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Processing, characterization and stability of Brazil nut (*Bertholletia excelsa*) extract

Processamento, caracterização e estabilidade do extrato de castanha do Brasil (*Bertholletia excelsa*)

Procesamiento, caracterización y estabilidad del extracto de castaña (*Bertholletia excelsa*)

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Cláudio Fernandes Cardoso
ORCID: https://orcid.org/0000-0002-1933-2190
Universidade Federal de Goiás, Brasil
E-mail: claudiofc.cardoso@gmail.com
Midiana Gusmão dos Santos
ORCID: https://orcid.org/0000-0002-5466-887X
Universidade Federal de Goiás, Brasil
E-mail: midianagsantos@hotmail.com
Yasmini Portes Abraham Silva
ORCID: https://orcid.org/0000-0001-5575-0819
Universidade Federal de Goiás, Brasil
E-mail: yasminiportes@gmail.com
Marianny Silva Canedo
ORCID: https://orcid.org/0000-0002-1437-0577
Universidade Federal de Goiás, Brasil
E-mail: mariannycanedo@hotmail.com
Lismaíra Gonçalves Caixeta Garcia
ORCID: https://orcid.org/0000-0002-8508-8982
Instituto Federal de Educação, Ciência e Tecnologia Goiano – Campus Rio Verde, Brasil
E-mail: lismairagarcia@hotmail.com
Flávio Alves da Silva
ORCID: https://orcid.org/0000-0002-3619-755X
Universidade Federal de Goiás, Brasil
E-mail: flaviocamp@gmail.com

Resumo

O objetivo deste trabalho foi obter extrato de castanha do Brasil solúvel em água em escala piloto, determinar as características físico-químicas, microbiológicas, sensoriais e reológicas dos produtos obtidos e determinar a estabilidade microbiológica, físico-química e física do extrato em presença e ausência de luz e conservantes. Trata-se de um estudo quantitativo, realizado em laboratório. Os extratos foram caracterizados em termos físico-químicos (composição próxima, perfil mineral, pH, acidez titulável, cor), reológicos (viscosidade aparente, índice de fluxo, coeficiente de consistência), microbiológicos (leveduras e bolores, *Salmonella* sp., Coliformes a 35ºC e 45ºC e Staphylococcus coagulase positivo) e sensorial (odor, cor, sabor, impressão global, intenção de compra). Além disso, o efeito da adição de conservantes (500 mg/L de ácido cítrico, 0,15% p:v benzoato de sódio e 0,1% p:v sorbato de potássio) e da exposição à luz sobre o físico-químico (pH, acidez titulável), físico (variação de cor) e estabilidades microbiológicas durante o armazenamento foram avaliadas. Os extratos apresentaram alto conteúdo energético, devido ao seu alto teor de proteínas e gorduras, além de constituir uma fonte valiosa de selênio. Os extratos foram caracterizados como fluidos pseudoplásicos não newtonianos e foram bem aceitos na análise sensorial, com até 60% dos consumidores indicando disposição para comprar o produto. A presença de conservantes dobrou pelo menos o prazo de validade dos produtos, de 9 para 21 dias. A incidência de luz não teve efeito na estabilidade dos extratos.

Palavras-chave: Minerais; Reologia; Análise sensorial; Líquido pseudoplásico.

Abstract

The aim of this work was to obtain Brazil-soluble Brazil nut extract on a pilot scale, to determine the physical-chemical, microbiological, sensory and rheological characteristics of the products obtained and to determine the microbiological, physical-chemical and physical stability of the extract in presence and absence light and preservatives. This is a quantitative study, carried out in the laboratory. Extracts were characterized in terms of physicochemical (proximate composition, mineral profile, pH, titratable acidity, color), rheological (apparent viscosity, flow index, consistency coefficient), microbiological (yeast and molds, *Salmonella* sp., coliforms at 35ºC and at 45ºC and coagulase positive Staphylococcus), and sensory (odor, color, taste, global impression, buying intention) parameters. Furthermore, the effect of the addition of preservatives (500 mg/L citric acid, 0.15% w:v sodium benzoate and 0.1% w:v potassium sorbate) and of the exposure to light on the physicochemical (pH, titratable acidity), physical (color variation) and microbiological stabilities throughout storage were assessed. The extracts had high energy content, due to its high protein and fat contents, besides being a valuable source of selenium. The extracts were characterized as non-Newtonian pseudoplastic fluids, and were well accepted in sensory analysis, with up to 60% of the consumers indicating willingness to buy the product. The presence of preservatives at least doubled the shelf-life of the products, from 9 to up to 21 days. The incidence of light had no effect in the stability of the extracts.

Keywords: Minerals; Rheology; Sensory analysis; Pseudoplastic fluid.

