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Original Article

Silviculture

Fruits, Seeds and Oil of Brazil Nuts Produced in Mato Grosso State

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ABSTRACT

Bertholletia excelsa (Brazil nut) is the only representative of the Bertholletia genus, but presents great phenotypic variability. The objective of this study was to characterize the fruits, seeds and crude oil of Brazil nuts from the state of Mato Grosso, Brazil. Fruit and seeds from adult trees were collected at the municipalities of Alta Floresta, Cotriguaçu, Itaúba and Juína. Physical characteristics, yield and composition of the seeds, as well as crude oil characteristics were evaluated. The fruits produced in Itaúba were the smallest and had the lowest number of seeds, while those from Juína presented larger size and mass. The nuts produced in Itaúba had the lowest mean unit mass and ten-seed mass values. The seeds produced in Juína presented higher lipid content and lower ash content. The selenium content of the nuts was higher in Cotriguaçu. There were no significant differences in fruit peel thickness, yield and protein content of the nuts or characteristics of the crude oil.

Keywords: *Bertholletia excelsa* bonpl., quality, physicochemical properties, post-harvest.

1. INTRODUCTION

The Brazil nut (*Bertholletia excelsa* H.B.K) stands out for having numerous uses. The fruits (known as *ouriços* in Portuguese) have a very hard woody shell and a spherical or slightly flattened shape, being used in the making of handicrafts by extractivists (Faustino & Wadt, 2014). Each fruit shell contains 12 to 24 seeds (known as Brazil nuts) (Yang, 2009). These seeds (or nuts) contain significant amounts of proteins (15-20%) and lipids (60-70%) (Santos et al., 2011), in addition to being sources of fibers, complex B1, B2 and B3 vitamins, pro vitamins A and E, and minerals such as calcium, magnesium, iron, potassium, sodium and selenium, highlighting the latter, which is a micronutrient with possible anticarcinogenic effects (Mahan & Escott-Stump, 2002; Freitas et al., 2008). The oil is appreciated for its emollient action in the cosmetics industry (Funasaki et al., 2016). Currently, the Brazil nut is used on a large scale in the cosmetics industry for manufacturing oils, perfumes, soaps and other derivatives (Barbosa & Moret, 2015).

In 2014, the national production of Brazil nuts reached 37,499 t, with the North region accounting for approximately 95.93% of this production, and the state of Acre being the most important (36.50%). The Central-West region, represented by the northern region of Mato Grosso, was responsible for the 4.07%, completing the national production (IBGE, 2014).

Due to the wide distribution of Brazil nut trees throughout the Amazon region, differences in morphological and phenological aspects, such as flowering period, number of fruits and yield per tree, have already been observed (Wadt et al., 2005; Tonini et al., 2008). Differences in the seed composition of the same species can also be found, depending on the dispersion period and the place of origin, since several chemicals are synthesized and stored in the seeds, aiming to guarantee the success of the next plant life cycle in its interaction with the environment (Larcher, 2004; Sreenivasulu & Wobus, 2013).

Thus, the physical-chemical characterization of fruits and seeds is important for acquiring knowledge on the nutritional value, as well as adding value to the product and allowing its quality to be maintained during post-harvest operations (Yahia, 2010). In addition, the characterization may contribute to studies of genetic diversity and pre-improvement of Brazil nuts, because characteristics such as appearance, size, fruit and seed shape and nutritional quality, among others, should be considered to satisfy the requirements of the national and international markets (Viana et al., 2013).

Regarding edible oils, the Ministry of Health regulates production through ANVISA, from the Resolution of Collegiate Board of Directors (RDC) nº. 270, dated September 22, 2005 (Brasil, 2005), and defines that the acid index and the peroxide index are reference parameters for determining the quality of oil conservation.

Thus, the objective of this study was to characterize fruits, seeds and crude oil of Brazil nuts produced in municipalities of Mato Grosso state, Brazil.

