WC2). Teresina, Piauí, Brazil, 2020.

Formulation	Mean grades
SC	7,23[a]
WC2	7,45[b]

[a,b]Different lowercase letters indicate that there is a significant difference between the means by the Tukey test at the level of 5% ($p \leq 0,05$) with 95% CI.

In QDA, the advisors characterized WC2 formulation as having the whole-grain appearance, light brown toasted color, nutty and soursop flavor, nutty aroma, and crunchy texture. Nut and residue addition improved the cookie crunchiness. A spider graph was obtained with the mean values of the WC2 characteristics. The zero point of the scale is represented in the center of the graph, and intensity increases from the center to periphery. The mean value of each characteristic is marked on the corresponding axis [Figure 2 near here].

The microbiological data in Table 4 indicate that the products showed results compatible with those required by resolution RDC No. 12 (Brasil, Ministério da Saúde. Agência Nacional de Vigilância Sanitária, 2001) [Table 4 near here].

The information on the centesimal composition and TEV of SC and WC2 are presented in Table 5. Only TEV showed no significant difference $p \geq .05$ between SC and WC2 [Table 5 near here].

WC2 had a mean moisture content of 5.78% and SC 2.39%, within the standards established by the Brazilian legislation (Brasil, Ministério da Saúde. Agência Nacional de Vigilância Sanitária, 2005), which suggests a maximum value of 14% moisture in cookies. As for the ash content, which represents the

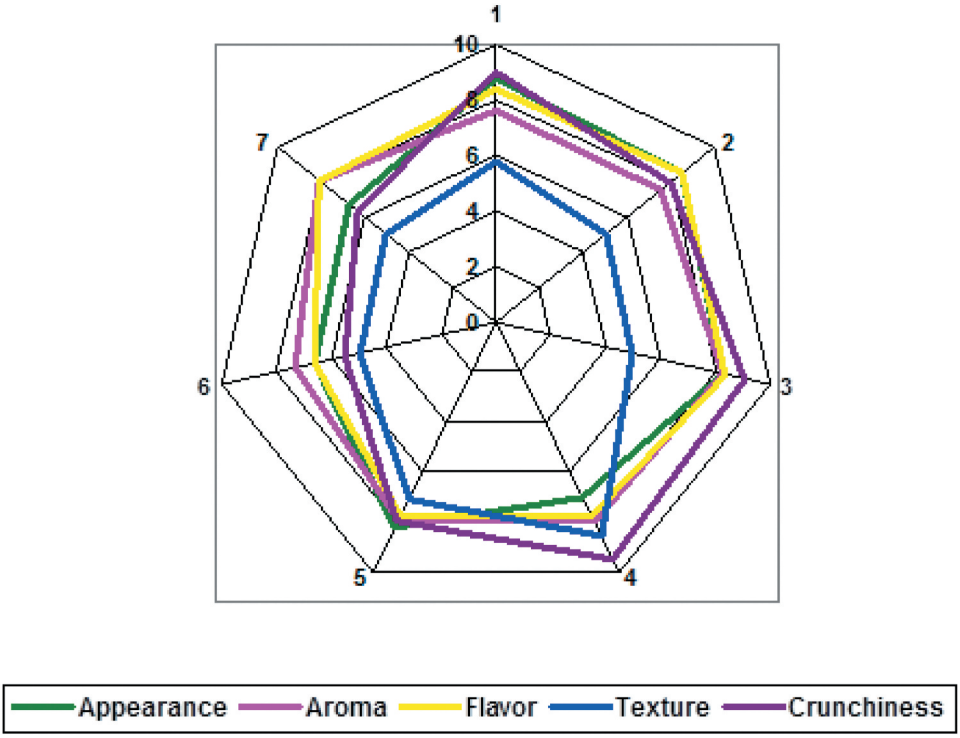

Figure 2. QDA spider graph of the cookie with added soursop residue and gurguéia nut flours (WC2).

Table 4. Microbiological analysis of standard cookie (SC) and the cookie with added soursop residue and gurgueia nut flours (WC2).

Microbiological analysis	SC	WC2	Microbiological Standard
Coliforms at 35°C (*MPN/g)	<3.0	7.4	-
Coliforms at 45°C (MPN/g)	<3.0	<3.0	10 (MPN/g)
Coagulase positive *Staphylococcus*	$<1.0 \times 10^1$	$<1.0 \times 10^1$	5×10^2(CFU/g)
Salmonella spp. (Absent 25 g)	Absent	Absent	Absent in 25 g

*MPN/g: Most Probable Number per gram of analyzed sample.
**CFU/g: Colony Forming Unit per gram of analyzed sample.

Table 5. Centesimal composition and Total Energy Value (TEV) of standard cookie (SC) and the cookie with added soursop residue and gurgueia nut flours (WC2).