Resumen

El objetivo de este trabajo fue obtener extracto de nuez de Brasil soluble en Brasil a escala piloto, determinar las características físico-químicas, microbiológicas, sensoriales y reológicas de los productos obtenidos y determinar la estabilidad microbiológica, físico-química y física del extracto en presencia y ausencia Luz y conservantes. Este es un estudio cuantitativo, realizado en el laboratorio. Los extractos se caracterizaron en términos físico-químicos (composición cercana, perfil mineral, pH, acidez titulable, color), reológicos (viscosidad aparente, índice de flujo, coeficiente de consistencia), microbiológicos (levaduras y mohos, *Salmonella* sp., Coliformes a 35ºC y 45ºC y Staphylococcus coagulasa positivo) y sensoriales (olor, color, sabor, impresión global, intención de compra). Además, el efecto de la adición de conservantes (500 mg / L de ácido cítrico, 0,15% p: v de benzoato de sodio y 0,1% p: v de sorbato de potasio) y la exposición a la luz en el químico (pH, acidez titulable), física (variación de color) y estabilidad microbiológica durante el almacenamiento fueron evaluados. Los extractos tenían un alto contenido de energía, debido a su alto contenido de proteínas y grasas, además de ser una fuente valiosa de selenio. Los extractos se caracterizaron como fluidos pseudoplásticos no newtonianos y fueron bien aceptados en el análisis sensorial, con hasta el 60% de los consumidores indicando su disposición a comprar el producto. La presencia de conservantes ha duplicado al menos la vida útil de los productos, de 9 a 21 días. La incidencia de la luz no tuvo efecto sobre la estabilidad de los extractos.

Palabras clave: Minerales; Reología; Análisis sensorial; Líquido pseudoplástico.

1. Introduction

The Brazil nut tree (*Bertholletia excelsea* H. B. K.) is native to the Amazon region in South America. Its fruits are spherical or capsular, each containing an average of 12 to 24 nuts or seeds composed of the edible kernels, a dark brown pellicle surrounding it, and a woody

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external shell (Nogueira et al., 2014). The production and extraction of "Brazil nut", the kernel after removal of the shell, only occurs in the Amazonian biome (Freitas-Silva & Venancio, 2011), making this product an important source of income for the native populations.

Brazil nuts contain approximately 60-70% oil and 17% proteins, and are a good source of micronutrients such as selenium, magnesium, phosphorus, thiamine, niacin, vitamin E, vitamin B6, calcium, iron, potassium, zinc and copper, for this reason being associated with many health benefits, such as decrease of cholesterol levels, antioxidant and anti-proliferative activities (Yang, 2009). This nut is considered the food with the highest content of selenium – a mineral required to regulate the thyroid and immunological functions, and action against prostate, liver and lung cancers – in amounts ranging from 0.03 to 512 mg/g (Santos et al., 2017).

Brazil nut is locally consumed in various ways, such as raw, dehydrated, salted, toasted or with diverse coatings such as chocolate, caramel and honey. It is also processed to a flour, and then used as an ingredient in the preparation of other food products, such as granola, ice-cream, chocolates, cakes and cookies. Considering its chemical composition, other co-products could be further commercially explored, such as oils, bran or cake and the water-soluble extract (Santos et al., 2012). The water-soluble Brazil nut extract (BE) is an important alternative that could be economically explored, as it can be easily obtained through a process involving the steps of removal or not of the kernel pellicle, extraction with water, separation of the insoluble residue, additions of other ingredients, heat treatment and packaging (Felberg et al., 2009), and has a good nutritional value for human nutrition (Cardarelli & Oliveira, 2000).

Considering the above, the objective of this work was to obtain BE in a pilot plant scale, to determine the physicochemical, microbiological, sensory and rheological characteristics of the products obtained, and to determine the microbiological, physicochemical and physical stability of the extract in the presence and absence of light and preservatives.

2. Material and Methods

The present work is a quantitative study, carried out in a laboratory (which includes the physical-chemical, rheological, microbiological and sensory characterization and data analysis) (Pereira et al., 2018).

