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Impact of Ferric Chloride–Induced Oxidation on the Physicochemical Characteristics of Chicken Fat and Its Biodiesel

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Abstract

Background/problems: Biodiesel from animal fats is vulnerable to metal-catalyzed oxidation that degrades fuel quality because of the unsaturated fatty acid content. This study investigates the oxidative stability and physicochemical changes of chicken fat and its biodiesel upon $FeCl_3$*-catalyzed oxidation. Methods: Chicken fat was transesterified to produce biodiesel; chicken fat (CF) and chicken-fat biodiesel (CF-BD) were then subjected to an* $FeCl_3$ *oxidative condition. Acid value (AV), iodine value (IV), peroxide value (PV), kinematic viscosity at 40 °C, and density were determined using standard titrimetric and physical methods; FTIR assessed spectral changes. ANOVA compared group means. Findings: Oxidation significantly increased AV and PV for both matrices, with a marked PV rise in CF-BD (6.17 ± 0.29 to 88.17 ± 5.35 meq/kg), and decreased IV (CF-BD 84.3 ± 4.51 to 48.7 ± 4.32 g* I_2*/100 g). Viscosity and density increased modestly. Visually, CF-BD shifted from clear yellow-green to brown and turbid with sediment, consistent with polymer formation and methyl-ester degradation. FTIR showed attenuation/shift of ester carbonyl and aliphatic bands. Conclusion:* $FeCl_3$*-induced oxidation measurably deteriorates CF-BD quality, breaching key limits in ASTM/SNI for acid value and viscosity. Impact: Results underscore the need to control trace metals and apply antioxidant/metal-deactivator strategies in storage and distribution, and validate the* $FeCl_3$ *assay as a practical accelerated-degradation model for stability and additive screening.*

Keywords: Biodiesel oxidation; chicken fat biodiesel; $FeCl_3$; physicochemical biodiesel; transesterification

Introduction

The use of alternative energy derived from plant- and animal-based feedstocks has gained increasing attention due to the depletion of fossil fuel reserves and growing concerns about greenhouse gas emissions. Biodiesel has been recognized as one of the most promising renewable fuels because it can be produced by transesterifying triglycerides with alcohol. Triglycerides are not limited to vegetable oils; they can also be derived from animal fats, which are often considered waste but have high lipid content. One notable example is chicken offal fat, a solid byproduct of poultry processing that remains underutilized despite its abundant availability (Kusumawardani et al., 2023). Converting this waste into biodiesel not only helps diversify renewable energy sources but also supports environmentally sustainable waste management practices.

However, biodiesel derived from chicken fat exhibits relatively poor oxidative stability (de Menezes et al., 2022). Biodiesel, rich in unsaturated fatty acids, especially oleic and linoleic acid, readily undergoes autoxidation when exposed to heat, light, or trace transition metals (Longanesi et al., 2022). The oxidation process initially forms hydroperoxides as primary products, which are commonly evaluated using peroxide value measurements (Pratiwi & Meikapasa, 2025). Primary hydroperoxides form first and then break down into aldehydes, ketones, and carboxylic acids that drive gum/sediment formation, raise viscosity, and ultimately degrade fuel quality and engine reliability (Mantovani et al., 2025).

The adoption of biodiesel from waste lipids continues to expand. However, it remains constrained by oxidative instability, particularly in fuels rich in unsaturated esters and exposed to heat, light, or transition metals (Longanesi et al., 2022). Transition-metal ions accelerate peroxide formation and subsequent secondary reactions that elevate acidity, viscosity, and deposit-forming species, compromising engines and storage systems (Song et al., 2023; Mantovani et al., 2025). Despite numerous storage-stability studies, metal-catalyzed oxidative

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models specific to animal-fat biodiesel are less represented, and the role of Fe^{3+} as a practical pro-oxidant for rapid screening merits closer examination (Sui et al., 2021; Fazal et al., 2022).

