



## Amazon Oils from Andiroba (*Carapa* sp.) and Babassu (*Orbignya* sp.) for Preparation Biodiesel by Enzymatic Catalysis

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### Abstract:

**Background:** Biodiesel represents an alternative energy source with economic, social, environmental, and technological advantages over nonrenewable fuels.

**Objective:** This study investigated two oils derived from plants of the Brazilian Amazon, andiroba (*Carapa* sp.) oil and babassu (*Orbignya* sp.) oil, as possible triacylglyceride sources for the production of biofuels by lipase B from *Candida antarctica* (CALB).

**Methods:** The production of biodiesel ethylic by enzymatic catalyst was carried out in 3-mL Erlenmeyer flasks containing 150 mg (154  $\mu$ L) of either babassu or andiroba oil, 475  $\mu$ L of ethanol, and 15 mg of CALB.

**Results:** The physical, chemical, and spectroscopic properties of these oils and their resulting biofuels were also reported. CALB exhibited high activity in the alcoholysis of babassu and andiroba oils under mild conditions and produced biodiesel ethyl esters in high yields (90% and 94%, respectively). The oils and biodiesel were characterized using <sup>1</sup>H NMR, Chromatography Gas–Mass Spectrometry (GC-MS), Fourier Transform Infrared Spectroscopy (FT-IR), and thermogravimetric analysis (TGA) being the main findings of the study.

**Conclusion:** Ethanolysis of babassu and andiroba oils by catalysis enzymatic (CALB), showed to be a successful route in the synthesis of ethylic biodiesel in high yield. The corresponding biodiesels were characterized by <sup>1</sup>H NMR spectra, GC-MS, FT-IR, and TGA analyses. Results described here provide relevant information for the further research uses andiroba and babassu oils, the alternatives to other oils in synthesis of biodiesel.

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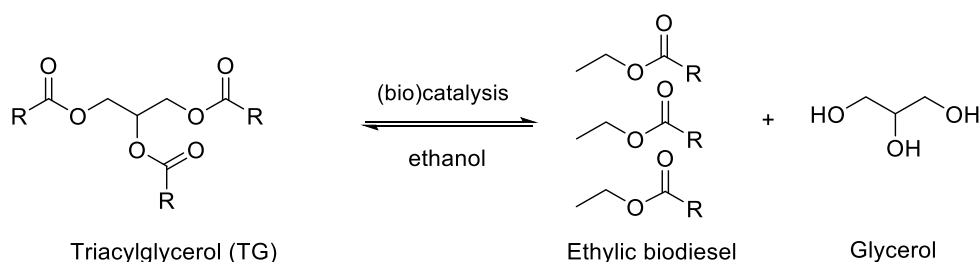
### 1. INTRODUCTION

Biodiesel has emerged as an environmentally-friendly and renewable alternative fuel to petroleum-based fuels [1], chemically is defined as a monoalkyl ester-based diesel fuel that is derived from vegetable oils or animal fats and exhibits properties similar to those of mineral-based diesel fuels

[2-4]. Therefore, biodiesel can serve as an ideal candidate either as a blending component for gasoline or as a direct replacement of diesel fuel. It plays an important role in the area of novel fuels [3, 5]. The high oxygen content in biodiesel allows the complete combustion of biodiesel in engines, thus exhaust emissions have lower amounts of particulates, hydrocarbons, gases like CO, CO<sub>2</sub> and SO<sub>x</sub>, making this fuel environmentally-friendly [1, 6].

Currently, biodiesel is produced on a commercial scale largely *via* catalysis with alkali metal hydroxides, mainly sodium hydroxide or sodium methoxide (Scheme 1). However,

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**Scheme (1).** (Bio)transesterification reaction of triacylglycerols with ethanol for the production of biodiesel.

this method has some process limitations that are considered drawbacks of chemical biodiesel [7]. Indeed, this synthetic route is normally associated with the side-formation of free fatty acids in quantities greater than 0.5%, which can lead to soap formation, and water exceeding 0.3%, which results in a reduction of the reaction yield [8]. Saponification can also lead to the formation of emulsions that reduce the overall ester yield and make it difficult to recover biodiesel [9].

Acid catalysts are also utilized in the synthesis of biodiesel, typically when the fatty acid content of the reactant oil is high. However, compared to alkali hydroxide catalysts, acid catalysts tend to produce lower yields, require longer reaction time, and require high reaction temperatures [10].

These drawbacks of commercial catalysis routes can be minimized through the use of enzymes as catalysts in transesterification reaction [7] (Scheme 1). Synthesis of biodiesel using enzyme catalysts is attracting the interest of researchers and biodiesel producers as it is a green approach to produce renewable fuel by environmental benign biocatalyst [1, 11-13]. Among the enzymes used in the production of biodiesel, the lipases are the most employed.<sup>13</sup> Lipases are hydrolases (E.C. 3.1.1.3) that act on carboxyl ester bonds in triglycerides to yield fatty acids and glycerol [14]. Consequently, this enzymes can be a route to biodiesel production with a number of environmental and economic advantages over chemical synthesis routes [15, 16]. These advantages include milder reaction temperatures, use of lower alcohol to oil ratios, elimination of the treatment costs associated with the recovery of chemical catalysts, reuse and low water consumption in the purification of the biodiesel [17, 18]. A variety of biolipid sources can be used for the production of biodiesel such as soybean oil [19] castor oil [12], and less common oils such as palm, jatropha [20], microalgae [21, 22] and karanja oil [23], consequently the biodiesel can be produced locally using various feedstocks depending upon its availability in as particular region and thus provides energy security. There are currently very few studies on enzyme catalysis of babassu and adiroba oils for the formation of biodiesel [24, 25].

The present paper addresses the use of Brazilian Amazon babassu (*Orbignya* sp.) and andiroba (*Carapa* sp.) oils as sources of biodiesel *via* enzymatic reactions. Physicochemical and spectroscopic techniques were used to characterize the fatty acid ethyl esters (FAEE) formed by CALB. The chemical composition of the oils and their physical-chemical properties were also studied.

