

Low-pressure solvent extraction of oil from macauba (*Acrocomia aculeata*) pulp: characterization of oil and defatted meal

Extração por solvente a baixa pressão do óleo da polpa de macaúba (*Acrocomia aculeata*): caracterização de óleo e farelo desengordurado

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ABSTRACT

This study aimed to extract the oil from macauba pulp using a low-pressure solvent extraction, as well as characterisation of the extracts and defatted meal obtained using different solvents (*n*-hexane, ethyl acetate and isopropanol). Results reported higher yields in oil, 27.43%, using isopropanol ($P < 0.05$), wherein the highest levels of β -carotene in the extract, 348.30mg 100g⁻¹, were obtained with this solvent. In the composition of the extracts, monounsaturated fatty acids were predominantly found, and it was reported that the type of solvent had no significant influence ($P > 0.05$) on the composition; however, in the quantification of free glycerol compounds, isopropanol showed higher levels, 104.15mg 100g⁻¹, of these compounds ($P < 0.05$). Meals had higher ash, protein and fibre content when compared to pulp. Thus, oil removal can be stated to promote water retention capacity; however, it has no influence on the other technological characteristics evaluated.

Key words: *Acrocomia aculeata*, vegetable oil, defatted meal.

RESUMO

O presente trabalho objetivou a extração do óleo da polpa de macaúba utilizando extração por solvente a baixa pressão, bem como a caracterização dos extratos e farelos obtidos, utilizando diferentes solventes (*n*-hexano, acetato de etila e isopropanol). Os resultados reportam maiores rendimentos em óleo, 27,43%, na utilização do isopropanol ($P < 0,05$), sendo que os maiores teores em β -caroteno no extrato, 348,30mg 100g⁻¹, foram obtidos com este solvente. Na composição dos extratos, predominam os ácidos graxos monoinsaturados e relata-se que o tipo de solvente não apresentou influência significativa ($P > 0,05$) na composição, no entanto, na quantificação dos compostos livres de glicerol, o isopropanol apresentou os maiores teores, 104,15mg 100g⁻¹, desses compostos ($P < 0,05$). Os farelos apresentaram

maiores teores de cinzas, proteína e fibras, quando comparados à polpa. A remoção do óleo favorece a capacidade de retenção de água, no entanto, não tem influência nas demais características tecnológicas avaliadas.

Palavras-chave: *Acrocomia aculeata*, óleo vegetal, farelo desengordurado.

INTRODUCTION

Macauba (*Acrocomia aculeata*) has high oil content (~30%) (CICONINI et al., 2013); the pulp is around 45% of the fruit composition (RAMOS et al., 2008) and provides oil that is orange-yellow in colour, rich in bioactive compounds and fatty acid composition, and contains high levels of oleic and palmitic acids (HIANE et al., 2005; AMARAL et al., 2011; COIMBRA & JORGE, 2011). Among the bioactive compounds, there is β -carotene, a powerful antioxidant, which is a precursor of vitamin A in human and animal metabolism, and flavonoids, which belong to the group of phenolic compounds, and are especially known for their antioxidant activity and several biological effects (HUBER & RODRIGUEZ-AMAYA, 2008).

Among the methods available for the extraction of vegetable oils, extraction by pressing and solvent extraction are the conventional techniques.

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Recently, studies have been developed as alternatives to these traditional methods using solvents that are acceptable in the food industry such as ethyl acetate (ALMEIDA et al., 2012; TIAN et al., 2013), and isopropanol (SETH et al., 2007; DUTTA et al., 2014; RAMLUKAN et al., 2014), among others. Among the techniques applied using such solvents, the researches of JESUS et al. (2013) and OLIVEIRA et al. (2013) reports pressurized liquid extraction and low-pressure solvent extraction for the extraction of palm and passion fruit seed oil, respectively, with good yields of oil. Regarding the oil obtained from Macauba pulp, in the literature there are reports of the use of organic solvent extraction in a Soxhlet (HIANE et al., 2005; RAMOS et al., 2008; COIMBRA & JORGE, 2011) and pressing (SILVA & ANDRADE, 2013).

