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RESEARCH

Chemical Characterization of the American Oil Palm from the Brazilian Amazon Forest

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ABSTRACT

American oil palm [*Elaeis oleifera* (Kunth) Cortes] is known to present a much slower palm oil deterioration rate than African oil palm (*E. guineensis* Jacq.). In the present study, fatty acid composition, total carotenoids, oil content, and free fatty acid content were characterized for five accessions (Careiro, Anori, Manicoré, Coari, and Autazes) belonging to an American oil palm collection maintained as field gene bank by Embrapa. This germplasm collection was originally collected in different areas in the Brazilian Amazon forest. These accessions were subjected to different storage times (1, 7, and 14 d) before processing to evaluate the quality of the oil produced. Oil content of the dried pulp was found to range between 31.36 and 50.34%. Gas chromatographic analysis revealed that oleic acid is the predominant monounsaturated fatty acid in all five accessions. After 14 d of fruit storage, these accessions were found to maintain low acidity (free fatty acid content), between 1.33 and 2.66%. Total carotenoid content was presented in high concentrations in all five accessions (>1500 mg kg^{-1}). The estimation of genetic parameters showed, in general, high heritability values (>80%), and the estimated genetic gains were generally very high. Coari and Careiro presented the highest oil yields, whereas Manicoré and Coari presented the lowest acidity. These two characteristics are the most important ones when selecting the best parental donor to develop superior interspecific hybrids.

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Abbreviations: AOCS, American Oil Chemists' Society; FAME, fatty acid methyl esters; FFA, free fatty acid; FID, flame ionization detector.

The oil extracted from the mesocarp of oil palm (*Elaeis guineensis* Jacq.) fruits (pulp oil) is currently the major source of edible oil in the world, with 60 Tg produced in 2016–2017. Altogether, palm oil and palm kernel oil accounted for 67 Tg of the vegetable oil consumed worldwide in 2016–2017 (Statista, 2018). This amount represents a little over 30% of the total world production of oils and fats. It is expected that the oil palm market share will reach at least 93 Tg to meet the demand for vegetable oil predicted by 2050 (Corley, 2009).

The genus *Elaeis* comprises two species, *E. guineensis* and *E. oleifera* (Kunth) Corts (Rees, 1965). *Elaeis guineensis*, the predominant species in commercial plantations, originates from West Africa and is known in Brazil as Dendê. *Elaeis oleifera* originates from Central and South America and is found in the Brazilian Amazon forest, where it is known as Caiaué.

Palm oil breeding programs elsewhere are developing interspecific hybrids between these two species. The main desired agronomic traits looked for in these hybrids are high yield, slow growth, and resistance to the oil palm lethal yellowing disease. In addition, the oil extracted from the interspecific hybrids has being reported as being of quality in terms of low acidity (free fatty acid [FFA] content) and higher contents of unsaturated fatty acids, sterols

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and vitamins (Hardon et al., 1985; Bergamin Filho et al., 1998; Sambanthamurthi et al., 2000b; Rios et al., 2012).

A germplasm collection of *E. oleifera* was established by Embrapa (Brazilian Agricultural Research Corporation) and IRHO (Institut de Recherches pourles Huiles et Oleagineux) in the 1980s in the municipality of Rio Preto da Eva in the state of Amazonas. The accessions in this germplasm bank were grouped, according to the place of origin, into 19 subpopulations, comprising a total of 246 accessions. These populations are commonly found throughout humid areas, close to the rivers in the Amazon forest (Rios et al., 2012). Despite the advances in the agronomic evaluation of these populations over the years, little is known about their pulp oil chemical variation. In addition, the effects of subjecting the fruit bunches to different postharvest storage periods on oil quality have not been studied.

The quality of the oil palm is highly affected by the hydrolytic action of mesocarp lipases (triacylglycerol acylhydrolase, EC 3.1.1.3) that releases FFAs in bruised ripe fruits during the postharvest period of storage and transportation, when the enzyme comes into contact with the substrate (triacylglycerides) (Henderson and Osborne, 1991; Sambanthamurthi et al., 2000a; Ebongue et al., 2008). These compounds generate rancidity and other technological issues that deteriorate the oil for diverse industrial purposes. According to the Codex Alimentarius (FAO and WHO, 1999), levels of FFA >5% are considered to be unsuitable for human consumption. The FFA content is expressed as acidity value, which is the main parameter to measure the oil palm quality and to determine its commercial price.

The objective of this study was to evaluate chemical variations related to oil quality parameters of the pulp oil extracted from fruits of five different accessions from the American oil palm germplasm bank. A genetic diversity identified based on these parameters can provide useful information for the selection of superior genotypes with low FFA, high yield, and better oil quality to be used in the breeding program at Embrapa or elsewhere.