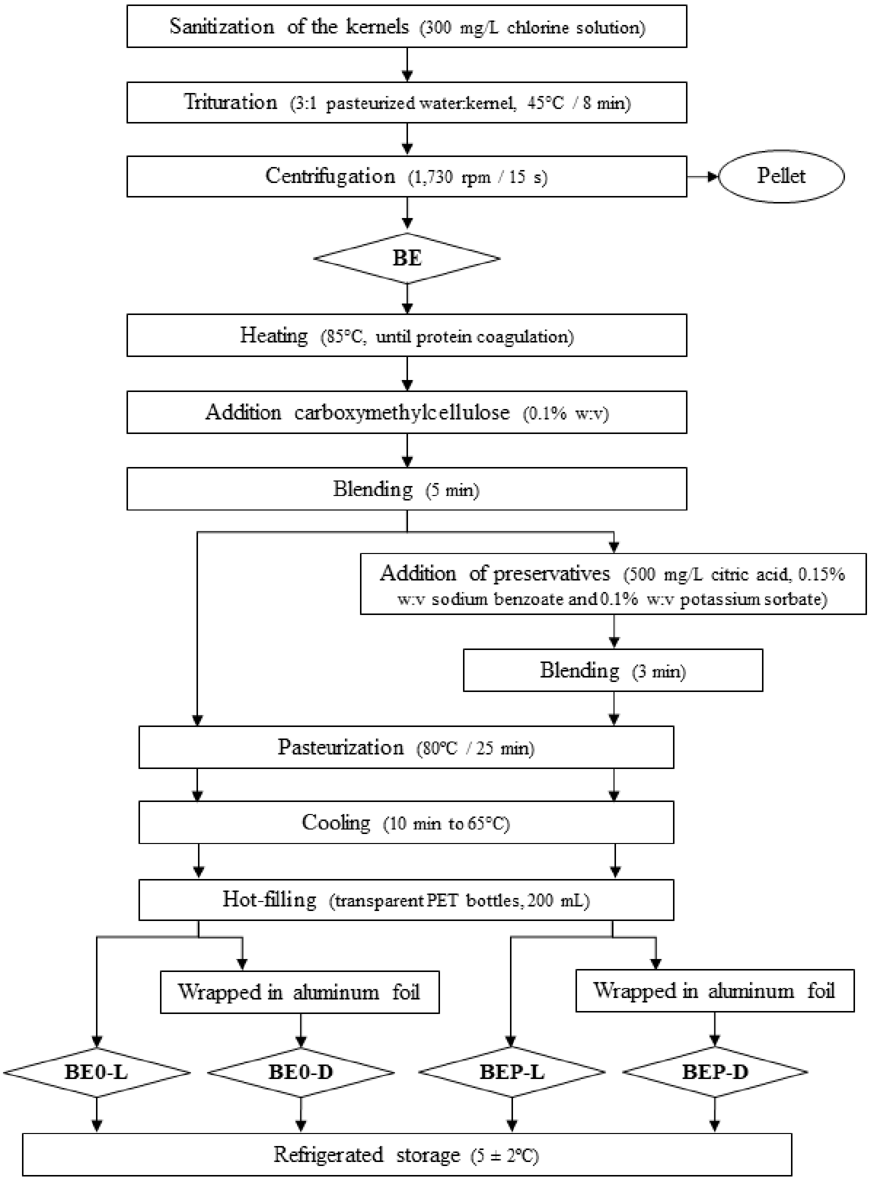
2.1. Material

Twenty kg of Brazil nuts (2014 harvest) were obtained in Rio Branco (Acre), Brazil, vacuum packed in flexible plastic packages laminated with aluminum, and stored at room temperature (25 ± 3 C°) until processing.

2.2. Processing of water-soluble Brazil nut extract

The pilot-scale process used to obtain the BEs is shown in Figure 1.

Figure 1: Process to obtain water-soluble Brazilian nut extracts (BEs).



Source: Author (2020).

The preservative used was 500 mg/L citric acid, 0.15% w:v sodium benzoate and 0.1% w:v potassium sorbate. Each formulation (BE0, without preservatives, and BEP, with added preservatives) was prepared in duplicate, in random order, and all extracts were filled into transparent PET bottles. For the extracts characterization (physicochemical, rheological and microbiological analyses), and sensory analysis, four bottles of each formulation were taken immediately after processing, frozen in ultra-fast freezer (HCFC 22, Irinox, Treviso, Italy) and stored at –18°C until the analyses.

The stability analyses were then carried out using a 2-factors block design with repetitions. The two factors were: X_1 = extract formulation and X_2 = exposure to light. Half of the extracts of each formulation were wrapped in aluminum foil to block light, these representing the storage in the dark (BE0-D and BEP-D), and the other half was stored without coverage (storage in light, BE0-L and BEP-L). The extracts were then analyzed at regular intervals (section 2.5).

2.3. Extract characterization

2.3.1. Nutritional composition

The proximate composition of the extracts (fat, crude protein, dry matter and ash contents) was determined according to the methods established by AOAC (2010). The total carbohydrate content was determined by difference. The energy content of the product was then calculated using the conversion factors of 4.0, 9.0 and 4.0 kcal/g for the energy-yielding substrates protein, fat and carbohydrate, respectively (FAO, 2003). The mineral content was determined by atomic absorption spectrophotometry *(3110, Perkin Elmer, Waltham, Massachusetts, USA) for the minerals* phosphorus (P), selenium (Se), calcium (Ca), magnesium (Mg), copper (Cu), iron (Fe), manganese (Mn), zinc (Zn), lead (Pb), chromium (Cr), cadmium (Cd) and nickel (Ni)*, and by flame photometry (FC180, Celm, São Caetano do Sul, São Paulo, Brazil) for minerals p*otassium (K) and sodium (Na).

2.3.2. Physicochemical and physical characterization

The pH was determined in an automatic pH-meter (MPA210, Instrutemp, São Paulo, Brazil), and total acidity was determined through titration with 0.1 M sodium hydroxide, using phenolphthalein as an indicator, and results are expressed as g of acid oleic per 100g of sample (IAL, 2008). The color parameters of the extracts were measured in the CIELab

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system (Colorquest II, Hunter Associates Laboratory Inc, Reston, Virginia, USA), with determination of parameters L* (luminosity, 0–100), a* (greenness (–) to redness (+)) and b* (blueness (–) to yellowness (+)).

