Composition of fatty acids, tocopherols, tocotrienols and β -carotene content in oils of seeds of Brazilian Sapindaceae and Meliaceae species

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Abstract This study analyzes the lipid composition and the oxidative stability of oils of Sapindaceae and Meliaceae seeds. The oil content ranged from 14.7% (*Guarea guidonia*) to 30.1% (*Allophylus puberulus* and *Paullinia elegans*). Ten fatty acids were identified in seed oils. *Guarea guidonia* seeds accumulated mainly oleic (44.9%) and linoleic (24.1%) acid, whereas the unusual gondoic and paullinic acids were identified in *A. puberulus* (15.8%; 8.9%) and *P. elegans* (14.4%; 44.2%), respectively. The oil of *P. elegans* had the highest oxidative stability (16.2 h.). Tocopherol predominated in *A. puberulus* (10.4 mg/100 g) and tocotrienol predominated in *G. guidonea* and *P. elegans* (2.6 mg/100 g). The vitamers α -tocopherol, γ -tocopherol, and γ -tocotrienol were found in the studied oils. β -carotene was predominantly detected in the oil of *A. puberulus* seeds (22.4 μ g/g). *Guarea guidonia* seed oil has a high potential in food due to richness in essential fatty acids. In turn, *A. puberulus* and *P. elegans* oils could be suggested for other industrial purposes (e.g., biodiesel,

varnishes, paints, soaps, or oleochemicals) due to their content of long-chain fatty acids.

Keywords Oilseeds · Antioxidants · Tocopherols · Tocotrienols · Oxidative stability

Introduction

Brazil has a great plant diversity, and several species are oleaginous (Luzia and Jorge 2013; Pinho et al. 2009). The economic potential of some oleaginous crops is attributed to the oil content and the presence of several chemical compounds, such as vitamin E, carotenoids (especially β -carotene), phytosterols, and fatty acids. Such compounds have been widely correlated to antioxidant, anti-inflammatory, antidiabetic, anti-obesity, anticarcinogenic, and antimutagenic activities (Alvarez and Rodríguez 2000; Hoed 2010). Therefore, the study of these properties and their composition may be potentially useful to the cosmetic and pharmaceutical industries, in addition to food consumption.

Some angiosperms (e.g., Meliaceae and Sapindaceae) have a wide distribution in different Brazilian ecosystems. They are economically exploited because of their high seed oil content associated to different bioactive properties. The anti-inflammatory and analgesic properties of *Carapa guianensis* Aubl. (Meliaceae) has aroused the interest of the pharmaceutical industry (Penido et al. 2006). Neem (*Azadirachta indica* A. Juss.) oil is traditionally used as a repellent and insecticide (Roychoudhury 2016). The seed of *Khaya senegalensis* A. Juss. also has oils with an antimicrobial action (Idu et al. 2014), and therapeutic properties (Taikin et al. 2013).

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The Sapindaceae family also has species with a high seed oil content. Long-chain fatty acids and cyanolipids are present. Compounds with insecticide and repellent properties were detected in the following Brazilian species: *Paullinia meliifolia* Juss., *Urvillea uniloba* Radlk., and *Cardiospermum grandiflorum* Sw. (Spitzer 1996). Fruits of *Allophylus edulis* (A. St-Hil., Cabess. & A. Juss.) Radlk. and *Sapindus esculentus* A. St-Hil are widely appreciated as food and exert an antioxidant activity (Oliveira et al. 2012). In our previous work, we studied some seeds of Brazilian species of the genera *Allophylus* and *Paullinia* focusing on their high oil content and fatty acid composition (Coutinho et al. 2015).

Species of these families have a wide range of applications. However, considering the economic potential of such families and the great diversity of tropical environments, few species of Sapindaceae and Meliaceae have been used for such purposes. This study presents the contents of oil, fatty acids, β -carotene, tocopherols, and tocotrienols, and the oxidative stability of oils of *Allophylus puberulus*, *Paullinia elegans* (Sapindaceae) and *Guarea guidonia* (Meliaceae) seeds.

