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GC–MS CHARACTERIZATION OF FATTY ACID ALKYL ESTERS IN BIODIESEL PRODUCED FROM WASTE COOKING OIL

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Abstract

This study aims to identify and characterize the major compounds in biodiesel produced through the transesterification of vegetable oils using Gas Chromatography–Mass Spectrometry (GC–MS). The analysis was conducted to evaluate the composition of fatty acid esters—specifically Fatty Acid Methyl Esters (FAME) and Fatty Acid Ethyl Esters (FAEE)—which play a crucial role in determining biodiesel quality. Three biodiesel samples were analyzed, revealing dominant peaks at retention times between 15 and 17 minutes, identified as methyl palmitate ($C_{17}H_{34}O_2$), methyl oleate ($C_{19}H_{36}O_2$), and methyl stearate ($C_{19}H_{38}O_2$). The total content of these major esters ranged from 85% to 92%, indicating an efficient conversion of triglycerides into esters. The resulting chemical profile exhibited a balanced proportion of saturated and unsaturated compounds, providing high oxidative stability and favorable cold flow properties. These findings demonstrate that the produced biodiesel meets the requirements of international standards ASTM D6751 and EN 14214, confirming its potential as an environmentally friendly alternative fuel.

Keywords: Biodiesel, GC–MS, Methyl Ester, Transesterification, Fatty Acid.

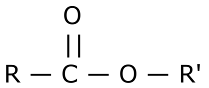
Abstrak

Penelitian ini bertujuan untuk mengidentifikasi dan mengkarakterisasi senyawa penyusun utama biodiesel hasil proses transesterifikasi minyak nabati menggunakan teknik *Gas Chromatography–Mass Spectrometry* (GC–MS). Analisis dilakukan untuk mengevaluasi komposisi senyawa ester asam lemak (*Fatty Acid Methyl Ester*, FAME, dan *Fatty Acid Ethyl Ester*, FAEE) yang berperan penting dalam menentukan kualitas biodiesel. Tiga sampel biodiesel dianalisis dengan hasil menunjukkan adanya puncak dominan pada waktu retensi antara 15 hingga 17 menit, yang diidentifikasi sebagai metil palmitat ($C_{17}H_{34}O_2$), metil oleat ($C_{19}H_{36}O_2$), dan metil stearat ($C_{19}H_{38}O_2$). Kandungan total ester utama mencapai 85–92%, menandakan konversi trigliserida menjadi ester berlangsung efisien. Profil kimia yang terbentuk memperlihatkan keseimbangan antara senyawa jenuh dan tak jenuh, yang memberikan kestabilan oksidatif tinggi serta sifat aliran dingin yang baik. Hasil ini menunjukkan bahwa biodiesel yang dihasilkan memenuhi karakteristik sesuai standar internasional ASTM D6751 dan EN 14214, serta berpotensi digunakan sebagai bahan bakar alternatif ramah lingkungan.

Keywords: Biodiesel, GC-MS, Metil Ester, Transesterifikasi, Asam Lemak.

1. Introduction

Biodiesel has emerged as a prominent alternative fuel sourced from renewable and environmentally friendly resources, aimed at reducing reliance on fossil fuels and diminishing greenhouse gas emissions, which are principal contributors to global climate change (Musharraf et al., 2012). It is characterized by its biodegradable nature and non-toxic properties, allowing it to be utilized directly in conventional diesel engines or blended with petroleum-based diesel without requiring extensive modifications (Mandari & Kumar, 2021). Chemically, biodiesel is primarily composed of long-chain fatty acid esters produced through the transesterification of triglycerides found in vegetable oils or waste cooking oils (Bhuiya et al., 2014). The chemical formula of long-chain fatty acid ester was shown in Figure 1.



$$R - \overset{\overset{O}{\|}}{C} - O - R'$$

Figure 1. The chemical formula of long-chain fatty acid ester.

This transesterification process converts triglycerides into Fatty Acid Alkyl Esters (FAAE) using short-chain alcohols such as methanol or ethanol, catalyzed by a base catalyst like sodium hydroxide (NaOH) or potassium hydroxide (KOH) (Kurdi et al., 2021). The primary end products are Fatty Acid Methyl Esters (FAMEs) when methanol is used and Fatty Acid Ethyl Esters (FAEEs) when ethanol is applied, with glycerol produced as a by-product (Chhetri et al., 2008). The composition and ratio of these esters are crucial as they directly influence the physicochemical properties and combustion performance of biodiesel (Khan et al., 2021).

