

Composition, antioxidant activity, thermal and oxidative stability of *Lecythis tuyrana* oil

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Summary

Oil from *Lecythis tuyrana* oilseeds is extracted for its potential use in food industry. The aim of this study was to gain knowledge on composition, antioxidant activity and thermal oxidative stability of this type of oil. Compositional parameters were evaluated. Antioxidant activity was evaluated by oxygen radical absorbance capacity and 2,2-diphenyl-1-picrylhydrazyl radical. Oxidative stability was evaluated by peroxide value, thiobarbituric acid reactive substances values and oxidative stability index (*OSI*). Thermal stability was determined by thermogravimetric assay and differential scanning calorimetry. Almonds were found to have a high oil content of 732 g·kg⁻¹. Total carotenoids, tocopherols and phenols were also quantified. *OSI* showed an induction time of 3.47 h and it was thermostable up to 200 °C. It was possible to improve the stability of oil by 27.5 % under accelerated conditions; using butylated hydroxytoluene as an antioxidant. *L. tuyrana* is a good source of unsaturated fatty acids (oleic acid 369.0 g·kg⁻¹, linoleic acid 272.0 g·kg⁻¹, palmitic acid 171.0 g·kg⁻¹, stearic acid 149.0 g·kg⁻¹, arachidonic acid 10.0 g·kg⁻¹, palmitoleic acid 7.0 g·kg⁻¹), which are oxidatively stabilized by the presence of carotenoids, phenols and tocopherols. In this way, the new oil with interesting nutraceutical properties is proposed to be used in the food, cosmetic and pharmaceutical industry.

Keywords

edible oil; *Lecythis tuyrana*; oxidative stability; thermal stability; oilseeds

Demand for new sources of vegetable oils with a high content of unsaturated fatty acids, as oleic, linoleic and linolenic, has promoted the study of new species, Lecythidaceae family being one of the most promising. Genus *Lecythis* consists of about 26 species distributed from Nicaragua to Brazil's Amazon with 73 % of species found there [1]. In Colombia, this genus is found in altitudes of 200 m above sea level, in particular on banks of the Pacific and Caribbean rivers. *Lecythis tuyrana* is a species of large tree, which is threatened by deforestation for its use as timber [2]. In their natural habitat, *L. tuyrana* trees can reach up to 60 m of height, they have large, woody fruits, which are also dehiscent and subglobose (9.5–14.5 cm × 10–17.5 cm), the seeds being oblong of 4.5–7 cm × 2–3 cm. The fruits are used as antidiarrheal medicine and are delivered by the Kuna Indians [3]. For certain birds, these trees are very important as a shelter and food. Moreover, the seeds of

this tree are desired by several rodent species [4]. However, reports on *L. tuyrana* from the point of view of the generation of agronomic crops in Colombia, uses in traditional medicine, composition and stability of the oil extracted from the almonds and their potential use in food products with a nutritional function, are very few.

The quality of oils is determined by its chemical composition and the presence of antioxidant bioactive substances, which act directly on the oil's stability, providing a greater added value. Carotenoids, tocopherols and polyphenolic compounds are among these bioactive substances [5]. Two studies report that the seeds of some species of *Lecythis* genus, namely, *L. ollaria* and *L. curranii*, can be pressed to obtain edible oils with a high content of unsaturated fatty acids. For example, Brazil nuts contain ω-3 fatty acid (α-linolenic acid) in a portion of approximately 70 % of total lipids. Here, half of it is oleic acid, followed by 25 %

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polyunsaturated lipids (linoleic acid, ω -6), and approximately 19 % saturated fat such as palmitic and stearic acids [6, 7].

However, the processing and incorporation of oils in food matrices, specifically the highly unsaturated oils, generates oxidative deterioration and affects the organoleptic characteristics and shelf life. Therefore, it is necessary to conduct studies on the thermal and oxidative stability of oils [8].

Differential scanning calorimetry (DSC) is a thermal analysis technique employed in the measurement of temperatures and heat flows associated with many physical and chemical properties of oils. Furthermore, DSC thermal profile provides information on oxidative stability of various fats and oils [9]. Thermogravimetric analysis (TGA) is an important parameter on the stability of edible oils, providing information about the overall compositional change of the sample [10].

To inhibit oxidative deterioration, the oils are supplemented with natural or synthetic antioxidants, which retard rancidity by trapping free radicals generated by reactive oxygen species. The synthetic antioxidant 2,6-di-*tert*-butyl-4-hydroxytoluene (BHT) is the most used in food matrices rich in polyunsaturated fatty acids, used at a recommended content of 100 mg·kg⁻¹ to avoid harmful effects on human health [11].

The aim of this research was to study composition, antioxidant activity and thermal as well as oxidative stability of oil extracted from *L. tuyrana* seeds, for the first time, as a new source of oil with high nutritional value and potentially beneficial to human health.