The defatted Macauba meal, after oil extraction and drying, can be utilised as dietary fibre or for animal feed. Oil removal can enhance the technical characteristics of the meal due to the increased protein and dietary fibre content, and determination of these characteristics is essential for establishing the potential for incorporation of this by-product. Dietary fibre can also impart some functional properties to foods, e.g., increase water holding capacity, oil holding capacity, emulsification and/or gel formation. Indeed, the dietary fibre incorporated into food products (bakery products, dairy, jams, meats, soups) can modify textural properties, avoid syneresis (the separation of liquid from a gel caused by contraction), stabilise high fat food and emulsions, and improve shelf-life (ELLEUCH et al., 2011).

According to the described context, this study aimed to evaluate solvents for the extraction of Macauba pulp oil, with experiments conducted at low pressure, and the characterization of the extracts and defatted meals obtained.

MATERIALS AND METHODS

The fruits of *Aculeata acrocomia* were collected, about 20kg, in Cariri - Ceará. Fruits were sanitized; separation of the parts was carried out and pulp, with 29.51 ± 0.48 °Brix, was dried at 60°C for 8h, with humidity determined to be 3.6 ± 0.1 %. Pulp was ground using an electric mill (Marconi) and classified using sieves series Tyler (Bertel, ASTM) to produce particles with an average diameter of 0.5mm. For the extraction, the following solvents were used: ethyl acetate (99.5%, F Maia), *n*-hexane (98.5%, Synth) and isopropanol (99.8%, JT Baker). Analytical standards methyl heptadecanoate, 5 α -cholestane and β -carotene, and derivatising BF₃-methanol and

N,O-bis (trimethylsilyl) trifluoro-acetamide-BSTFA with trimethylchlorosilane-TMCS were obtained from Sigma-Aldrich with purity >99%.

Oil extraction was performed at 40°C, 40rpm and 4 hours on orbital shaker (Marconi, MA 839/A), where 5g of pulp was placed in flasks with a glass cover (250ml) together with solvents in the ratio 1:8 (weight of pulp: volume of solvent). The amount of solvent was selected after preliminary tests indicate that the increase in the ratio of 1:2 to 1:8 increases the extraction of oil; however, the use of higher ratios than 1:8 gave no significant differences in yield. After the extraction period, the samples were filtered and excess solvent in the filtrate was evaporated via the evaporator route (Marconi, MA120), and the remainder kept in an oven (Marconi, MA035) to constant weight. The extracts were stored under refrigeration (Consul) until the time of analysis. The yield was calculated as the ratio of mass of oil extracted and the mass of the pulp used.

The concentration of β -carotene in the extracts was determined at 450nm using a UV spectrophotometer (Femto 700 plus). Samples were prepared in *n*-hexane and β -carotene content was determined using calibration curve for concentrations of 1.0-100.0mg L⁻¹, which showed a regression coefficient of 0.998. The analysis of total flavonoids was performed according to the procedure reported by FRANCIS (1982). To determine the composition of fatty acids and free glycerol compounds, gas chromatographs coupled to a mass spectrophotometer were used (Thermo-Finnigan), with a capillary column Agilent HP-5MS (30m x 0.250mm x 0.25 μ m), injection 0.4 of μ L in split mode 1:10 and helium as a carrier gas, at a flow of 1mL min⁻¹. Identification of components present in the samples was performed by Xcalibur® software (Thermo Electron).

For determination of the fatty acids, the samples were derivatised following the method of AOCS Ce 2-66 (WALKER, 1990) and analysed using the following chromatographic conditions: initial temperature of the column 120°C, maintained at this temperature for 5 minutes, increasing to 180°C at a rate of 15°C min⁻¹ and for 240°C at a rate of 5°C min⁻¹, staying for five minutes. The free glycerol compounds were converted to silylated compounds, as outlined by FREITAS et al. (2008), with modifications. Approximately 30mg of oil was derivatised with 20 μ L of BSTFA/TMCS with the subsequent addition of analytical standards 5 α -cholestane and methyl heptadecanoate to quantify the phytosterols and free fatty acids, respectively. The solution was stirred and remained 30 minutes at 60°C. The analyses were performed under the following conditions: initial

temperature of the column 100°C, maintained at this temperature for 6 minutes, increasing to 230°C at a rate of 5°C min⁻¹ and 280°C at 15°C min⁻¹, where it was held for 15 minutes.