Materials and methods

Description of samples

Mature seeds of *G. guidonia* (L.) Sleumer (Voucher UFP 75.205) (Meliaceae), and *Allophylus puberulus* (Cambess.) Radlk. (Voucher UFP 74.178) and *P. elegans* Cambess. (Voucher UFP 74.737) (Sapindaceae) were obtained between March and May 2013 from three locations in the city of Recife, Pernambuco state, Brazil. Voucher specimens are deposited at the Geraldo Mariz Herbarium (UFP), Federal University of Pernambuco.

Oil extraction and analysis

The oil was extracted from 5 g of powdered fresh seeds in a Soxhlet apparatus by using anhydrous sodium sulfate to remove moisture. The extraction was performed using *n*-hexane (approximately 68 °C) for 8 h. The oils were stored in amber glass vials at − 20 °C until analysis (Coutinho et al. 2016).

The hydrolysis and transesterification of oils were performed according to Milinsk et al. (2008), with modifications. In brief, 3 mL of *n*-heptane were added to 25 mg of oil, and vortexed for 20 s. Subsequently, 1.5 mL of KOH in methanol (2 M) was added and stirred for 30 s. The samples were centrifuged (10 min, 731 g), and the supernatant was collected and stored in amber flasks at − 20 °C until analysis. FAMES were identified using a GC/MS

(Agilent Technologies 7820A, 5975C, Waldbronn, Germany) by comparison with authentic standards (Supelco FAME mix C4-C24, Bellefonte, PA, USA) and by comparing their mass spectra with the Wiley database 229 (Wiley, New York) and the NIST/EPA/NIH Mass Spectral Library (NIST 08). GC analysis was performed in a HP-5MS capillary column with an initial column temperature of 135 °C (1 min) and a heating rate of 3 °C/min up to 215 °C, and then kept at 215 °C for 20 min. The injector and detector temperatures were 230 °C and 240 °C, respectively. Helium was used as the carrier gas (0.59 mL/min), and the injection volume was 2 μ L with a split ratio of 1:100. The FAME relative amount was expressed as percentage of the total area of all peaks obtained by the GC/FID (flame ionization detector), GC2010Plus, and the AOC20i autoinjector (Shimadzu, Kyoto, Japan) under the same conditions as described above.

Evaluation of oxidative stability

Oxidative stability was measured using a Rancimat 892 apparatus (Metrohm Ltd., Harisau, Switzerland). The extracted oil (3 g) was heated to 110 ± 1.6 °C with an air flow of 20 L/h until reaching the induction time (Amaral et al. 2003). The volatile compounds released were collected, and the conductivity inflection point was recorded. The results were expressed in hours (h), representing the induction time. All analyses were carried out in triplicate.

β -Carotene content and vitamin E profile

The β -carotene content and vitamin E profile (α -, β -, γ -, δ -tocopherols and the respective tocotrienols) of the oils were simultaneously analyzed by normal phase HPLC with a diode array and fluorescence detection (Alves et al. 2009). The internal standard tocol (20 μ L, 1 mg/mL) was added to 20 mg of oil, and the final volume was completed to 1 mL with *n*-hexane. The samples were injected into a HPLC system equipped with an AS-2057 Plus automatic injector, a PU-2089 Plus pump, a CO-2060 Plus column oven, a MD-2018 multiwavelength diode array detector (DAD), and a FP-2020 Plus fluorescence detector (Jasco, Japan). The compounds were separated in a Zorbax Rx-SIL column (4.6 cm $\times$ 250 mm, 5 μ m, Agilent Technologies, Waldbronn, Germany) operating at 21 °C. An isocratic elution with *n*-hexane:1,4-dioxane (92:8 v/v) was performed at a flow rate of 1.5 mL/min. The fluorescence detector was set with $\lambda_{\text{excitation}} = 290$ nm and $\lambda_{\text{emission}} = 330$ nm (gain 10) for vitamin E analysis. For the β -carotene analysis, chromatograms were recorded at 450 nm.

The compounds were identified by a direct comparison with retention times and the UV–Vis spectra of authentic

standards of β -carotene (Fluka, Sigma-Aldrich, St. Louis, Missouri, USA), tocopherols, and tocotrienols (Supelco, Bellefonte, PA, USA) under the same conditions of analysis. The quantification of compounds was performed based on the internal standard method. Chromatographic data were analyzed using ChromNAV 1.17.01 Chromatography Data System (Jasco, Japan). The results for tocopherols and tocotrienols were expressed as mg/100 g of seeds, and β -carotene was expressed as $\mu\text{g/g}$ of oil.