Table 1. The Criteria for Good Biodiesel (ASTM International, 2023)

Parameter	Standard Limit	Importance for Biodiesel Quality
Density (40° C)	0.86 – 0.90 g/cm^3	Influences fuel injection and atomization efficiency
Kinematic viscosity (40° C)	1.9 – 6.0 mm^2/s	Affects fuel flow and combustion characteristic
Cetane number	≥ 47 (ASTM) or ≥ 51 (EN)	Indicates ignition quality and combustion efficiency
Acid value	≤ 0.5 mg KOH/g	High acidity can cause corrosion in engines
Flash point	≥ 120° C	Indicates fuel safety during storage and transport
Water content	< 500 ppm	Excess water promotes microbial growth and corrosion
Sulphur content	≤ 15 ppm	Reduces SO_x emissions and environmental pollution
FAME content	≥ 96.5%	Indicates the purity of biodiesel product
Oxidation stability	≥ 6 h	Determines resistance to degradation during storage
Free glycerol	≤ 0.02% wt	Prevents injector deposits and engine fouling

Several key factors dictate the quality of biodiesel, which show in Table 1. These include the choice of feedstock, reaction conditions such as the molar ratio of alcohol to oil, temperature, reaction time, and the concentration of the catalyst, alongside the purity of the final product (Park et al., 2019). The distribution and content of FAME and FAEE compounds are particularly significant in determining crucial biodiesel characteristics like density, viscosity, cetane number, oxidative stability, and low-temperature flow properties (Poudel et al., 2017). Consequently, analyzing the chemical composition of biodiesel is essential to confirm that it meets international quality standards such as ASTM D6751 and EN 14214 (Emmanouilidou et al., 2023). Gas Chromatography-Mass Spectrometry (GC-MS) is among the most reliable and widely used analytical techniques for identifying and quantifying biodiesel constituents (Peters et al., 2022). This method enables the separation and detection of fatty acid ester compounds based on their retention times and mass spectral data, allowing for the identification of the compounds that significantly influence biodiesel quality (Khan et al., 2021). Moreover, GC-MS analysis provides insights into the effectiveness of the transesterification process, the efficiency of triglyceride conversion, and the presence of possible impurities such as unreacted triglycerides, diglycerides, and monoglycerides (Park et al., 2019).

Gas chromatography–mass spectrometry (GC–MS) is a critical analytical technique employed to identify and quantify the major chemical constituents of biodiesel derived from the transesterification of vegetable oil or waste cooking oil. This method provides detailed chromatograms that showcase the retention times and peaks of various chemical components within a sample. Specifically, chromatograms for biodiesel samples exhibit well-resolved peaks in a retention time range of approximately 10–35 minutes, thereby confirming the presence of distinct fatty acid methyl esters (FAMEs) as the primary components constituting biodiesel (Musharraf et al., 2012; (Mandari & Kumar, 2021; Bhuiya et al., 2014). The identification of these components is not only crucial for quality control but also plays a significant role in ensuring compliance with biodiesel fuel standards. Research indicates that biodiesel comprised primarily of FAMEs is appreciated for its renewable nature and lower emissions profile compared to conventional fossil fuels (Mandari & Kumar, 2021; Kurdi et al., 2021). The biogenic synthesis of these esters through transesterification signifies a transformation of lipid-rich raw materials into a viable energy source, highlighting the environmental sustainability aspect of such processes (Chhetri et al., 2008; Bhuiya et al., 2014).

Moreover, recent studies have shown that the specific composition of these esters can vary based on the feedstock type and processing conditions, with waste cooking oil yielding notable amounts of specific fatty acids like methyl oleate and methyl linoleate (Khan et al., 2021; Park et al., 2019). For instance, the biodiesel synthesized from waste cooking oils can exhibit variations in ester content when compared to those derived from fresh oils or algal sources, emphasizing the necessity for careful characterization using techniques like GC–MS to tailor biodiesel production according to required specifications (Poudel et al., 2017; Park et al., 2019). In the context of biodiesel production, the successful application of GC–MS allows for the meticulous analysis and optimization of biodiesel formulations, thereby ensuring efficacy and sustainability in biodiesel use across various applications (Emmanouilidou et al., 2023), (Peters et al., 2022). The clear identification of FAMEs serves as a benchmark for assessing biodiesel quality and its performance in differing engine types while ensuring compliance with established fuel standards such as EN 14214 (Mandari & Kumar, 2021; Emmanouilidou et al., 2023).