MATERIALS AND METHODS

Materials

The oil was obtained as described further. Fluorescein, 2,4,6-tripyridyl-*s*-triazine (TPTZ), 2,2-diphenyl-2-picrylhydrazyl free radical (DPPH[•]), gallic acid, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), β -carotene, thiobarbituric acid, methyl- β -cyclodextrin and α -tocopherol were purchased from Sigma-Aldrich (St. Louis, Missouri, USA). Methanol, ethanol, *n*-hexane, butanol, acetone, dichloromethane, Folin-Ciocalteu reagent, sodium carbonate and trichloroacetic acid (analytical grade) were purchased from Merck (Darmstadt, Germany). Ethyl ether, chloroform and octane, were purchased from Carlo Erba (Cornaredo, Italy).

Plant material

Almonds were obtained from *L. tuyrana* cul-

tivars grown in the area of Urabá, department of Antioquia, Colombia, cultivated from January to May 2017. A voucher specimen was left at Botanical Garden “Joaquín Antonio Uribe”, Medellín, Colombia (voucher number 2330). The seeds were selected in the best condition of maturity, which was indicated by spontaneous opening of the fruit shell. These were washed and dried at room temperature and then stored at 4 °C in sealed polypropylene bags until analysis.

Oil extraction

The oil was obtained by cold pressing the almonds with a manual mill (Piteba, Schiedam, Holland), controlling the pressing temperature at 45 °C. The oil obtained was filtered through a metallic sieve (opening 1.00 mm, 18 mesh) and centrifuged at room temperature at 4000 \times g for 15 min (centrifuge Z206A; Hermle, Wehingen, Germany) and then stored in an amber glass at -18 °C. The acid, saponification and iodine numbers were determined according to the methods recommended by the American Oil Chemists' Society (AOCS): AOCS Cd 3d, AOCS Cd 1-25 and AOCS Cd 3-25, respectively [12].

Composition of oil

Fatty acids

To determine the fatty acid profile of oil, a gas chromatograph 6890N GC coupled to a 5973N MS selective detector and equipped with a split/splitless injector (Agilent Technologies, Santa Clara, California, USA) was used. The injector temperature was (300 \pm 1) °C. Samples were processed by adding 1 ml 0.5 mol·l⁻¹ potassium hydroxide in methanol to 50 mg of sample in 1 ml *n*-hexane and subsequent vigorous shaking for 60 s, then neutralized with 1 mol·l⁻¹ HCl and centrifuged. The upper *n*-hexane layer containing fatty acids methyl esters (FAME) was collected, filtered and automatically injected (3.0 μ l) to the chromatograph in the splitless mode. An HP-5 MS (5.0 % phenylmethylsiloxane) 30 m, 0.25 mm column, (Agilent Technologies), 0.25 μ m film thickness, and a maximum temperature of 325 °C were used. The database NIST 98 (National Institute of Standards and Technology, Gaithersburg, Maryland, USA) was used to identify fatty acids [13]. The quantity of fatty acids was expressed as grams of fatty acid per kilogram of oil.

Total carotenoids

Quantification of total carotenes was carried out as described by Biswas et al. [14]. A sample of 100 mg was taken into a test tube, to which

4 ml of cold acetone were added and, after stirring vigorously for 120 s, it was allowed to stand 15 min at 4 °C. Then, it was centrifuged at 4000 $\times g$ for 10 min and the supernatant was transferred to another test tube. The above procedure was repeated until the sample was exhausted. Finally, both of the acetone extracts were combined and filtered. The absorbance of the solution at 449 nm was determined, using acetone as blank, in a spectrophotometer UV/Vis Multiskan Spectrum (Thermo Scientific, Waltham, Massachusetts, USA). Results were expressed as milligrams of β -carotene per kilogram of crude oil, using a calibration curve with β -carotene standard.

Total polyphenols

The total polyphenols content was determined by the adapted Folin-Ciocalteu assay. Briefly, 10 g of oil were extracted with 150 ml of methanol in a separation funnel and, when the phases separated, the methanolic layer was centrifuged at 4000 $\times g$ for 15 min. The remaining oil was removed and the solution was distilled under reduced pressure. The extract obtained was mixed with Folin-Ciocalteu reagent and sodium carbonate solution (71 g \cdot kg⁻¹). The mixture was stirred and stored at room temperature for 0.5 h in the darkness. The absorbance was measured at 760 nm against a blank containing only Folin-Ciocalteu reagent, methanol and sodium carbonate. A standard calibration curve of gallic acid was used and results were expressed as milligrams of gallic acid equivalent per kilogram of crude oil [15].