Statistical analysis

The analyses were performed in triplicate. Data were first verified for normality and homogeneity. Significant statistical differences were analyzed using one-way ANOVA followed by post hoc Tukey HSD test ($p < 0.05$). Statistical analyses were carried out using the STATISTICA (version 8.0) software (Statsoft Inc., Tulsa, OK, USA).

Results and discussion

Oil content and fatty acid profile

The seed oil content ranged from 14.7 (*G. guidonia*) to 30.1% (*P. elegans* and *A. puberulus*) (Table 1). The oil content of *G. guidonea* was analyzed for the first time and was similar to that of Mexican species of this genus, such as *G. chichon* (15.9%) and *G. excelsa* (14.5%) (Sotelo et al. 1990). The oil content of Sapindaceae species is similar to that found in other representatives of this family (Azam et al. 2005; Coutinho et al. 2015, 2016). This confirms the plant as a good oil source for industrial purposes.

Ten fatty acids were identified. MUFA (monounsaturated fatty acids) predominated in all species (44.9–74.3%), while SFA (saturated fatty acids) were the second major FAMES in *G. guidonea* (29.0%) and *A. puberulus* (24.0%). PUFA (polyunsaturated fatty acids) were also representative in *P. elegans* (12.6%) (Table 1).

Six major mass fragments were observed in each seed oil (Table 1). The base peak in all saturated FAMES was observed at m/z 74. Three other peaks (m/z 43, 55, 87) were common to saturated FAMES. In contrast, the base peak of MUFA was at m/z 55, besides the peaks at m/z 67, 69, 83. Paullinic and gondoic acids have a similar pattern of fragmentation, including a base peak at m/z 55 and molecular ion (M^+ 324). However, they could be differentiated by the greater intensity of the peak at m/z 292 in the paullinic acid spectrum (NIST/EPA/NIH Mass Spectral Library).

Palmitic, stearic, oleic, linoleic, and linolenic acids were the most frequent. The oleic (44.9%) and linoleic (24.1%) acids were the most abundant in *G. guidonia*. Comparatively, this fatty acid profile was found in oils of other

Meliaceae species, such as *Carapa guianensis* Aubl. (Polonini et al. 2012), which is used in cosmetics. *G. excelsa* and *G. chichon* have a high content of oleic and linoleic acids (Sotelo et al. 1990).

For *A. puberulus*, oleic (28.8%) and gondoic (*cis*-11-eicosenoic acid) (15.8%) were the main fatty acids in seeds. In *P. elegans* seeds, paullinic (*cis*-13-eicosenoic acid) (44.2%) and gondoic acids (14.4%) (Table 1) were the most abundant. This fatty acid profile has been previously observed in this species (Spitzer 1996) and in others of the genera *Paullinia*, *Cardiospermum* and *Urvillea* (Spitzer 1996). High contents of isomers of eicosenoic acid have been identified in seed oils of *Cardiospermum*, *Serjania* and *Paullinia* species (Azam et al. 2005; Coutinho et al. 2015).

Due to the presence of unusual fatty acids in the oils of the studied Sapindaceae species, they may be unsuitable for consumption. Moreover, the substantial quantity of long-chain fatty acids deserves special attention because of their value for industrial purposes, such as production of bio-diesel, varnishes, paints, soaps, or oleochemicals (Coutinho et al. 2016).

Vitamin E and β -carotene contents and oxidative stability

Concerning vitamin E, *A. puberulus* had the highest content of tocopherols, while *G. guidonea* and *P. elegans* had the highest content of tocotrienols (Table 2). α -Tocopherol, γ -tocopherol and γ -tocotrienol were mainly found in the studied oils. The highest content of γ -tocopherol occurred in *A. puberulus*, while α -tocopherol and γ -tocotrienol had a higher content in *P. elegans* and *G. guidonia*, respectively (Table 2).

Vegetable oils are rich sources of vitamin E, and α - and γ -tocopherol are major vitamers, which are often associated to high levels of unsaturated fatty acids for the protection against lipid oxidation (Bramley et al. 2000). The predominance of γ -tocopherol was reported for other Brazilian oil seeds (Luzia and Jorge 2013) and traditional oils, such as corn, palm, peanut, and soybean oils (Bramley et al. 2000; Tuberoso et al. 2007).