To date, limited information is available regarding the effects of $FeCl_3$ on the oxidative behavior of chicken fat–based biodiesel. Most previous studies have focused on natural storage stability or antioxidant use (Masudi et al., 2023; Longanesi et al., 2022; de Sousa et al., 2021) without a detailed analysis of the chemical changes induced by catalytic oxidation. However, no study has systematically evaluated $FeCl_3$-catalyzed oxidation as an accelerated model for chicken fat-based biodiesel, combining physicochemical parameters and FTIR analysis.

Several methods have been employed to assess the oxidative stability of biodiesel, including the Rancimat test and accelerated aging at elevated temperatures. Although many studies address storage stability and antioxidant use, comparatively fewer works explicitly simulate metal-catalyzed oxidation; ferric chloride ($FeCl_3$) is a practical Fe^{3+} source that can participate in Fenton-like Fe^{3+}/Fe^{2+} redox cycles to generate reactive radicals and accelerate ester degradation. Evidence also indicates that soluble Fe(III) interacts with oxidized biodiesel to form catalytic iron–carboxylate species, reinforcing the role of iron in promoting secondary reactions (Song et al., 2023; Sui et al., 2021). This mechanism may effectively accelerate peroxide formation in biodiesel, providing a controlled model for studying oxidative degradation.

This study quantifies the impact of $FeCl_3$-induced oxidation on the physicochemical quality of chicken fat and its biodiesel by comparing AV, IV, PV, viscosity, density, and FTIR signatures before and after oxidation, and by benchmarking against ASTM D6751/SNI 7182 criteria. We further evaluate $FeCl_3$ oxidative challenge as a rapid screening model for stability and additive studies.

Methods

Materials

The primary feedstock used in this research was broiler chicken offal obtained from Bersehati Market, Manado, Indonesia. All chemicals used in this study were of analytical grade (pro analysis, Merck, Germany). The reagents included ethanol (CH_3CH_2OH), potassium hydroxide (KOH), ferric chloride ($FeCl_3$), potassium iodide (KI), glacial acetic acid (CH_3COOH), chloroform ($CHCl_3$), sodium thiosulfate pentahydrate ($Na_2S_2O_3 \cdot 5H_2O$), phenolphthalein, starch, Wijs solution, and sodium hydroxide (NaOH), with distilled water supplied by WaterOne. Laboratory glassware and reflux apparatus were employed for synthesis and purification procedures. Viscosity measurements were performed using a digital viscometer (Icon Scientific, England), while density was determined with standard glass pycnometers. Fourier Transform Infrared (FTIR) spectra were recorded on a Bruker Invenio S (Germany) spectrometer.

Chicken fat Separation

The chicken offal was cleaned, and the associated glands were removed. The fat was then gently heated over low heat until no oil was released. The extracted fat was separated from water by decantation, filtered through filter paper, and stored in air-tight containers for further use.

Transesterification

Transesterification is following Maanari et al. (2023). 500 mL of chicken fat was heated to 50°C and mixed with a solution containing 4.8 g of KOH dissolved in 410 mL of ethanol. The mixture was then refluxed at 70–72°C for 2 h under continuous stirring. After the reaction was complete, the product was washed with distilled water until a neutral pH (~7) was achieved and subsequently dried to remove residual moisture.

Oxidation reaction using $FeCl_3$

Two grams of the sample were mixed with 20 mL of a chloroform: glacial acetic acid solution (1:1, v/v), followed by the addition of 0.5 mL of saturated KI. The mixture was gently shaken and kept in the dark for 5 min. Then, 30 mL of distilled water was added, and the solution was titrated with $Na_2S_2O_3$ until a pale-yellow color appeared. One milliliter of 1% starch indicator solution was added, and titration continued until the solution became colorless (AOAC, 1997). The peroxide value was calculated as follows:

$$PV(meq\ O_2/kg) = \frac{(V \times N) \times 100}{m} \tag{1}$$

Where V is the volume of $Na_2S_2O_3$ in liters, N is the normality of $Na_2S_2O_3$, and m is the sample mass in grams.