Determination of vitamin E

The content of vitamin E (α -tocopherol) was determined by high-performance liquid chromatography (HPLC), according to the modified protocol of GLISZCZYŃSKA-ŚWIGŁO et al. [16]. An aliquot of the oil was diluted in the mobile phase, filtered through a membrane filter (pore size 0.20 μ m) and then injected into the chromatograph. An LC-20AD liquid chromatograph (Shimadzu, Tokyo, Japan) was used, equipped with a SIL-20A auto injector/HT, a communication module CBM-20A and diode array detector (PDA) at a wavelength of 295 nm. Quantification of vitamin E was conducted on a LiChrospher RP-18 column (250 mm \times 4.5 mm, particle size 5 μ m; Merck). A mixture of methanol and dichloromethane (85:15, v/v) was used as a mobile phase. The flow rate of the mobile phase was 0.8 ml \cdot min⁻¹, column temperature was kept at 35 °C and isocratic flow conditions were used. Concentration of α -tocopherol was calculated from the calibration curve and results were ex-

pressed as milligrams of tocopherols per kilogram of crude oil.

Antioxidant activity of oil

Oxygen radical-absorbance capacity

The oxygen radical-absorbance capacity (ORAC) assay was carried out using the following methodology. A volume of 3 ml of the following solution was prepared: 21 μ l of a 10 μ mol \cdot l⁻¹ solution of fluorescein, 2899 μ l of 75 mmol \cdot l⁻¹ phosphate buffer (pH 7.4), 50 μ l of 600 mmol \cdot l⁻¹ 2,2'-azobis(2-amidinopropane) dihydrochloride (AAPH) and 30 μ l of oil, which was dissolved in a 70 g \cdot l⁻¹ solution of methyl- β -cyclodextrin. Fluorescence was recorded on a spectrofluorometer with a thermostated multicell (model LS55; Perkin Elmer, Waltham, Massachusetts, USA). The ORAC value was calculated from a calibration curve using Trolox as an antioxidant standard and expressed as micromoles of Trolox equivalents (TE) per kilogram of oil [17].

DPPH radical-scavenging activity

DPPH radical-scavenging assay described by LEE et al. [18] was used with some modifications. This procedure was performed using 10 μ l of the extract and 990 μ l of the DPPH radical, in octane solution (20 mg \cdot l⁻¹). After 0.5 h of reaction at room temperature in the dark, the absorbance was read at 517 nm. For each studied sample, the percentage inhibition of the radical concentration was calculated and the results were expressed as millimoles of BHT equivalents per kilogram of oil, by the construction of a calibration curve using BHT as a standard.

Oxidative stability of oil

Using freshly extracted oil, the oxidative quality indices peroxide value (PV), thiobarbituric acid reactive substances value (TBARS), total polar compounds (TPC) and oxidative stability index (OSI) were determined. The effect of the antioxidant BHT to increase the useful life of the oil (oxidative stability) was measured by the active oxygen method (AOM).

Peroxide value

Peroxide value (PV) was determined by the International Dairy Federation (IDF) method [19]. This method is based on the ability of lipid peroxide to oxidize Fe²⁺ to Fe³⁺. A volume of 3.5 ml of a mixture of chloroform and methanol (7:3) was stirred for 10 s and, subsequently, 50 μ l of a solution of FeSO₄ (0.144 mol \cdot l⁻¹), BaCl₂ in HCl (0.4 mol \cdot l⁻¹) and 50 μ l of a solution of NH₄SCN (0.44 mol \cdot l⁻¹) were added to 1 ml of

the above solution. This mixture was incubated for a period of 20 min in darkness and, after this time, absorbance was determined at a wavelength of 510 nm. Results were expressed as milliequivalents of oxygen per kilogram of oil.

Anisidine value

Anisidine value (*AV*), which quantifies aldehydes as final degradation products, was determined according to IUPAC 2504 standard [20]. The oil samples, dissolved in octane, were allowed to react with *p*-anisidine solution in acetic acid (2.50 g·l⁻¹) for 10 min at room temperature. The absorbance of the resulting coloured complex was measured at 350 nm and results were expressed according to Eq. 1.

$$AV = 1.25 \times \frac{(1.2A_1 - A_2)}{m} \quad (1)$$

where A_1 is the sample absorbance, A_2 the absorbance of the blank and m is weight of sample (expressed in grams).

Total oxidation value

The total oxidation value (*TV*) was calculated through Eq. 2 [21].

$$TV = 2PV + AV \quad (2)$$

Thiobarbituric acid reactive substances

The end product of lipid peroxidation, malonaldehyde (MDA), reacts with 2-thiobarbituric acid (TBA) to produce a fluorescent complex that can be measured at 500 nm excitation and 520 nm emission wavelengths. The fluorescence was read by an LS-55 spectrofluorometer. Volumes of 80 µl of trichloroacetic acid (TCA, 10 g·l⁻¹) and 160 µl of TBA (60 g·l⁻¹) were added to 500 µl of oil. This mixture was incubated for a period of 20 min at 90 °C and then submerged to cold water for 10 min. After this period, 600 µl of butanol was added to it. The sample was stirred and the respective measurements were taken. TBA reactive substances values (*TBARS*) were expressed as millimoles of malondialdehyde per kilogram of crude oil using a calibration curve with MDA as a standard [22].