2. MATERIAL AND METHODS

The fruits, seeds and crude oil extracted from Brazilian nuts from the 2012/2013 crop were analyzed. The fruits were collected between November/2012 and March/2013, in areas of native forest in the municipalities of Alta Floresta (9°59’48” S and 56°17’31” W), Cotriguaçu (9°49’15” S and 58°17’22” W), Itaúba (11°05’53” S and 55°02’11” W) and Juína (11°36’19” S and 58°36’38” W), in Mato Grosso state. One hundred and forty fruits from 28 adult trees were collected (DBH > 30 cm).

The experiment followed a completely randomized design with 4 treatments (collection municipalities), with 5 replicates. The data were submitted to analysis of variance and the means were compared by the Tukey test at 5% probability using the Sisvar® statistical program (Ferreira, 2011).

2.1. Fruit characterization

Transverse and longitudinal shell diameters and thickness were measured in three points of the equatorial region of the fruit. These metric measurements, as well as all the others in this work, were performed with a digital caliper with 0.01 mm resolution. The fruit mass was also determined on a digital scale with a resolution of 0.01 g, which was also used in the other mass evaluations for the fruits and seeds. The number of seeds was obtained by counting.

Five replicates were performed for each evaluated parameter, with each repetition being composed on average of 28 fruits.

2.2. Nut characterization

To determine the physical characteristics of Brazil nuts, all fruits from each location were opened and the seeds homogenized. Approximately 1.5 kg sample of seeds from each area was removed and sent for analysis. It should be noted that seeds refer to whole nuts, with its integument or peel and almond; and nut only refers to the edible part, without the presence of the integument.

The following parameters were determined in 5 repetitions:

- *External characteristic dimensions*: by determining the seed length, width and thickness. Each replicate was composed of 12 seeds;
- *Average unit mass:* was obtained after determining the mass of the fruit seeds divided by the number of seeds;
- *Yield*: 40 seeds were weighed with and without integument for each replicate, and the yield was calculated by mass difference, given as a percentage;
- *Tissue thickness*: was obtained from the average of three measurements, one on each face in the equatorial region of the seed. Ten seeds were used for each replicate;
- *Mass of ten seeds*: was obtained from the mass measurement of 10 seeds for each replicate, in duplicate.

For determining the centesimal composition, four replicates of 50 seeds were peeled and milled. All analyzes were performed in triplicate, being:

- *Moisture*: determined in greenhouse with forced air circulation at 105 ± 3 °C (Brasil, 2009).
- *Protein*: the total nitrogen content was determined and multiplied by the conversion factor of 6.25 according to the micro Kjeldahl method, described in the physical-chemical methods for food analysis of the Adolfo Lutz Institute (IAL, 2005);
- *Lipids*: lipid quantification was performed with a Soxhlet extractor using n-hexane as solvent, according to physicochemical methods for food analysis of the Adolfo Lutz Institute (IAL, 2005);
- *Ashes*: were determined according to the methodology described in the physicochemical methods for food analysis of the Adolfo Lutz Institute (IAL, 2005) using 0.5 g of sample, which was taken to a muffle oven for calcination at 550 °C for about 7 hours or until the ashes were completely white;
- *Total carbohydrates*: calculated by the difference between 100 and the sum of percentages of water, protein, total lipids and ashes;
- *Selenium*: 0.5 g of sample of each site in triplicate was digested with 5 mL HNO_3 concentrated in teflon (PTFE) tubes and allowed to stand for 12 hours for pre-digestion. Then the pre-digested samples were taken to a microwave oven (CEM, model Mars 5, CEM Corporation, Matthews, NC, USA) and digested at 0.76 MPa for 10 min. After cooling the extracts, 5 mL of distilled water was added. The selenium determination was performed using an atomic absorption spectrophotometer with electro-thermal atomization by a graphite furnace (PerkinElmer Analyst 800) with cross-heating and automatic sampler. A selenium hollow cathode lamp was used as the radiation source operating at 6.0 mA, with a wavelength of 196.0 nm, a gap of 2.0 nm, and using an atomization temperature of 1900 °C.