The defatted Macauba meal was dried in an oven with forced air circulation at 60°C (MA 035, Marconi). Dried material was ground in a knife mill type Willye (SL-031, Solab) and passed through a set of sieves with particle separation as a result of being subjected to vibration for 10min. Defatted meal with particle size of 100µm was used in the analysis. Hydration properties of the defatted meal were determined as per the procedure reported by ROBERTSON et al. (2000) and SEIBEL & BELIA (2009). Water retention capacity (WRC) was defined as the quantity of water that remains bound to the hydrated fibre following the application of an external force. Swelling capacity (SC) is a measure of the ratio of volume occupied when the sample is immersed in an excess of water and after equilibration to the actual weight (RAGHAVENDRA et al., 2006). Oil absorption capacity (OAC) was determined according to RAGHAVENDRA et al. (2006) and was calculated as the ratio of quantity of fat compared to the initial dry weight of residue. Emulsification activity (EA) was investigated using the method described by DENCH et al. (1981) and was expressed as a percentage. The physicochemical analyses were carried out according to the methodologies of AOAC (1990) and used the colorimeter (Minolta CR-400) to determine colour values, L*, a* and b*.

All experiments and analyzes were performed in triplicate and data were subjected to one-way ANOVA and Tukey tests (P>0.05), using Excel® 2010 software, to evaluate differences in the results when using different solvents.

RESULTS AND DISCUSSION

The results in oil yield using different solvents, β-carotene and flavonoids content in the

extracts obtained are shown in table 1. The results demonstrate higher yield for the use of isopropanol and reports that yield using ethyl acetate and *n*-hexane showed no significant difference (P>0.05). Oil extraction may be attributed to the polarity of the solvents (TAKEUCHI et al., 2009; RAMLUCKAN et al., 2014), the ethyl acetate has a higher polarity, followed by isopropanol, and *n*-hexane had apolar properties. The use of polar solvents allows high yields to be obtained due to the ability of these solvents to break up the forces that bind lipids in the vegetable matrix (AQUINO et al., 2011). The apolar solvents selectively extract surface lipids (RAMLUCKAN et al., 2014); since the isopropanol, as an alcohol, may have bipolar characteristics, with superior performance compared to other solvents, this is suitable for extraction of active compounds (ALMEIDA et al., 2012) and lipid fraction (RODRIGUEZ-SOLANA et al., 2014). Literature reports that the isopropanol was more efficient at removing soybean oil flakes (SETH et al., 2007) and seeds of *Crotalaria juncea* (DUTTA et al., 2014) compared to *n*-hexane. It can be seen in table 1 that oil yields between 23 and 27.43% were obtained, and the literature reports contents of 18.7% to 29% oil in the pulp of *Acrocomia aculeata* (COIMBRA & JORGE, 2012; CICONINI et al., 2013).

It was observed that ethyl acetate gave an extract with a higher concentration of flavonoids, followed by isopropanol and *n*-hexane, which is an expected result since flavonoids are polar molecules (HUBER & RODRIGUEZ-AMAYA, 2008). Regarding β-carotene content, isopropanol and *n*-hexane showed greater efficiency in the extraction of this compound, but there was no significant difference between the solvents. Carotenoids present in general molecules with nonpolar characteristics are easily extracted by *n*-hexane (JAIME et al., 2010; AQUINO et al., 2011; MUSTAPA et al., 2011); however, alcohols may have bipolar characteristics, dissolving easily nonpolar organic substances

Table 1 - Results from oil yield, β-carotene and flavonoids contents in pulp and extracts obtained with different solvents. Means followed by same letters (in each row) did not differ statistically (P>0.05).

