

UNIVERSIDAD SAN FRANCISCO DE QUITO USFQ

Colegio de Ciencias e Ingenierías

**Production and analysis of Biodiesel from the fruits of
*Roystonea oleracea***

Artículo académico

Nicolás Andrés Cabrera Moreta

Ingeniería Química

Trabajo de titulación presentado como requisito
para la obtención del título de
Ingeniero Químico

Quito, 26 de julio de 2016

UNIVERSIDAD SAN FRANCISCO DE QUITO USFQ
COLEGIO POLITÉCNICO

HOJA DE CALIFICACIÓN
DE TRABAJO DE TITULACIÓN

Production and analysis of Biodiesel from the fruits of *Roystonea Oleracea*

Nicolás Andrés Cabrera Moreta

Calificación:

Nombre del profesor, Título académico

Jose Ramón Mora, Ph.D.

Firma del profesor

Nombre del profesor, Título académico

Valeria Ochoa-Herrera, Ph.D.

Firma del profesor

Quito, 26 de julio de 2016

Derechos de Autor

Por medio del presente documento certifico que he leído todas las Políticas y Manuales de la Universidad San Francisco de Quito USFQ, incluyendo la Política de Propiedad Intelectual USFQ, y estoy de acuerdo con su contenido, por lo que los derechos de propiedad intelectual del presente trabajo quedan sujetos a lo dispuesto en esas Políticas.

Asimismo, autorizo a la USFQ para que realice la digitalización y publicación de este trabajo en el repositorio virtual, de conformidad a lo dispuesto en el Art. 144 de la Ley Orgánica de Educación Superior.

Firma del estudiante: _____

Nombres y apellidos: Nicolás Andrés Cabrera Moreta

Código: 00107445

Cédula de Identidad: 1714864582

Lugar y fecha: Quito, 26 de julio de 2016

Production and analysis of Biodiesel from the fruits of *Roystonea oleracea*

Nicolás A. Cabrera

¹ Universidad San Francisco de Quito USFQ, Colegio de Ciencias e Ingeniería - El Politécnico. Calle Diego de Robles y Vía Interoceánica, Campus Cumbayá, Edif. Newton. Quito, Ecuador
Correspondence should be addressed to Nicolás Cabrera; nicolas.cabrera@estud.usfq.edu.ec

Producción y análisis de Biodiesel a partir de los frutos de *Roystonea Oleracea*

Abstract

Biodiesel was produced from neutral lipids from *Roystonea Oleracea* fruits, which were extracted by three different methods. Biodiesel produced from commercial soybean oil was employed as a reference, which have been analyzed and reported in the literature. Fatty acid methylation was carried out by the transesterification reaction with methanol and potassium hydroxide. Several methods have been developed to study fatty acids (FA) like Thin-Layer Chromatography (TLC), Gas chromatography coupled to Mass Spectrometry (GC/MS) and Flame Ionization Detector (GC/FID). This research presents a qualitative and quantitative analysis of Fatty Acid Methyl Esters (FAME) of *Roystonea Oleracea* fruit biodiesel (ROFB). TLC analysis was carried on plates of silica plates and compared retention factors (Rf) with soybean biodiesel. FAMES identification was done on using a GC/MS with a low-polarity SH-Rxi™-5Sil MS column. ROFB was composed of methyl caprate (C_{10:0}), methyl myristate (C_{14:0}), methyl palmitate (C_{16:0}), methyl linoleate (C_{18:2}), methyl oleate (C_{18:1}) and methyl stearate (C_{18:0}). FAME percentage composition in ROFB was done with GC/FID using a TR-WAX-20M column and the standard internal method. A calibration curve with a relation between m_{Ei}/m_{EI} vs. A_{Ei}/A_{EI} demonstrated that relative response factor (RRF) is 1:1 for components with 14 to 20 carbons chain. The mass of the analyte components in the sample was calculated. In conclusion, the direct transesterification method (DTM) is a good way to produce ROFB with a yield of 1, 7.54 and 6.61% with 0.05, 0.1 and 0.2 grams of KOH, respectively.

Keywords. Transesterification, Biodiesel, Biomass, *Roystonea Oleracea*, Fatty Acid Methyl Esters.

Resumen

Biodiesel fue producido a partir de los lípidos neutros del fruto de la *Roystonea Oleracea*, los cuales fueron extraídos por tres métodos distintos. El biodiesel generado a partir de aceite de soya fue utilizando como referencia el cual ha sido analizado y reportado en la literatura. La metilación de los ácidos grasos se llevó a cabo por transesterificación con metanol e hidróxido de potasio. Algunos métodos de análisis se han desarrollado para estudiar los ácidos grasos (AG) como Cromatografía de Capa Fina (CCF), cromatografía de gases acoplada a Espectrometría de Masas (GC/MS) y Detector de Ionización de Llama (GC/FID). En este trabajo, se presenta un análisis cualitativo y cuantitativo de los Ésteres Metílicos de Ácidos Grasos (EMAG) en el Biodiesel producido a partir del fruto de la *Roystonea Oleracea* (BFRO). El análisis por CCF se lo realizó en placas de sílice y comparados con los factores de retención (RF) del biodiesel de soya. EMAG identificación se realizó en GC-MS con una columna de baja polaridad SH-Rxi™-5Sil MS. El BFRO está compuesto por caprato de metilo (C_{10:0}), el miristato de metilo (C_{14:0}), palmitato de metilo (C_{16:0}), linoleato de metilo (C_{18:2}), oleato de metilo (C_{18:1}) y estearato de metilo (C_{18:0}). La composición porcentual de EMAG en RFBO se llevó a cabo con GC/FID utilizando una columna TR-WAX-20M y el método de estándar interno. Una curva de calibración con una relación entre m_{Ei} / m_{EI} vs. A_{Ei} / A_{EI} demostró que el factor de respuesta relativa (FRR) es igual a 1 para los componentes cadenas de 14 a 20 átomos de carbono. La masa de los componentes de analito en la muestra fue calculada. En conclusión, el método de transesterificación directa (MTD) es una buena manera de producir RFBO. Los rendimientos fueron 1, 7.54 y 6.61% con 0,05, 0,1 y 0,2 gramos de KOH, respectivamente.

Palabras clave. Transesterificación, Biodiesel, biomasa, *Roystonea Oleracea*, Ésteres metílicos de ácidos grasos .