## 2. MATERIALS AND METHODS

### 2.1. Materials

Samples of andiroba and babassu oils were purchased at a popular market in the Brazilian Amazonian region (Altamira, Pará, Northern Brazil) in April 2011 and stored at  $-4^{\circ}\text{C}$  until use. Ethanol (Spectroscopic grade) was purchased from Tedia (USA) and *n*-hexane and ethyl acetate (PA grade) were obtained from Quimis (Brazil) and used without further purification. Lipase acrylic resin from *Candida antarctica* (CALB  $\geq 20.000$  U/g) was purchased from Sigma-Aldrich.

### 2.2. Physicochemical Analysis of the Vegetable Oils

The acidity index, saponification number, ester content, peroxide, and moisture content found in andiroba and babassu oils were analyzed by the methods described by the Standards of Analytical Adolfo Lutz Institute (Lutz 2005) [28].

### 2.3. Production of Biodiesel by Enzymatic Ethanolysis

The biodiesel ethylic from enzymatic catalysis, by CALB, was carried out in 3-mL Erlenmeyer flasks containing 150 mg (154  $\mu\text{L}$ ) of either babassu or andiroba oil, 475  $\mu\text{L}$  of ethanol, and 15 mg of CALB. The mixtures were incubated at  $32^{\circ}\text{C}$  on a 130-rpm orbital shaker (Tecnal model, TE-421, Brazil) [26]. The reaction progress was monitored by thin-layer chromatography (TLC) using aluminum-backed pre-coated silica gel 60 F<sub>254</sub> layers with *n*-hexane and ethyl acetate (9:1) as eluent. The plates were visualized through iodine vapor exposure. After 24 h, the reaction was completed, the reaction mixture filtered over, the organic phase dried over sodium sulfate, and the solvent removed under reduced pressure. The product (biodiesel) was purified by gel silica column chromatography with a mixture of *n*-hexane and ethyl acetate (9:1) as eluent and characterized using  $^1\text{H}$  NMR and GC-MS analysis.

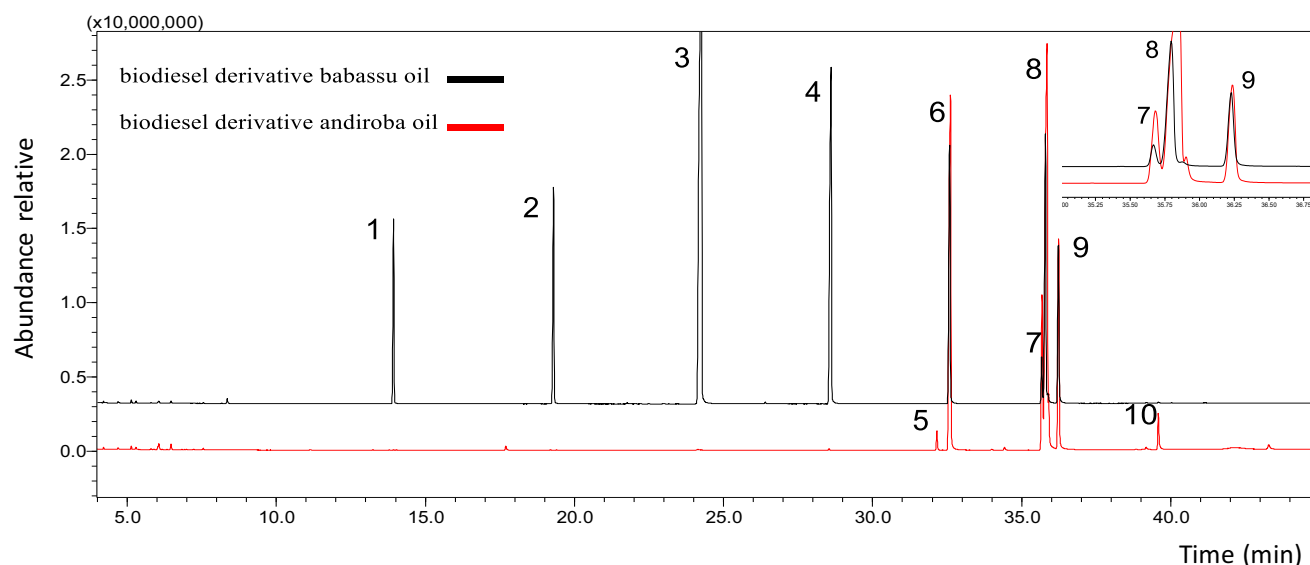
### 2.4. Biodiesel Characterization

#### 2.4.1. Fourier Transform-Infrared (FT-IR)

FT-IR spectra were recorded on a SHIMADZU IRAffinity-1 spectrometer. The samples were prepared as thin liquid films, and spectra were recorded between  $4000\text{--}400$   $\text{cm}^{-1}$  with a resolution of  $2$   $\text{cm}^{-1}$  and averaged over 64 scans.

#### 2.4.2. Gas Chromatography-Mass Spectrometer (GC-MS)

Crude oils from andiroba and babassu were analyzed as ethyl esters for the determination of the fatty acid content *via* gas chromatography (GC-MS) using a Shimadzu GC2010



**Fig. (1).** Gas chromatograms (GC-MS) of biodiesel derived from babassu and andiroba oils *via* CAL-B catalysis *via* CALB catalysis (See Table 2).

Plus Gas Chromatography system equipped with a mass spectrometer detector (Shimadzu MS2010 Plus) with electron ionization (70 eV) and a DB-5 Agilent J&W column (30 m  $\times$  0.25 mm  $\times$  0.25 mm). Helium was used as a carrier gas at a pressure of 55 kPa, with the following parameters: split ratio, 1:10; injection volume, 1  $\mu$ L; injector temperature, 250°C; detector temperature, 250°C; and oven temperature programmed to ramp from 50°C to 250°C at 5°C/min over the total time of analysis (45 min) [26].

#### 2.4.3. Proton Nuclear Magnetic Resonance ( $^1\text{H}$ NMR)

$^1\text{H}$  NMR spectra were recorded on an Agilent Technologies 500/54 Premium Shielded. All samples (0.020 mL) were prepared by dissolving in 0.6 mL of deuterated chloroform ( $\text{CDCl}_3$ , Cambridge Isotope Laboratories, USA) and TMS as the internal standard. The chemical shifts were expressed in ppm and coupling constants ( $J$ ) in Hz.