2.3.3. Rheological characterization

Rheological measurements were performed using a Physica MCR 101 rheometer (Ostfildern, Germany). The measurement temperature (25ºC) was maintained on the lower plate using the Peltier system, with plate-plate geometry and a 50 mm diameter. The flow curves were determined using three scanning consecutive continuous ramps with a deformation rate from 0 to 500 s^{-1}, using ascending, descending and ascending cycles to eliminate thixotropy. The curves prepared using the data from the third ascending ramp were chosen, since they best illustrated the rheological behavior of the extracts. The value of apparent viscosity (μ, Pa.s) and shear stress (τ, Pa) were obtained from the RheoPlus software. Parameters *K*, consistency index, and *n*, flow behavior index, were then determined using with the data from the third ascending ramp, after elimination of thixotropy from the samples, according to the Power Law (Steffe, 1996), shown in Equation (1), where: τ is the shear stress (Pa), *K* the consistency index ($Pa.s^n$), γ the strain (deformation) rate (s^{-1}), and *n* the flow behavior index (dimensionless).

$$\tau = K\gamma^n \tag{1}$$

2.3.4. Microbiological analyses

The microbiological standards for coconut milk were adopted as reference, since there are no microbiological standards for Brazil nut extract in the Brazilian legislation (Brazil, 2001). Therefore, the following analyses were conducted: coliforms at 35ºC, coliforms at 45ºC, coagulase positive Staphylococcus, and presence of *Salmonella* in 25g. Furthermore, a yeast and mold count was performed for evaluation of hygiene conditions of the products. All microbiological analyses followed the procedures of the American Public Health Association (APHA, 2001).

2.4. Sensory analysis

The sensory analysis involving human beings was carried out after approval of the research by the Ethics in Research Committee from the Federal University of Goiás (Brazil), process nº 556.159. An acceptance test with a 9-point hedonic scale was applied to assess the attributes of appearance, odor, flavor and global impression of the extracts. The consumer buying intention was evaluated using the following question: “Would you buy this product if it was commercialized?”, with the answer options “Yes” or “No”. The buying potential index was defined as the percentage ratio of consumers showing intention of buying (replied “Yes” to the previous question) divided by the total number of consumers. The tests were carried out with 50 non-trained consumers after completion of the microbiological assays (IAL, 2008).

2.5. Evaluation of stability

Stability of the extracts was assessed through microbiological and physicochemical analyses at 0, 3, 6, 9, 12, 15 and 21 days of refrigerated storage (5 ± 2°C), or until the end of shelf-life, which was defined as the number of days until the growth of yeasts and molds was observed. To verify the effects of ultraviolet radiation on the stability of the product, samples were exposed to light with an intensity of 1,800 lm at a distance of 1.80 m for a daily period of 13 h (Walter, 2010) throughout the shelf-life study, with BE0-L and BEP-L samples stored in transparent PET bottles, and BE0-D and BEP-D samples stored in the same conditions but with aluminum foil covering the bottles to protect from light. One bottle from each treatment was taken on the pre-defined days to carry out the following stability analyses.

2.5.1. Microbiological stability and determination of shelf-life

The same microbiological analyses were performed as described in 2.4.3. The standard used to determine the end of the shelf-life of the Brazil nut extracts was the growth of yeasts and molds.

2.5.2. Physicochemical and physical stability

The physicochemical stability of the extracts during the shelf-life was assessed through analyses of pH and titratable acidity, as described in section 2.4.1. The physical stability was evaluated as the total color difference value (ΔE*) in relation to day 0 (immediately after processing), according to Equation (2), where L_2^*, a_2^* and b_2^* are the color parameters of the sample measured at the different days of storage, and L_1^*, a_1^* and b_1^* are these same parameters measured at day 0, as in section 2.4.1.

$$\Delta E^* = \sqrt{(L_2^* - L_1^*)^2 + (a_2^* - a_1^*)^2 + (b_2^* - b_1^*)^2} \quad (2)$$

2.6. Statistical data analysis

All analyses were performed in triplicate, and results are expressed as average ± standard deviation. Analysis of variance (ANOVA) and Tukey test were performed to compare effects of the treatments, with a significance level of 5% ($p < 0.05$). Statistical analyses were carried out using the ASSISTAT program (Silva, 2013).

3. Results and Discussion

3.1. Nutritional composition

Table 1 shows the proximate composition, physicochemical characteristics and mineral composition of the formulations BE0 and BEP, immediately after processing.