2.3. Oil characterization

Each replicate of the crude oil for analysis from each municipality was obtained by grinding 1.0 kg of nuts in a cold vegetable extracting press (Scott Tech, Model ERT40), resulting in approximately 500 mL of crude oil. The oil was centrifuged at 3500 rpm for 15 min and then analyzed in triplicate for the following parameters:

- *Acidity index*: by titration with 0.1 mol L^{-1} sodium hydroxide, according to the Adolfo Lutz Institute (IAL, 1985);
- *Iodine index*: by titration with 0.1 mol L^{-1} sodium thiosulphate solution, according to AOCS 1 - 25 (AOCS, 2000);
- *Peroxide index*: determined according to AOCS (2000), Method Cd 8-53;
- *Relative density at 25 °C*: according to AOCS 10a-25 (AOCS, 2000) using a pycnometer.

3. RESULTS AND DISCUSSION

Fruits collected in Itaúba had the smallest longitudinal diameters (99.37 mm) and the largest transversal diameters, as well as the fruits of Juína. This fact describes an intrinsic characteristic of the place for the collectors (extractivists) from Itaúba, which is known to produce small and flat fruits. Juína fruits had a larger longitudinal diameter (109.44 mm) and a higher fruit mass (674.54 g) (Table 1).

The values found for the longitudinal and transverse diameters are higher than those found by Santos et al. (2006) when analyzing fruits harvested in Belém, Pará state, which obtained 96.4 and 97.5 mm, respectively. The mean mass value of fruits collected in three of the four municipalities varied between 565.46 and 674.54 g, being higher than those found by Camargo et al. (2010), who obtained an average value of 537.62 g of fruit mass collected in Cotriguaçu.

The fruits of Itaúba and Cotriguaçu had a lower number of seeds per fruit when compared to other sites. Borges et al. (2016) and Camargo et al. (2010) verified similar values to those of this study, of about 17 and 17.18 seeds per fruit collected in the municipalities of Itaúba and Cotriguaçu, respectively. However, the number of seeds per fruit was generally higher than that found by Tonini (2013), who found 15 seeds per fruit when analyzing Brazil nut fruits from the Caracaraí municipality, in Roraima state, Brazil.

There was no significant difference when the thickness of the fruit shell was evaluated between the sites, with average thickness 9.28 mm. Teixeira et al. (2015) obtained a considerably higher average value of 15 mm for the thickness of the brazil nut fruit shells from the *Campo Experimental Confiança* belonging to Embrapa Roraima in the Cantá municipality, Roraima state.

The seeds of all evaluated sites have an average of 26.31 mm of mean axis (b), thus corroborating with Ferreira et al. (2006) and Santos et al. (2006), who found mean axis values of 27.77 mm in Brazil nuts from the municipality of Laranjal do Jarí, Amapá state, and 28.12 mm in nuts from Belém, Pará state, respectively, being close to those obtained in this study. However, the Itaúba seeds presented the lowest values for the highest (a) and lowest (c) dimension characteristics of the seeds, meaning the length and thickness, respectively, as presented in Table 2. In studying the physical properties of Brazil nut seeds harvested in the municipality of Itaúba, Nogueira et al. (2014) found values higher than these, being about 39.35 mm for the largest (a) and 17.88 mm for the smaller (c) characteristic sizes of the seeds.

It was observed that the lowest values were found for the mean unit mass (6.09 g) in Itaúba, and consequently the lowest value for the ten-seed mass (26.29 g). The highest values were found for Juína, being 8.57 g, 37.28 g and 1.21 mm for unit mass, ten-seed mass and shell thickness, respectively. The unit mass varied from 6.09 to 8.57 g, being close to that observed by Tonini (2013), of 7.63 g. For the Brazil nut seed yield, it was observed that there was no significant statistical difference (Table 2), with an average yield of 45.08%. Nogueira et al. (2014) obtained a superior yield in their study, around 51.01%.