MATERIALS AND METHODS

Plant Material

Ten fresh fruit bunches were collected from five different *E. oleifera* accessions belonging to the American oil palm germplasm bank maintained by Embrapa Western Amazon at the Rio Urubu Experimental Station, located 140 km from Manaus in the municipality of Rio Preto da Eva, Amazonas, Brazil (2°35′ S, 59°28′ W, 200 m asl). These accessions belong to five different regions (Careiro, Anori, Manicoré, Coari and Autazes) in the course of the Amazon, Solimões, and Madeira Rivers, spread over the Brazilian Amazon forest (Fig. 1). These accessions were selected based on results of previous genetic diversity studies performed in our laboratory (Pereira, 2015).

Two bunches were collected per accession, one per tree. Each bunch was spiked separately, and the fruits were collected from spikelets at the base, middle, and tip to have a representative sample of the bunch. Each sample was composed of nine fruits, including three fruits from each spikelet. The harvest was done in triplicate. Fruits were submitted to different storage times after harvest (1, 7, and 14 d) under environmental conditions, with the subsequent sterilization by autoclaving the samples at 120°C and 101.325 kPa for 20 min to stop the enzymatic hydrolysis reaction. In the palm oil mill industry, bunches are processed within 48 h after harvesting, so one storage time was chosen within (24 h) and two outside (7 and 14 d) of the range normally used. Fruits stored for 7 and 14 d were maintained connected to the spikelets to simulate the same conditions as found in the industry.

Oil Content

Sterilized fruits were pulped and dried for 72 h at 60°C to determine the weight of the air-dried sample, according to the method reported by Nogueira et al. (1998). This parameter is important because it allows calculation of oil content in wet basis. Dry matter content was determined to convert the analytical results obtained to a dry basis, using an oven overnight with forced air at 105°C (Sluiter and Sluiter, 2010). The ethereal extract content was determined, according to Method Am 5-04 of the American Oil Chemists' Society (AOCS, 2005), in an Ankon XT15 extraction system, using petroleum ether as the extraction solvent. The extracted compounds were predominantly triglycerides.

Oil Extraction for Quality Tests

To determine oil quality (carotenoids content, fatty acid profile, and acidity), the oil was first extracted from the pulp mesocarp of the dried fruits using a Dionex Accelerated Solvent Extractor (ASE) 350. Petroleum ether was used as the extraction solvent. Equipment parameters were adjusted in the following configuration: five cycles of heating for 5 min at 70°C, rinsing with 100% volume, purging for 60 s. After extraction, evaporation of the ether was finished in a rotary evaporator at 35°C. The oil was kept in a freezer at −20°C prior to determination of quality parameters, except for FFA determination, which was performed on the same day as extraction.

Total Carotenoids Content

Total carotenoid content was determined by spectrophotometry using an adapted protocol derived from the method described by Achir et al. (2010). Briefly, 20 mg of palm oil was dissolved in 25 mL of acetone, followed by 5 s of vortexing. An aliquot was then transferred to a 1-cm path-length cuvette, and the absorbance at 450 nm (i.e., the maximum wavelength of absorption for all-trans-carotene) was determined using a Lambda 35 ultraviolet-visible spectrophotometer (PerkinElmer) with acetone as a blank control. All steps were performed under subdued light to minimize oxidation of the carotenoids, and the total carotenoids concentration was calculated considering the absorption coefficient of β-carotene in acetone equal to 2620 (Meléndez-Martínez et al., 2007), according to Rodriguez-Amaya and Kimura (2004):

$$\text{total carotenoids } (\mu g\ g^{-1}\ \text{oil}) = [A_{450\ nm} \times \gamma\ (mL) \times 10^4] / (A^{1\%}_{1cm} \times W_{oil})$$