Table 1: Proximate composition (g/100g dry weight – d.w.), total solids (g/100g fresh weight – f.w.), energy content (kcal/100g f.w.) and mineral content (mg/100g d.w.) of water-soluble Brazil nut extracts without (BE0) and with added preservatives (BEP).*

Parameter	Treatment	
Proximate composition (g/100g d.w.)	BE0	BEP
Fat	74.10 ± 1.21[a]	74.51 ± 1.15 [a]
Proteins	16.11 ± 2.15 [a]	12.06 ± 0.69 [b]
Carbohydrates	6.78	10.06
Ashes	2.89 ± 0.22 [b]	3.37 ± 0.67 [a]
Total solids (g/100g f.w.)	21.88 ± 3.46 [a]	20.92 ± 1.87 [a]
Energy content (kcal/100g f.w.)	165.9	158.8
Minerals (mg/100g d.w.)	BE0	BEP
P	1,053.25 ± 69.39[a]	990.75 ± 148.06[a]
K	850.00 ± 41.63[b]	965.00 ± 34.16[a]
Na	165.00 ± 30.00[b]	320.00 ± 23.09[a]
Se	205.40 ± 40.51[a]	274.96 ± 70.56[a]
Mg	265.00 ± 5.77[a]	252.50 ± 17.08[a]
Ca	110.00 ± 8.16[a]	102.50 ± 5.00[a]
Fe	15.70 ± 1.78[a]	15.88 ± 0.96[a]
Mn	7.63 ± 0.22[a]	7.48 ± 0.57[a]
Cu	3.60 ± 0.23[a]	3.70 ± 0.22[a]
Pb	0.38 ± 0.05[a]	0.40 ± 0.00[a]
Cr	0.30 ± 0.00[a]	0.30 ± 0.00[a]
Cd	0.10 ± 0.00[a]	0.10 ± 0.00[a]
Ni	0.10 ± 0.00[a]	0.10 ± 0.00[a]

Note: *In each line, averages followed by different letters indicate significant difference ($p < 0.05$). Source: Author (2020).

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The BE0 and BEP extracts presented high total solids (21.88 and 20.92 g/100g d.w.), protein (16.11 and 12.06 g/100g d.w.) and fat (74.10 and 74.51 g/100g d.w.) contents. Due to the majority of the dry component being fat, the extracts showed a high energy content (165.9 and 158.8 kcal/100g f.w.) and can therefore be considered as products of high energetic value.

Regarding the mineral composition, phosphorus was the most abundant mineral, followed by potassium, sodium, selenium, magnesium and calcium, all these present in amounts greater than 100 mg/100 g (d.w.). There was a significant difference between the treatments only for Na and K contents, due to the preservatives used in the BEP formulation. The variation in the amounts of these minerals in the Brazil nuts, as a function of the location of the trees, the climate and specially the type of soil (Silva et al., 2010), influence the amount of minerals found in products obtained from Brazil nuts (Felberg et al., 2009) The selenium doses found in 100g of the fresh products (44.9 and 57.5 mg) are well above the recommended daily intakes, which varies between 6 and 42 μg/day depending on the population group (FAO, 2001), evidencing the high nutritional value and the important selenium composition of the extracts produced.

3.2. Physicochemical characterization

Table 2 shows physicochemical properties and color parameters of the extracts.

Table 2: Physicochemical properties, color parameters and rheological parameters estimated by the Power Law of water-soluble Brazil nut extracts without (BE0) and with added preservatives (BEP).*

Parameter	Treatment	
	BE0	BEP
Physicochemical properties		
pH	6.34 ± 0.06^a	5.87 ± 0.08^b
Titratable acidity (g linoleic acid/100g)	0.30 ± 0.01^b	0.53 ± 0.02^a
Color parameters		
L*	86.82 ± 0.14^a	84.06 ± 0.14^b
a*	0.33 ± 0.26^a	0.48 ± 0.30^a
b*	5.43 ± 0.41^a	6.15 ± 0.46^a
Rheological parameters		
Apparent viscosity (μ, Pa.s)$^+$	0.028 ± 0.01^a	0.020 ± 0.00^b
Flow behavior index (n)	0.872^b	0.922^a
Consistency index (K, Pa.s^n)	0.059^a	0.033^b
R^2 (Power law equation)	0.9998	0.9999

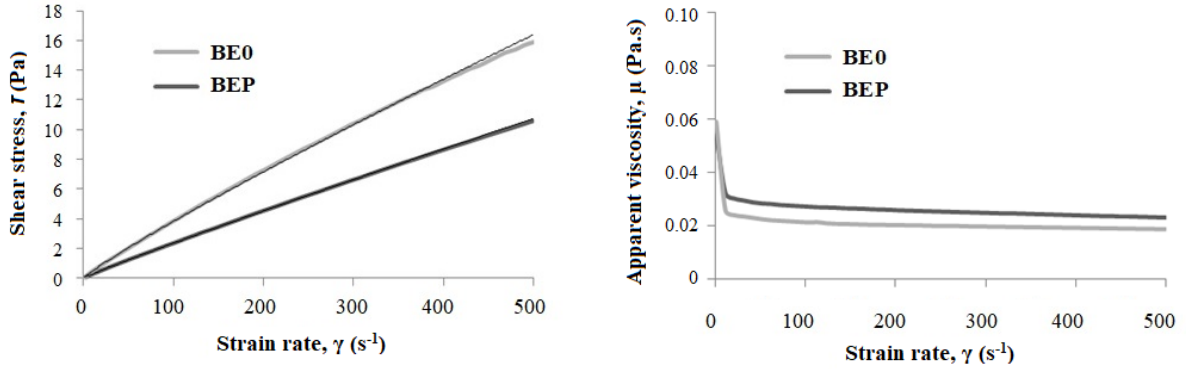
Note: *In each line, averages followed by different letters indicate significant difference ($p < 0.05$). $^+$Deformation stress at 500^{-s}. Source: Author (2020).