1. INTRODUCTION

Chemical process industries have been controlled by the production and use of petroleum. Additionally, oil reserves are running out fast. As a result, renewable energy is requested. The use of renewable energy reduces carbon CO₂, SO_x, and HC emissions and helps to protect the environment [1, 2]. Worldwide consumption of renewable energy has been increasing in the last years. In fact, in 2011 16.7% of the total global energy consumption was from renewable sources. Alternative sources of energy include sun, wind, water and biomass [3, 4]. Bioenergy produced by biomass has been proposed as one of the solutions to replace fossil fuels and contribute to produce clean energy. The uses of promising sources to produce biodiesel such as algae, non-edible sources are expected to increase by 2020. Biofuel produced from biomass is carbon neutral, the carbon absorbed by biomass, is offset by CO₂ produced when it is combusted. The production of biofuels do not generate hazardous compounds, in fact toxics chemicals and toxics solvents are avoided on bioconversion processes. In the short term, biofuels could be the unique renewable energy that can be used in the transportation industry without replacing the vehicles [5]. Biodiesel (BD) is composed of methyl esters or ethyl esters. They are derived from vegetable or animal oils [6, 7]. Transesterification (TE) with alcohol in a catalytic medium of vegetable and animal oils is an effective process to produce biodiesel. Biodiesel is typically composed of methyl ester with carbon chains from C_{8:0} to C_{22:1}. Each component influence on fuel properties such us viscosity, lubricity, oxidative, stability, etc [8, 9, 10, 11]. Transesterification has three reversible reactions (i) conversion of triglyceride to diglyceride (ii) diglyceride to monoglyceride

(iii) monoglyceride to glycerol. In this process, three moles of alcohol are needed to get one mole of monoalkyl ester (product) and one mole of glycerol (byproduct). TE is affected by the molar ratio of alcohol to oil, the type of alcohol used, the temperature of the reaction, the stirring rate, the type and amount of catalyst [12-15]. To remove lipids from biomass is recommended to use non-polar organic liquids, such aliphatic hydrocarbons petroleum ethers, or n-hexane as solvents [16].

First generation biofuels are produced by edible food like corn, sugar cane, sorghum, etc. and vegetable oils and animal fats. Second generation biofuels are produced by non-food feedstock's like jatropha and algae[17]. Third generation biofuels are produced from no eatable vegetable oil which is the case of biodiesel from *Roystonea Oleracea* (RO). RO has been cultivated with decorative purposes. This palm is considered invasive in the swamps of Guiana[18], panama[19], in the Atlantic Forest of Brazil[20] and Isla Santay in Guayaquil[21]. Leaves of RO and reproductive structures decompose slowly, and develop a sandy layer on the ground. Due to the height of RO, heavy leaves modify habitat conditions and the recruitment of native species [22, 23]. When palm leaves are discarded into the environmental, there is a potential raw material for the production of biofuels [24]. The main objective of this work is to evaluate the use of RO fruits to produce biodiesel and control this invasive palm.

2. METHODS

This investigation was done in Environmental Laboratory (LIA-USFQ) and Chemical Engineering Laboratories at Universidad San Francisco de Quito. The experiment consists of three sections: (i) extraction of lipids, (ii) transesterification of

lipids to produce biodiesel and (iii) characterization of biodiesel.

2.1 Samples

All samples of fruits were collected from Isla Santay in Guayaquil City. A total of two kilograms were harvested from four individual palms trees. The fruits were washed, crushed and sieved on a mesh #20. 150 grams of the samples were packing on a hermetic bags and keeping at 15°C. Before analysis, the fruits were dried in an oven at 100°C for 1 h.

2.2 Materials

n-Hexane (C₆H₆), potassium hydroxide (KOH), hydrochloric acid (HCl), sodium chloride (Na Cl), potassium hydroxide (KOH), petroleum ether, diethylether ((C₂H₅)₂O) analytical grade were purchased from reactivos H.V.O. Acetic acid (CH₃COOH) (100% purity) was obtained from Merk. Methanol (CH₃OH) was purchased from HR Representaciones., Methyl palmitate (C_{16:0}) (99% purity) was obtained from MP Biomedicals. , Methyl linoleate (C_{18:2}) (99% purity) was purchased from Acros Organics and methyl dodecanoate (C_{12:0}) (99% purity) was obtained from Alfa Aesar.

2.3 Lipids Extraction

2.3.1 Soxhlet-hexane extraction (SHE)

The neutral lipids of fruits were extracted with Soxhlet, using hexane as solvent [25].

2.3.1.1 Single sample

Lipids were extracted from 15 grams of dried RO fruit using n-hexane, for 3 hours. The solvent was recovered with a rotary

evaporator and lipids were placed at the bottom of Florence flask.

2.3.1.2 Multiple samples

Lipids were extracted from four samples of 15 grams of RO fruit dried using n- hexane and changed each 3 hours. The solvent was added to keep the same volume. At the end, it was recovered with a rotary evaporator and lipids were placed at the bottom of Florence flask.

2.3.2 Hexane extraction

20 grams of dried RO fruit were placed on an Erlenmeyer with 40 mL of hexane for 1 day. Then the solution was mixed for 24 hours with a small magnetic stirring bar and mixing with ultrasound for 1 hour. N-hexane was recovered in a rotary evaporator and lipids were placed at the bottom of Florence flask.

2.4 Transesterification reaction

2.4.1 Soxhlet-hexane lipids (SHL)

5 grams of lipids were precisely weighed with 1.4 mL CH₃OH and 0.05 grams KOH [26]. The solutions were stirring for 3 and 6 hours. The solution was transferred to separatory funnels and washed 3 times with 2 mL NaCl (6M) and 2 times with 2 mL of distillate water [27].

2.4.2 Hexane extraction (HE)

3.1 grams of lipids were precisely weighed with 3 mL CH₃OH and 0.03 grams KOH. The solutions were stirring for 3 h. It was transferred to separatory funnels and washed 3 times with 2 mL NaCl (6M) and 2 times with 2 mL of distillate water.

2.4.3 Direct Transesterification Method (DTM)

5 grams of the sample were precisely weighed with 5mL CH₃OH and 0.05, 0.10 and 0.20 KOH. The solutions were stirring for 3 hours. 10mL n-hexane was added and the solution was transferred to separatory funnels and washed 3 times with 2 mL NaCl (6M); and 2 times with 2 mL of distillate water.

2.5 Characterization of biodiesel

2.5.1 Thin-Layer Chromatography (TLC)

Thin-layer chromatography (TLC) is a simple, inexpensive and commonly used separation method for analyzing samples [28]. The biodiesel was dissolved with hexane and put on the plate. The plate of size 3 cm x 6 cm was used, a line was drawn at 0.5 cm from the bottom using a graphite pencil. Silica was used as a stationary phase and a solution of petroleum ether, diethyl ether and acetic acid (80:19:1) as a mobile phase [26]. The beaker size and the volume of the mobile phase ensure that the liquid level is below the line. TLC plate was staining with iodine vapor and refraction factors (RF) from the sample biodiesel were compared with biodiesel from soybean oil.

2.5.2 Chromatography Analysis

Fatty acid methyl esters (FAME) were quantified in a Shimadzu GCMS-QP2010 Ultra with autoinjector AOC-20i for liquid samples, mass spectrometry detector (GC-MS) and flame ionization (GC-FID).

2.5.2.1 Mass Spectrometry (GC-MS)

The analysis was conducted in a GC-MS (Shimadzu Technologies, Japan) with a low

–polarity phase (CrossbondTM silarylene phase 1,4-bis (dimethylsiloxy) phenylene dimethyl polysiloxane SH-RxiTM-5Sil MS) column (30 m length, 0.25 mm ID, 0.25 um film thickness). The software used was GCMS solution Version 4.11 SU2 equipped with commercial mass spectral libraries (NIST 11). The injection volume was 0.2 µL and a program was used in the split mode 1:10. Injection temperature was 250 °C. The column oven program was configured as follows: the initial temperature was 150 °C; ramp to 250 °C at a rate of 25 °C/min; ramp to 253 °C at a rate of 1°C/min; ramp to 275 °C at 25°C/min, held isothermally at 275 °C for 2 min. The pressure of helium carrier gas was 99.5 kPa and the velocity was 13 mL/min.