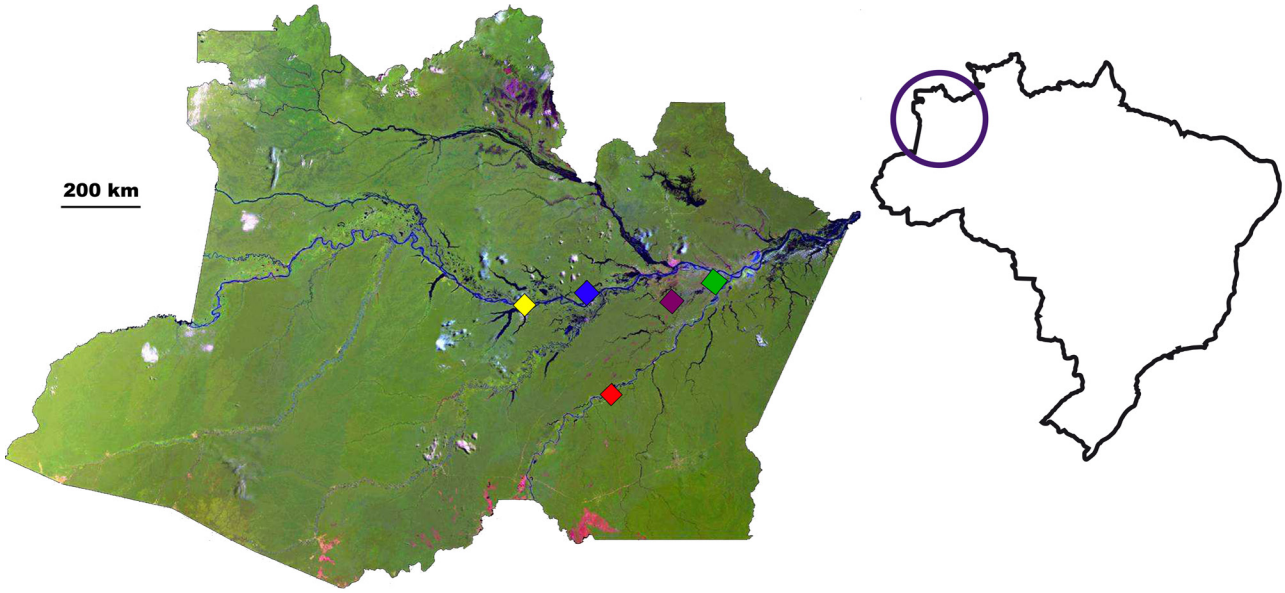

Fig. 1. Geographic distribution of the *Elaeis oleifera* accessions in the state of Amazonas, Brazil, used in the study: Coari (yellow), Manicoré (red), Careiro (purple), Autazes (green), and Anori (blue). Source of the satellite picture: www.cnpm.embrapa.br/projetos/cdbrasil/txt/meto.htm.

where $A_{450\ nm}$ represents the oil absorbance, γ (mL) is the total volume of extract, $A^{1\%}_{1cm}$ is the coefficient of absorption of β-carotene in acetone, and W_{oil} represents the oil weight in powdered form.

Fatty Acid Composition

The fatty acids were converted to fatty acid methyl esters (FAME) by heating 20 mg of palm oil within 10% BF_3–methanol, according to the procedure reported by the AOCS Methods Ce 1-62 and Ce 2-66 (AOCS, 2005). The FAME profile was separated on a Shimadzu GC-2010, equipped with flame ionization detector (FID).

The samples of trans-esterified oils were injected in gas chromatograph (Agilent Type 7890A) through an automatic injection system split-type injection (1:100), with the use of a HP-88 column (60 m × 0.25-mm i.d. × 0.2 m), which uses the FID-type detecting system. The carrier gas was He_2 at 1 mL min^{-1} (constant flow), and the chromatographic run time for each sample was ∼60 min. The oven was operated with an initial temperature of 140°C and a final temperature of 240°C with a ramp of 4°C min^{-1}. The injector and detector temperature was 260°C. The fatty acid compositions of the different species were determined by comparing the retention times with a 37-component FAME standard (Sigma Code 47885, Supelco), which was run on the same column under the same conditions to facilitate identification.

Free Fatty Acid Content

The acid content was measured for each accession by sampling 1 g of oil, and the FFA released from the lipase hydrolysis reaction was titrated with NaOH (0.1 mol L^{-1}; AOCS, 2005) using an automatic titrator (Titrando 907, Metrohm) according to the AOCS Method Cd 3d-63 (AOCS 2005). This assay was performed the same day of oil extraction. The values were converted to a percentage of oleic acid, the main fatty acid present in American palm oil, by dividing the total value of the acidity (mg KOH g^{-1} oil) by two (AOCS, 2005).

Statistical Analysis

All chemical analyses were performed in triplicate, and mean values were calculated by pooling the results of three composed samples collected from the two bunches from each accession. The results obtained are presented as means ± SD. The mean differences were considered significant at the $p > 0.05$ level. Two factors were considered in statistics analysis: variation of acidity and oil content during storage and different origin. The data were submitted to ANOVA, and the means were grouped by the Scott–Knott test at 5% of significance using the statistical program SISVAR 5.6 (Ferreira, 2011). The genetic parameters of broad-sense heritability (h^2), the coefficients of genetic (CV_g) and environmental (CV_e) variation, and the ratio of coefficients of genetic and environmental variation (CV_g/CV_e) were estimated from the variance components, for the characters studied, using GENES software (Version X) (Cruz, 2013).