It was observed that the Juína seeds contain the highest lipid content and lowest amount of ashes. However, the seeds from Alta Floresta and Itaúba had the lowest values for lipids and the highest values for the ash content (Table 3). Moreover, the average lipid content of the seeds, regardless of location, is close to those reported by Santos et al. (2006), which was 66.24% in Brazil nuts from the state of Pará. Regarding ash content, Queiroga et al. (2009) found a higher value of about 4.32% in Brazil nuts obtained in the market of Belém, Pará.

It was identified that the amount of protein present in the seeds did not vary between sites, with a mean value of 14.86% (Table 3). Balbi et al. (2014) found

Table 1. Averages of longitudinal diameter (LD), transverse diameter (TD), mass (Mf), number of seeds (Ns), and shell thickness (Stf) of Brazil nut fruits for each municipality.

Location	LD (mm)	TD (mm)	Mf (g)	Ns	Stf (mm)
Alta Floresta	103.42b	103.53b	568.39b	17.87a	9.17a
Cotriguaçu	102.91b	102.72b	565.46b	17.05ab	9.30a
Itaúba	99.37c	107.88a	529.66b	16.50b	9.19a
Juína	109.44a	109.23a	674.54a	18.00a	9.45a
Mean	103.79	105.84	584.51	17.35	9.28
CV[1] (%)	9.84	12.81	34.92	19.73	15.51

Means followed by the same letter in the column do not differ from each other by the Tukey test at 5% significance. [1]CV: coefficient of variation.

Table 2. Mean characteristic dimensions (*a*: largest, *b*: average, *c*: lowest), mean unit mass (Mus), ten-seed mass (TsM), shell thickness (Sts) and yield (Y) of Brazil nut seeds for each municipality.

Location	*a* (mm)	*b* (mm)	*c* (mm)	Mus (g)	TsM (g)	Sts (mm)	Y (%)
Alta Floresta	40.06 b	25.76^{ns}	17.57 b	7.35 b	34.65 bc	0.98 a	44.19^{ns}
Cotriguaçu	41.91 c	26.73	17.48 b	7.64 b	33.14 b	1.06 b	43.16
Itaúba	35.73 a	26.60	16.57 a	6.09 a	26.29 a	0.83 a	45.95
Juína	42.65 c	26.13	18.10 b	8.57 c	37.28 c	1.21 c	47.03
Mean	40.09	26.31	17.43	7.41	32.84	1.02	45.08
CV[1] (%)	9.68	50.21	11.52	27.33	5.87	26.45	7.03

Means followed by letters in the column do not differ from each other by the Tukey test at 5% probability. [1]CV: coefficient of variation.

Table 3. Averages of moisture content (M), lipids (Lip), proteins (Pro), ash (Ash), carbohydrates (Carb), and selenium (Se) of Brazil nuts for each municipality.

Location	M (%)	Lip (%)	Pro (%)	Ash (%)	Carb (%)	Se (mg kg^{-1})
Alta Floresta	4.64 bc	62.36 a	15.33 ^{ns}	3.73 c	13.94 b	3.89 b
Cotriguaçu	4.39 ab	67.70 c	15.62	3.35 b	8.95 a	12.10 c
Itaúba	4.70 c	65.13 b	14.98	3.67 c	11.52 ab	2.08 a
Juína	4.26 a	69.44 c	13.49	3.03 a	9.78 a	2.89 a
Mean	4.50	66.16	14.86	3.45	11.05	5.49
CV[1] (%)	2.30	1.11	6.92	1.39	1.20	6.18

Means followed by letters in the column do not differ from each other by the Tukey test at 5% probability; ns = not significant. [1]CV: coefficient of variation.

similar protein value (14.28 g 100 g^{-1}) in nuts sold in Curitiba, Paraná state. Santos et al. (2011) verified higher values for the protein content (18.58 g 100 g^{-1}) in Brazil nuts from the state of Pará. The moisture, which exerts influence on these characteristics, was higher for Itaúba and Alta Floresta, with the latter being explained by the rainfall that occurred during the harvesting and transportation of the fruits.