2.5.2.1 Flame Ionization Detector (GC-FID)

This analysis was carried on a GC-FID (Shimadzu Technologies, Japan). The injection volume is 0.2 µL into TR-WAX column (30 m length, 0.25 mm ID, 0.25 um film thickness). The injection port and detector temperature were kept at 260 and 280°C, respectively. The column oven program was as follows: the initial temperature was 190°C; ramp to 200°C at 4 °C/min; ramp to 225°C at 2°C/min; ramp to 260° at 15°C/min, held isothermally at 260°C for 2 min. The pressure of the Helium was 142.6 kPa and the velocity was 3mL/min [29]. Dodecanoic acid (E1) was used based on the internal standard method [30]. Palmitic (E1) and linoleate (E2) were used as standards to calculate to calculate the yield.

3. RESULTS AND DISCUSSION

Some experiments were carried with three different ways to extract lipids with the Soxhlet-hexane extraction (SHE) with

simple and multiple samples and three different amounts of NaOH (0.05, 0.1, 0.2g) and methanol was maintained constant in the DTM. The mean mass percentage of neutral lipids on *Roystonea Oleracea* fruits was 12.48 ± 1.02 %. Qualitative and quantitative analysis were carried out to characterize biodiesel produced from RO.

3.1 TLC analysis

TLC was applied for the first qualitative analysis of the formation and composition of FAME in the samples. As a reference, we used Retention Factor (Rf) of soybean oil. FAME's retention factor are presented in Table 1.

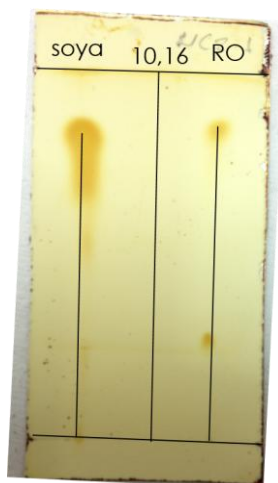


Figure 1 TLC analysis on the right side soybean oil biodiesel on the right SHL RO biodiesel

Table 1 Rf values of soybean and RO biodiesel

Substance	Rf
Soybean oil biodiesel ^[28]	0.82
Experimental	
Soybean oil biodiesel	0.82
DTM RO biodiesel	0.84
SHL RO biodiesel	0.80

3.2 GC/MS analysis

GC/MS analysis was used to identify the methyl esters that were present in the RO

biodiesel. The retention time, molecular mass, name and chain length of the principal components reported on GC/MS are shown in Table 2 and 3. Unfortunately, C_{18:n} components were not separated because is necessary a high polar column [31].

3.2.1 Soybean Oil

4 different samples of soybean oil were analyzed with GC/MS. Five peaks were observed in the analysis (Fig. 2) and reported on Table 2. The mass spectrum of each peak is shown in Figure 4. GC/MS analysis confirmed the presence of methyl palmitate (C_{16:0}), methyl linoleate (C_{18:2}), methyl oleate (C_{18:1}) and methyl stearate (C_{18:0}). Those components were in accordance with the reported composition of soybean oil. The mean mass percentage of neutral lipids on soybean is 19.4% [32, 33]. For saturated methyl esters, the common fragment ions at m/z showed that the strongest peak was at 74(-(CH₂)₄CH₃, pentyl), followed by 87(-(CH₂)₅CH₃, hexyl) 43(-(CH₂)₂CH₃, propyl), 143(-(CH₂)₉CH₃). For unsaturated compounds, the common peaks are at 41, 55, 67, 81, and 123. The strongest peak is methyl linoleate C_{18:2} at 67(M-(CH₂)₁₅CH₃) and methyl oleate C_{18:1} at 55 (M-(CH₂)₁₅CH₃).

Table 2 Retention time of compounds reported on soybean oil biodiesel samples analyzed by GC/MS

N°	Name	Chain Length	Retention Time	m/z
1	Methyl laurate	C _{12:0}	2.870	214
2	Methyl palmitate	C _{16:0}	4.426	270
3	Methyl linoleate	C _{18:2}	5.240	294
4	Methyl oleate	C _{18:1}	5.272	296
5	Methyl stearate	C _{18:0}	5.388	298