RESULTS AND DISCUSSION

Oil Content on a Dry and Wet Basis

For analysis of oil content on a dry basis, it was observed that the oil production of the accessions remained constant during the two storage times used (24 h and 7 d, Table 1). This indicates that there was no synthesis or degradation of oil during the postharvest period for all accessions. According to the ANOVA, it was observed that the accessions are statistically different regarding oil content on a dry basis (Table 2), where Anori has the lowest oil content, followed by Autazes, and the remaining three accessions as a group (Table 1).

The oil content on a wet basis, which is correlated to the industrial yield of the extraction, varied between 19.77 and 39.79% (Table 1). According to the ANOVA, the storage times evaluated are statistically different regarding this variable; the same is true of the accessions (Table 2).

Table 1. Chemical properties of five accessions from the Brazilian germplasm bank of *E. oleifera* after 1, 7, and/or 14 d of storage.

Accession	Oil content on a dry basis		Oil content on a wet basis		Free fatty acid content			Total carotenoid content
	Day 1	Day 7	Day 1	Day 7	Day 1	Day 7	Day 14	Day 1
	%							mg kg^{-1}
Careiro	47.25 ± 8.00Aa†	45.67 ± 5.77Aa	33.60 ± 6.69Aa	35.49 ± 3.48Aa	0.53 ± 0.11Bc	1.12 ± 0.18Ab	1.90 ± 0.62Ba	2200.63 ± 242.67B
Anori	31.36 ± 2.90Ba	34.59 ± 2.39Ca	19.77 ± 2.57Cb	29.21 ± 3.11Ba	0.88 ± 0.14Ac	1.31 ± 0.21Ab	2.66 ± 1.09Aa	3343.56 ± 411.61A
Manicoré	46.82 ± 4.60Aa	45.62 ± 1.67Aa	31.79 ± 4.33Ab	37.68 ± 2.30Aa	0.52 ± 0.03Bc	0.86 ± 0.11Bb	1.33 ± 0.35Ca	1694.93 ± 202.85C
Coari	48.54 ± 2.18Aa	50.34 ± 8.07Aa	31.51 ± 2.01Ab	39.79 ± 3.38Aa	0.53 ± 0.10Bc	0.88 ± 0.18Bb	1.55 ± 0.55Ca	1526.68 ± 225.25C
Autazes	42.96 ± 2.74Aa	41.46 ± 1.99Ba	26.47 ± 3.93Bb	31.78 ± 3.47Ba	0.63 ± 0.10Bc	1.14 ± 0.29Ab	1.93 ± 0.92Ba	1737.43 ± 202.11C

† Means in each row (lowercase letters) or column (uppercase letters), followed by different letters, were significantly different by Scott–Knott test ($p < 0.05$ level).

Table 2. Analysis of variance for two storage times for five accessions from the Brazilian germplasm bank of *E. oleifera*.

Source	df	Sum of squares	Medium square	*F*	*P* (>*F*)
Oil content on a dry basis					
		%			
Time	1	0.3362	0.3362	0.016	0.9013
Accession	4	1966.6712	491.6678	22.72	9.7×10^{-11}***
Time × accession	4	59.0402	14.76	0.682	0.6077
Residue	50	1081.9966	21.6399	–	–
Total	59	3108.0442	–	–	–
Oil content on a wet basis					
		%			
Time	1	569.091	569.091	40.612	5.87×10^{-8}***
Accession	4	1098.1453	247.5363	19.592	9.47×10^{-10}***
Time × accession	4	103.0271	25.7568	1.838	0.1362
Residue	50	700.6408	14.0128	–	–
Total	59	2770.9042	–	–	–
Air-dried sample					
		%			
Accession	4	312.1	78.04	5.935	0.0017**
Residue	25	328.7	13.15	–	–
Total	29	640.8	–	–	–
Total carotenoid content					
		mg kg^{-1}			
Accession	4	13,084,769	3,271,192	45.3	4.2×10^{-11}***
Residue	25	1,805,245	72,210	–	–
Total	29	14,890,014	–	–	–
Free fatty acid content					
		%			
Time	2	72.2806	36.1403	191.923	$<2 \times 10^{-16}$***
Accession	4	16.2408	4.0602	21.562	2.5×10^{-15}***
Time × accession	8	6.4282	0.8035	4.267	7.9×10^{-5}***
Residue	255	48.0182	0.1883	–	–
Total	269	142.9677	–	–	–

,* Significant at the 0.01 and 0.001 probability levels, respectively.

It was verified that Anori presented the smallest amount of oil, regardless of the time elapsed for the sterilization of the fruits. Coari, Manicoré, and Careiro presented the highest oil contents (Table 1).

