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Bioactive compounds and composition of *Elaeis oleifera* mesocarp oil extracted by hydraulic pressing

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SUMMARY: *Elaeis oleifera* oil has stood out for its high contents of carotenoids, tocochromanols, oleic and linoleic acids and low content of saturated fatty acids. The content of these bioactive compounds depends on the oil extraction process and the data available is related to solvent extraction. Pressing is the current industrial oil extraction process for palm fruits. *E. oleifera* bunches were harvested on two different days at an interval of two months. They were then sterilized and the fruits were submitted to hydraulic pressing and the oil from the mesocarp was evaluated. The main fatty acids of two samples were C18:1 (54.72 – 55.69%), C16:0 (23.72 – 24.23%) and C18:2 (15.47 – 16.95%). The alpha- and beta-carotene contents were 620-725 µg/g and 1,358– 1,403 µg/g, respectively. Gamma-tocotrienol accounted for 799 and 1,066 µg/g. The oil is a concentrate of pro-vitamin A and rich in gamma-tocotrienol and its low oil acidity allows for the consumption of the crude oil.

KEYWORDS*: Acidity; Carotenoids; Liquid chromatography; Pro-vitamin A; Tocotrienols.*

RESUMEN: ***Compuestos bioactivos y composición del aceite de mesocarpio de Elaeis oleifera extraído por prensado hidráulico***. El aceite de *Elaeis oleifera* destaca por su alto contenido en carotenoides, tococromanoles, ácido oleico y linoleico y bajo contenido en ácidos grasos saturados. El contenido de estos compuestos bioactivos depende del método de extracción del aceite y los datos disponibles están relacionados con la extracción con solventes. El prensado es el proceso actual de extracción de aceites de los frutos de la palma. Los racimos de *E. oleifera* fueron esterilizados y los frutos fueron sometidos a prensado hidráulico en dos días diferentes con un intervalo de dos meses y se evaluó el aceite de mesocarpio. Los principales ácidos grasos de las muestras fueron C18:1 (54,72 – 55,69%), C16:0 (23,72 – 24,23) y C18:2 (15,47 – 16,95%). El contenido de alfa y beta-caroteno fue de 620-725 µg/g y de 1358-1403 µg/g, respectivamente. El gamma-tocotrienol presentó valores de 799 y 1.066 µg/g. El aceite es un concentrado de provitamina A y rico en gamma-tocotrienol y su baja acidez permite el consumo del aceite crudo.

PALABRAS CLAVE*: Acidez; Carotenoides; Cromatografia líquida; Pro-vitamina A; Tocotrienoles.*

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1. INTRODUCTION

Caiaué (*Elaeis oleifera* (Kunth) Cortés) is a species of palm which is native to Central and South America and grows spontaneously throughout humid areas of the Amazon. Also known as American palm oil, it is exploited for domestic consumption by traditional communities but has so far not been cultivated on a commercial scale, as it shows significantly lower oil production compared to the main cultivated African palm (*Elaeis guineensis*). *Elaeis oleifera* (Eo) is grown at experimental stations since its germoplasm represents a highly valuable genetic resource to breed interspecific hybrids with *E. guineeensis* (Eg) due to some interesting traits such as slow yearly height increment, disease resistance and great oil quality (Choo *et al*., 1997; Mohd *et al*., 2000; Rios *et al*., 2011).

Despite exhibiting a lower production capacity, Caiaué mesocarp oil is more unsaturated than Eg

mesocarp oil and presents higher oleic and lower palmitic acid contents. In this way, Caiaué mesocarp oil meets the increasing requirements for healthier oils/fats used in the food industry like conventional palm oil and its derivatives are rich in saturated fatty acids. However, there is currently a great deal of controversy about the reduction of saturated fats in the diet and its effect on various diseases, as well as in relation to the replacement strategy (Astrup *et al.*, 2020).

Crude palm oil (*Eg*) is the richest natural source of beta-carotene (carotenoids 500-700 ppm) and shows high contents of tocochromanols (500–1000 ppm), phytosterols, squalene, phenolics and coenzyme Q10. The alpha- and beta-carotene are pro-vitamin A and carotenoids have been related to other benefits to human health such as cardioprotective effects in ischemic heart disease, antiatherogenic, antihemorrhagic, antihypertensive and anticancer (Tan *et al.*, 2021). However, España *et al*. (2018) reported the total carotene of *E. oleifera* oil from five accessions (Coari, Manicore, Anori, Autazes and Careiro) to vary from 1,527 to 3,344 ppm, indicating higher contents than palm oil (Eg).