#### 2.4.4. Thermogravimetric Analysis (TGA)

Thermogravimetric analyses were performed on a TA Instruments Q-600 under a dynamic nitrogen atmosphere using a 50 mL/min flow rate and a heating rate of 10°C/min at 1000°C.

#### 2.4.5. Fluorescence Spectroscopy

Fluorescence contour maps (emission/excitation) of the samples were recorded using a bench fluorescence spectrophotometer (Cary Eclipse, Varian). The excitation wavelength was scanned between 300 and 450 nm, and emission was monitored from 300 to 800 nm at intervals of 2 nm with excitation and emission with slits of 10 nm. Fluorescence was measured at a 90° angle relative to the excitation light (right-angle geometry). All counter maps were recorded at room temperature (296 K), using 10-mm path length quartz cells. The samples were used to obtain the contour maps.

### 3. RESULTS AND DISCUSSION

The quality and physicochemical properties, including fatty acid profile, saponification index, ester index, peroxide index, and moisture content of the feedstock influence the transesterification reaction by enzymatic catalysis. These metrics dictate feedstock quality and, therefore, influence the transesterification reaction and ultimately the quality of the biodiesel generated [27].

The feedstock used in this study was characterized according to the methodology recommended by official methods of the Adolfo Lutz Institute (2005) [28]. The relevant physicochemical properties of babassu and andiroba oils are summarized in Table 1.

**Table 1.** Physicochemical characteristics of babassu and andiroba oils determined by the Standards Analytical Adolfo Lutz Institute (Brazil) [28].

Physicochemical Analysis	Oils	
	Andiroba	Babassu
Acidity index (mg KOH/g)	12.5	0.9
Saponification index (mg KOH/g)	208	174
Ester index (mg KOH/g) (%)	195 (94)	173 (99)
Peroxide index (meq/kg)	7.5	3
Moisture content (% $\text{H}_2\text{O}$ )	0.4	0.2

It has been found that an excess of free fatty acids in the presence of base-catalyzed transesterification leads to competing saponification reactions. The acidity index of the babassu oil was found to be 0.9 mg KOH/g, which is condu-

cive for processing the oil into biodiesel and should not compromise the enzymatic transesterification reaction. Silva *et al.* [29] found a similar acidity index of 0.6 mg KOH/g for babassu oil (obtained from the Brazilian Amazonian region), which is less than the upper limit allowed by Brazilian National Agency of Petroleum, Natural Gas and Biofuels (*Agência Nacional do Petróleo, Gás Natural e Biocombustíveis - ANP*). The acidity index of the andiroba oil was found to be 12.5 mg KOH/g, which is high for base-catalyzed transesterification; however, it was lower than 36.1 mg KOH/g, which was reported by Lha *et al.* [30] for andiroba oil (obtained from Amazon region). A high acidity index can promote intramolecular reactions that affect the thermal stability of biodiesel, besides, damages caused to the engine.

The values of the saponification index for the andiroba and babassu oils were 174 and 208 mg KOH/g, respectively, which are close to those reported by Andrade *et al.* [31] (194 mg KOH/g for andiroba oil) and Carvalho *et al.* [22] (205 mg KOH/g for babassu oil). The triglyceride values found for the andiroba and babassu oils were 94% and 99%, respectively [23].

Using the Karl Fisher method, we found that the babassu oil contained 0.2% H<sub>2</sub>O by mass, while andiroba oil contained 0.4% H<sub>2</sub>O. One of the advantages of enzymatic catalysis for the transesterification reaction is the maintenance of high yield of the reaction even in the presence of some amount of water in oil, this peculiar behavior can be explained by lipase catalyzes this reaction at the lipid-water interface, for example, Rossett *et al.* [12] demonstrated that lipase-catalyzed transesterification reactions are less affected by sample water content and varying the amount of water from 0.1-4% in the CALB-catalyzed transesterification of soybean oil did not substantially influence the reaction yield (86-89%, respectively).

The peroxide value found for babassu oil was 2.59 meq/kg, which is much lower than the value of 7.48 meq/kg determined for andiroba oil. Bataglion *et al.* [32] recently reported a much lower peroxide value of 1.2 meq/kg for andiroba oil, this discrepancy may be related to the differences in the management of oil extraction and conservation.

The ethanolysis of babassu oil by lipase from CALB (15% w/w) gave an overall 97% yield (w/w) after 24 h (132 rpm, 32°C). The biodiesel derived from andiroba oil was obtained in 90% yield (w/w) after purification *via* a similar route. Paiva *et al.* [33] studied the ethanolysis the babassu using alkaline hydrolysis (1.0%) under ultrasound and showed that KOH is preferred over NaOH because better conversions were obtained with KOH and the purification steps were simpler.

The fatty acid profile of the babassu and andiroba-derived biodiesels was determined by GC-MS analysis. Each peak in the chromatograms (Fig. 1) was analyzed and the fatty acids were identified using an MS database (NIST 5.0).

The relative percentage of fatty acid esters was calculated from the total ion chromatography by integrator, and the results are summarized in (Table 2 and Fig. 1).

The babassu-derived biodiesel was found to contain five saturated fatty acid ethyl esters (FAEEs), namely caprylic, capric, lauric, palmitic, and stearic, and saturated linolenic, oleic, and palmitoleic FAEEs. However, the biodiesel derived from andiroba oil was composed of saturated myristic, stearic, and arachidic FAEEs and unsaturated, specifically, oleic, linoleic, and palmitoleic FAEEs. Saturated lauric (39%) and palmitoleic (18%) FAEEs were the major components of the babassu oil-derived biodiesel, while the biodiesel derived from andiroba oil was rich in unsaturated oleic (43%) and palmitic (30%) FAEEs (Table 2).