Macronutrients/TEV g/100 g/Kcal/Kjoule/100	SC (Mean ±SD)	WC2 (Mean ± SD)
Humidity	2.39 ± 0.01[a]	5.78 ± 0.24[b]
Ashes	1.02 ± 0.01[a]	1.49 ± 0.00[b]
Proteins	3.81 ± 0.09[a]	4.42 ± 0.35[b]
Lipids	13.55 ± 0.30[a]	17.09 ± 0.09[b]
Carbohydrates (by difference)	79.226[a]	71.214[b]
TEV	454.06[a]	456.34[a]

Mean of three repetitions. Similar lowercase letters in a row indicate no significant difference between means by the Tukey's test at 5% level ($p > 0.05$) with CI95%.

Table 6. Bioactive compounds and antioxidant activity of standard cookie (SC) and the cookie with added soursop residue and gurgueia nut flours (WC2).

	Types of cookie	
	SC	WC2
Bioactive compounds	Mean ± SD	Mean ± SD
Total phenolic (mg gallic acid g^{-1})	52.54 ± 0.00[a]	444.57 ± 0.00[b]
Total flavonoids (mg de quercetina.g^{-1})	N.D.	145.67 ± 0.00[b]
Condensed tannins (mg catechin g^{-1})	76.11 ± 0.00[a]	195.66 ± 3.36[b]
Vitamin C (mg vit. C/ g^{-1})	Unrealized	13,98 ± 0,67
Antioxidant activity (µmol Trolox.g^{-1})	690.80 ± 0.00 [a]	1217.35 ± 8.55[b]

Mean of three repetitions. Similar lowercase letters in a row indicate no significant difference between means by one-way ANOVA test: post Hoc multiple comparisons with the Tukey's test at 5% level ($p > 0.05$) with CI95%.

inorganic matter in the product, the value was 1.49% for WC2 and 1.02% for SC. The legislation recommends a maximum of 3% ash; thus, the percentage obtained in this research is within the recommended values.

WC2 had a protein content of 4.42%, which was higher than that of SC 3.81%, and similar to the value reported by Brito (2018) in whole-grain cookies using chichá and gurguéia nut flour 4.04%. As for lipid content, WC2 and SC showed values of 17.09% and 13.55%, respectively. WC2 had a carbohydrate content of 71.21%, which was lower than that of SC 79.23%. For total calories, WC2 and SC provided 456.34 and 454.06 calories, respectively.

Table 6 features the levels of total phenolic compounds, flavonoids, tannins, vitamin C, and antioxidant activity in SC and WC2 [Table 6 near here].

WC2 achieved higher levels of all bioactive compounds and antioxidant activity compared to SC.

Discussion

The cookies developed in this study showed excellent acceptance; WC2 was chosen since it displayed a higher percentage of acceptance and lower rejection grades compared to SC (Table 2). Vasconcelos et al. (2018) also reported high acceptance (86%) of a cookie developed with bacuri flour.

In the result of the test showing the intention to buy these cookies, there was a statistical difference between WC2 and SC. This data was different from the result obtained by Piovesana, Bueno, and Klajn (2013) in the cookies that were prepared by adding oat and grape bagasse flour, where there was no significant difference in the intention to buy test.

Figure 2 illustrates the averages of the attributes obtained. It was found that in WC2 cookies the attribute that got the highest score was the crispness 8.60, followed by the flavor 7.90, aroma 7.80, appearance 7.74 only the texture attribute obtained the lowest note 5.91. In cookies, crispness is an important characteristic for its acceptance because it is synonymous with new and quality product, and its loss determined by softening, is one of the causes of consumption rejection (Guimarães & Silva, 2009). Andrade (2013), in biscuit enriched with 10% of green banana flour obtained a similar result for the flavor attribute 7.92 and equal value for the aroma 7.80.

The microbiological analysis of the cookies showed satisfactory results, proving that they were properly prepared. According to Franco and Landgarf (2013), assessing the types of microorganisms and their quantities in food is essential to verify hygiene conditions in food production, as it may pose a risk to consumer health. It is also important to highlight that the national microbiological standards were met in the present study.

WC2 and SC showed moisture content within the standards established by the Brazilian legislation (Brasil, Ministério da Saúde. Agência Nacional de Vigilância Sanitária, 2005), which advocates maximum moisture of 14% in cookies. Cookies with low moisture content have a better shelf-life. Santiago, Silva, Conceição, and Aquino (2016) obtained higher moisture levels in diet cookies added with passion fruit peel flour, with values ranging from 9.35% to 11.54%. Uchoa et al. (2009) reported that cookies supplemented with cashew powder presented a moisture content of 4.9%, and those containing guava power showed 4.1%, levels, which are lower than those reported in this study.

As for the ash contents, which represents the inorganic matter in the product, the values were also within the aforementioned legislation recommendation, which requires a maximum of 3% of ash (Table 5). Clerici, Oliveira, and Nabeshima (2013), when studying about the partial replacement of wheat flour by sesame flour, obtained a variation of 1.33% to 1.83% and reported that the higher the sesame flour concentration, the higher the ash contents (fixed mineral residue).