Regarding the β -carotene content, *P. elegans* had the highest concentrations (22.4 $\mu\text{g/g}$) (Table 2). This value was higher than those of traditional oilseeds, such as flaxseed, maize, peanuts, soybeans, and sunflower after refining, which do not have more than 1 $\mu\text{g/g}$ of β -carotene (Tuberoso et al. 2007). Natural antioxidants from oilseeds (vitamin E and β -carotene) have several advantages: antioxidant activity, safety, availability, effects on sensory value, and price when compared to synthetic antioxidants (Tuberoso et al. 2007).

Table 1 Oil content and fatty acid composition of seeds from native Brazilian species

Oil and fatty acids (%)	Mass fragments	Family/Species		
		Meliaceae	Sapindaceae	
		<i>G. guidonia</i>	<i>A. puberulus</i>	<i>P. elegans</i>
Oil content		14.7 ± 0.2 ^b	30.1 ± 4.3 ^a	30.1 ± 1.5 ^a
Palmitic acid (C16:0)	43,55,74,87,227,270	19.9 ± 0.1 ^a	6.7 ± 0.1 ^b	2.4 ± 0.1 ^c
Palmitoleic acid (C16:1)	41,55,69,83,232,268	–	2.9 ± 0.1 ^a	2.9 ± 0.1 ^a
Stearic acid (C18:0)	43,55,74,87,255,298	9.1 ± 0.1 ^a	3.3 ± 0.1 ^b	1.1 ± 0.1 ^c
Oleic acid (C18:1 <i>cis</i> -9)	41,55,69,83,263,296	44.9 ± 0.1 ^a	28.8 ± 0.1 ^b	12.8 ± 0.1 ^c
Linoleic acid (C18:2 <i>cis</i> -6)	55,67,81,95,263,294	24.1 ± 0.1 ^a	6.8 ± 0.1 ^b	3.1 ± 0.1 ^c
Linolenic acid (C18:3)	67,79,93,95,194,292	2.0 ± 0.1 ^c	12.2 ± 0.1 ^a	9.5 ± 0.1 ^b
Arachidic acid (C20:0)	43,55,74,87,143,326	–	13.2 ± 0.1 ^a	5.8 ± 0.2 ^b
Gondoic acid (C20:1 <i>cis</i> -11)	55,69,83,97,292,324	–	15.8 ± 0.1 ^a	14.4 ± 0.1 ^b
Paullinic acid (C20:1 <i>cis</i> -13)	55,69,83,97,292,324	–	8.9 ± 0.1 ^b	44.2 ± 0.1 ^a
Behenic acid (C22:0)	43,55,74,87,143,354	–	0.8 ± 0.1	–
Other fatty acids		–	0.6 ± 0.1 ^b	3.8 ± 0.1 ^a
∑ SFA		29.0 ± 0.1 ^a	24.0 ± 0.1 ^b	9.3 ± 0.1 ^c
∑ MUFA		44.9 ± 0.1 ^c	56.4 ± 0.1 ^b	74.3 ± 0.1 ^a
∑ PUFA		26.1 ± 0.1 ^a	19.0 ± 0.1 ^b	12.6 ± 0.1 ^c

Values are expressed as the mean ± SD (n = 3). Different lowercase letters in the same line show significant differences among species by one-way ANOVA and Tukey's test ($p < 0.05$)

– not detected, *SFA* saturated fatty acids, *MUFA* monounsaturated fatty acids, *PUFA* polyunsaturated fatty acids

During the oxidative stability test, *P. elegans* had the highest induction time (16.2 h) and *G. guidonea* had the lowest induction time (0.1 h) (Table 2). *P. elegans* oil had a higher oxidative stability than edible oils such as canola, corn, peanut, soybean, and sunflower (Tan et al. 2002). The differences between the induction times of *P. elegans* and

G. guidonea may be due to their different β -carotene content (Table 2).

Economic potential of oils

Based on results of this study, some economic applications can be suggested. The oil of *G. guidonia*, for example, is

Table 2 Tocopherols, tocotrienols, β -carotene and oxidative stability of seeds oils from native Brazilian species