Tocochromanols are a group comprising of eight naturally occurring lipid-soluble compounds, four tocopherols and four tocotrienols. These micronutrients show activity in the form of vitamin E besides antioxidant activity in oils and *in vivo* and neuroprotective, anti-cancer, anti-inflammatory, anti-hyperlipidemic, anti-osteoporotic, anti-hyperglycemic and cholesterol lowering properties (Ahsan *et al*., 2015). The content of tocochromanols of the hybrid of *E. guineeensis* × *E. oleifera* (Eo×Eg) and Eo oils is expected to be higher than palm oil (Eg), according to Jalani *et al.* (1997), but the method of oil extraction was not reported. According to Irias-Mata *et al*. (2017), the tocochromanol content from Eo mesocarp oil obtained by solvent extraction (hexane) was 2.5 times higher than screw-pressed oil. The results available for fatty acid, carotene and tocochromanol profile of Eo were obtained for solvent extraction on lab scale (Lieb *et al*., 2017; España *et al*., 2018; Chaves *et al*., 2018).

There is little information about the fatty acid profile and carotenoid and tocochromanol contents in Caiaué oil obtained by pressing. As far as we know, there is only one study about Eo mesocarp oil extracted by pressing (Irias Mata *et al*., 2017), and it only reported results on tocotrienols.

At present, crude oils for human consumption are obtained by pressing (screw or hydraulic pressing) and the oils from solvent extraction are submitted to oil refining. Refining is the usual process for obtaining low acidity and light-colored oils but this process destroys the carotenes and reduces the amount of tocochromanols (Basiron, 2005). Pressing is the usual process for olive, palm oil, palm kernel oil, cocoa butter, nuts etc. In Brazil, there is a market for oils from pressing including crude palm oil.

Recently, some Brazilian companies started extracting Caiaué oil from mesocarp expecting to obtain a more nutritionally attractive composition when compared to African palm oil. Therefore, the objective of this study was to evaluate the crude pressed Caiaué oil regarding its quality parameters, fatty acid composition, carotenoids and tocochromanol contents.

2. MATERIALS AND METHODS

The study has been registered in the National System of Genetic Resource Management and Associated Traditional Knowledge (SisGen) of Brazil under number AF170FD. The bunches of Eo palms were harvested at the field of the company Denpasa, Dende do Pará S/A (Para State, Brazil) on two different days with an interval of two months. The identification of the accession of Eo had not yet been evaluated at this time.

2.1. Oil extraction

The criterion for bunch harvesting is the same for African palm oil and the ripe bunches were collected when they had 5–10 detached fruits that had fallen to the ground. After harvesting, a heat treatment was carried out on twelve bunches with boiling water for one hour. The fruits were detached and pressed using a hydraulic press consisting of a stainless-steel square box of 40×40×40 cm, specially developed for Eo fruits. The separation of oil and water was carried out by centrifugation. The oil was kept frozen until the analysis. The assays were performed on two different days at an interval of two months and the samples were identified as 1 (day 1) and 2 (day 2) and each sample was collected in triplicate, and the analyses were performed in triplicate or quadruplicate for each plastic bottle totaling nine or twelve replicates.

2.2. Physical characteristics and quality analysis

Free fatty acid (FFA), expressed as percentage of oleic acid, was determined according to method Ca 5a-40 (AOCS, 2009). The refractive index (RI) was determined by refractometry at 40 °C on a Bausch & Lomb Abbe refractometer, according to method Cc7-25 (AOCS, 2009). Relative Density was determined at 20 °C by a digital densimeter Anton Paar DMA-46.

2.3. Fatty acid profile

The analysis of the fatty acid profile of oil is accredited to ISO/IEC 17025. The methyl esters were obtained according to Antoniassi *et al*. (2018b) and analyzed by gas chromatography on an Agilent 7890, equipped with a flame ionization detector operating at 280 °C and a HP FFAP capillary column (25 m x 0.2 mm x 0.30 μm). The oven temperature was programmed as follow: initial temperature of 150 °C for 1 min; from 150 to 180 °C at a rate of 30 °C/min; from 180 to 200 °C (20 °C/min); 200 to 230 °C (3 °C/min) and held at the final temperature of 230 °C for 10 min. A pressure ramp was used as described: initial pressure 15 psi for 10 min, from 15 to 25 psi with a 5 psi/min ramp rate and a final pressure of 25 psi for 11 min. The injector was set to 250 °C and operated in a 1:50 split mode and 1 μL of sample was injected. Identification was performed by comparing retention times with the standards of NU-CHEK PREP, Inc. (Elysian, MN, USA) and Supelco (Bellefonte, PA, USA) and quantification was performed by internal normalization.