Total polar compounds

The content of total polar compounds (*TPC*) was determined according to IUPAC 2507 standard [20]. Polar and non-polar fractions were separated from 100 mg of oil dissolved in 2 ml of *n*-hexane using silica cartridge for solid-phase extraction (Sep-pak; Waters, Milford, Massachusetts, USA). The non-polar fraction was eluted

with 20 ml hexane : ethyl ether (90 : 10, v/v), the solvent was passed through while the sample was retained on the column. The second fraction (containing *TPC*) was eluted with 20 ml diethyl ether. The efficiency of separation by silica cartridges was checked by thin layer chromatography (plates of Silica Gel 60; 5 cm × 10 cm, 0.25 mm thickness; Merck) using *n*-hexane : diethyl ether : acetic acid (80:20:1 v/v/v) for plate development. Dried plates were exposed to iodine vapor to visualize the separation of two fractions. Solvents were removed at reduced pressure in a rotary evaporator and both fractions were weighed. The results were expressed as grams of polar compounds per kilogram of oil.

Oxidative stability index

Oxidative stability index (*OSI*) was evaluated by Rancimat model 679 (Metrohm, Herisau, Switzerland) following AOCS Official Method Cd 12b-92 [12]. Oil (2.5 g) was heated to a temperature of 120 °C and oxidized by bubbling air through the sample (20 l·h⁻¹). Volatile secondary products from lipid oxidation were carried by the air into a tube with distilled water, where conductivity was monitored. *OSI* is the time until the maximum rate of conductivity increase is achieved.

Active oxygen method

The AOM method was performed according to ANWAR et al. [23]. A volume of 30 ml of oil was supplemented with BHT at 100 mg·l⁻¹, which is a concentration accepted by Food and Agriculture Organization (FAO) [11]. In addition, a control sample was used. The test was conducted under accelerated conditions (100 °C ± 1 °C with an air flow of 1 150 ml·min⁻¹), until a time in which *PV* of 100 meq·kg⁻¹ oil was reached.

Thermal stability of oil

Differential scanning calorimetry

A sample of 5.0 mg of oil was submitted to heating at a constant rate of 10 °C·min⁻¹ from room temperature to 500 °C with a flow of air of 40 ml·min⁻¹. Heat fluxes were measured on Q20 instrument (TA Instruments, New Castle, Delaware, USA).

Thermogravimetric analysis

The thermal stability of the oil was evaluated by thermogravimetry on a thermogravimetric analyser Q50 (TA Instruments) by heating the crude oil sample in air, from 100 °C to 800 °C at a rate of 20 °C·min⁻¹. The Platinum Software (TA Instruments) was used to export and analyse graphical and numerical data.

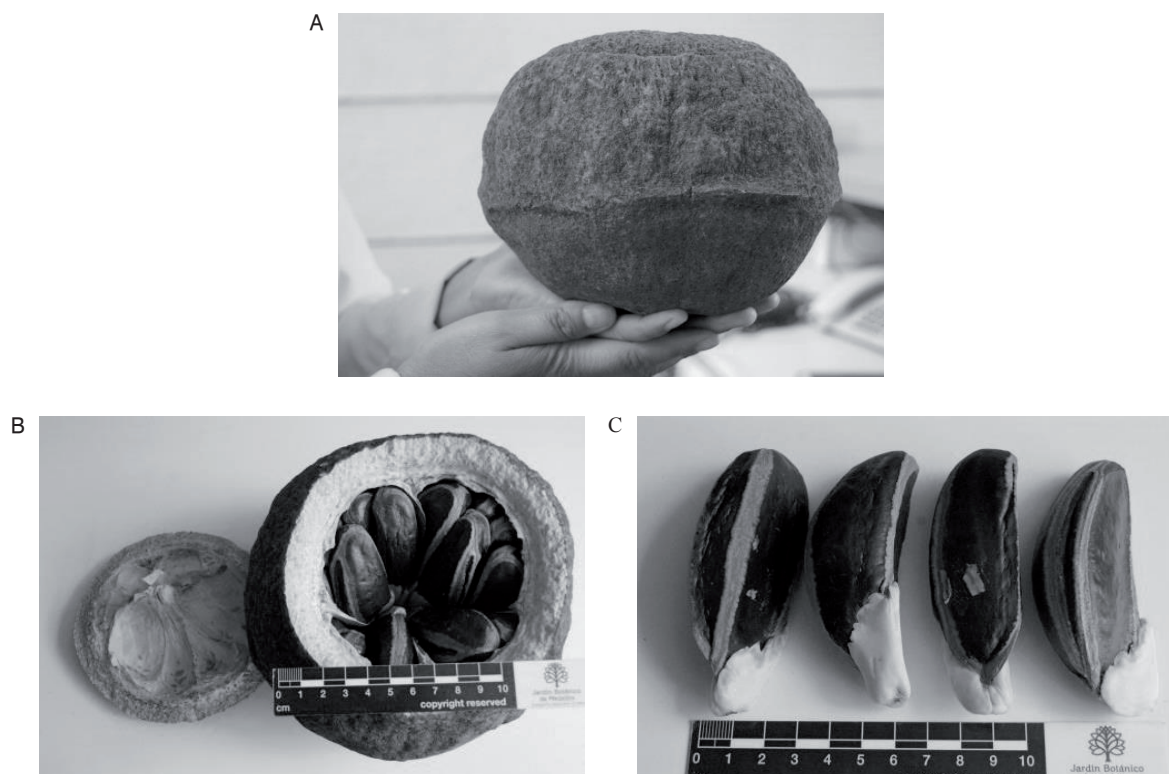


Fig. 1. Photographs of *Lecythis tuyrana* fruits.