The use of preservatives in the formulation had a significant effect in the contents of ash, pH, titratable acidity, which was expected since the preservatives were acidic and contained K (potassium sorbate) and Na (sodium benzoate) in their formulations. BEP showed a higher titratable acidity and consequently lower pH value than BE0, due to the presence of these acidic preservatives. The color parameters are an indicative of the opaque white color of the extracts, as indicated by the high value of the parameter L*, close to the upper limit of +100 (white), and the low values of a* and b*, indicating the absence of other hues (green/red or blue/yellow) in the samples.

3.3. Rheological characterization

Figure 2 shows the rheograms of the Brazil nut extracts.

Figure 2: (a) Flow curve (shear stress, τ × strain rate, γ) and (b) viscosity (apparent viscosity, μ × strain rate, γ) of Brazil nut extracts without (BE0) and with (BEP) added preservatives.



Source: Author (2020).

The flow behavior varied with the deformation rate, the shear stress τ showing a linear relationship with the strain rate γ (Figure 2(a)) and the apparent viscosity μ decreasing with increase in the strain rate γ (Figure 2(b)), thus characterizing the extracts as non-Newtonian pseudoplastic fluids, similar to other vegetable extracts such as coconut milk (Simuang et al., 2004; Tipvarakarnkoon et al., 2010). Figure 2(b) shows that apparent viscosity μ was strongly reduced at strain rates of up to 25 s^{-1}, then changed slightly and remained constant at higher rates. This could be related to a reduction in size of the colloidal aggregates with the increase in deformatioin rate (Ibanoglu, 2002). The Brazil nut extract is a colloidal system containing oil droplets dispersed in an aqueous phase. When shear stress is applied, the particles can reorganize themselves in a direction parallel to the shear stress, and the larger particles can break into smaller ones. As a result of this particle-particle interaction, the resistance is reduced, thus the particles can flow more easily, resulting in a decrease in viscosity.

Table 2 shows the values for apparent viscosity and the parameters estimated by the Power Law equation for the Brazil nut extracts. This model fitted the flow curves of the Brazil nut extracts very well, with very high values for the correlation coefficients ($R^2 > 0.99$). The formulations showed significantly different rheological behaviors. BEP presented higher values of flow index (n) and apparent viscosity (μ), while BE0 showed a greater consistency coefficient (K). The consistency coefficient K is an indicator of the viscous nature of the system, and the flow index n an indicator of its fluid nature (Chen & Opara, 2013). These results indicate that the addition of preservatives, and thus the difference in pH between the two formulations, affected the rheological behavior, with the formulation BE0 with the higher pH value (6.34), showing higher viscosity. Viscosity depends on the pH value and is independent of the type of acid present, since the acidity of the product affects the molecular interactions among the components, and therefore their flow behavior (Hirashima et al., 2005). The values found for n (0.923) and K (0.034 $P.s^n$) for formulation BE0 in the present work are comparable to those found for coconut milk (Simuang et al., 2004; Tipvarakarnkoon et al., 2010).

3.4. Microbiological characterization

None of the microorganisms investigated were found, thus conforming to standards for coconut milk adopted as parameters for the present study (Brazil, 2001) and indicating adequate processing conditions. The absence of coliform groups at 35ºC and 45ºC indicates the efficiency of the pasteurization and the absence of recontamination, since the presence of these organisms in processed foods is a useful indicator of post-sanitization and post-processing contamination (Kornacki & Johnson, 2001). The samples were therefore considered suitable for sensory analysis, as follows.

3.5. Sensory analysis

The mean scores attributed by the panelists are shown in Table 3, indicating that the addition of preservatives did not interfere with consumer acceptance, since no significant differences were found in the mean scores attributed to the samples.

Table 3: Sensory analysis of water-soluble Brazil nut extracts without (BE0) and with added preservatives (BEP).*

Attribute +	Treatment	
	BE0	**BEP**
Odor	6.59 ± 1.54[a]	6.43 ± 1.63[a]
Color	7.17 ± 1.38[a]	7.20 ± 1.44[a]
Taste	6.35 ± 1.55[a]	6.17 ± 1.85[a]
Global impression	6.80 ± 1.29[a]	6.64 ± 1.37[a]
Buying intention	60%	57%

Note: *In each line, averages followed by different letters indicate significant difference ($p < 0.05$). +Attributes evaluated in a 9-point hedonic scale. Source: Author (2020).