The iodine value and saponification values were calculated based on the fatty acid composition.

2.4. Carotenes and tocochromanols

The analyzes were performed simultaneously using a Waters Technologies Alliance 2695 liquid chromatograph (HPLC) (Milford, Massachusetts, USA), equipped with a quaternary pump system, on-line degasser, column furnace, automatic injection Rheodyne valve (Rheodyne LCC, Rohnert Park, USA), connected to Waters 2998 photodiode array (PDA) and Waters 2475 fluorescence detector. The oil was dissolved with acetone. The compounds were separated on a YMC C30 reverse-phase analytical column (0.25 m × 3.0 mm I.D., 5.0 μm particle size) (Kyoto, JAPAN) conditioned to 35 °C. Samples were kept to 15 °C. The separation was performed using a gradient (flow 0.8 mL/min) with the mobile phase methanol/methyl tert-butyl ether (9/1, v/v) for 20 min, methanol/methyl tert-butyl ether (1/9, v/v) for 5 min, and methanol/methyl tert-butyl ether (9/1, v/v) for 3 min totaling 28 min. The identification of the compounds was made by comparing the sample retention time with the peak retention time of the carotenoid and tocochromanol standards and by the absorption spectra of the carotenes (Davis, 1976). Chromatograms were processed at 290 nm (excitation) and 330 nm (emission) at the fluorescence detector for tocopherols; and, between 200 and 800 nm, in scan mode, in the PDA detector and quantification at 450 nm. The quantification of tocopherols was done through external standardization. The concentration of each tocochromanol stock solution was calculated according to AOCS (2009). For total carotenes, quantification was performed by spectrophotometry considering the wavelengths, molar absorptivities and the solvent used in the dilution, according to Davies (1976). The calculation of Retinol Activity equivalent was calculated based on the conversion factors reported by Charrondière *et al*. (2012).

2.5. Statistics

Analysis of variance and Tukey test were performed using Statgraphics (Statgraphics Technologies Inc) at a significance level of 0.05.

3. RESULTS AND DISCUSSION

3.1. Oil composition

The main fatty acids were C18:1 (54.72 – 55.69%), C16:0 (23.72 – 24.23), C18:2 (15.47 – 16.95%) and C18:0 (1.91 – 1.97%), which accounted for about 97% of total fatty acids. The C18:1 comprised oleic (C18:1 *cis*-9) and *cis*-vaccenic (C18:1 *cis*-11). C18:3 was quantified up to 0.7% and minor fatty acids below 0.5% were C14:0, C16:1, C17:0, C17:1, C20:0 and C20:1. C22:0 and C24:0 were quantified as traces. There was no difference among the caiaué oil samples ($p < 0.05$) except for C18:0 (1.97 – 1.91%) and C20:0 (0.18 – 0.15%). The samples were obtained within a two-month interval from mature bunches available at the time and not neces-

sarily from the same plants. However, the low variation observed among samples indicated low variability among different plants whose accession had not been identified at this time. (Table 1).

The fatty acid profile of pressed Caiaué oil was in the ranges reported for the oil obtained by solvent extraction from the dried mesocarp of five Brazilian accessions of Eo cultivated in Brazil (España *et al*., 2018). In addition, the results were consistent with the variation observed for Eo accessions from Panama, Suriname, Brazil, Ecuador, Costa Rica, Colombia grown in Costa Rica (Lieb *et al*., 2017) and for genotypes from Brazil, Peru and Colombia grown in Colombia (Chaves *et al*., 2018), both by solvent extraction. The same was observed for the results reported by Mohd *et al.* (2000) for progenies of Eo from Colombia, Panama, Costa Rica and Honduras cultivated in Malaysia, although the method of extraction was not reported. The oil extraction with solvents such as hexane for industrial extraction or for laboratory purposes (e.g. Soxhlet extraction with petroleum ether) are very effective methods for oil recovery since the meal presents less than 1% oil. However, pressing is the usual process for oil extraction from palm fruits and the fiber from palm oil processing may contain 5-6% residual oil (Basiron, 2005). Therefore, the fatty acid profile of pressed oil is more reliable for nutritional purposes. As far as we know, this is the first report on the fatty acid from *E. oleifera* oil obtained by pressing.