Improper disposal of waste cooking oil (WCO) poses significant environmental and public health concerns. When discharged into drainage systems or natural water bodies, WCO forms an oil layer on the water surface that inhibits oxygen transfer and increases chemical oxygen demand, which can lead to oxygen depletion and damage to aquatic ecosystems. In addition, the accumulation of oil in sewer systems may cause pipe blockages, unpleasant odors, and increased maintenance costs in wastewater infrastructure. WCO disposed into soil can also infiltrate groundwater and reduce soil permeability, potentially affecting terrestrial ecosystems. From a health perspective, repeated heating and improper reuse of cooking oil can generate harmful degradation products such as aldehydes, polymers, and free radicals that are associated with various health risks. Therefore, proper management and valorization of WCO into value-added products such as biodiesel represent an important strategy to mitigate environmental pollution while promoting sustainable energy production (Bhatia et al., 2020; Mannu et al., 2020; Tsai, 2019).

This study aims to utilize the GC-MS technique to identify and quantify fatty acid ester compounds present in biodiesel derived from the transesterification of vegetable oils or waste cooking oils. Such analysis is expected to offer further elucidation on the chemical composition of biodiesel and its correlation with the ensuing fuel quality, thereby contributing valuable information to the field of biodiesel research (Fakai et al., 2024). In line with sustainable developmental goals (SDGs) point 7, affordable and clean energy, the demand of sustainable energy and the environmental challenges associated with waste cooking oil disposal have intensified interest in biodiesel production from waste-derived feedstocks. A comprehensive GC-MS characterization of biodiesel produced from waste cooking oil, focusing on the identification and distribution of both fatty acid methyl esters (FAME) and fatty acid ethyl esters (FAEE) was provided in this study. The comparison of several biodiesel samples demonstrates differences in ester composition and conversion efficiency, providing important insights into the chemical characteristics and quality evaluation of biodiesel produced from waste-derived feedstocks.

2. Methodology

2.1 Materials

The materials used in this study included vegetable oil (or pretreated and filtered waste cooking oil) as the lipid feedstock, methanol (CH_3OH) and ethanol (C_2H_5OH) as transesterification agents, and sodium hydroxide (NaOH) as the homogeneous alkaline catalyst. *n*-Hexane was employed as an extraction and dilution solvent during sample preparation prior to GC–MS analysis. The experimental apparatus comprised a mechanically agitated, temperature-controlled transesterification reactor, a high-speed centrifuge for efficient phase separation, a rotary evaporator for solvent recovery, and a Shimadzu QP-2010 Plus gas chromatography–mass spectrometry (GC–MS) system for comprehensive qualitative and quantitative characterization of the biodiesel chemical composition.

Biodiesel Synthesis Procedure

C3H5(OOCR)3 + 3 R'OH → 3 RCOOR' + C3H5(OH)3
Triglyceride Alcohol Fatty Acid Ester Glycerol

Biodiesel was produced via an alkali-catalyzed transesterification reaction using an alcohol-to-oil molar ratio of 6:1 and sodium hydroxide (NaOH) at 1 wt% relative to the oil. The reaction mixture was heated to 60 °C and maintained for 60 min under continuous stirring. Upon completion of the reaction, phase separation occurred, yielding an upper biodiesel phase and a lower glycerol phase. The biodiesel layer was carefully separated, repeatedly washed with warm distilled water until a neutral pH was achieved, and subsequently dried at 105 °C to remove residual moisture.

2.2 GC–MS Analysis

Purified biodiesel samples were analyzed using a gas chromatography–mass spectrometry (GC–MS) system. Chromatographic separation was carried out using a Rtx-5MS capillary column (30 m × 0.25 mm i.d. × 0.25 μm film thickness), with helium employed as the carrier gas at a constant flow rate of 1.0 mL min^{-1}. The mass spectrometer was operated in electron impact (EI) ionization mode at 70 eV. The oven temperature program was set to an initial temperature of 60 °C (held for 2 min), ramped at 10 °C min^{-1} to 300 °C, and held for 10 min. Sample injection was performed in split mode with a split ratio of 1:50 and an injection volume of 1 μL.
Mass spectra were processed using the instrument's integrated software, and compound identification was achieved by matching the obtained spectra with those in the NIST Mass Spectral Library. The analytical parameters evaluated included retention time (RT), peak area percentage, and compound identity for each chromatographic peak. The resulting data were used to determine the relative proportions of fatty acid methyl esters (FAME) and fatty acid ethyl esters (FAEE) in each sample and to assess the overall quality of the produced biodiesel.