The extracts were accepted by the majority of panelists with scores between “liked slightly” and “liked moderately” (above 6). With respect to buying intention, the majority of the panelists said they would buy the extracts, with buying intention indexes of 60% for BE0 and 57% for BEP. The acceptance of the water-soluble Brazil nut extract as a food product could be further increased by the addition of other flavoring ingredients, such as sugar and sweeteners (Felberg et al., 2002), or by adding other vegetable extracts to elaborate a mixed beverage (Arévalo-Pinedo et al., 2013; Felberg et al., 2009).

3.6. Evaluation of stability during storage

3.6.1. Microbiological stability and determination of shelf-life

All processed extracts (day 0) conformed to the microbiological standards established for coconut milk (Brazil, 2001), showing no growth of any of the microorganisms evaluated. No coliforms at 35ºC and at 45ºC, coagulase positive *Staphylococcus*, or *Salmonella* sp. were observed at any time during the shelf-life of the products. These results indicate the efficiency of the thermal pasteurization treatment to prevent development of these microorganisms, potentially found in this type of product.

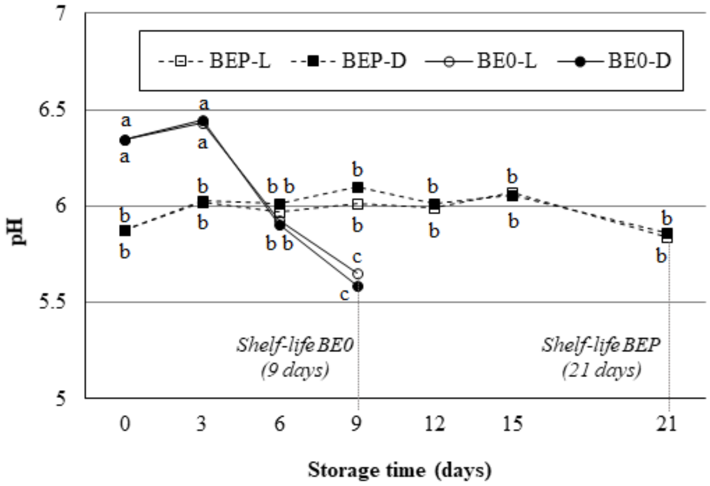
Depending on the type of product under study, various criteria may be used to determine the end of its shelf life, such as microbiological (fungal growth, a high bacterial count or the presence of potentially toxic microorganisms), physical (color changes) or chemical (reduction of the level of a given nutrient) parameters, or sensory evaluations (Valero et al., 2012). In the present study, the growth of yeasts and molds was adopted as parameter to determine the end of shelf-life. During evaluation of stability, yeasts and molds did not develop in the BE0 formulations until day 9, which was then determined to be the shelf-life of the formulations without preservatives. The exposure to light had a marginal effect, with higher count found in the BE0-L (1.1×10^2 CFU/mL) than in the BE0-D (6.0×10^1 CFU/mL) formulation. Both formulations containing preservatives showed no growth until the last day of the study (21 days), irrespective of exposure to light, therefore, for samples BEP-L e BEP-D the shelf-life was of at least 21 days. These results indicate that the addition of preservatives had an important effect in delaying the development of yeasts and molds, since it more than doubled the shelf-life of the products. Since their presence of these additives did not affect negatively the consumer acceptance, it is thus recommended the use of preservatives to increase shelf-life of the Brazil nut extracts. These results were highly satisfactory since the shelf-life of refrigerated pasteurized products is generally short, from seven to ten days (Lewis & Heppell, 2000). The addition of the preservatives extended the storage period, since these compounds prevent or delay the microbial or enzymatic deterioration of the samples, increasing their shelf-life (Wedzicha, 2003), as was presently observed, with the BEP formulations presenting a shelf-life twice as longer as BE0.

3.6.2. Physicochemical and physical stability

Color variation (ΔE*) was insignificant during storage for all formulations, with an absolute value below 1.3 for all measures, indicating that physicochemical changes did not have a strong impact on the physical stability of the product, and the exposure to light did not affect color.

Figure 3 shows the mean values obtained for the pH of the extracts during storage.

Figure 3: pH of water-soluble Brazil nut extracts without (BE0) and with (BEP) added preservatives throughout refrigerated storage under light (BE0-L and BEP-L) and in the dark (BE0-D and BEP-D). Letters indicate significant difference ($p < 0.05$).