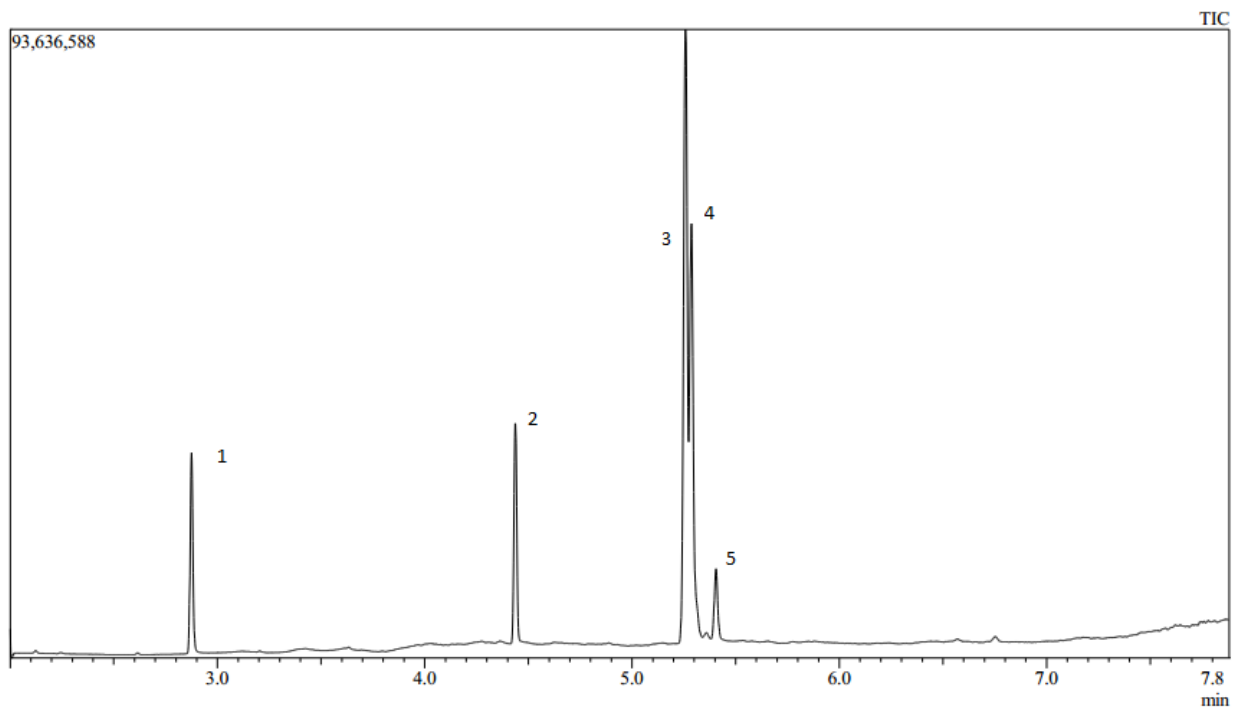


Figure 3 Soybean oil biodiesel GC/MS chromatogram

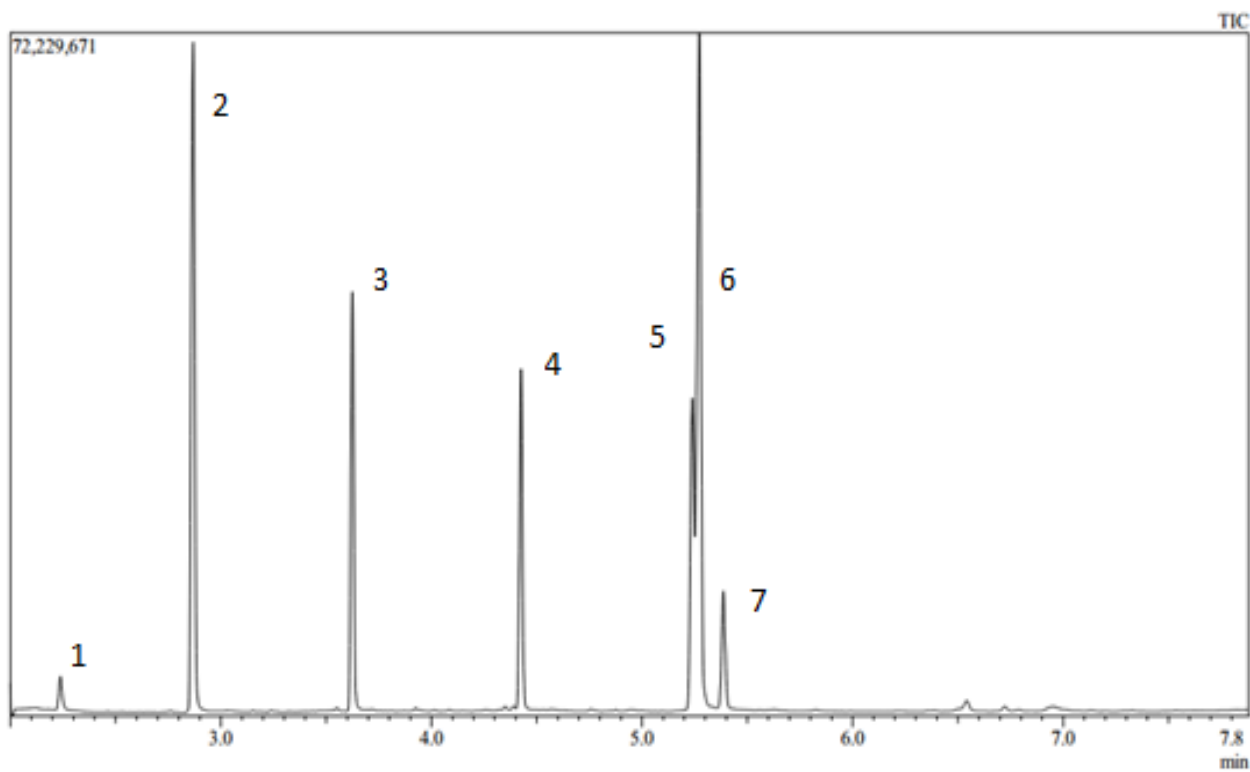


Figure 2 ROFB GC/MS chromatogram

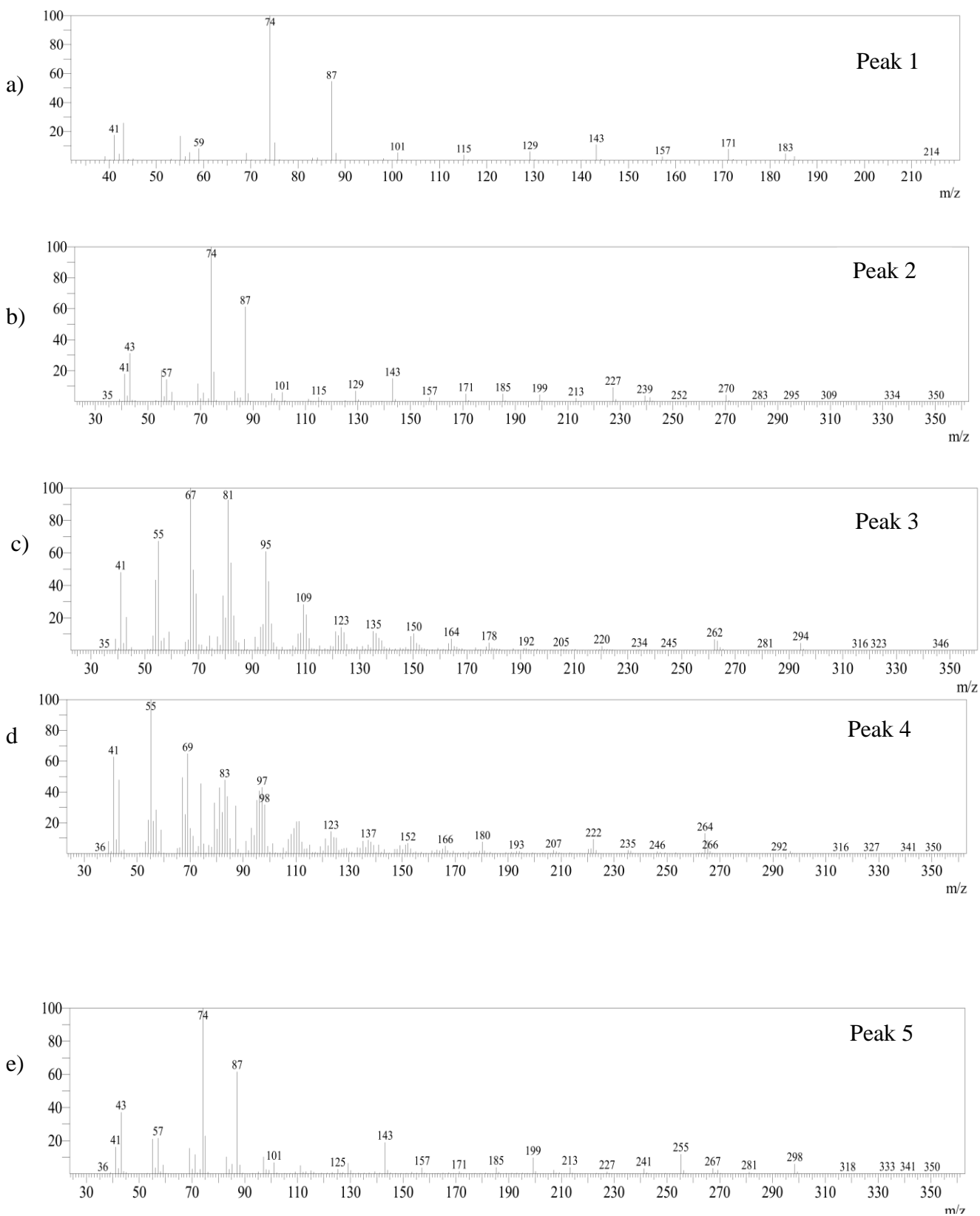


Figure 4 Spectrum of soybean oil biodiesel i) Methyl laurate ii) Methyl palmitate, iii) Methyl linoleate iv) Methyl oleate v) Methyl stearate