A – whole fruit, B – fruit interior, C – almonds.

Statistical analysis

All experiments were carried out over triplicate samples and their mean values were reported. Software Statgraphics Centurion XVI (Statpoint Technologies, The Plains, Virginia, USA) was used for statistical analysis.

RESULTS

Fig. 1 shows three photographs of the fruits of *L. tuyrana* used in this research. The fruits reach a size of almost 18 cm of diameter, and a height near 15 cm. The whole fruit inside has three compartments where the seeds are found, can contain up to 18 almonds, which have brown testa. To obtain the oil, this layer was removed.

In almonds of *L. tuyrana*, a lipid content of 732.0 ± 27.1 g·kg⁻¹ was found. The extracted oil had an acid value of 2.70 ± 0.30 g·kg⁻¹ (expressed as oleic acid), saponification value 194.70 ± 14.54 g·kg⁻¹ oil (expressed as KOH), iodine value 64.02 ± 4.73 g·kg⁻¹ oil (expressed as I₂ equivalent). These results are similar to those reported for other seeds in the *Lecythis* genus [24–26]. Hydrophilic secondary metabolites, such

as total polyphenols, with antioxidant characteristics in the *Lecythis* oil seeds, were quantified in the polar extract of oil and were found at a level of 62.20 ± 3.50 mg·kg⁻¹ oil, expressed as gallic acid equivalent.

In the lipophilic phase of oilseeds, total carotenoids were found to be present at 3.00 ± 0.10 mg·kg⁻¹ of oil (expressed as β-carotene equivalent). In general, very low amounts were found in *Lecythis* almond oils, lower values have been reported in oilseeds and other nuts such as *Pistacia vera*. Carotenoids (of the type of β-carotene) and lutein were found at 4.0–10.0 mg·kg⁻¹ and 15.0–96.0 mg·kg⁻¹ of oil, respectively [27]. α-Tocopherol in *L. tuyrana* oil was found in a content of 32.20 ± 0.40 mg·kg⁻¹ oil, which was similar to data reported by BOLLING et al. for pecan (*Carya illinoensis*) and the Brazil nut (*Bertholletia excelsa*) [28].

Fig. 2 shows the fatty acid profile of *L. tuyrana* oil, with a high content of oleic acid (ω-9) 369.0 g·kg⁻¹, linoleic acid (ω-6) 272.0 g·kg⁻¹, palmitic acid 171.0 g·kg⁻¹, stearic acid 149.0 g·kg⁻¹ and, to a lesser extent, unsaturated acids like arachidonic acid (ω-6) 10.0 g·kg⁻¹, palmitoleic acid (ω-7) 7.0 g·kg⁻¹, and very low amounts of satu-

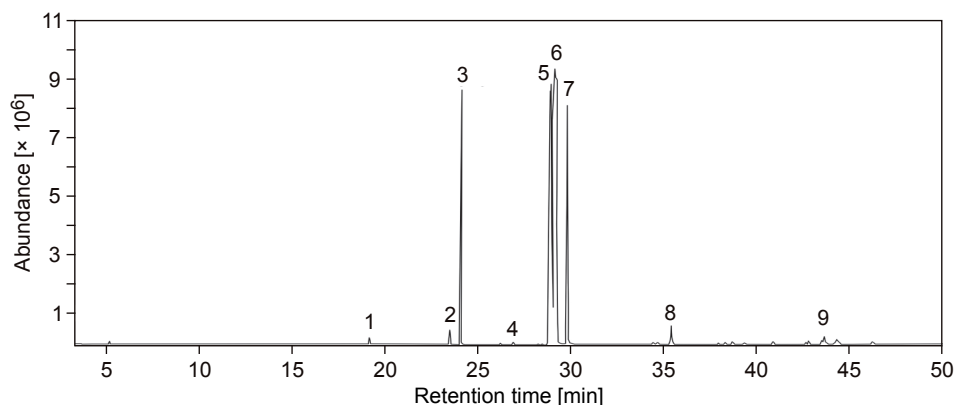


Fig. 2. Analysis of fatty acids methyl esters from *Lecythis tuiyana* oil.