As shown in Table 5, the protein content of WC2 pointed to higher levels than the SC, which is similar to that reported by Brito (2018) in whole-grain cookies using chichá and gurguéia nut flour (4.04%), and higher than the mean content (0.17%) obtained by Silva and Silva (2015) in cookies produced with dehydrated mango by-products. A similar result was also reported by Pereira, Oliveira, Almeida, and Feitosa (2016), wherein they obtained mean protein values of 4.36% when evaluating buttered cookies based on jatobá flour.

Regarding the contents of lipids Table 5, WC2 had higher contents than the SC, which could be due to the presence of fatty ω-9 acids (oleic acid) in gurguéia nut oil. According to Costa (2011), gurguéia nut oil consists of

65.59% oleic acid (C18:1 n-9), which is its main monounsaturated fatty acid that decreases total cholesterol and low-density lipoprotein (LDL) without reducing the high-density lipoprotein (HDL) (Lopez, More, & Serra, 2009). However, Vasconcelos et al. (2018) obtained a higher lipid content of 19.86% in their studies with bacuri flour cookies compared to the authors of these research results.

WC2 reached lower carbohydrate content than SC (Table 5). Compared to this study, lower levels of carbohydrates were reported by Vasconcelos et al. (2018) in bacuri cookies (54.67%) and by Melo, Oliveira, Feitosa, Feitosa, and Oliveira (2017) in cookies incorporated with cashew flour and sweetener (49.8%). Thus, the formulated cookie provides a source of carbohydrates for the human body. As for the total calories shown in Table 5, WC2 and SC were found to provide 456.34 and 454.06 calories, respectively. This calorie content was close to the one reported by Vasconcelos et al. (2018) in bacuri cookies (467.22 calories). Leite, Feitosa, and Rocha (2017) developed two formulations of buttered cookies with jatobá flour, where they observed similar values (468.11 and 457.94 calories, respectively) to those reported in this study. This data was also in agreement with Neiva and Moreira-Araújo (2015), who obtained gurguéia nut cookies with 463.73 calories.

WC2 cookie showed higher levels in all bioactive compounds and antioxidant activity compared to the SC. A study by Brito (2018) on whole-grain cookie involving addition of chichá and gurguéia nut flour obtained lower levels of total phenolic compounds (68.92 mg of gallic acid. g^{-1}), flavonoids (2.91 mg of quercetin. g^{-1}), and antioxidant activity (77.25 μmol Trolox. g^{-1}). In cocoa bran cookies developed by Barros (2019), the total phenolic contents, which varied from 149.05 to 264.97 mg EAG 100 g^{-1}, were lower compared to WC2. A study by Fioravante, Hiane, Campos, and Candido (2016) with cookies made from caraguatá, a typical plant of the Brazilian Cerrado, reported lower levels of total phenolics (7.49 mg of galic acid. g^{-1}) and antioxidant activity (53.12 μmol Trolox. g^{-1}) in relation to the study presented.

Lemos, Cavalcanti, Cândido, Guimarães, and Siroma (2019) verified a lower total tannin contents of 0.35 mg of EAT g^{-1} in cookies produced with beer bagasse and baru nuts than those obtained in this study. Aquino et al., (2010) obtained a total vitamin C values of 2172.6 mg. g^{-1} in cookies made with acerola residue flour, values higher than that achieved in this study. While 2018) developed cookies with pequi almonds and reached levels of 8.46 of vitamin C, lower than the value obtained in this research.

The discrepant differences in values obtained from bioactive compounds in the analysis of the same fruits can probably be explained, because of their environmental cultivation characteristics, genetic factors, fruit variety, ripeness stage, as well as production and conservation conditions (Melo & Guerra, 2002).

Phenolic, flavonoid, and vitamin C compounds are sources of natural antioxidants, and their consumption inhibits the formation of free radicals, also called reactive substances, which are associated with a lower incidence of oxidative stress diseases (Singh, Singh, Kaur, & Singh, 2018).

Antioxidants act on different levels of organism protection, such as by acting through the defense mechanism against free radicals and preventing their formation mainly by chain reactions with iron and copper. Antioxidants can capture the free radicals produced by cellular metabolism or by exogenous sources, preventing attacks on lipids, proteins, double bonds in polyunsaturated fatty acids, and DNA bases, and inhibiting lesions and loss of cellular integrity (Verruck, Prudencio, & Silveira, 2018).

Conclusions

The cookies formulated with the addition of the soursop residue and gurguéia flours (WC2) had higher humidity, protein, ash, and lipid contents when compared to that of SC. As for the result from the intention to buy test, high scores were obtained, along with great sensory acceptance, showing that there is a potential market for the product; moreover, there is a possibility for the use of a by-product that is discarded in nature. WC2 was sensorially characterized as a whole-grain looking cookie, with light brown toasted color containing nut pieces, nut and soursop flavor, brown aroma, and crunchy texture.

According to microbiological results, the food produced was fit for consumption based on health quality standards. WC2 showed higher levels of bioactive compounds and antioxidant activity compared to the SC. Thus, the cookie produced can be a new option for obtaining a healthier product, using regional raw materials and by-products of the fruit pulp industry.

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Disclosure statement

No potential conflict of interest was reported by the authors.

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