The variation observed among studies for the fatty acid profile of Eo could be due to genetic factors, growing conditions and according to the extraction protocols employed. The fatty acid profile of Eo oil is expected to be more unsaturated than palm oil and the Eo×Eg hybrid according to Jalani *et al*. (1997), Choo *et al*. (1997) and Yap *et al.* (1991). According to Rios *et al.* (2011), the highest contents of oleic and

Table 1. Fatty acids composition (g fatty acid/total fatty acids) of two samples of pressed Caiaué oils[1] (*Elaeis oleifera*)

Fatty acid	*Elaeis oleifera*		Codex Alimentarius (FAO, 2022)		
	Sample 1	Sample 2	Palm olein	Palm Superolein	Palm oil with a higher oleic acid
C14:0	0.31±0.03[a]	0.33±0.01[a]	0.5–1.5	0.5–1.5	<0.8
C16:0	23.72±0.72[a]	24.23±0.13[a]	38.0–43.0	30.0–39.0	23.0–3.08
C16:1	0.51±0.03[a]	0.53±0.03[a]	<0.6	<0.5	
C17:0	Nd-0.07	Nd-0.07			
C17:1	Nd-0.06	ND-0.06			
C18:0	1.97±0.03[a]	1.91±0.01[b]	3.5–5.0	2.8–4.5	1.5–4.5
C18:1	54.72±1.10[a]	55.69±0.0.05[a]	39.8–46.0	43.0–49.5	48.0–60.0
C18:2	16.95±1.41[a]	15.47±0.01[a]	10–13.5	10.5–15.0	9.0–17.0
C18:3	0.68±0.07[a]	0.72±0.00[a]	<0.6	<0.5	<0.6
C20:0	0.18±0.01[a]	0.15±0.00[b]			
C20:1	0.12±0.01[a]	0.11±0.02[a]			
C22:0	tr	nd			
C24:0	tr	nd			
SFA	26.27±0.61[a]	26.65±0.13[a]			
MUFA	55.36±1.08[a]	56.34±0.04[a]			
PUFA	17.63±1.48[a]	16.19±0.01[a]			
SFA/UFA	0.4±0.01	0.4±0.00			
Iodine value[2] (g/100g)	79	77	≥56	≥60	58–75
Saponification Value[2] (mg KOH/g)	196	196	194–202	180–205	189–199

[1]Results expressed as the average and standard deviation of nine replicates

[2] Calculated. Different Means with different lowercase letters in the same row are significantly different ($P < 0.05$) by the Tukey test.

SFA – Saturated fatty acids; MUFA – Monounsaturated Fatty acids; PUFA – Polyunsaturated fatty acids; UFA – Unsaturated Fatty acids

linoleic acids and the lower content of palmitic acid of Eo mesocarp oil are useful characteristics to transmit to interspecific hybrids with Eg. In addition, Eo is useful for breeding due to its resistance to Bud rot-type (fatal yellowing) and red ring diseases, besides the smaller size palm tree.

The saturated fatty acids (SFA) comprised 26.4 and 26.7% and the SFA/UFA (unsaturated fatty acids) ratio was 0.4 and in the range of 0.3 to 0.5 reported by Lieb et al. (2017) for Eo genotypes and 1.2 for Eg. The high UFA concentration in Caiaué oil increases its fluidity and minimizes the formation of stearin, a high melting point fraction responsible for the semi-solid physical state of similar oils during storage that could be undesirable for some food applications.

The fatty acids of pressed Caiaué oil were in the range reported for "Palm oil with higher oleic acid" derived from the fleshy mesocarp of hybrid palm fruit (Eo × Eg) by Codex Alimentarius (FAO, 2022) which covered the genetic variation presented for different countries. However, the saturated and unsaturated fatty acids of pressed Caiaué oil were in the lower and higher limits of these reported ranges, respectively (Table 1). Palm olein and Palm super olein are commercial products from palm oil fractionation for obtaining liquid and clear oil with a lower content of saturated fatty acids than palm oil. Caiaué oil showed significantly lower content of saturated (C16:0 and C18:0), higher content of mono- and polyunsaturated fatty acids (C18:1 and C18:2) and higher Iodine value (77-79 g/100 g) than these palm oleins (Table 1). In addition, Caiaué oil is liquid at room temperature, indicating that it can meet the same food applications as these two products. The saponification value was in the range of palm oil and its oleins and Eo×Eg oils.

No differences wereas observed for Refractive index or Relative density between the two samples of pressed Eo oil (Table 2). The Refractive index depends on the fatty acid profile and the results were in the range described for palm oil with higher oleic acid by Codex Alimentarius (FAO, 2022), and higher than palm olein and palm superolein. The relative density was similar to palm olein. The usual temperature for liquid oils analysis is 40 ºC while 50 ºC is suitable for fats. In this way, the comparison is not possible.