Chromatogram was obtained by gas chromatography – mass spectrometry. The y-axis represents total ion counts. Retention: 1 – myristic acid (19.538 min), 2 – palmitoleic acid (ω -7, 23.734 min), 3 – palmitic acid (24.376 min), 4 – behenic acid (27.053 min); 5 – linoleic acid (ω -6, 29.050 min), 6 – oleic acid (ω -9, 29.264 min), 7 – stearic acid (29.924 min), 8 – arachidonic acid (ω -6, 35.351 min), 9 – margaric acid (43.348 min).

rated fatty acids, such as myristic acid $3.0 \text{ g}\cdot\text{kg}^{-1}$, behenic acid $2.0 \text{ g}\cdot\text{kg}^{-1}$ or margaric acid $1.0 \text{ g}\cdot\text{kg}^{-1}$.

The antioxidant activity evaluated by free radical-scavenging assays is a mechanism of hydrogen atom transfer (HAT) and, for *L. tuiyana* oil, it was evaluated by the capacity of trapping peroxy radical (ROO^\bullet) by ORAC method with a result of $3641 \pm 323 \mu\text{mol}\cdot\text{kg}^{-1}$ (expressed as TE). This value falls within a range determined by NINEALI et al. [29] with 33 olive oils from various regions of Italy and Spain, while phenol and tocopherol values were similar to those for *Lecythis* oil. Another important test to trap free radicals (DPPH^\bullet) implemented in recent years is a lipophilic DPPH assay to determine oxidative stability of oils and fats. In *Lecythis* oil, the obtained result was $14.70 \pm 0.50 \text{ mmol}\cdot\text{kg}^{-1}$

oil (expressed as BHT), which is similar to the values for olive oil ($15.425 \text{ mmol}\cdot\text{kg}^{-1}$), greater than canola oil ($7.19 \text{ mmol}\cdot\text{kg}^{-1}$) and less than the soybean oil ($18.365 \text{ mmol}\cdot\text{kg}^{-1}$) [18]. Low values of *PV* ($3.64 \pm 0.45 \text{ meq}\cdot\text{kg}^{-1}$) and *TBARS* ($2.69 \pm 0.21 \mu\text{mol}\cdot\text{kg}^{-1}$) were determined for *L. tuiyana* oil.

Oxidative and thermal stability

Oxidative stability of oils determined as *OSI* is completely dependent on the temperature and oxygen flow used. In this case, due to the oil composition, a temperature of 120°C with an air flux of $20 \text{ l}\cdot\text{h}^{-1}$ was used, and an *OSI* value was 3.47 h (Fig. 3).

The AOM method facilitates characterization of oil regarding degradation kinetics, under ac-

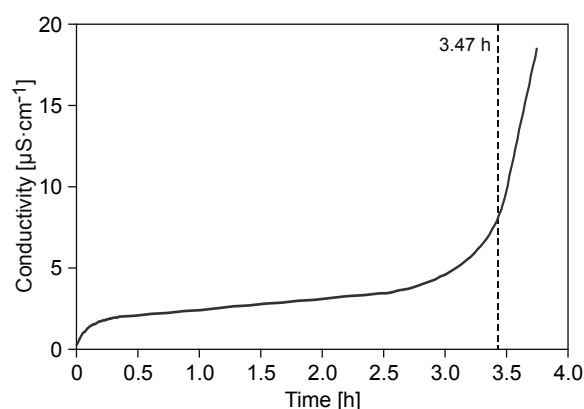


Fig. 3. Time course of conductivity of *Lecythis tuiyana* oil at temperature of 120°C with air flux of $20 \text{ l}\cdot\text{h}^{-1}$.

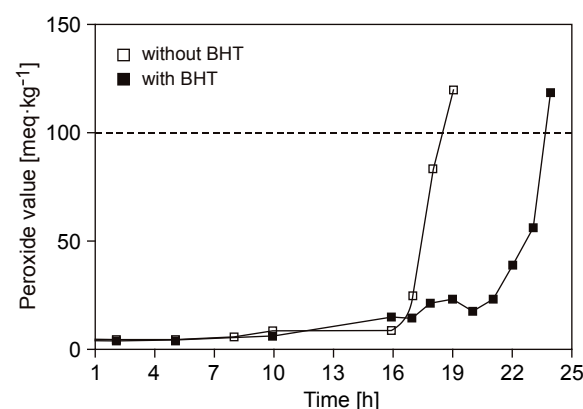


Fig. 4. Formation of peroxides in *Lecythis tuiyana* oil supplemented with and without butylhydroxytoluene. BHT – butylhydroxytoluene ($100 \text{ mg}\cdot\text{l}^{-1}$).

celerated conditions. *PV* is used to indicate primary oxidative deterioration of fats and oils. The *PV* measurements were performed over time until reaching 100 meq·kg⁻¹ (accepted as the total oxidative rancidity of an oil). Eq. 3 and Eq. 4 show the kinetic model for the control (oil without antioxidant) and for oil supplemented with BHT 100 mg·l⁻¹, respectively.

$$PV = e^{(11.2338 + 0.849749t)}; R^2 = 94.0 \% \quad (3)$$

$$PV = e^{(6.78086 + 0.479106t)}; R^2 = 96.6 \% \quad (4)$$

where *t* is time in hours.