**Table 2. Fatty Acid Ethyl Ester (FAEE) composition of biodiesel samples derivative babassu oil and andiroba oils as determined by GC-MS analysis.**

Fatty Acid <sup>a</sup>	Molecular Formula <sup>a</sup>	wt.% of FAEE in Biodiesel			
		Babassu	Retention Time (min)	Andiroba	Retention Time (min)
Caprylic (1)	C <sub>8</sub> H <sub>16</sub> O <sub>2</sub>	6.2	13.9	-	-
Capric (2)	C <sub>10</sub> H <sub>20</sub> O <sub>2</sub>	8.0	19.3	-	-
Lauric (3)	C <sub>12</sub> H <sub>24</sub> O <sub>2</sub>	38.9	24.3	-	-
Palmitoleic (4)	C <sub>16</sub> H <sub>30</sub> O <sub>2</sub>	17.8	28.6	-	-
Myristic (5)	C <sub>14</sub> H <sub>28</sub> O <sub>2</sub>	-	-	0.8	32.1
Palmitic (6)	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	10.5	32.5	29.7	32.5
Linoleic (7)	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	1.5	35.6	10.1	35.6
Oleic (8)	C <sub>18</sub> H <sub>34</sub> O <sub>2</sub>	11	35.8	42.9	35.8
Stearic (9)	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	5.71	36.2	12.6	36.2
Arachidic (10)	C <sub>20</sub> H <sub>40</sub> O <sub>2</sub>	-	-	1.7	39.5

<sup>a</sup> % of FAEE corresponding fatty acid.

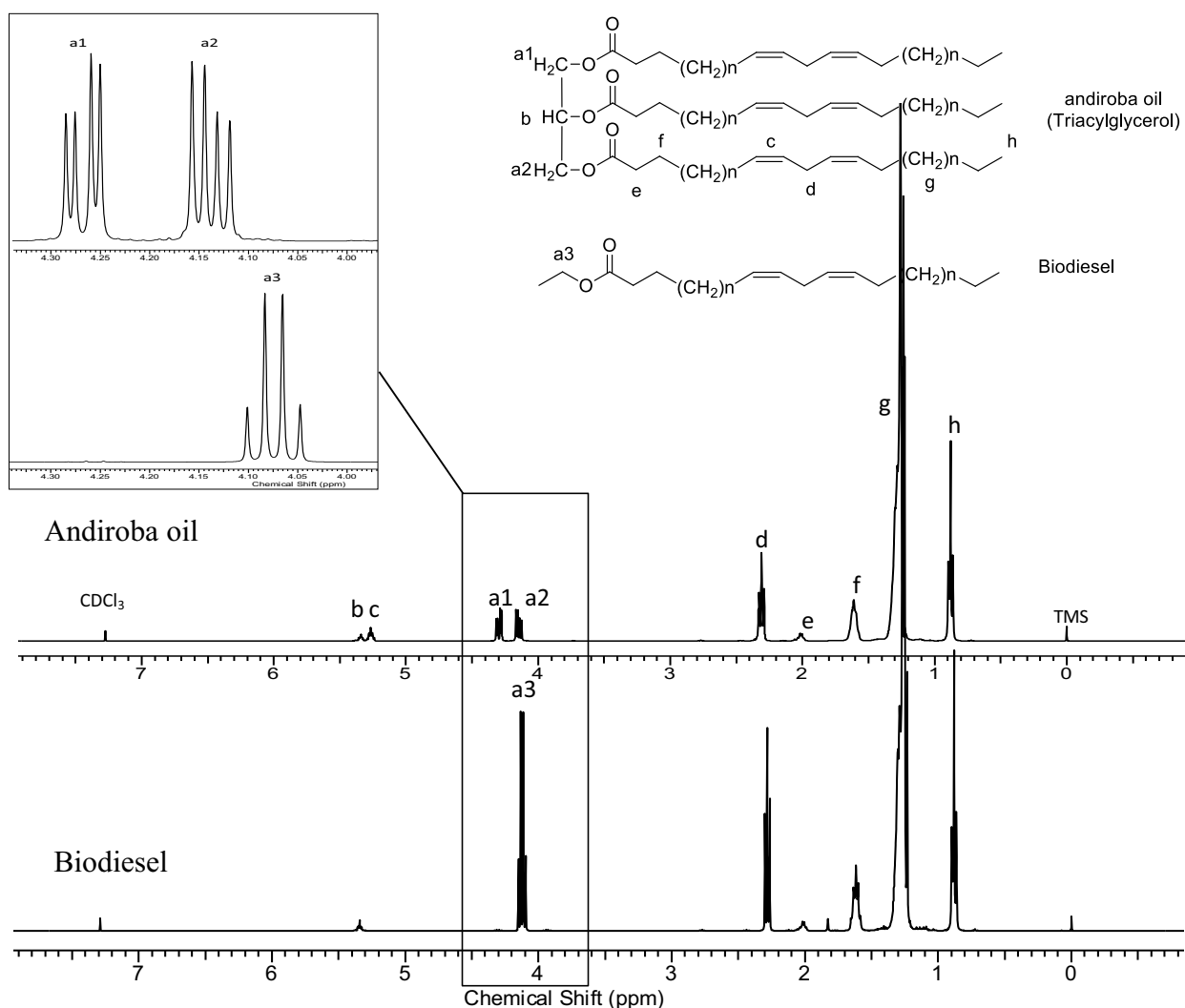
This high lauric acid content makes babassu oil an excellent candidate for biodiesel production and derives from a large quantity of short-chain lauric ester in the babassu oil, which undergoes more efficient catalysis by CALB than long-chain esters [34]. Babassu oil biodiesel is nonconventional biodiesel that is characterized by short ethyl ester chains, mostly C12:0 and C16:0, which results in less dense biodiesel [35]. This result also suggests that using babassu oil as a replacement for current feedstock in the synthesis of biodiesel from heterogeneous catalysts may result in higher yields [7]. However, biodiesel from andiroba oil was rich in the unsaturated, oleic (43%) and palmitic (30%) FAEs.