The samples presented low free fatty acid (FFA) content (1.06 and 1.33%, as oleic acid) (Table 2). The acidity of Caiaué oil met the limit (2%) reported by Brazilian Regulation for edible oil obtained by pressing (Brasil, 2021). Oil acidity is related to the action of lipases that promote triacylglycerol hydrolysis. After harvest and due to the cell disruption of mesocarp by handling or processing, the enzymes are activated. Therefore the heat treatment is useful for palm fruits with high moisture to control the oil's acidity (Basiron, 2005). Udeh and Obibuzor (2017) found a FFA range of 0.41 – 1.48% (as palmitic acid) for eight samples of Eo oil while a wider range was described for Eo×Eg hybrids and palm oil (2-2.2 and 3.2-3.6%, respectively) by Mozzon *et al.* (2013). The oil acidity imparts a flavor that may be undesirable in crude oils and in the oil refining process, the higher the acidity, the greater the loss of neutral oil, reducing the yield of the process. Then, Eo genotypes with low lipase activity associated with good practices of bunch harvest followed by heat treatment are desirable to produce oil with low acidity (Basiron, 2005).

Table 2. Physical characteristics and quality parameters of pressed Caiaué oil[1] ***(Elaeis oleifera)***

Samples	Refractive index (40 °C)	Relative density (40 °C /20 °C)	Free fatty acid (as % oleic acid)
E. oleifera Sample 1	1.4615±0.000[a]	0.899±0.000[a]	1.06±0.06[a]
E. oleifera Sample 2	1.4618±0.0000[a]	0.898±0.000[a]	1.33±0.07[b]
Palm oil[2]	1.454- 1.456 (50ºC)	0.891-0.899 (50ºC/20 ºC)	
Palm oil with a higher oleic acid[2]	1.459-1.462	0.896– 0.910 (50ºC/20 ºC)	
Palm olein[2]	1.458-1.460	0.899-0.920	
Palm superolein[2]	1.459-1.460	0.900-0.925	

[1]Results expressed as the average and standard deviation of nine replicates for acidity and three replicates for refractive index and density
[2] Codex Alimentarius (FAO, 2022)
Different means with different lowercase letters in the same column are significantly different ($P < 0.05$) by the Tukey test.

3.2. Carotenes

The carotenes of pressed Caiaué oil showed significant differences between the two samples analyzed as reported in Table 3 ($p < 0.05$). Alpha-carotene and beta-carotene accounted for approximately 92% of the total carotenoids, with averages of 620-725 µg/g and 1,358– 1,403 µg/g of oil, respectively, while the total carotene content was 2,145–2,330 µg/g of oil. The differences observed between the two samples could be due to maturity at harvest, harvesting and post-harvest handling, processing, and storage. Ripening is a main factor for enhanced carotenogenesis and carotenoids increase markedly in number and quantity (Rodriguez-Amaya *et al.,* 2008). The comparison between the two samples regarding carotenes, tocochromanols and acidity will be addressed in the next section.

The long-established function of carotenoids in terms of human health is the activity of provitamin A. In recent years, the focus has been on reduction of the risk of developing chronic degenerative diseases (Tan *et al*., 2021). The Recommended Dietary Allowance (RDA) of vitamin A for men and women is 900 and 700 µg retinol activity equivalents (RAE)/day, respectively (Institute of Medicine, 2001). Regarding the contribution of pressed Caiaué oil as a source of vitamin A, the consumption of 8 mL is equivalent to 1000 µg of RAE and meets the RDA for both men and women. The RAE of the two samples was 139 and 147 µg/g (Table 3). The composition of oils from native palm fruits makes them a new source of high-added-value phytochemicals. In addition, lipids are known to stimulate the absorption of carotenoids, so they may have the advantage of greater bioavailability (Rodriguez-Amaya *et al*., 2008; Santos *et al.*, 2015).

The results were higher than all fruits, vegetables and Brazilian foods reported by Rodriguez-Amaya *et al.* (2008). The analysis of the carotenoid content in mesocarp oils from different Brazilian palms (*Astrocaryum vulgare, Bactris gasipaes, Maximiliana maripa, Oenocarpus bacaba* and *Mauritia flexuosa*) showed large variations ranging from 3 to 567 µg/g of beta-carotene and negligible amounts of alpha-carotene and beta-cryptoxanthin (extraction by Soxhlet with ethyl ether and the HPLC method) (Santos *et al.*, 2015). Speranza *et al.* (2016) reported 781 µg/g of beta-carotene for *Mauritia flexuosa* (spectrophotometric method). Although there is potential for the application of Brazilian palm mesocarp oils, the pressed Caiaué oil is a remarkable source of pro-vitamin A.