The model to reach 100 meq·kg⁻¹ was an exponential type model, and its findings were *PV* values of 18.64 h for the control and 23.77 h for the sample with BHT. These results showed that BHT can inhibit the oxidative process of *L. tuyrana* oil by 27.5 % at a temperature of 100 °C (Fig. 4).

Fig. 5 shows the results of TGA for *Lecythis* oil. Thermal degradation started when the oil temperature was of approximately 200 °C and the mass loss became continuous up to 550 °C, being very significant at 340 °C. Similar results were reported for vegetable oils and it was suggested that a continuous breaking of the hydrocarbon chains in molecules, such as carbon dioxide and monoxide, volatilize carbon easily. This is favoured by the proportion of unsaturated fatty acids in *Lecythis* oil, which is 64 % [30]. Further mass loss occurs at 420 °C and 530 °C due to degradation of saturated fatty acids and polymers formed during the final stages of peroxidation [31].

Results of DSC assay are shown in Fig. 6. An increase in the heat flux, characteristic of exothermic reactions, can be seen above 200 °C.

DISCUSSION

Lipids belong to main nutrients of the human diet, and they are of great importance for consumers and food industry. Seed oils constitute a significant part of the typical human diet because they are a source of essential nutrients such as fatty acids, tocopherols and phytosterols. Health conscious consumers require from the industry to provide oilseeds and oils which are rich in beneficial compounds including ω-3, ω-6, ω-9 fatty acids, tocopherols, carotenoids and polyphenols.

In *L. tuyrana* oil, mainly 9 fatty acids are found, with carbon chains from 14 to 22 carbon atoms, with a ratio of percentage of saturated and unsaturated (*I/S*) of 2.0 %. This index shows a high proportion of unsaturated fatty acids, and oleic and linoleic acids as key components. Oleic acid is the

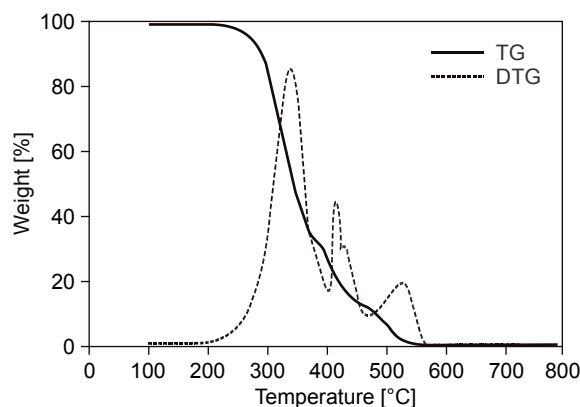


Fig. 5. Thermogravimetric analysis for crude *Lecythis tuyrana* oil.

TG – curve of weight loss, DTG – first derivative of TG curve.

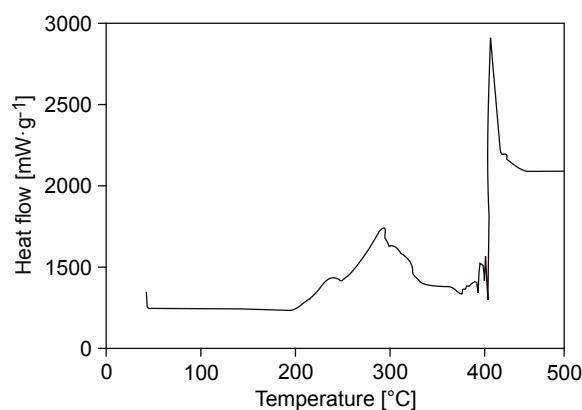


Fig. 6. Differential scanning calorimetry for *Lecythis tuyrana* oil.

mono-unsaturated fatty acid (MUFA) most commonly used in Western diet and is present in most oils of animal or plant origin. Also, oleic acid is an indicator of quality of oils for culinary use, such as olive, sunflower and canola oil. It has multiple health benefits, including a decrease of the risk of heart disease by 20–40 %, primarily through the reduction of low density lipoproteins (LDL) cholesterol. Beneficial effects on cerebrovascular risk factors, such as thrombogenesis, and on insulin sensitivity were reported [32–34].

The linoleic acid content is similar to that reported for *Lecythis pisonis* [26], higher than olive and soybean oil [35], and lower than those reported for some types of sunflower and canola oils [37, 37]. Linoleic acid is an essential fatty acid with many health effects like the ability to inhibit proliferation of colorectal cancer and decrease growth of atherosclerotic plaque [38]. Palmitoleic acid, which is a ω-7, a minor fatty acid, known for

its beneficial action on the skin and mucous membranes [39].

Content of vitamin E in *L. tuyrana* almonds and oil was similar to other massively commercialized nuts. An increase in the consumption of this type of products is beneficial for health due to an important role in protection of polyunsaturated fatty acids (PUFA) and other components of cell membranes, as well as protection of LDL from oxidation by free radicals. Vitamin E is the main lipid-soluble antioxidant in the cellular antioxidant defense system and is retrieved exclusively from the diet. Its consumption must be such as to ensure a 2.25-times higher plasma concentration than that of cholesterol [40].