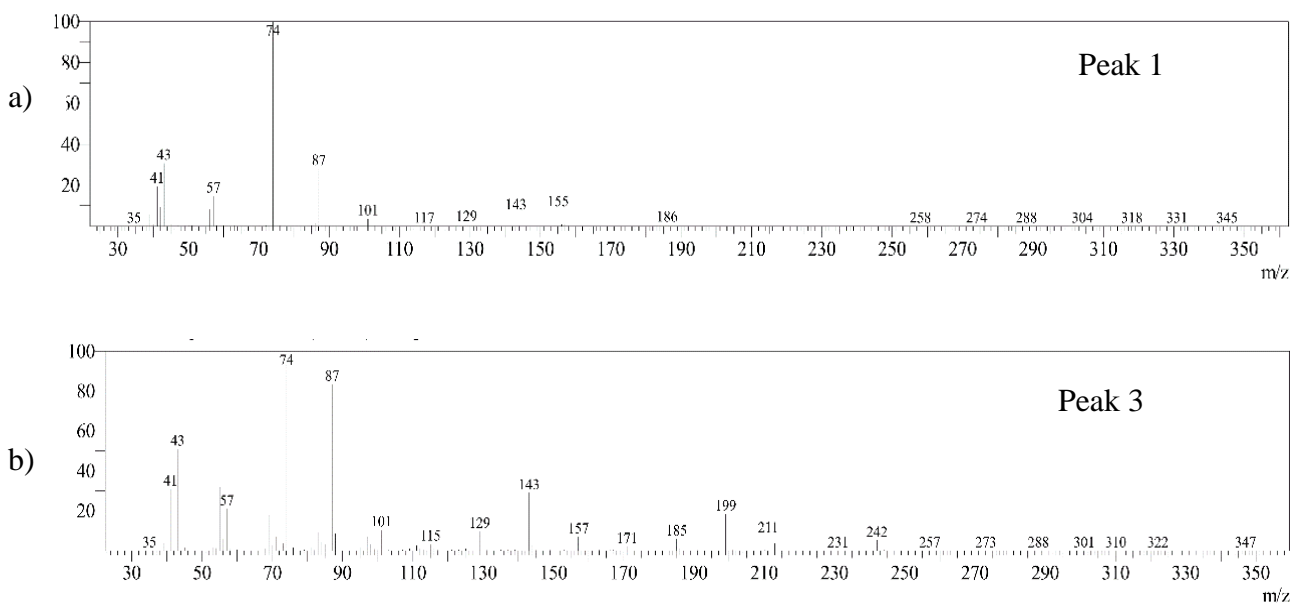


Figure 5 Spectrum of ROFB i) Methyl caprate ii) Methyl myristate

expensive and less time consuming method [31].

3.2.2 *Roystonea Oleracea* fruit biodiesel

4 different samples of RO biodiesel produced by the direct transesterification method were analyzed with GC/MS. Seven peaks were observed in the chromatograms (Fig. 3) and they are reported in Table 3. GC/MS analysis confirmed the presences of methyl caprate (C_{10:0}), methyl myristate (C_{14:0}), methyl palmitate (C_{16:0}), methyl linoleate (C_{18:2}), methyl oleate (C_{18:1}) and methyl stearate (C_{18:0}). m/z ratios for saturated compounds are similar to soybean oil biodiesel at 74, followed by 87, 43 and 143.

3.3 GC/FID analysis

To quantify FAME in the samples and yield for different transesterification reactions. For this purpose, TR-WAX column was used to analyze FAME in biodiesel components with a chain length between C₁₄ and C₂₄ can be separated using a less

Table 3 Retention time of compounds reported on ROFB samples analyzed by GC/MS

N°	Name	Chain Length	Retention Time	m/z
1	Methyl caprate	C _{10:0}	2.239	186
2	Methyl laurate	C _{12:0}	2.870	214
3	Methyl myristate	C _{14:0}	3.625	242
4	Methyl palmitate	C _{16:0}	4.426	270
5	Methyl linoleate	C _{18:2}	5.240	294
6	Methyl oleate	C _{18:1}	5.272	296
7	Methyl stearate	C _{18:0}	5.388	298

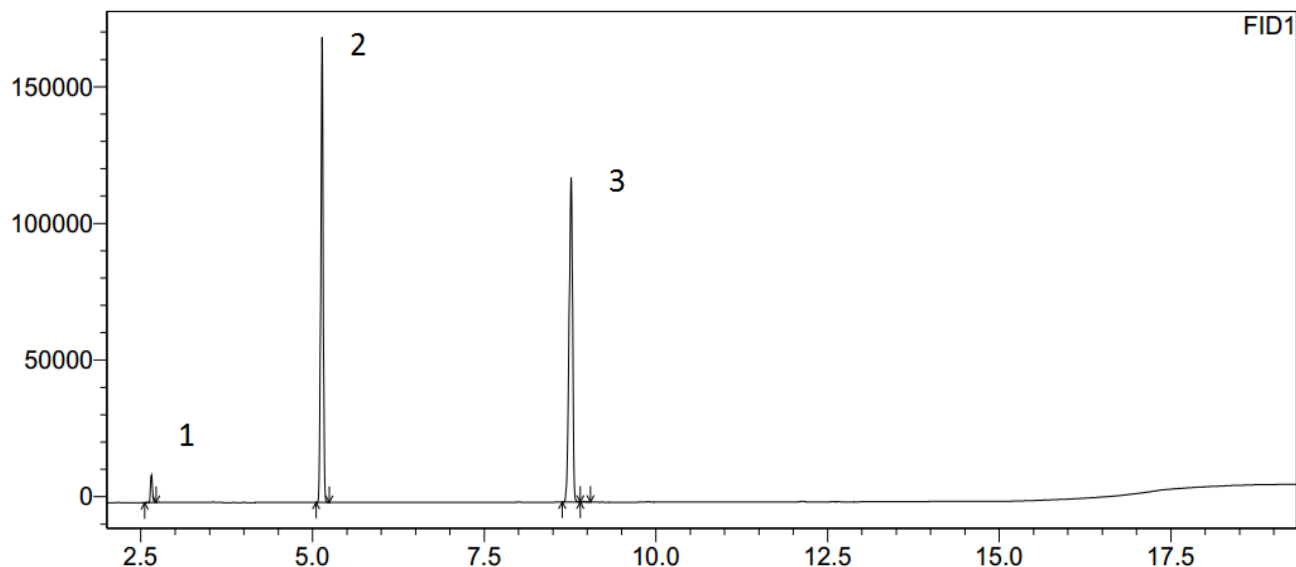


Figure 6 Standard solution GC/FID

3.3.1 Internal standard selection and calibration curve

Methyl palmitate and methyl linoleate standards were employed to quantify the yield of the transesterification process based on GC/MS analysis. The dodecanoic methyl ester was used as an internal standard because it has similar physicochemical properties and it is not present in the sample. Seven points calibration curves ranging from 100 to 500 microliters from internal standard and from 400 to 2000 μL from palmitate (E1) (Fig. 7) and linoleate (E2) (Fig. 8) with a concentration of 10 mg/mL. Standard solutions were analyzed on GC-FID. Then the calibration curve showed a linear relationship between m_{Ei}/m_{EI} vs. A_{Ei}/A_{EI} then with a slope close to 1. The curves were obtained with a correlation coefficient greater than 0.99 ($R^2 > 0.992$). When the same mass of the standards $C_{16:0}$ and $C_{18:2}$ were injected their areas were approximately equal (Fig. 9, Table 4). The relation $m_{C_{16:0}}/m_{C_{18:2}}$ and $A_{C_{16:0}}/A_{C_{18:2}}$ are close to 1. Then the response factor between methyl palmitate and methyl linoleate is 1:1.