	-----Solvent-----			
	Pulp	Ethyl acetate	<i>n</i> -hexane	Isopropanol
Yield (%)	25.72±0.02 ¹	22.97±0.33 ^a	23.39±0.15 ^a	27.43±0.33 ^b
β-carotene (mg 100g ⁻¹)	124.16±0.46	308.38±0.60 ^a	334.05±0.47 ^b	348.30±0.31 ^c
Flavonoids (mg 100g ⁻¹)	25.34±0.28	16.97±0.11 ^a	13.78±0.09 ^b	14.78±0.05 ^c

¹Obtained by Soxhlet with petroleum ether as solvent at reflux for 8 hours.

(TAKEUCHI et al., 2009). As a result, the β -carotene can be extracted by appropriate solvents such as isopropanol. Furthermore, SACHINDRA et al. (2006) evaluated the solubility of carotenoids in different solvents and reported a high solubility of this compound in isopropanol.

Table 2 shows the composition of extracts in terms of fatty acids and free glycerol compounds. Regarding fatty acid composition, it appears that, in general, the levels were not affected ($P>0.05$) by the solvent extractor. As indicated by the results presented in table 2, the extracts showed high contents of oleic acid (~60%) and palmitic acid (~27%), and low concentrations of linoleic acid (~6%) and linolenic (~3%). Other authors report similar results for the composition of Macauba pulp oil, such as the research of HIANE et al. (2005), which reported the predominance of acids oleic (62.2%) and palmitic (18.5%), and the study of AMARAL et al. (2011), which showed oleic (69.07%), palmitic (12.18%) and linoleic (6.77%) acids to be the main compounds in the oil composition.

To verify the results of quantification of free glycerol compounds shown in table 2, it appears that the oil extraction using isopropanol generated higher levels of these compounds ($P<0.05$). Results obtained by ELLER et al. (2010) suggested a greater extraction of free glycerol compounds using a polar solvent. Percentage of free fatty acids (FFAs) in the extracts ranged from 1.13 to 1.34%. COIMBRA & JORGE (2011) and HIANE et al. (2005) reported levels of free fatty acids in the Macauba pulp oil extracted by Soxhlet of 9.43% and 0.83%,

respectively. Differences in the levels reported in the literature and obtained in this study may be explained due to the different regions, climatic conditions, drying times and temperatures to which the pulp was exposed before the oil extraction, even when the extraction method is the same (CICONINI et al., 2013). Regarding phytosterols, campesterol, stigmasterol and β -sitosterol have been identified in macauba pulp oil, with total contents ranging from 76.85 to 104.15mg 100g⁻¹. According to our findings, there probably has been no research reporting the quantification of phytosterols in Macauba pulp oil.

Physic-chemical characterization of pulp (without oil removal) and meal obtained as coproducts of the extraction with different solvents is shown in table 3. It can be seen that the removal of oil made possible to increase the levels of ash, protein and fibre in the meal, without any statistically significant difference between the different solvents used. Comparing the colour results shown in table 3, it is clear that there is a decrease of b^* (ranging from yellow/+ to green/-) with the removal of oil. Similarly, bran has a sharp white colour (L^*) and the red tone (a^*) is reduced with oil extraction.

Regarding the technological characteristics, it was reported that the oil removal increased water retention capacity and did not affect the other technological characteristics; also, the type of solvent does not influence the technological characteristics of the meal under the conditions evaluated in this study, as can be seen in table 1. The water retention capacity of Macauba defatted meal (3.98g g⁻¹ dry matter) was higher compared to other dietary fibres such

Table 2 - Quantification of fatty acids, free fatty acids (FFA) and phytosterols in the Macauba pulp oil obtained with different solvents. Means followed by same letters (in each row) did not differ statistically ($P>0.05$).