The oil contents of fruit mesocarp, both on a dry and wet basis, are important parameters in the evaluation of the genetic material of oil palm, due to their relation to oil yield. In an assay described in a previous study, oil content on a wet basis and dry basis for five accessions of *E. oleifera* varied from 19.8 to 33.6 and 31.4 to 48.5%, respectively. Cadena et al. (2013) reported oil contents on a dry and wet basis for five families of *E. oleifera* grown in different locations in the central zone of Colombia. The average values of oil content on a wet basis were lower (13.6 ±

2.3%) than the values reported in the present study with five accessions of *E. oleifera* from the Brazilian Amazon forest. The same behavior was observed in oil content on a dry basis (26.3 ± 4.0%). This can be attributed to existing climatological and geographical conditions or genotypic differences. Cadena, Prada et al. (2013) also reported oil contents on a wet and dry basis for commercial varieties of *E. guineensis* var. *tenera* and interspecific hybrids (*E. oleifera* var. Coari × *E. guineensis* La Mé). Results of oil content on a wet basis of *tenera* (54.7 ± 2.1%) and interspecific hybrids (47.0 ± 1.6%) were higher than in *E. oleifera* from Colombia, and similar to the results of the present work. Similar results were observed for dry oil content, which presented mean values of 78.0 ± 1.9% for *E. guineensis*, 71.5 ± 0.8% for interspecific hybrids, and 21.4 ± 0.9% for Colombian *E. oleifera*. These results, as expected, indicate that *E. oleifera* palms contain lower pulp oil content than the African species. Despite the fact that *E. oleifera* presents lower oil content in the mesocarp, it is known that this species presents lower values of acidity, an important parameter in palm oil quality. Hybrids presenting higher oil content and still a low FFA content are one of the main targets for an oil palm breeding program.

Total Carotenoid Content and Air-Dried Sample

Total carotenoid content of oil extracted from the mesocarp of different *E. oleifera* accessions, sterilized after 24 h of harvest, varied between 1526.68 and 3343.56 mg kg^{-1} (Table 1). Analysis of variance showed that there was statistical difference among the accessions (Table 2); this information was confirmed by the Scott–Knott means comparison test with a 5% level of significance. The broad-sense heritability amongst the averages found for the studied accessions, which are members of half-sib families, showed great possibility of success in the selection for this variable (97.79%). This high-magnitude value reflects the considerable presence of the genetic component in expression of this character. Results obtained from the genetic parameter analysis indicate that the CV_g/CV_e ratio is >1.0 for this parameter. This indicates that there is a greater contribution of the genetic factors than the environmental factors in estimating the phenotypic correlations between the characters studied. This information can be useful to the oil palm breeding program at Embrapa when using these accessions for the development of interspecific hybrids.

Anori contains the highest concentration of carotenoids (3343.56 mg kg^{-1}) amongst the five accessions studied. Manicoré, Coari, and Autazes showed statistically similar values of carotenoids (1526.68, 1694.93, and 1737.43 mg kg^{-1}, respectively). Studies with *E. oleifera* show that this species contains a higher carotene concentration (4600 mg kg^{-1}) than *E. guineensis* (800 mg kg^{-1}) (Yap et al., 1991). Carotenoids content of crude African oil palms from Malaysia and Zaire varies between 500 and 700 mg kg^{-1}; larger amounts (800–1600 mg kg^{-1}) from Dura species and interspecific hybrids have been reported from Nigerian sources (Goh et al., 1985).

It was also observed, according to the ANOVA, that the variable air-dried sample is statistically different among the accessions (Table 2). The broad-sense heritability among the averages found for the studied accessions was 83.15%, and the CV_g/CV_e ratio was slightly lower than 1.0 for this parameter.

A typical analysis of carotenoid composition shows that α- and β-carotenes are the major components (∼90%), and the rest are γ-carotene, lycopene, phytoene, ζ-carotene, δ-carotene, neurosporene, phytouene, α-zeacarotene, β-zeacarotene, and xanthopylls (Choo et al., 1992). Studies on the composition of *E. oleifera* oil report that the main carotenes present are α- and β-carotenes, participating with 40.38 and 54.08 mg kg^{-1} of the total carotenoid composition, respectively, followed by lycopene (7.81 mg kg^{-1}), phytoene (2.49 mg kg^{-1}), phytofluene (1.24 mg kg^{-1}), *cis*-β-carotene (0.15 mg kg^{-1}), *cis*-α-carotene (0.86 mg kg^{-1}), ζ-carotene (2.00 mg kg^{-1}), δ-carotene (2.00 mg kg^{-1}), γ-carotene (1.16 mg kg^{-1}), neurosoprene (0.77 mg kg^{-1}), β-zeacarotene (0.56 mg kg^{-1}), and α-zeacarotene (0.30 mg kg^{-1}) (Yap et al., 1991).