The municipality of Alta Floresta has nuts containing higher amounts of carbohydrates when compared to other locations. However, the carbohydrate values found in the nuts, regardless of the place of origin, were higher than that found by Queiroga et al. (2009) (5.69%) in Brazil nuts sold in Belém, Pará state. The average carbohydrate values of the four sites were close to that observed by Felberg et al. (2009) (11.61%).

Brazil nuts from the municipality of Cotriguaçu have a higher amount of selenium when compared to nuts from other municipalities (Table 3). The selenium content in this municipality was much higher than that found by Balbi et al. (2014), who observed a value of 0.425 mg 100 g^{-1}; however, very close to the content found in the nuts from the other locations.

Parekh et al. (2008) state that selenium values vary widely in Brazil nuts from different regions, and that this difference is due to the soil conditions of each region where the plant developed, with larger or smaller amounts of selenium. Brazil nuts are a naturally rich selenium source (Balbi et al., 2014), and the levels found in this study were higher than the daily dose (34 µg day^{-1}) recommended by the National Agency of Health Surveillance (ANVISA) for an adult (Brasil, 2005).

Regarding the oil quality, no significant difference of the evaluated characteristics was observed between the municipalities. The average values found for the acidity index were 1.51 mg KOH g^{-1}, acidity in oleic acid of 0.76%, 99.85 mg 100 mg^{-1} iodine index and relative density of 0.869 g mL^{-1} (Table 4).

It was observed that the average of the indices for the determining the oil quality was inferior to the maximum that is permitted by the RDC/ANVISA of nº. 270 (Brasil, 2005), meaning that the oil extracted from Mato Grosso state Brazil nuts has good quality.

In general, the nuts produced in Juína presented better attributes related to the physical and production quality, and Cotriguaçu nuts were distinguished by their chemical quality with high selenium content.

Although the Brazil nut is the only representative of the Bertholletia genus, there is great phenotypic

Table 4. Relative density at 25 °C (RD), acidity index (AI), acidity in oleic acid (AO), iodine index (II), and peroxide index (PI) of Brazil nut crude oil compared to the maximum values of ANVISA (Brasil, 2005).

Analyses	Results	Max values (ANVISA)
RD (g mL^{-1})	0.870	n.d.[2]
AI (mgKOH g^{-1})	1.515	4.00
AO (%)	0.7625	n.d.
II (mg 100 mg^{-1})	99.853	n.d.
PI (meq kg^{-1})	< DL[1]	15.00

[1]DL = detection limit; [2]n.d. = not determined.

variability, which is presented by the physical and chemical characteristics of the seed and fruit composition and also in the production. These variations are mainly morphological, and can be influenced by genetic characteristics, geographic distribution and edaphoclimatic conditions (Camargo et al., 2010). Soil, climatic conditions, water and nutrient availability, plant density and composition influence the plant's healthy development and consequently the productivity and phenotypic plasticity of its nuts (Zuidema, 2003; Kainer et al., 2007).

The physical and physicochemical composition of Brazil nuts is an important step to favor its use in technological processing as a resource source for the industry, as well as a joint form of promoting the economic and social growth of this sector (Santos et al., 2011), in addition to being necessary for knowledge of Brazilian biodiversity. Brazil nut biometry studies also aid in genetic improvement programs, in understanding physiology and in developing techniques for propagating this species.

Thus, it is important that this type of work is continued and repeated in different harvests, which allows a value aggregation through certification or tracking, since the populations of Brazil nut trees present morphological and phenological diversity in different regions of the Brazilian Amazon Basin (Sujii et al., 2015).

4. CONCLUSIONS

1. There is a difference in the shape and size of Brazil nut fruits in the Mato Grosso state;
2. Some parameters, such as fruit shell thickness, yield, protein content and oil characteristics do not vary with the production site;
3. The Brazil nut fruits collected in Juína are larger and have more seeds;
4. Cotriguaçu nuts have the highest selenium content among the municipalities evaluated in the state of Mato Grosso;
5. The oil extracted from Mato Grosso Brazil nuts is high quality.

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