The oil from the mesocarp of fruits of *Elaeis* palms is the main source of pro-vitamin A carotenoids. Trujillo-Quijano *et al.* (1990), Yap *et al*. (1991), Jalani *et al*. (1997) and Choo *et al.* (1997), compared the total amount and profile of carotenoids in oils and found a higher level of carotenoids (up to 4,600 µg/g of oil) for Eo than Eg and the hybrid Eo×Eg. However, the content of carotenoids depends on the extraction method, which was not informed by Jalani *et al*. (1997) and Choo *et al*. (1997), while Yap *et al*. (1991) reported extraction by Soxhlet (hexane). Trujillo-Quijano *et al*. (1990), reported a higher content of carotenoids (1577 µg/g) for the oil from Eo and up to 6-fold than Eg, although the extraction was performed by the Bligh and Dyer method (Bligh and Dyer, 1959). Silva *et al.* (2020) employed three different protocols for oil extraction and the highest content of beta-carotene (spectrophotometric method) was found for Eo compared to Eg and their interspecific hybrid Eo×Eg, but a significant difference was observed among the

Table 3. Carotenoid composition (µg/g)[1] and Retinol Activity equivalent[2] of pressed Caiaué oil (*Elaeis oleifera*)

Carotene	*E. oleifera*	
	Sample 1	**Sample 2**
All-trans-alpha-carotene	620±35[a]	725±8[b]
All-trans-beta-carotene	1,358±32[a]	1,403±17[b]
Total carotene	2,145±54[a]	2,330±21[b]
Retinol Activity equivalent (µg/g)	139	147

[1]Values represent means ± standard deviation of twelve replicates

[2] Calculated for the average of each sample based on the conversion factors reported by Charrondière *et al.* (2012). Different means with different lowercase letters in the same row are significantly different ($P < 0.05$) by Tukey's test.

extraction methods. Then, the results of the carotene content in oil obtained by solvent extraction from mesocarp is useful for genotype comparison but the industrial processing of palm bunches requires sterilization (heat treatment) for enzyme inactivation and the temperature of screw pressing and centrifugation usually reach 90 ºC (Basiron, 2005). As far as we know, this is the first report on the carotenes (HPLC method) of pressed *E. oleifera* oil.

The comparing among Eo genotypes for five Brazilian accessions of *E. oleifera* cultivated in Brazil revealed that the carotene content varied from 1527 to 3344 µg/g (España *et al.,* 2018). These results were obtained by spectrophotometric analysis from solvent-extracted oil.

The comparison among available results is difficult. Besides the differences in oil extraction protocols, there are different strategies to perform carotene quantification. The HPLC method provides separation, then the quantification of individual carotenoids is feasible. The spectrophotometric method requires the molar absorptivity of the predominant carotene as an estimate but the result should be expressed as total carotenes instead of content of beta-carotene.

The difference observed between the sum of alpha- and beta-carotene and total carotenes (Table 3 and Figure 1) is due to the isomerization of the original carotenes present in the mesocarp and these compounds showed no pro-vitamin A activity. The isomerization is consistent with the heat treatment of the bunches and it was observed for the screw-pressed Eo×Eg hybrid palm oil by Antoniassi *et al.* (2018a).

3.3. Tocochromanols

The two samples of *Eo* oil presented the same profile of tocochromanols, three forms of tocotrienols (alpha, delta and gamma) and two forms of tocopherols (alpha and gamma) (Table 4). Significant differences ($p < 0.05$) between the tocochromanol concentrations of the two samples were observed for alpha-tocotrienol, gamma-tocotrienol and total tocochromanol contents. Gamma-tocotrienol was the most abundant of the isomers (circa 80% of the total tocochromanols), followed by alpha-tocotrienol (14.0–17.6%). Total tocochromanols accounted for 979 and 1330 µg/g and gamma-tocotrienol corresponded to 799 and 1066 µg/g.

The screw-pressed oils from Costa Rica presented 599 and 211 µg/g of gamma-tocotrienol for Eo samples, while for Eg, it varied from 70 to 90 µg/g and EoxEg hybrid showed an intermediate amount of 146 µg/g, but lower than the results obtained in this work. Solvent extraction yielded 2 to 2.5 times more tocochromanols than the screw-press method (Irias-Mata *et al.*, 2017).