The  $^1\text{H}$  NMR spectra of andiroba oil and the corresponding biodiesel exhibited many overlapping peaks, as shown in (Fig. 2). Two doublet of doublets, **a1** and **a2**, were present in the  $^1\text{H}$  NMR spectra of andiroba oil at  $\delta = 4.30$  ( $J = 11.9$  Hz) and  $\delta = 4.15$  ( $J = 11.9$  Hz), respectively (Fig. 2, top); these signals corresponded to hydrogen atoms of the  $-\text{CH}_2$  group in triacylglycerol. However, the  $^1\text{H}$  NMR spectra of the andiroba biodiesel (Fig. 2, bottom) exhibited only a quartet (**a3**) in the same spectral range, with  $\delta = 4.12$  ( $J = 7.04$  Hz), corresponding to methylene protons. The absence of other peaks

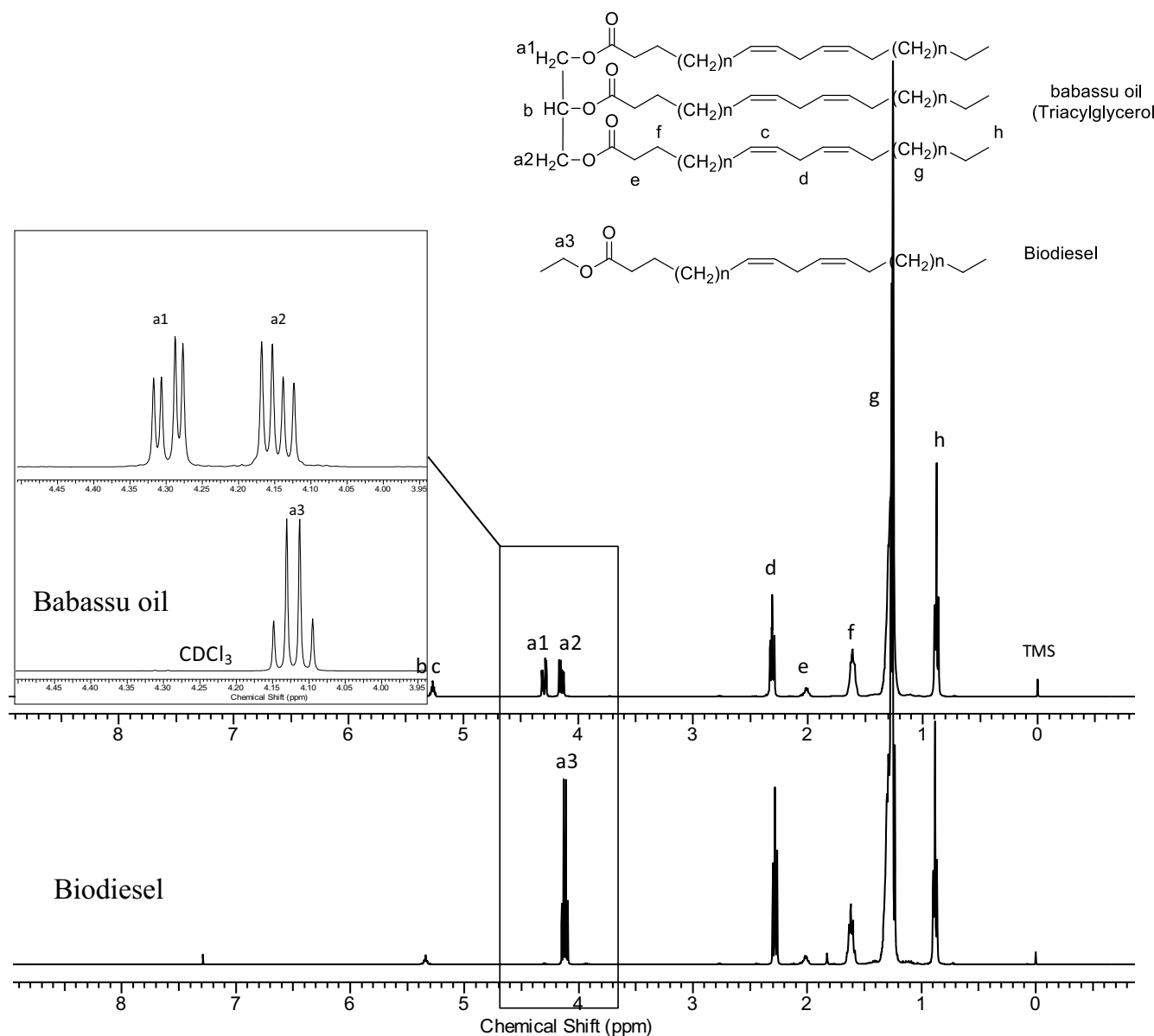
corresponding to mono-, di-, and triglycerides confirmed the total conversion of fatty acids to their respective ethyl esters. The multiplet at  $\delta = 5.36$  arises from the central protons (**b**) of triacylglycerol and the multiplet at 5.31 ppm was due to the olefinic protons (**c**) (Fig. 2, top). The bis-allylic proton signal of polyunsaturated fatty acid ethyl esters in the biodiesel (such as linoleic acid) generally appear as the triplet signal at  $\delta = 2.28$  with  $J = 7.53$  Hz (**d**, Fig. 3, bottom).

The multiplet signal at  $\delta = 1.98$  to  $2.07$  corresponded to the  $\alpha$ -methylene protons of the ethyl esters (**e**), while the  $\beta$ -methylene protons (**f**) appeared as a multiplet signal at  $\delta = 1.58$  to  $1.65$ . Multiplet signals at  $\delta = 1.23$  to  $1.35$  were expected for the protons of the methylene backbone (**g**) of the long fatty acid chain, while the terminal methyl protons (**h**) at  $\delta = 0.86$  to  $0.90$  appeared as a distorted triplet for both the andiroba oil and the corresponding biodiesel.

Fig. (3) shows the  $^1\text{H}$  NMR spectra of babassu oil and its biodiesel; again, the spectra were very similar, apart from the absence of signals **a1** (doublet of doublets,  $\delta = 4.30$ ,  $J = 11.8$  Hz) and **a2** (doublet of doublets,  $\delta = 4.15$ ,  $J = 11.8$  Hz) in the biodiesel spectrum, as these peaks corresponded to hydrogen atoms of the triacylglycerol  $-\text{CH}_2$  group.



**Fig. (2).**  $^1\text{H}$  NMR spectra (500 MHz,  $\text{CDCl}_3$ ) of andiroba oil (triacylglycerol) and the biodiesel obtained via CAL-B catalysis.