Solvent		Ethyl acetate	<i>n</i> -hexane	Isopropanol
Fatty acids ¹	Caprylic	0.27±0.03 ^a	0.13±0.01 ^b	0,09±0,01 ^b
	Capric	0.04±0.01 ^a	0.06±0.01 ^a	0,03±0,01 ^b
	Lauric	0.09±0.01 ^a	0.11±0.01 ^a	0,11±0,01 ^a
	Myristic	0.22±0.01 ^a	0.18±0.02 ^a	0,18±0,01 ^a
	Palmitic	27.77±0.29 ^a	27.39±0.12 ^a	27,24±0,42 ^a
	Stearic	1.81±0.29 ^a	1.80±0.14 ^a	2,48±0,46 ^a
	Oleic	61.03±0.48 ^a	61.15±0.15 ^a	60,17±0,51 ^a
	Linoleic	5.79±0.03 ^a	6.34±0.19 ^b	5,72±0,07 ^a
	Linolenic	2.96±0.11 ^{ab}	2.81±0.05 ^a	3,09±0,07 ^b
FFA ¹		1,13±0,01 ^a	1.15±0.02 ^a	1.34±0.01 ^b
Phytosterols ²		82,45±0,71 ^a	76.85±0.48 ^b	104.15±0.73 ^c

¹Results in g 100g⁻¹ of oil; ²Results in mg of PT 100g⁻¹ of oil.

Table 3 - Physicochemical and technological characteristics and color of Macauba pulp and defatted meal (dry basis). Means followed by same letters (in each row) did not differ statistically ($P>0.05$).

	-----Solvent-----			
	Pulp	Ethyl acetate	<i>n</i> -hexane	Isopropanol
Ash	2.42±0.03 ^a	3.96±0.12 ^b	3.86±0.24 ^b	4.63±0.44 ^b
Crude fiber	8.76±0.50 ^a	11.84±0.04 ^b	11.41±0.15 ^b	11.39±0.34 ^b
Total lipids	26.31±0.02 ^a	1.80±0.20 ^b	1.80±0.39 ^b	0.87±0.09 ^b
Crude protein	3.79±0.07 ^a	6.58±0.04 ^a	6.71±0.03 ^a	6.65±0.06 ^a
Color	*L	67.04±0.30 ^a	80.85±1.19 ^b	80.17±0.66 ^b
	*a	4.29±0.04 ^a	1.15±0.02 ^b	0.94±0.0 ^c
	*b	56.69±1.06 ^a	21.80±0.84 ^b	21.72±0.91 ^b
WRC (g water g ⁻¹ dry matter)	2.69±0.05 ^a	3.61±0.26 ^b	3.89±0.05 ^b	3.98±0.05 ^b
OAC (g oil g ⁻¹ dry matter)	2.25±0.39 ^a	2.86±0.03 ^a	2.88±0.05 ^a	3.01±0.24 ^a
SC (mL g ⁻¹ dry matter)	3.75±0.35 ^a	3.75±0.35 ^a	4.0±0.0 ^a	4.75±0.35 ^a
EA (%)	100 ^a	100 ^a	100 ^a	100 ^a

WRC: water retention capacity; OAC: oil absorption capacity; SC: swellin capacity; EA: Emulsification activity.

as apple, pea, wheat and carrot (values ranged 2.5 to 3.5g g⁻¹ dry matter) (THEBAUDIN et al., 1997). The oil absorption capacity (3.01g g⁻¹ dry matter) and emulsification activity (100%) were higher than those reported for defatted soy flour 2.4g g⁻¹ dry matter and 56.8%, respectively (SIEBEL, 2009). Ability of defatted meal to absorb and retain water and oil may help to improve the binding capacity, enhance flavour retention, improve mouth feel and reduce moisture and fat losses of extended meat products (KAUR et al., 2013). Values of technological characteristics found in this research suggested some possibilities about the use of Macauba defatted meal as ingredients in food products, such as meat products, mayonnaise and bakery products.

CONCLUSION

The Macauba pulp oil obtained in this research used different solvents and proved to be rich in β -carotene, flavonoids and monounsaturated fatty acids. Isopropanol showed higher oil yield, β -carotene and free glycerol compounds. Ethyl acetate provided extracts with higher concentrations of flavonoids. Results showed that Macauba oil with good quality, before the evaluated characteristics, can be obtained by extraction technique adopted. The meals presented increased water retention capacity when compared to the pulp resulting from the change in centesimal composition and presents technological characteristics for use in food formulations.

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