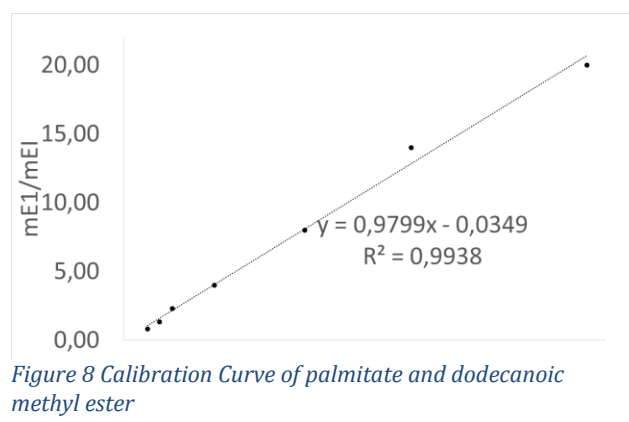


Figure 8 Calibration Curve of palmitate and dodecanoic methyl ester

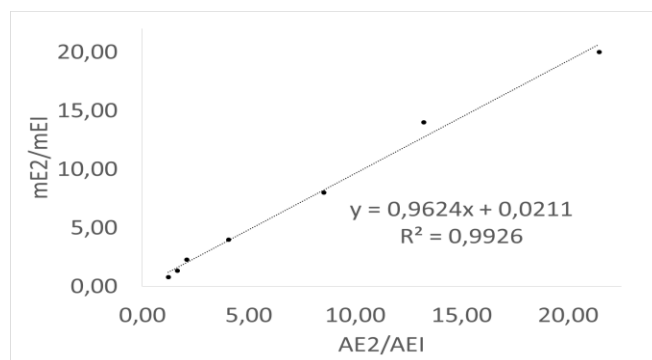


Figure 7 Calibration Curve of linoleate and dodecanoic methyl ester

Table 4 Areas of compounds reported on Standard solution analyzed by GC/FID

N	Name	Chain Length	Area	Mass injected
1	Methyl laurate	C _{12:0}	19636	4.5 mg
2	Methyl Palmitate	C _{16:0}	414969	6 mg
3	Methyl Linoleate	C _{18:2}	421413	6 mg

The two following equations are proposed to calculate the mass composition of (C_{16:0}), methyl linoleate (C_{18:2}) based on C_{12:0} mass:

$$m_{C_{16}} = \left(\frac{A_{C_{16}}}{A_{EI}} 0.9799 - 0.0349 \right) m_{EI}$$

$$m_{C_{18:2}} = \left(\frac{A_{C_{18:2}}}{A_{EI}} 0.9624 - 0.0211 \right) m_{EI}$$

Where:

m_{EI} , is the mass of internal standard.

$m_{C_{16}}$, is the mass of palmitate standard.

$m_{C_{18:2}}$, is the mass of linoleate standard.

A_{EI} , is the area of internal standard.

$A_{C_{16}}$, is the area of palmitate standard.

$A_{C_{18:2}}$, is the area of linoleate standard.

Table 5 Retention time of compounds reported on soybean oil biodiesel analyzed by GC/FID

N°	Name	Chain Length	Retention Time
1	Methyl laurate	C _{12:0}	2.664
2	Methyl palmitate	C _{16:0}	5.156
3	Methyl stearate	C _{18:0}	7.729
4	Methyl oleate	C _{18:1}	8.039
5	Methyl linoleate	C _{18:2}	8.784
6	Methyl linolenate	C _{18:3}	9.916

To demonstrate that the response factor to other components is 1:1 for biodiesel composition from soybean oil reported [32] was compared with soybean oil obtained in the laboratory (Table 6).

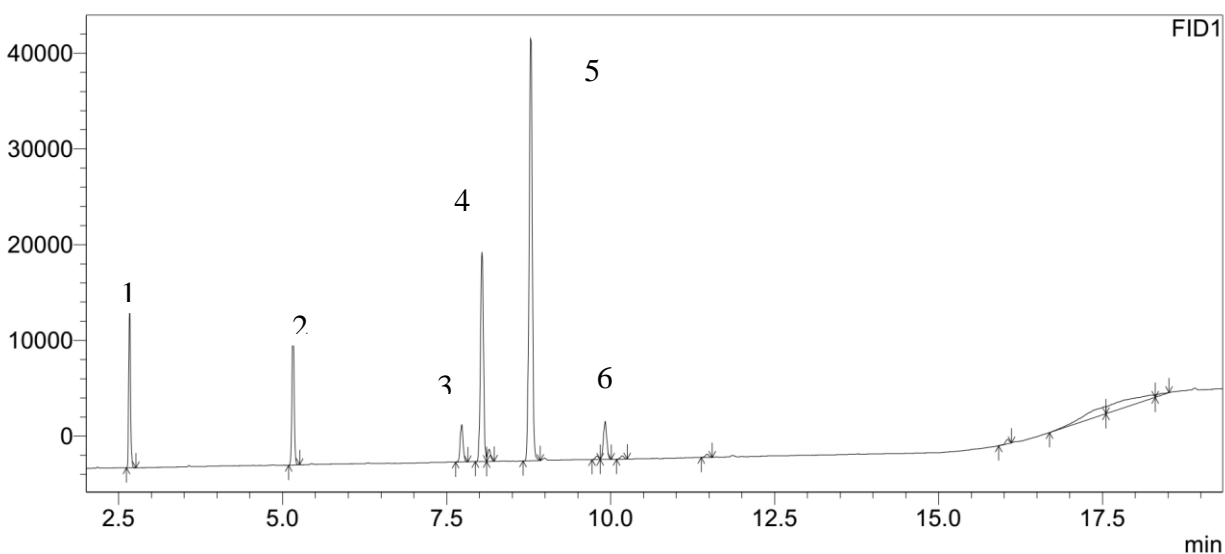


Figure 9 Soybean oil biodiesel analysis on GC/FID

Table 6 Soy oil ester composition

	Soybean ester composition ^[32]		Soybean ester Composition	
	Mean	Desv	Mean	Desv
Palmitate	11,60	2,00	12.71	0.18
Oleate	23,70	2,40	25.49	1.65
Linoleate	53,80	3,50	57.36	1.58
Linolenate	5,90	2,60	6.00	0.90
Sterate	3,90	0,80	4.44	0.11

The linear regression between experimental and reported SB has a slope of 0.9978 and $R^2 = 0.9961$ (Fig 10). As a result, the slope and the correlation coefficient are close to 1. This demonstrates that the response factor is equal to 1:1 for esters with 14 to 20 carbons chain [30].