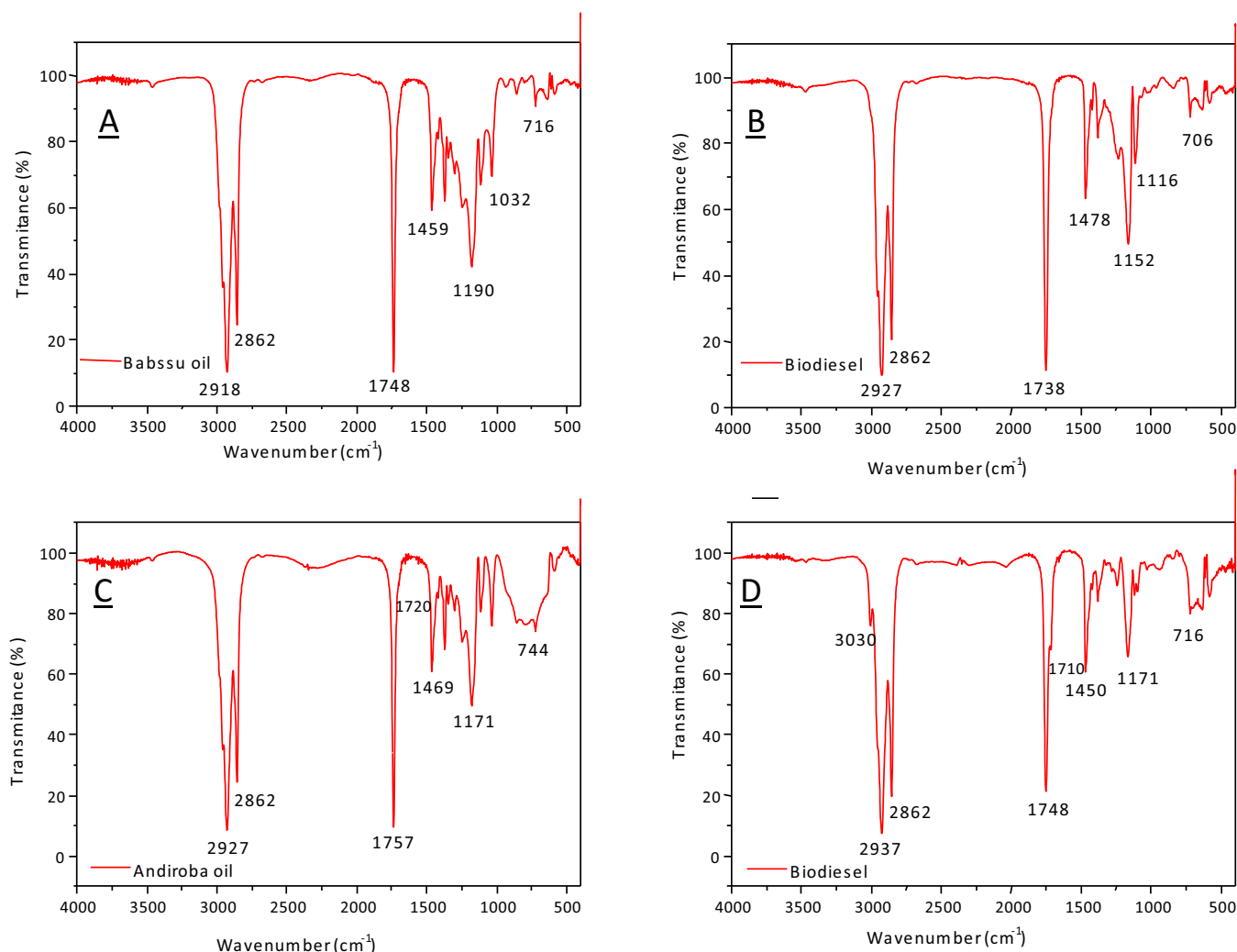
**Fig. (3).**  $^1\text{H}$  NMR spectra (500 MHz,  $\text{CDCl}_3$ ) of babassu oil (triacylglycerol) and biodiesel obtained *via* CAL-B catalysis.

FT-IR spectroscopy was used to identify specific functional groups in each of the samples. For the babassu oil, an intense band at  $1757\text{ cm}^{-1}$  (Fig. 4A) corresponded to  $\text{C}=\text{O}$  stretching and the  $\text{C}=\text{O}$  stretch for the biodiesel was found at  $1748\text{ cm}^{-1}$  (Fig. 4B), confirming the conversion of the oil to biodiesel. Absorptions at  $1720\text{ cm}^{-1}$  for the oil and  $1710\text{ cm}^{-1}$  for the biodiesel corresponded to  $\text{C}=\text{C}$  stretches for each sample. For the biodiesel, these peaks were related to the presence of unsaturated fatty acids, which was confirmed by GC-MS and  $^1\text{H}$  NMR analyses.

Similar peaks were observed by FT-IR characterization of the andiroba oil (Fig. 4C). The two intense bands at  $2918\text{ cm}^{-1}$  and  $2862\text{ cm}^{-1}$  were asymmetric and symmetric  $-\text{CH}_2$  stretching vibrations, respectively, and were also observed for the biodiesel at  $2927\text{ cm}^{-1}$  and  $2862\text{ cm}^{-1}$  (Fig. 4B). The band at  $1748\text{ cm}^{-1}$  (Fig. 4C) was assigned to the  $\text{C}=\text{O}$  stretching vibration of the carboxylic groups of the andiroba oil, and the new peak at  $1738\text{ cm}^{-1}$  for the biodiesel (Fig.

4D) confirmed the conversion. This frequency corresponded to that of ethyl esters and triglycerides. The bands at  $1459\text{ cm}^{-1}$  (Fig. 4C) and  $1478\text{ cm}^{-1}$  (Fig. 4D) were assigned to  $-\text{CH}_2$  scissor deformation vibrations. The region in the spectra at  $1459$  to  $1232\text{ cm}^{-1}$  (Fig. 4C) and  $1478$  to  $1116\text{ cm}^{-1}$  (Fig. 4D) indicated the presence of  $\text{C}=\text{C}-\text{C}-\text{O}$  vibrations.