These compounds present in the palm oil from Brazilian *E. oleifera* in high values are therefore potent actives for use in the pharmaceutical industry with broad medicinal and nutritional properties. In particular, α- and β-carotenes are known for the activity of their provitamin A, because they can be converted into vitamin A in vivo. In addition, the lycopene compound is an effective antioxidant. Epidemiological studies strongly associated β-carotene with the prevention of certain types of cancer, such as oral, pharyngeal, pulmonary, and stomach cancer (Suda et al., 1986; Temple and Basu, 1988; Murakoshi et al., 1989; Gaziano et al., 1990). Unfortunately, all this carotene is destroyed by the present refining process, which traditionally favors colorless oils for major consumers (Goh et al., 1985).

Fatty Acid Composition

The main fatty acids found in fruit mesocarp oil of the five accessions of *E. oleifera* used in this study are shown in Table 3. Oleic acid (the accessions presented values of 45.91–53.56%), palmitic acid (25.73–29.26%), linoleic acid (13.45–18.26%), stearic acid (1.40–3.63%), and *cis*-vaccenic acid (1.32–3.18%). The other fatty acids (myristic, palmitoleic, and linoleic acid) were found in traces amounts (concentration < 1% of total fatty acids). Analysis of variance showed that there is a significant difference ($p < 0.05$) between the fatty acid composition of *E. oleifera* accessions. In addition, genetic parameter analyses

Table 3. Fatty acid composition of the fruit pulp oil for five accessions from the Brazilian germplasm bank of *E. oleifera*.

Fatty acid	Acid name	Formula	Total fatty acids				
			Careiro	Anori	Manicoré	Coari	Autazes
			%				
Saturated fatty acids							
Myristic	Tetradecanoic acid	$C_{14:0}$	Tr†	Nd‡	0.14 ± 0.18a§	Nd	Nd
Estearic	Octadecanoic acid	$C_{18:0}$	2.53 ± 0.46b	1.40 ± 0.12c	1.65 ± 0.05c	1.48 ± 0.03c	3.63 ± 0.28a
Palmitic	Hexadecanoic acid	$C_{16:0}$	25.73 ± 3.38a	29.26 ± 2.12a	28.29 ± 1.18a	27.95 ± 0.56a	27.27 ± 0.24a
Monounsaturated fatty acids							
Palmitoleic	9-Hexadecanoic acid	$C_{16:1}$	1.27 ± 0.22a	1.16 ± 0.38a	0.8 ± 0.02b	1.29 ± 0.24a	0.64 ± 0.25b
Oleic	9-Octadecanoic acid	$C_{18:1}$	53.56 ± 6.73a	45.91 ± 0.72b	50.91 ± 2.64a	49.56 ± 0.29b	47.90 ± 0.48b
cis-Vacenic	9-Octadecanoic acid	$C_{18:1}$	2.48 ± 0.23c	3.18 ± 0.13a	2.47 ± 0.04c	2.79 ± 0.20b	1.32 ± 0.04d
Polyunsaturated fatty acids							
Linoleic	9,12-Octadecadienoic acid	$C_{18:2}$	13.45 ± 3.21b	17.81 ± 1.11a	15.00 ± 1.35b	16.41 ± 0.40a	18.26 ± 0.69a
Linolenic	9,12,15-Octadecatrienoic acid	$C_{18:3}$	0.90 ± 0.28b	0.98 ± 0.06b	0.75 ± 0.02c	0.53 ± 0.18d	0.98 ± 0.04b

† Tr, traces (concentration < 0.06% of the total fatty acids).

‡ Nd, not detected.

§ Means in each row followed by different letters were significantly different ($p < 0.05$ level).

indicated that most of the fatty acids ($C_{16:1}$, $C_{18:0}$, $C_{18:1}$, $C_{18:2}$, and $C_{18:3}$) found in *E. oleifera* oil had CV_g/CV_e ratio values >1.0, suggesting that there is a greater contribution from genetic factors than environmental factors in the estimates of the phenotypic correlations among the studied characters. This indicates a favorable condition for selection, with a high accuracy.

Together, myristic saturated fatty acid (found only in Manicoré), stearic acid, and palmitic acid accounted for 28.26 to 30.09% of the total fatty acids in the oil of all *E. oleifera* accessions evaluated. Monounsaturated palmitoleic, oleic, and *cis*-vaccenic monosaturated fatty acids were detected in all accessions, varying between 49.86 and 57.31% of the total fatty acid. Two polyunsaturated fatty acids with 18 carbons (linoleic and linolenic [in traces]) accounted for between 14.35 and 19.24% of the total fatty acid (Table 3).