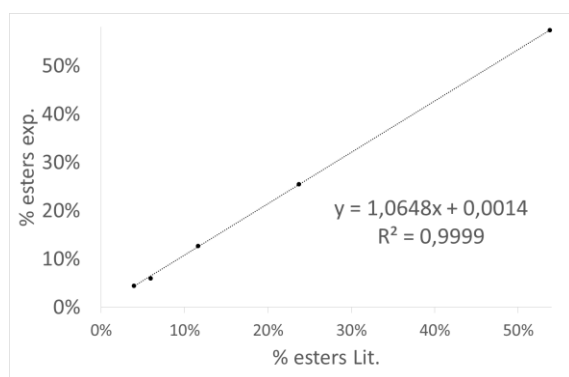


Figure 10 Correlation between % esters reported and % esters on laboratory

Soy ester composition was compared between the correlation and bibliographic composition. As response factor is equal to 1 mass of components from 14 to 20 carbons chain, three equations are proposed to calculate the unknown mass esters based on Palmitate and Linoleate mass.

$$m_{C14:0} = \left(\frac{A_{C14:0}}{A_{C16:0}} \right) m_{C16:0}$$

$$m_{C18:0} = \left(\frac{A_{C18:0}}{A_{C18:2}} \right) m_{C18:2}$$

$$m_{C18:1} = \left(\frac{A_{C18:1}}{A_{C18:2}} \right) m_{C18:2}$$

$$m_{C18:3} = \left(\frac{A_{C18:3}}{A_{C18:2}} \right) m_{C18:2}$$

Where:

m_{C14} , is the mass of methyl myristate.

m_{C16} , is the mass of methyl palmitate.

$m_{C18:0}$, is the mass of methyl sterate.

$m_{C18:1}$, is the mass of methyl oleate.

$m_{C18:2}$, is the mass of linoleate standard.

$m_{C18:3}$, is the mass of linolenate standard.

A_{C14} , is the area of methyl myristate.

A_{C16} , is the area of methyl palmitate.

$A_{C18:0}$, is the area of methyl sterate.

$A_{C18:1}$, is the area of methyl oleate.

$A_{C18:2}$, is the area of linoleate standard.

$A_{C18:3}$, is the area of linolenate standard.

3.3.2 Biodiesel Analysis

3.3.2.1 Soybean biodiesel yield

Using equations showed on Section 3.3.1 the yield of DTM from 5 grams of soybean biomass from different methods are showed in Table (7).

Table 7 Yield of SB with different methods

Method	Time	g KOH	Yield
DTM 1	3h	0.05	3.27%
DTM 2	3h	0.1	9.46%
DTM 3	3h	0.2	9.27%

The most accurate way to produce biodiesel is DTM 2 from soybean biomass. The esters percentage mass composition can be understood by the mass variation of KOH on DTM from the biomass showed on Table 8.

Table 8 Ester compositions on soybean biomass biodiesel

Name	DTM1	DTM2	DTM3
Methyl palmitate(C _{16:0})	12.28	12.40	12.61
Methyl linolenate(C _{18:3})	8.26	7.10	6.95
Methyl linoleate(C _{18:2})	62.00	60.45	60.91
Methyl oleate (C _{18:1})	14.96	17.04	16.58
Methyl stearate (C _{18:0})	2.51	3.01	2.95

On biodiesel from soybean, the composition of esters does not vary at all. These values are not the same as esters from soybean oil biodiesel but methyl linoleate is the most abundant followed by methyl oleate, palmitate, linolenate and the less abundance is methyl stearate. Abundance in all esters corresponds to reported methyl esters from soybean oil biodiesel.

3.3.2.2 Roystonea Oleracea fruit biodiesel yield

The yield of 5 grams of Roystonea Oleracea fruit to ROFB from different methods is showed on Table 9.

Table 9 Yield on ROFB with different methods

Method	Time	g KOH	Yield
SHL	3h	0.05	Failed
SHL	6h	0.05	Failed
HE	3h	0.03	<1%
DTM1	3h	0.05	1%
DTM2	3h	0.1	7.54%
DTM3	3h	0.2	6.61%

As soybean biodiesel in ROFB the best procedure to produce biodiesel is DTM2. Percentage mass composition of esters is showed in Table 10.

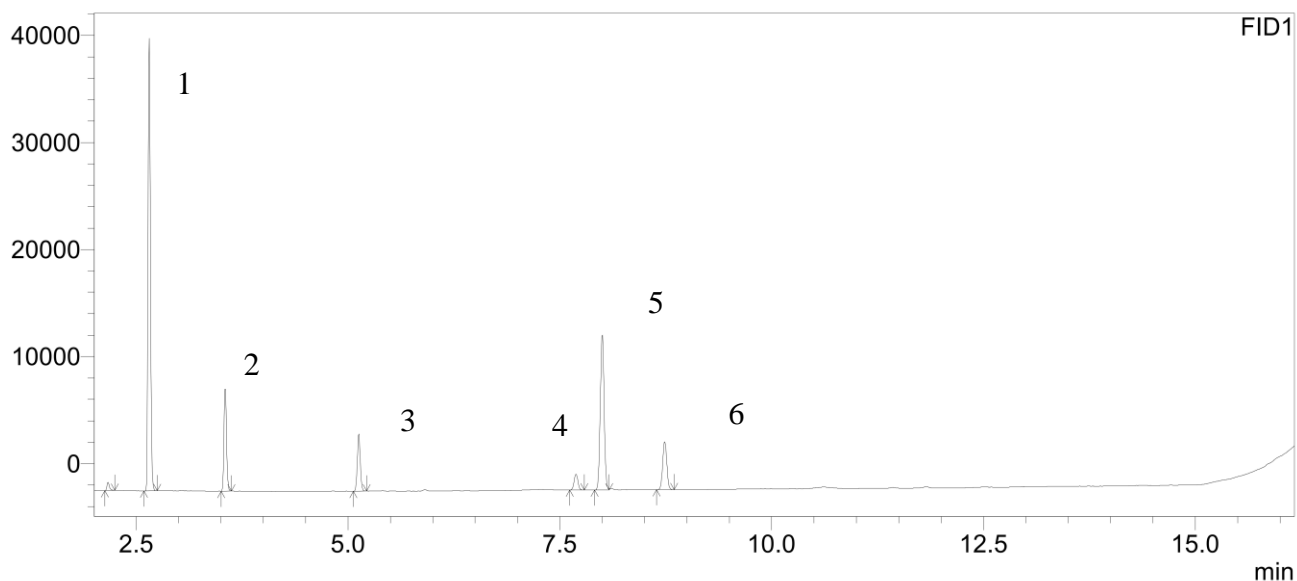


Figure 11 ROFB analysis by GC-FID

Table 10 Ester compositions on ROFB

Name	DTM1	DTM2	DTM3
Methyl myristate (C _{14:0})	24.72	18.92	22.80
Methyl palmitate(C _{16:0})	10.98	12.85	12.46
Methyl linoleate(C _{18:2})	45.26	47.67	45.21
Methyl oleate (C _{18:1})	19.03	16.81	16.20
Methyl stearate(C _{18:0})	<1	3.75	3.33

Table 11 Retention time of compounds reported on ROFB biodiesel analyzed by GC/FID

N°	Name	Chain Length	Retention Time
1	Methyl laurate	C _{12:0}	2.664
2	Methyl myristate	C _{14:0}	3.550
3	Methyl palmitate	C _{16:0}	5.156
4	Methyl stearate	C _{18:0}	7.729
5	Methyl oleate	C _{18:1}	8.039
6	Methyl linoleate	C _{18:2}	8.784

DTM 1, 2 and 3 show different composition of esters. In any case, methyl linoleate is the most abundant ester followed by methyl myristate, methyl oleate, methyl palmitate and the less abundant is methyl stearate. In order to get methyl myristate (C_{14:0}), which is not produce on soybean biodiesel, is useful to employ DTM 1 and 3.