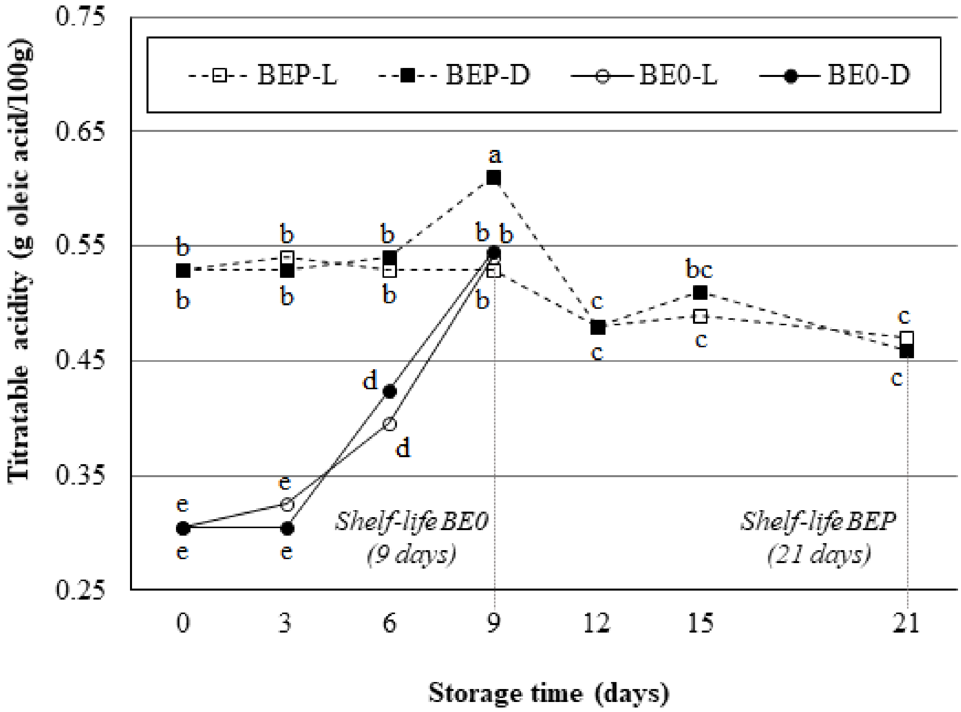


Source: Author (2020).

Although not regulated by Brazilian legislation, pH is of extreme importance in beverage formulations. Lower pH values (higher acidity) are preferred by the industry since they do not favor enzyme activity and inhibit the development of pathogenic microorganisms (Karastogianni et al., 2016). Irrespectively of the exposure to light, the treatments formulated with preservatives (BEP-L and BEP-D) initially showed lower pH values than the treatments without preservatives (BE0-L and BE0-D) and maintained their pH values practically stable throughout storage, only presenting a slight decrease at the end of storage (21 days). On the other hand, the extracts without preservatives showed considerable decreases in pH after the third day of storage until the end of their shelf-life determined at 9 days, which was probably related to the development of acid-producing yeasts and molds in the BE0 formulations. When extracts of the same formulation were compared (BE0-L × BE0-D and BEP-L × BEP-D), no significant difference was observed ($p > 0.05$), indicating that light did not influence the stability of the extracts, therefore a transparent plastic package is sufficient to store this type of product.

Figure 4 shows the values obtained for the titratable acidity of the Brazil nut extracts during storage.

Figure 4: Titratable acidity of water-soluble Brazil nut extracts without (BE0) and with (BEP) added preservatives throughout refrigerated storage under light (BE0-L and BEP-L) and in the dark (BE0-D and BEP-D). Letters indicate significant difference ($p < 0.05$).



Source: Author (2019).

A similar behavior to the pH was observed, since these two variables are strongly correlated. The acidity of extracts containing preservatives (BEP) did not vary significantly, remaining at about 0.50% throughout the 21 days of storage, and until day 6 it was higher than the mean acidity of those without preservatives (BE0), due to the presence of the preservatives added. However, during storage the acidity of BE0 extracts increased until the end of their shelf-life (day 9), from about 0.30% to between 0.50% and 0.60%, reaching values comparable to the BEP values. This behavior of the BE0 extracts is probably due to the concomitant growth of acid-producing yeasts and molds observed in the same period, with the production of acids by these microorganisms during multiplication.

4. Conclusions

Brazil nut extracts showed elevated total solids, protein and fat contents, and can therefore be considered as products of high nutritional value, and were well accepted by panelists in sensory analysis. The extracts containing preservatives presented good physicochemical stability, with only slight oscillations in the values for acidity and pH being observed. The high pH value of the product could favor the growth of pathogenic microorganisms, therefore the use of preservatives is important to increase the shelf-life of this type of product.

In this study the addition of preservatives increased shelf-life from 9 to 21 days. The extract is light-stable, therefore there is no need to use a special package with a light barrier. Therefore, with appropriate formulation and packaging, Brazil nut extracts could be processed and stored under refrigeration for at least 21 days, allowing for distribution and consumption of the product.

The information regarding the characterization and stability of the water-soluble Brazil nut extract presented in this study may contribute to the development of viable alternatives of exploring the Brazilian nut tree for the production of a healthy, well-accepted and stable food product, while also contributing to the development of local economy.

It is suggested that in future works tests be made on an industrial scale and with the use of tetra pack.

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Porcentagem de contribuição de cada autor no manuscrito

Cláudio Fernandes Cardoso – 16%

Midiana Gusmão dos Santos – 20%

Yasmini Portes Abraham Silva – 16%

Marianny Silva Canedo – 16%

Lismaíra Gonçalves Caixeta Garcia – 16%

Flávio Alves da Silva – 16%