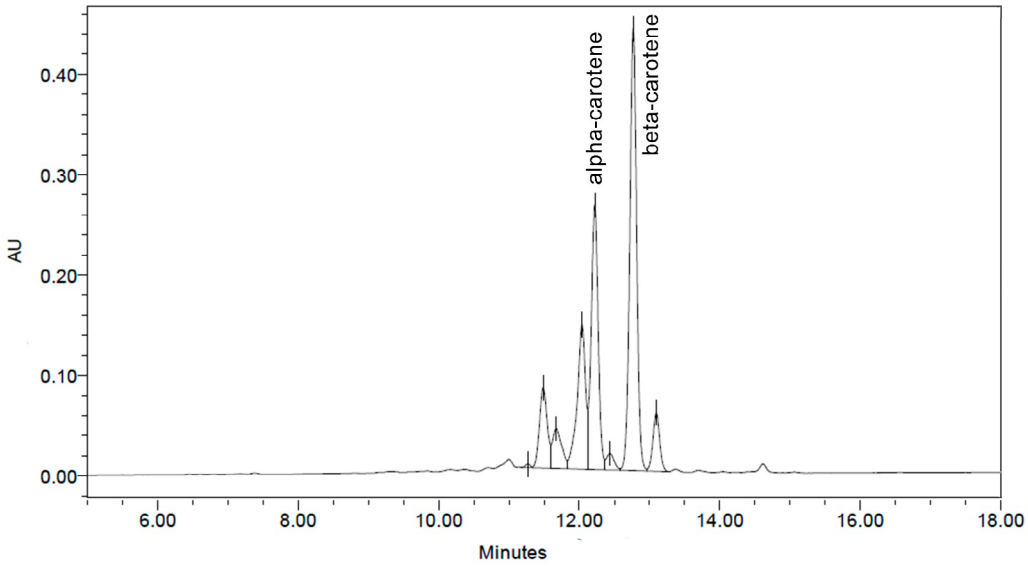

Figure 1. Chromatogram by HPLC of pressed Caiaué oil for carotenoids with Photo diode Array detector (450 nm).

Chaves *et al.* (2018) reported gamma-tocotrienol (376 to 827 µg/g) and alpha-tocotrienol (45–263 µg/g) for genotypes of Eo from Brazil, Peru and Colombia grown in Colombia but the oil was obtained by solvent extraction. Regarding the *Elaeis* species, Choo *et al.* (1997) reported a similar range of 700–1000 µg/g of tocochromanols. Jalani *et al.* (1997), quoted a lower range for Eg (600–1000 µg/g) and higher for Eo and Eo×Eg hydrid oils (600–1000 µg/g), although the extraction method was not mentioned.

Caiaué oil presented a higher content of gamma-tocotrienol and in the range of alpha-tocotrienol reported for Palm Olein, Palm Superolein and Eo×Eg hybrid oil by Codex Alimentarius (FAO, 2022) (Table 4). The comparison should be among crude oils because refining is a usual process for palm oil in order to obtain low acidity and light-colored oils. Refining destroys the carotenes and reduces tocochromanol content by 50%. This is an advantage of crude oils because tocopherols and tocotrienols

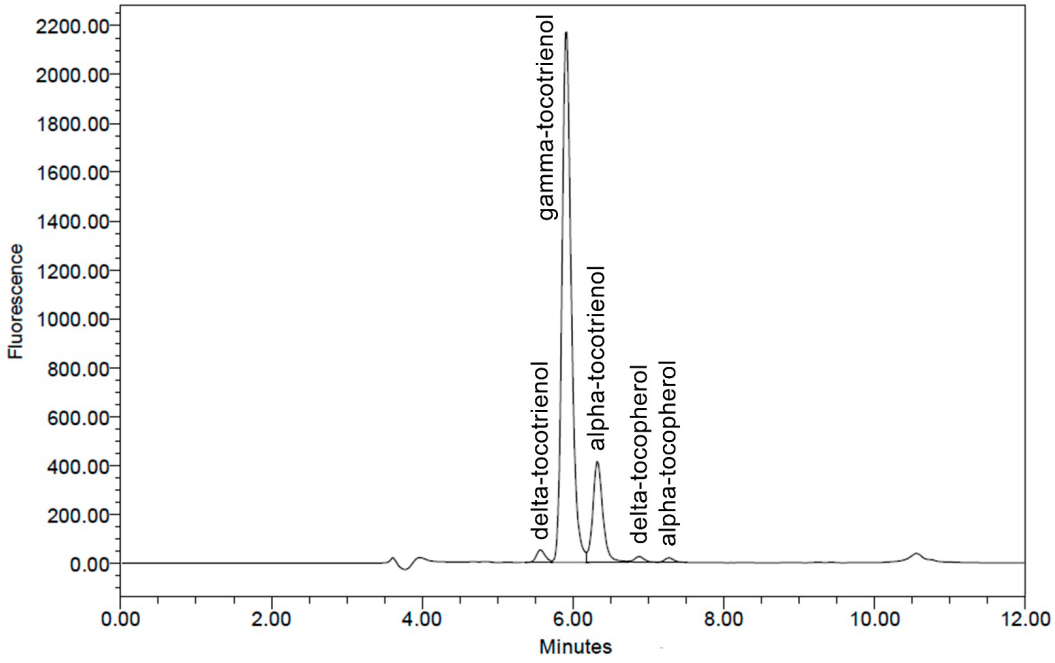

Figure 2. Chromatogram by HPLC of pressed Caiaué oil for tocochromanols with Fluorescence detector.