4. CONCLUSIONS

The production and characterization of biodiesel from RO was successfully conducted in this research. Lipids soxhlet-hexane extraction (SHE) with a simple sample and hexane extraction has a low yield whereas SHE with multiple samples to produce biodiesel was not successful. Direct transesterification method is the best way to produce ROFB with alkali-catalyzed reaction with a yield of 7.54% and 0.1 g of KOH, 5 mL of methanol and 5 grams o *Roystonea Oleracea* fruits.

5. REFERENCES

- [1] Ocal, O., & Aslan, A. (2013). Renewable energy consumption–economic growth nexus in Turkey. *Renewable and Sustainable Energy Reviews*, 28, 494-499.
- [2] Barnwal BK, Sharma MP. Prospects of biodiesel production from vegetable oils in India. *Renew Sust Energ Rev* 2005;9:363–78
- [3] Pao, H. T., & Fu, H. C. (2013). Renewable energy, non-renewable energy and economic growth in Brazil. *Renewable and Sustainable Energy Reviews*, 25, 381-392.
- [4] Esen, M., & Yuksel, T. (2013). Experimental evaluation of using various renewable energy sources for heating a greenhouse. *Energy and Buildings*, 65, 340-351.
- [5] Yue, D., You, F., & Snyder, S. W. (2014). Biomass-to-bioenergy and biofuel supply chain optimization: overview, key issues and challenges. *Computers & Chemical Engineering*, 66, 36-56.
- [6] Garcia-Sánchez, J., López-Martínez, J., & Flores, H. (2012). Life Cycle Analysis of Euro IV urban buses. *DYNA* 87 , 45-57.

- [7] Caye, M., Nhuan, P., & Terry, H. (2008). *Biofuels Engineering Process Technology*. USA: McGraw-Hill.
- [8] Srivastava A, Prasad R. Triglycerides-based diesel fuels. *Renew Sust Energ Rev* 2000;4:111–33.
- [9] Khan AK. Research into biodiesel kinetics and catalyst development. In: Department of Chemical Engineering, University of Queensland, Brisbane, Australia; 2002.
- [10] Kandhro, A., Sherazi, S. T. H., Mahesar, S. A., Bhangar, M. I., Talpur, M. Y., & Rauf, A. (2008). GC-MS quantification of fatty acid profile including trans FA in the locally manufactured margarines of Pakistan. *Food Chemistry*, 109(1), 207-211
- [11] Knothe, G. (2005). Dependence of biodiesel fuel properties on the structure of fatty acid alkyl esters. *Fuel processing technology*, 86(10), 1059-1070
- [12] Freedman B, Pryde EH, Mounts TL. Variables affecting the yield of fatty esters from transesterified vegetable oils. *J Am Oil Chem Soc* 1984;61:1638–43.
- [13] Ma F, Clements LD, Hanna MA. The effect of mixing on transesterification of beef tallow. *Bioresour Technol* 1999;69:289–93.
- [14] Chitra P, Venkatachalam P, Sampathrajan A. Optimisation of experimental conditions for biodiesel production form alkali catalyzed transesterification of *Jatropha curcas* oil. *Energ Sust Dev* 2005;9:13–8.
- [15] Patil PD, Deng S. Optimization of biodiesel production from edible and nonedible vegetable oils. *Fuel* 2009;88:1302–6
- [14] Hoeksema, S. D. (2000). U.S. Patent No. 6,166,231. Washington, DC: U.S. Patent and Trademark Office.
- [17] Meher LC, Dharmagadda VS, Naik SN. Optimization of alkali catalyzed transesterification of *pongamia pinnata* for production of biodiesel. *Bioresour Technol* 2006;97:1392–7.
- [18] Henderson, A., Galeano-Garces, G., & Bernal, R. (1997). *Field guide to the palms of the Americas*. Princeton University Press.
- [19] Svenning, J. C. (2002). Non-native ornamental palms invade a secondary tropical forest in Panama. *PALMS-LAWRENCE*-, 46, 81-8.
- [20] Svenning, J. C. (2002). Non-native ornamental palms invade a secondary tropical forest in Panama. *PALMS-LAWRENCE*-, 46, 81-8.
- [21] Herrera, I., Hernandez-Rosas, J., Cornejo, X., Suarez, C., Ayala, C., Amaya, A., y otros. (2015). Naturalización y distribución potencial de una palma exótica ornamental (*Roystonea*). *Revista do Jardim Botânico do Rio de Janeiro*, Review work.
- [22] Farris-Lopez, K., Denslow, J. S., Moser, B., & Passmore, H. (2004). Influence of a common palm, *Oenocarpus mapora*, on seedling establishment in a tropical moist forest in Panama. *Journal of Tropical Ecology*, 20(04), 429-438.
- [23] Zucaratto, R., & Pires, A. D. S. (2014). The exotic palm *Roystonea oleracea* (Jacq.) OF Cook (Arecaceae) on an island within the Atlantic Forest Biome: naturalization and influence on seedling recruitment. *Acta Botanica Brasilica*, 28(3), 417-421.
- [24] Werle, L. B., Garcia, J. C., Kuhn, R. C., Schwaab, M., Foletto, E. L., Cancelier, A., ... & Mazutti, M. A. (2013). Ultrasound-assisted acid hydrolysis of palm leaves (*Roystonea oleracea*) for production of fermentable sugars. *Industrial Crops and Products*, 45, 128-132.
- [25] Ferreira-Dias S., Valente D. and Abreu J. (2003). Comparison between ethanol and hexane for oil extraction from *Quercus suber* L. fruits. *Grasas y Aceites* 54(4); 378–383.

- [26] Geris, R., dos Santos, N. A. C., Amaral, B. A., de SMaia, I., Castro, V. D., & Carvalho, J. R. M. (2007). Biodiesel de soja-reação de transesterificação para aulas práticas de química orgânica. *Química Nova*, 30(5), 1369
- [27] Akers, S. M., Conkle, J. L., Thomas, S. N., & Rider, K. B. (2006). Determination of the heat of combustion of biodiesel using bomb calorimetry. A multidisciplinary undergraduate chemistry experiment. *J. Chem. Educ.*,83(2), 260.
- [28] P. E. Wall, *Thin-Layer Chromatography: A Modern Practical Approach*, Royal Society of Chemistry, Cambridge, 2005.
- [29] Moraes, M. S. A. (2008). Biodiesel de sebo: Avaliação de propriedades e testes de consumo em motor a diésel.
- [30] Cachet, T. (2011). Guidelines for the quantitative gas chromatography of volatile flavouring substances, from the Working Group on Methods of Analysis of the International Organization of the Flavor Industry (IOFI)
- [31]Ragonese, C., Tranchida, P. Q., Sciarrone, D., & Mondello, L. (2009). Conventional and fast gas chromatography analysis of biodiesel blends using an ionic liquid stationary phase. *Journal of Chromatography A*,1216(51), 8992-8997
- [32] Hoekman, S. Kent, et al. "Review of biodiesel composition, properties, and specifications." *Renewable and Sustainable Energy Reviews* 16.1 (2012): 143-169.
- [33]Wu, L. C., & Bates, R. P. (1972). SOY PROTEIN-LIPID FILMS. 1. Studies on the Film Formation Phenomenon. *Journal of Food Science*, 37(1), 36-39.