Compounds	Family/species		
	Meliaceae	Sapindaceae	
	<i>G. guidonia</i>	<i>A. puberulus</i>	<i>P. elegans</i>
Total tocopherols (mg/100 g)	6.0 ± 0.7 ^c	10.4 ± 0.1 ^a	8.6 ± 0.4 ^b
Total tocotrienols (mg/100 g)	2.6 ± 0.8 ^a	1.7 ± 0.1 ^b	2.6 ± 0.5 ^a
Vitamin E (mg/100 g)			
α -tocopherol	0.5 ± 0.1 ^c	0.8 ± 0.1 ^b	4.5 ± 0.4 ^a
α -tocotrienol	0.5 ± 0.9 ^a	–	1.0 ± 0.9 ^a
β -tocopherol	0.2 ± 0.1 ^a	0.3 ± 0.1 ^a	0.2 ± 0.1 ^a
β -tocotrienol	0.5 ± 0.1 ^a	0.7 ± 0.1 ^a	0.4 ± 0.1 ^a
γ -tocopherol	5.3 ± 1.2 ^b	9.1 ± 0.1 ^a	3.9 ± 0.3 ^c
γ -tocotrienol	1.5 ± 0.6 ^a	1.0 ± 0.2 ^a	1.2 ± 0.1 ^a
δ -tocopherol	–	0.2 ± 0.1	–
δ -tocotrienol	0.1 ± 0.1	–	–
β -carotene (μ g/g)	6.4 ± 0.9 ^c	13.5 ± 1.2 ^b	22.4 ± 1.5 ^a
Oxidative stability (h)	0.1 ± 0.1 ^c	4.9 ± 1.1 ^b	16.2 ± 2.9 ^a

Values are expressed as the mean ± SD (n = 3). – not detected. Different letters in the same line show significant differences among species by ANOVA one-way and Tukey's test ($p < 0.05$)

rich in essential fatty acids such as oleic and linoleic acids, besides possessing emollient and excipient properties (Dyer et al. 2008). Therefore, it can be used in several industrial applications, such as cosmetics, pharmaceutical, or food industries. Nevertheless, studies on toxicity are needed to confirm this potential. The oils of *A. puberulus* and *P. elegans* have gondoic and paullinic acids, which could be useful for the manufacture of industrial products (Spitzer 1996). In addition, *P. elegans* oil has a higher oxidative stability and β -carotene content, which provides greater resistance to oxidation (Alvarez and Rodríguez 2000; Dyer et al. 2008). In view of these characteristics, it is possible to state a high economic value for the formulation of several industrial products, such as cosmetics and pharmaceutical products (Silva and Jorge 2014).

In addition to our findings, other studies reinforce potentialities of using some Meliaceae and Sapindaceae species native to Brazil. The oil of *Carapa guianensis* Aubl. (Meliaceae), for example, is widely used in cosmetics and pharmaceutical products (Penido et al. 2006). The oil from seeds of *Carapa procera* D.C. has repellent and insecticide properties. It is also used for the production of candles and cosmetics such as soaps and shampoos (Djenontin et al. 2012).

Among Sapindaceae, *Paullinia meliifolia* Juss., *Urvillea uniloba* Radlk. and *Cardiospermum grandiflorum* Sw. (Spitzer 1996) have oils with repellent and insecticide properties (Díaz and Rossini 2012). Other Sapindaceae, such as “pitomba” (*Talisia esculenta* Radlk.) have edible seeds (Guarim Neto et al. 2013), while “guaraná” (*Paulinia cupana* Kunth.) is used to develop beverages and has several pharmacological properties (Marques et al. 2019).

Conclusion

This study develops the knowledge on the composition and antioxidant properties of native seed oils from Brazil, especially *G. guidonia*. Its oils were investigated here for the first time and have a potential to be used in food, cosmetic, and pharmaceutical industries. *A. puberulus* and *P. elegans* are suitable for industrial purposes (biodiesel, varnishes, paints, soaps, or oleochemicals). For food purposes, species of Sapindaceae with unusual fatty acids need to be evaluated as for toxicity. Our findings also reinforce the need for studies on agronomic and productive aspects necessary for a future economic establishment of these species as crops.

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