Another technique often used to confirm the conversion of oils into biodiesel was thermogravimetric analysis. The thermal properties of the oil samples were measured as a function of various reaction parameters, such as temperature, time, and heating rates [25]. Two thermal events were observed in the andiroba oil sample (Fig. 5A). The first event occurred at  $226^\circ\text{C}$ , with a mass loss of 13% followed by a mass loss of 86% at  $400^\circ\text{C}$ . The andiroba biodiesel was thermally stable up to  $225^\circ\text{C}$  and exhibited only one degradation step corresponding to a mass loss of 92%, which was attributed to a single substance or a mixture of compounds with small differences in the molecular weight (Fig. 5B).



**Fig. (4).** FT-IR spectra (A) of andiroba oil; (B) biodiesel from andiroba; (C) babassu oil; (D) biodiesel from babassu.

The andiroba oil also exhibited one step decomposition at 373°C with mass loss of 99% (Fig. 5C); the babassu biodiesel showed decomposition at 196 °C with a mass loss of 99% (Fig. 5D). This may be because the composition of fatty acids in babassu biodiesel was a predominantly short chain, which reduces the melting point of the FAEE (Table 1). Due to the possibility of increasing use of biodiesel, it is important to quantify the parameters of biodiesel pyrolysis.

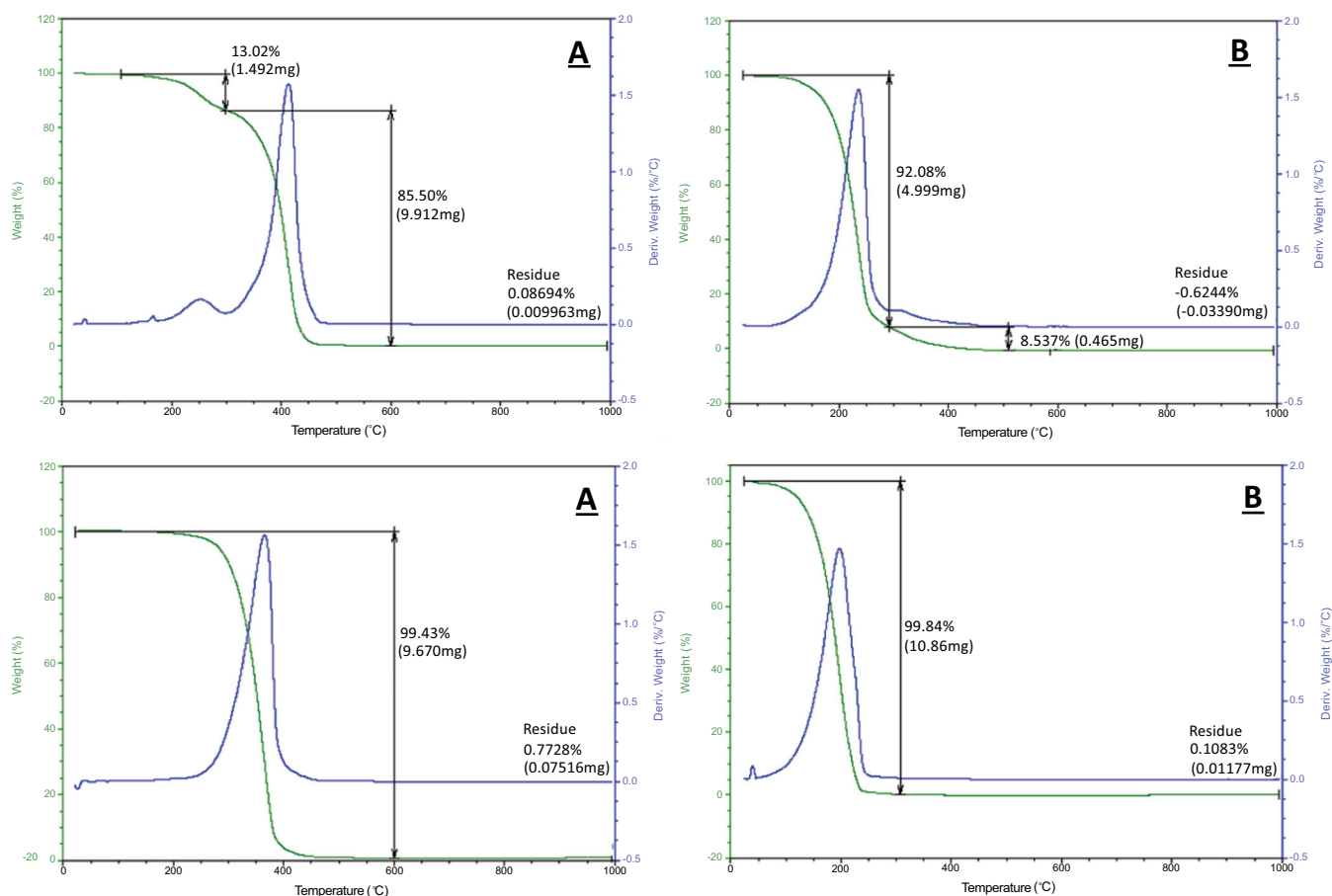
Fig. (6) shows fluorescence spectra of babassu and andiroba oil excited at 400 nm and emission collected between 400 and 800 nm. Both samples presented a maximum of emission about 475 nm, however, andiroba sample showed a small emission band around 670 nm.