3. Results and Discussion

3.1 Gas Chromatography–Mass Spectrometry (GC–MS) in Biodiesel Analysis

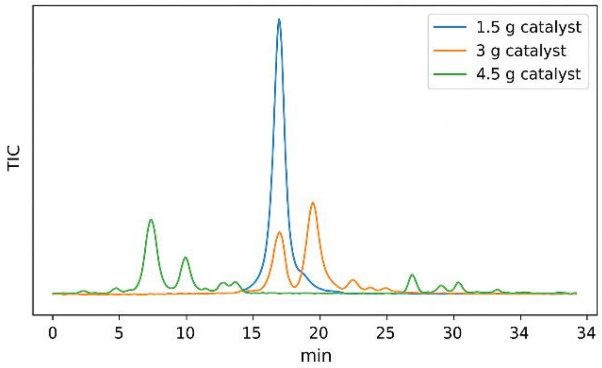


Figure 2. Representative GC–MS chromatograms of the biodiesel samples.

The analysis of biodiesel derived from waste cooking oil via gas chromatography-mass spectrometry (GC-MS) has shown that the dominant peaks in the chromatograms occur within the retention time range of 15.0–17.5 minutes. These peaks correspond to significant fatty acid methyl esters (FAMEs), namely methyl palmitate, methyl oleate, and methyl stearate. These methyl esters are the esterification products of long-chain saturated and monounsaturated fatty acids (C16–C18), which are prevalent in these feedstocks (Khan et al., 2021), (Park et al., 2019). The presence of methyl palmitate and methyl oleate, in particular, highlights the composition's alignment with common fatty acid profiles found in palm oil and waste cooking oil. Waste cooking oil, which is increasingly used for biodiesel production, contains substantial amounts of these specific fatty acids, emphasizing its potential as a cost-effective and sustainable feedstock (Fakai et al., 2024). In fact, studies have indicated that the biodiesel produced from waste cooking oil tends to exhibit a favorable composition of FAMEs, enhancing thermal stability and overall fuel performance (Fakai et al., 2024). Moreover, the chromatographic analysis indicated the absence of significant peak overlap or baseline disturbances, which implies effective purification techniques and satisfactory thermal stability of the synthesized biodiesel. This clear separation supports the notion that the biodiesel produced is not only rich in essential methyl esters but also of high purity, which is crucial for meeting quality standards in biodiesel production (Hsiao et al., 2018).

3.2 Composition of Major Compounds

GC–MS identification revealed that all samples exhibited similar chemical compositions, dominated by FAME and FAEE species. The major compounds identified in each sample are summarized in Table 1.

Table 2. Major compounds identified by GC–MS analysis

Sample	Major compound	Retention time (min)	Peak area (%)	Compound type
1.5 g	Methyl palmitate	15.09	33.18	Saturated FAME
	Methyl oleate	16.88	48.19	Monounsaturated FAME
	Methyl stearate	17.04	5.91	Saturated FAME
3 g	Methyl palmitate	15.09	34.60	Saturated FAME
	Methyl oleate	16.87	33.47	Monounsaturated FAME
	Methyl stearate	17.21	9.92	Saturated FAME
4.5 g	Methyl palmitate	15.10	33.73	Saturated FAME
	Methyl oleate	16.87	33.78	Monounsaturated FAME
	Methyl stearate	17.15	13.72	Saturated FAME
	Ethyl oleate	17.21	10.65	Monounsaturated FAEE

The total ester content (FAME + FAEE) ranged from approximately 87% to 92%, indicating a high conversion efficiency of triglycerides into alkyl esters. This result confirms that the transesterification reaction proceeded effectively, with 8-13% residual glycerides or impurities.