These results coincide with the fatty acid profile of *E. oleifera* fruit pulp oil described in a previous study (Montoya et al., 2013), where the total contents of saturated fatty acid (27.00%), monounsaturated fatty acid (57.3%), and polyunsaturated fatty acid (15.7%) were close to the values reported in this study. In a study of palm fruit pulp oil from palm trees in the state of Paraíba in the northeast region of Brazil, Bora et al. (2003) presented different concentrations in the fatty acid profile than the *E. oleifera* profile described in this study, indicating that the total content of monounsaturated fatty acid (45.73%) is close to the total content of saturated fatty acid (42.79%), with palmitic acid being responsible for 36.9% of the total. In the present study, oleic monounsaturated fatty acid (mean value of accessions = 49.56%) is the predominant compound in *E. oleifera*. Other studies have reported that the fatty acid composition of African palm fruit oil collected in Malaysia is ∼50% saturated, 40%, mono-unsaturated, and 10% polyunsaturated fatty acid (Tan and Oh, 1981).

The identity pattern of the fatty acid composition of palm oil established by the Codex Alimentarius is reported by FAO and WHO (1999), indicating that the oleic monounsaturated acid that was the predominant fatty acid in *E. oleifera* fruits can be found in values between 36 and 44%. In this study, the values of oleic acid content were higher than this standard value (between 45.91 and 53.56%). The codex also states that palmitic acid can be found in values between 39.3 and 47.5%; however, the content of this saturated fatty acid was found to be lower in Brazilian accessions of *E. oleifera* used in this study (25.73–29.26%). One of the big questions about the introduction of a new species in the world production of palm oil is the different profile of fatty acids. Based on the results reported in this present study for fatty acids in *E. oleifera*, this could be seen as a negative factor unless a change in the demand from the industry and/or consumers happens.

Among polyunsaturated fatty acids, the concentration of linoleic acid in pulp oil was higher in Autazes (18.26%) than in Careiro (13.45%). However, Careiro presented the highest total concentration of unsaturated fatty acids (71.66%), followed by Coari (70.58%). These oils also contained small amounts of linolenic acid (0.53–0.98%). Linoleic acid is sensitive to oxidation and thermal deterioration (Bora et al., 2003). Regarding the oleic/linoleic acid ratio, the oil that presented the best stability and useful life was the Careiro oil, with a ratio of ∼4 (Table 3).

The composition of the fatty acid profile can determine the final destination of the oil in the industry. Palm oil can be fractionated into two components after refining: olein (60%) and stearin (40%). Olein is liquid oil, mainly composed of fatty acids, and has emollient and lubricating properties. After processing for the purification of fatty acids, mainly by distillation, it has applications in the paint and thinner, food, cosmetic, and oleo chemical industries, in general. Stearin, due to its higher degree of saturation,

can be used as fat in cakes and biscuits. It also serves as a raw material for the manufacture of margarines, mayonnaise, and ice cream. In addition, it can replace sebum in the production of soaps (Mba et al., 2015). The results for *E. oleifera* pulp oil in all accessions showed a higher content of unsaturated fatty acids (>80%), conferring liquid state at room temperature, which could be of more interest to the first group of industries cited.

Free Fatty Acid Content

In this study, FFA content was evaluated in fruit samples from different accessions of *E. oleifera* subjected to three different storage durations at room temperature after harvest.

The effect of storage time on the FFA content is shown in Table 1. A continuous increase in FFA content was recorded for all accessions during the first 2 wk of storage. The highest increase rate was attributed to Careiro, which increased 258% from Day 1 to Day 14 of storage, starting at 0.53% and reaching 1.33% by the end of the period. However, the greatest increase in FFA content occurred between the first and seventh storage day (rate of increase = 111%). This behavior was observed in all accessions. Significant changes occurred among the first, seventh, and last days of storage in all accessions samples evaluated (Table 1).

According to the ANOVA, the storage times and the accessions evaluated, and the interaction between them, are statistically different regarding this variable (Table 2). These analyses were confirmed with the Scott–Knott averages comparison test at a significance level of 5%. The highest values for FFA content were recorded from the Anori samples, at all storage times evaluated. The Manicoré accession showed the lowest FFA value for all evaluated times.

The FFA content is the most commonly used criterion for determining palm oil quality, which should not exceed 5% expressed as palmitic acid in the case of *Elaeis guineensis* and, in the case of *E. oleifera*, expressed as oleic acid (because they are the majority fatty acid), according to the norms established in the Codex Alimentarius (FAO and OMS, 2005). Fatty acids are usually present in the oil as part of the triacylglycerol molecules. The presence of FFAs in palm oil is indicative of deterioration of oil quality. This process is essentially attributed to the activity of the lipases present in palm oil mesocarp, which are responsible for the hydrolysis of triacylglycerides (Cadena et al., 2013). Lipase is activated during fruit maturity or by injury or bruising.