Table 4. Concentrations of tocotrienols and tocopherols of pressed Caiaué oil (*Elaeis oleifera*)[1]

Tocochromanols	*Elaeis oleifera*				Codex Alimentarius (FAO, 2022)		
	Sample 1		Sample 2		Palm olein	Palm Superolein	Palm oil with a higher oleic acid
	µg/g	%	µg/g	%	µg/g	µg/g	µg/g
Delta-tocotrienol	23±15[a]	2.3	20±2[a]	1.5	40–120	60–120	33–86
Gamma-tocotrienol	799±73[a]	81.6	1,066±27[b]	80.2	20-700	230–420	406–887
Alpha-tocotrienol	137±17[a]	14.0	234±6[b]	17.6	50-500	170–300	74–256
Gamma-tocopherol	12±6[a]	1.2	7±2[a]	0.5	<100	<40	4–138
Alpha-tocopherol	9±4[a]	0.9	3±2[a]	0.2	30-280	170–300	49–188
Total tocromanols	979±104[a]	100.0	1,330±29[b]	100.0			

[1]Values represent means ± standard deviation of twelve replicates
[2] Total tocochromanols is the sum of delta, gamma and alpha-tocotrienols, alpha and gamma tocopherols.
Different means with different lowercase letters in the same row are significantly different (P < 0.05) by theTukey test.

are antioxidants and the synergism among tocochromanols, carotenes and the combination with the fatty acid profile provide high oxidative stability (Basiron, 2005). However, the acidity of *Elaeis oleifera* oil is expected to be lower than palm oil due to the low lipase activity and the result obtained below 1.3% allows its consumption as crude oil which is rich in pro-vitamin A and tocochomanols, and the oil is liquid at room temperature.

The sample 2 showed higher amounts for alpha-, gamma-tocotrienol, total tocochromanols, alpha-, beta-carotene and total carotenes and oil acidity as well, indicating that the bunches presented different maturity stages for the two assays (Rodriguez-Amaya *et al.,* 2008). Surely, the interval after harvest until the heat treatment was higher for sample 2 because of its higher acidity as observed by España et al (2018). Enzymes promote the hydrolysis of the cell wall and release oil and bioactive compounds and increase oil acidity.

Pressed Brazilian Caiaué oil is a valuable source of tocotrienols. *In vitro* studies suggested that tocotrienols display stronger antioxidant, anti-inflammatory and chemopreventive activities when compared with tocopherols. In addition, while various studies have indicated that alpha-tocotrienol is neuroprotective while gamma-tocotrienol exhibited the greatest anticancer effects (Azzi, 2019). The tocochromanols are effective antioxidants in oils but the difference among them depends on the type of oil, fat, emulsion or food system evaluated. Alpha-tocopherol has been tested and shown to prevent vitamin E deficiency disease according to Azzi (2019) and there are no conversion factors for tocotrienols to Vitamin E. The contribution of Caiaué as a source of Vitamin E is negligible, but some authors have shown the sum of tocochromanols as total vitamin E content.

4. CONCLUSIONS

Pressed Caiaué oil showed high levels of alpha-, beta-carotene, tocotrienols, low acidity and it is clear at room temperature, highlighting its use as a concentrate of pro-vitamin A and gamma-tocotrienol. It was estimated that 8 mL of this oil met the Recommended Dietary Allowance/day of vitamin A for men and women. The oil presented low levels of saturated fatty acids and high levels of oleic and linoleic acids when compared to African palm oil and its derivatives and most oil samples of *Elaeis oleifera*.

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DECLARATION OF COMPETING INTEREST

The authors of this article declare that they have no financial, professional or personal conflicts of interest that could have inappropriately influenced this work.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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AUTHORSHIP CONTRIBUTION STATEMENT

RA: Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Writing – original draft, Writing – review & editing. AEW: Methodology, Formal analysis; Writing – original draft. AMMG: Formal analysis, Investigation, Methodology, Writing – review & editing. AFFM: Investigation, Methodology, Formal analysis.

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