3.3 Interpretation of Chemical Composition

Gas chromatography-mass spectrometry (GC-MS) results illustrate that the predominant components of biodiesel are methyl oleate ($C_{19}H_{36}O_2$) and methyl palmitate ($C_{17}H_{34}O_2$), followed closely by methyl stearate ($C_{19}H_{38}O_2$) and methyl linoleate ($C_{18}H_{34}O_2$). This specific composition reflects the high concentration of oleic and palmitic acids, fatty acids that are characteristic of various oil feedstocks including palm oil and waste cooking oil (Tabatabai et al., 2018) Khan et al., 2021). The significant presence of these fatty acid methyl esters (FAMEs) is indicative of the feedstock quality, largely contributing to the physical and chemical properties of the resulting biodiesel.

Saturated FAMEs such as methyl palmitate and methyl stearate are known to enhance oxidative stability and improve cetane numbers, which in turn enhances combustion efficiency and storage stability (Fakai et al., 2024). These properties are essential for the long-term usability of biodiesel in various engine types. Methyl oleate, a monounsaturated FAME, positively influences fuel flow properties and significantly reduces the viscosity of biodiesel, which benefits performance particularly in low-temperature conditions (Knothe & Dunn, 2003; Knothe, 2002). The balanced composition of saturated and unsaturated esters plays a crucial role in biodiesel quality, ensuring high oxidative stability without compromising fluidity, thus allowing biodiesel to be effective under diverse climatic and operational scenarios (Tat et al., 2007).

3.4 Comparison among Samples

A comparative evaluation of the three samples revealed minor variations in the distribution of major ester components (Table 2).

Table 3. Comparison of major ester contents among samples

Compound	Chemical formula	Sample 1 (%)	Sample 2 (%)	Sample 3 (%)
Methyl palmitate	CH3–(CH2)14–C(=O)–O–CH3	33.18	34.60	33.73
Methyl oleate	CH3–(CH2)7–CH=CH–(CH2)7–C(=O)–O–CH3	48.19	33.47	33.78
Methyl stearate	CH3–(CH2)16–C(=O)–O–CH3	5.91	9.92	13.72
Ethyl oleate	CH3–(CH2)7–CH=CH–(CH2)7–C(=O)–O–CH2–CH3	–	–	10.65
Total major esters		87.28	78.00	91.88

Sample 1 exhibited the highest methyl oleate content (48.19%), suggesting superior cold-flow performance. Sample 2 showed a more balanced distribution between methyl palmitate and methyl oleate, whereas Sample 3 displayed increased levels of methyl stearate (13.72%) and ethyl oleate (10.65%). The presence of higher saturated FAME and FAEE contents in Sample 3 indicates a more complete transesterification reaction and suggests the involvement of ethanol as a co-reactant during synthesis.

These variations may be attributed to differences in processing parameters, such as alcohol-to-oil ratio, catalyst concentration, or reaction temperature. Sample 3 demonstrated the highest conversion efficiency, as reflected by a total ester content of 91.88%, implying improved thermal stability and potentially lower susceptibility to oxidative degradation.

3.5 Implications for Biodiesel Quality

The chemical composition of biodiesel directly influences its physicochemical properties and combustion performance. The methyl oleate contained in sample 1, sample 2, and sample 3 are 48.19%, 33.47%, and 33.78% respectively A high methyl oleate content enhances combustion efficiency, reduces carbon monoxide and particulate emissions, and improves fuel atomization within the combustion chamber. Conversely, the presence of methyl palmitate and methyl stearate reinforces oxidative stability and mitigates thermal degradation during long-term storage.

Overall, the predominance of C16–C18 FAMEs ensures compliance with international biodiesel specifications such as ASTM D6751 and EN 14214. An ester content exceeding 85% confirms the efficiency of the transesterification process, while the moderate presence of FAEE enhances biodiesel adaptability to varying ambient temperatures. Furthermore, a balanced ratio of saturated and unsaturated esters contributes to stable combustion behavior and reduced exhaust emissions, reinforcing the potential of biodiesel as a sustainable and environmentally friendly alternative fuel.

4. Conclusion

This study successfully demonstrated the synthesis of biodiesel from vegetable oil or waste cooking oil via alkali-catalyzed transesterification, followed by comprehensive chemical characterization using gas chromatography–mass spectrometry (GC–MS). The GC–MS analysis confirmed that the produced biodiesel predominantly consisted of fatty acid methyl esters (FAMEs) and fatty acid ethyl esters (FAEEs), with total ester contents ranging from approximately 87% to 92%, indicating a high conversion efficiency of triglycerides into alkyl esters.

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