In order to evaluate if the samples presented different spectral regions, the excitation wavelength was varied, monitoring the emission in different regions, obtaining a contour map. Figs. (6) shows the excitation/emission contour maps of the vegetable oils (andiroba and babassu). All samples exhibited fluorescence between 350 and 700 nm when excited between 325-450 nm. Raman and Rayleigh scattering was not omitted from the graphs. The results indicated that andiroba oil sample showed an emission maximum

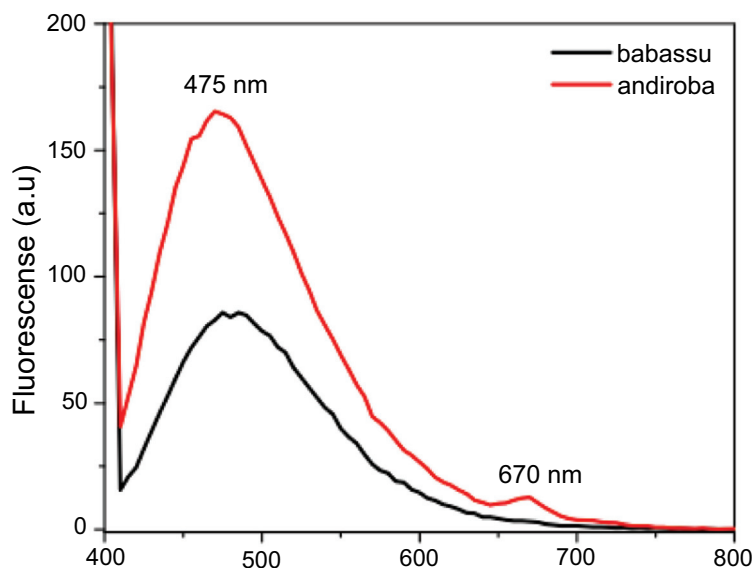
around 475 nm, with maximum excitation around 400 nm (Fig. 7). However, babassu sample showed a maximum of around 500 nm, with maximum excitation at 420 nm, indicating that measurements using only one single excitation wavelength cannot differentiate the evaluated oils (Fig. 7).

Magalhães and co-workers attributed the fluorescence of soybean oil and biodiesel in the region between 400 and 550 nm to conjugated tetraene molecules, which are endogenous products of these materials [36]. Both samples showed characteristic emission of conjugated tetraenes. Fluorescence in oils around 350 nm has been attributed to compounds derived from tocopherols, naturally present in vegetable oils [37, 38]. In addition, the andiroba oil sample when excited at 400 nm exhibits emission region between 600 and 700 nm due to fluorescence characteristics of porphyrins, such as chlorophyll  $\alpha$  and  $\beta$ , which may be present in the oils [37].

However, the babassu oil sample did not show the characteristic emission of porphyrins. The contour maps also indicated a difference in the fluorescence intensity for the samples, due tetraenes conjugated composition or products degradation of oleic, linoleic and linolenic acid. Table 2 indicated that the sample of babassu oil has a high concentration



**Fig. (5).** TGA data for (A) andiroba oil; (B) biodiesel from andiroba; (C) babassu oil and (D) biodiesel from babassu.

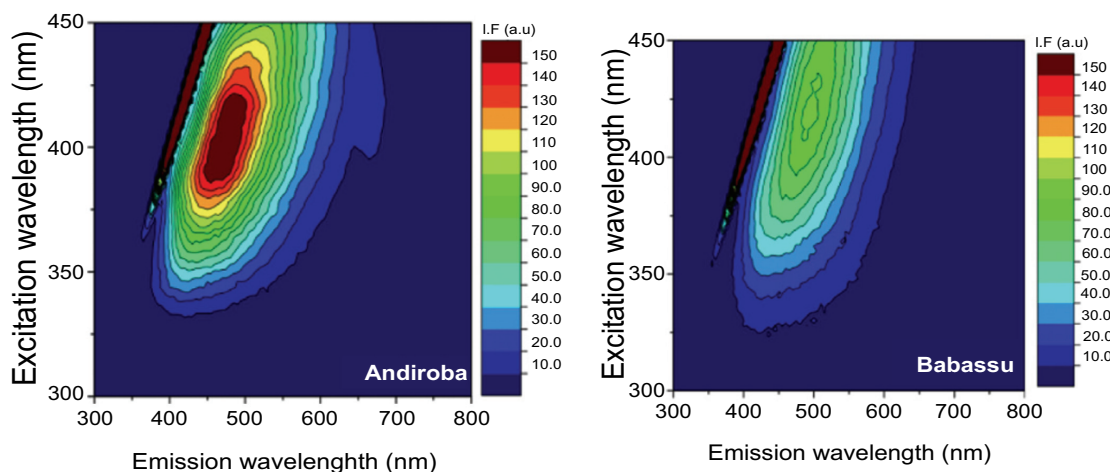


**Fig. (6).** Emission spectra of babassu and andiroba oil under excitation at 400 nm.

of saturated fatty acids, such as lauric acid. However, andiroba oil sample showed a high amount of unsaturated fatty acids such as linoleic and oleic acid, which can be precursors of chemical reactions that promote the formation of conjugated dienes, trienes and tetraenes [39] which favors the presence of fluorophores in the sample. In addition, the results of acidity index and peroxide index (Table 1)

indicated a greater presence of fatty acids in the andiroba oil, which may be due to the form of storage, extraction, collection period, or obtaining of this oil, thus favoring degradation processes of esters and formation of fluorescent by-products such as conjugated tetraenes. Intrinsic factors for each sample such as viscosity, fluorophores density (concentration), sample color and refractive index may





**Fig. (7).** Fluorescence contour map of an undiluted sample of andiroba oil and babassu oil.

promote changes in the excitation/ emission spectra [39, 40]. Fluorescence results indicated that the babassu and andiroba oil samples exhibited distinct fluorescence profiles due to the varied composition of unsaturated and conjugated methyl esters.

#### 4. CONCLUSION

In summary, synthesis of ethylic biodiesel by catalysis enzymatic (CALB), showed to be a successful route in the ethanolysis of babassu and andiroba oils in high yields. The corresponding biodiesels were characterized by  $^1\text{H}$  NMR spectra, GC-MS, FT-IR, and TGA analyses. Fluorescence measurements indicated that the babassu and andiroba oil samples showed emission corresponding to the presence of conjugated tetraenes, which may be due to the degradation of the oil or from the seed. In addition, andiroba oil exhibited characteristic fluorescence of compounds possibly derived from chlorophyll. Results described here provide relevant information for the further research uses andiroba and babassu oils the alternatives to other oils in the synthesis of biodiesel, considering the properties physics-chemical and cost of raw material. This, in turn, implies that maintaining the native forests of the Brazilian Amazon is clearly important in responding to demands in biodiesel production.

#### ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

#### HUMAN AND ANIMAL RIGHTS

No Animals/Humans were used for studies that are the basis of this research.

#### CONSENT FOR PUBLICATION

Not applicable.

#### CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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