Studies have verified that the activity of the endogenous lipases can vary between different lineages, identifying some with very low lipolytic activity. Crude palm oil extracted from the fruits of lineages with low lipolytic activity also showed a low FFA/oil acid content (Ebongue et al., 2008). FFA may also be generated by lipases contaminating microorganisms (Hiol et al., 2000, Abbas et al., 2002). To limit lipase activity, fresh fruit bunches should be processed quickly after harvesting. For all *E. oleifera* accessions used in this study, acidity values were below the 5% limit. These results confirmed the hyphotesis that *E. oleifera* oil has a better oil quality (Cadena et al., 2013). Rapid sterilization of the fruits at high temperatures rapidly inactivates lipases, thereby limiting the subsequent accumulation of FFA in palm oil. In some cases, collected bunches may be kept at room temperature for a week or more before being processed.

Other authors studied the formation of FFA in fruit oils from several accessions of *E. guineensis*, *E. oleifera* and interspecific hybrids submitted to low temperatures (5 and −20°C). It was observed that the maximum FFA formation occurred at 5°C, with formation of 47% and 29% in *E. guineensis* and in the interspecific hybrid, respectively. In the case of *E. oleifera*, these authors reported the formation of FFA < 1%, not observing changes at different temperatures. These results are in agreement with the data reported in this study for *E. oleifera* fruits submitted to sterilization, after 24 h of collection (Cadena et al., 2013). These authors also identified the highest levels of FFA in *E. guineensis* after storage of the fruits at 5°C (70%). In addition, Ebongue et al. (2006) showed the production of 55% of FFA, after submitting the fruits to freeze/thaw cycles. These authors suggest that the effect can be caused by the damage generated by the freezing temperatures in the mesocarp tissues of the fruits. The breakdown of oleosomes releases the enzymes associated with the membrane and increases the availability of triacylglycerides (substrate) stored in the same organelles (Ebongue et al., 2006).

The harmful effect of fermentation is the continuous FFA production in fruit mesocarp under lipase action. Since the fruits are processed, lipase is no longer active, but FFA content of the resulting palm oil may also increase during storage as a result of the autocatalytic hydrolysis. In this case, the FFA acts as catalysts for the reaction between triacylglycerols and water to generate more FFA (Ebongue et al., 2011). Table 1 clearly describes the process of increasing the FFA content in the American palm oil samples during the first 2 wk of storage, where the autocatalytic hydrolysis reaction may have occurred. Ebongue et al. (2011) reported that there is a positive correlation (r = 0.76) between the FFA content and the moisture content of *E. guineensis* fruits, suggesting to limit the FFA content and crude palm oil moisture before long-term storage. Autocatalytic hydrolysis is unlikely to occur below the 0.1% moisture level recommended by Codex Alimentarius standards (FAO and OMS, 2005).

The results showed that the FFA content was significantly different between the studied *E. oleifera* accessions, indicating that there is no phenotypic uniformity for this characteristic among the most representative accessions

of genetic diversity in the *E. oleifera* germplasm bank at Embrapa. Manicoré was the accession that presented the lowest FFA content over all storage times evaluated. On the other hand, it was observed that the FFA content increased continuously during the 2 wk (at rates >100%) of fruit storage, after subsequent sterilization.

In large-scale oil production, bunches are processed within 48 h after harvest and FFA content varies from 3 to 4%; on the other hand, in small-scale production from small farmers, this content may reach 9 to 15% (Gibon et al., 2007). Consequently, only large-scale producers can have access to refining processes, and small-scale palm oil may take several weeks to reach the extraction site, generating oil with high levels of FFA that is not suitable for human consumption. In this sense, palm oil produced by small farmers cannot be competitive, especially in international markets. These results clearly indicate that, although there is some variability among the accessions of *E. oleifera*, this species has a genetic background that allows longer postharvest storage times (up to 14 d) for the processing of the bunches. This genetic background is of major importance for breeding programs aiming the development of interspecific hybrids harboring this trait, by means of classical breeding (back-cross strategy) or genetic engineering.

CONCLUSIONS

Five *E. oleifera* accessions from the Brazilian Amazon forest were submitted to different storage times and characterized in terms of oil content (on a wet and dry basis), moisture content, acidity, total carotenoid content, and fatty acid profile. Results showed variability within these accessions for most of the analyzed parameters: oil content on a wet basis, acidity, fatty acid profile, and total carotenoid content. However, the results showed phenotypic uniformity for the variables oil content on a dry basis (regardless of storage time) and moisture content (these values were higher in autoclaved samples 7 d after collection, due to the loss of moisture in the environment during the storage period).

The ones that presented the best characteristics in terms of high yield were Coari and Careiro, and the best in terms of low acidity were Manicoré and Coari. These two characteristics are the most important ones when selecting the best genotypes for future breeding studies to develop superior interspecific hybrids.

Conflict of Interest

The authors declare that there is no conflict of interest.

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