The oxidative stability of oil is determined by the relationship between saturated fatty acids, unsaturated fatty acids and secondary metabolites present, and it is reflected by its antioxidant activity, specifically those measurable by lipophilic methodologies. *ORAC* and *DPPH* values determine the antioxidant potential of a food product, with protective properties on many systems such as lipid membranes and cell wall. *Lecythis* oil has values similar to many oils reported with beneficial health effects, such as olive and canola oils.

The oxidation quality of the freshly extracted oil is characterized by *PV* ($3.64 \text{ meq}\cdot\text{kg}^{-1}$), which registers the initial and intermediate products of lipid peroxidation. *TBARS* value ($2.69 \mu\text{mol}\cdot\text{kg}^{-1}$) and *TPC* ($60.0 \text{ g}\cdot\text{kg}^{-1}$) are associated with the presence of final oxidation compounds. In addition, *TV* of 3.54 shows that the oil did not suffer oxidative deterioration in the extractive process and its natural antioxidant ingredients were protected. These values are a particularly characteristic for each oil and they are indicators of its potential shelf life. The values found for each of the oxidative stages reflected a high stability of *Lecythis* oil.

OSI of oils and fats determined by the Rancimat assay is based on the induction time of oxidation of the sample by exposure to elevated temperatures and air flow in accelerated conditions. The result for *Lecythis* oil (*OSI* of 3.47 h) is comparable to those reported for maize oil and cottonseed oil [41]. *OSI* is a fundamental tool in technology assessment, quality and shelf life of oils and fats, which facilitates evaluation and optimization of stabilization strategies for oils and fats by antioxidant compounds [42].

The thermal stability of crude vegetable oils obtained by cold extraction processes is an indicator of quality for its use in different food matrices or cosmetics. *Lecythis* oil is characterized by a high proportion of unsaturated fatty acids (*I/S* = 2.02)

containing $370 \text{ g}\cdot\text{kg}^{-1}$ oleic acid and $270 \text{ g}\cdot\text{kg}^{-1}$ linoleic acid, which suggests a poor thermal stability in frying processes where temperatures reach 180–200 °C. Tests of TGA and DSC type, commonly used in the industry of fats and oils, were done to confirm the thermostability. These assays characterize the phenomena of decomposition, sublimation, reduction, oxidation and adsorption or desorption of gases or vapours, being a function of temperature change, thermal reactivity, initial temperature, final combustion (in the case of fuel oil) and degradation rate [43, 44].

DSC is a technique used to measure heat fluxes in respect to temperature changes associated with phase transitions or chemical reactions such as oxidation or hydrolysis. This technique was used to determine the thermal profile of the oil, the results of this process being reactions associated with formation of degradation products, such as peroxides or polar compounds, commonly formed in lipidic matrices due to the effects of diffusion of oxygen reached in the dynamic process of heating and aeration of the sample [45, 46].

To retard the oxidative deterioration of oils caused by radical oxygen species, antioxidants are used, which reduce the oxidation process and improve the organoleptic quality of the final product. Antioxidants in food determine shelf life and reduce nutritional losses. However, they never improve or regenerate the quality of a highly oxidized food product. Antioxidants to be used in food must meet certain requirements such as being odourless, tasteless, effective at low contents, easy to incorporate, supporting processing conditions and stable in the finished product [47].

The oxidation process of *Lecythis* oil under accelerated conditions ($100 \pm 1^\circ\text{C}$ with an air flow of $1150 \text{ ml}\cdot\text{min}^{-1}$) was reduced by 27.5 % using BHT as an antioxidant at $100 \text{ mg}\cdot\text{l}^{-1}$. Therefore, we were able to obtain a stabilized oil with high nutraceutical and functional potential that allows its use in food. In addition, research is under way on the use of natural antioxidants with a greater efficiency than BHT.

CONCLUSIONS

L. tuyrana oil obtained by pressing is a source of unsaturated fatty acids such as oleic acid ($369.0 \text{ g}\cdot\text{kg}^{-1}$) and linoleic acid ($272.0 \text{ g}\cdot\text{kg}^{-1}$), which are stabilized by carotenoids, lipophilic antioxidants ($3.00 \pm 0.10 \text{ mg}\cdot\text{kg}^{-1}$ β -carotene) and tocopherols ($32.20 \text{ mg}\cdot\text{kg}^{-1}$). This proposal is supported by oxidative stability values, namely, *TBARS*, *PV* and *TV* and, finally, by the low *TPC*

value. The oil was found very stable compared to many other newly extracted oils. TGA, DSC and OSI assays also demonstrated thermal stability for the oil, which, although not stabilized by additives, did not show thermal decomposition below 200 °C. However, the high presence of unsaturated fatty acids (close to 64.0 %) can cause significant oxidative damage. *Lecythis* oil is a novel oil with interesting nutraceutical properties, which is proposed to be used as an ingredient in fresh preparations, conventional frying, vacuum frying and baking processes.

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