Acid value (AV) determination

Two grams of the biodiesel sample were dissolved in 20 mL of hot ethanol. The solution was titrated with 0.1 N KOH using phenolphthalein as an indicator until a persistent pink color appeared for 30 s (AOAC, 1997). The acid value was calculated as follows:

$$AV(mg\ KOH/g) = \frac{V \times N \times 56.1}{m} \tag{2}$$

Where V is the volume of KOH in mL, N is the normality of KOH, and m is the sample mass in grams.

Iodine value (IV) determination

A 0.2 g sample was dissolved in 10 mL of chloroform, and 25 mL of Wijs iodine reagent was added. The mixture was then incubated in the dark for 30 min. Next, 20 mL of a 10% potassium iodide solution and 100 mL of distilled water were added. The solution was titrated with 0.1 N $Na_2S_2O_3$ until a pale yellow color appeared, and then the starch indicator was added until the blue color disappeared

(AOAC, 1997). The iodine value was calculated as follows:

$$IV(g\, I_2/100g) = \frac{V \times N \times 12.69}{m} \qquad (3)$$

Where V is the volume of $Na_2S_2O_3$ in mL, N is the normality of $Na_2S_2O_3$, and m is the sample mass in grams.

Viscosity, density, and FTIR analysis

The viscosity and density of the chicken fat and biodiesel samples were measured before and after oxidation. Viscosity was determined using a Viscosity Analyzer (Icon Scientific) at 40°C, and density was measured using a pycnometer at room temperature. Fourier-transform infrared (FTIR) spectra were obtained using Invenio S by Bruker to identify the changes in functional groups associated with oxidation at a scanning range of 400–4000 cm^{-1}.

Statistical analysis

Data was analyzed using analysis of variance (ANOVA) one-way ($p < 0.05$) with post hoc test by IBM SPSS Statistics 27. Each determination was performed in triplicate to ensure reliability, and the results were expressed as the mean ± standard deviation.

Results and Discussion

The extraction process in this study yielded broiler chicken fat at an average rate of 23.78 ± 4.3%. Subsequent transesterification of the extracted fat achieved a high conversion efficiency of 92.58 ± 2.1%, resulting in a substantial production of biodiesel. When considering the initial mass of broiler chicken as the baseline, the total yield of biodiesel corresponded to 22.01% of the starting material. These findings indicate that, for every 100 grams of broiler chicken processed, approximately 22 grams of biodiesel can be produced. However, a significant drawback associated with chicken fat-derived biodiesel is its pronounced susceptibility to oxidative degradation, primarily due to the high content of unsaturated fatty acids such as oleic and linoleic acids. The presence of these unsaturated components facilitates rapid autoxidation, which can adversely affect fuel stability and quality during storage (Binhweel et al., 2023).

A comparative analysis of the physicochemical properties of chicken fat and its derived biodiesel, both before and after accelerated oxidation using $FeCl_3$ as a prooxidant, is presented in **Table 1**.

Table 1. A comparative analysis of physicochemical properties of chicken fat and its derived biodiesel, both before and after accelerated oxidation using $FeCl_3$

Parameter	Chicken Fat Before Oxidation	Chicken Fat After Oxidation	Chicken Fat Biodiesel Before Oxidation	Chicken Fat Biodiesel After Oxidation	SNI 7182:2015
Acid value (mg KOH/g)	0.21 ± 0.11^c	1.80 ± 0.20^a	0.17 ± 0.05^c	0.89 ± 0.14^b	0.5 max.
Iodine value (g I_2/100g)	75.5 ± 6.12^a	68.0 ± 4.95^b	84.3 ± 4.51^a	48.7 ± 4.32^c	115 max.
Peroxide value (meq/kg)	2.50 ± 1.50^c	15.33 ± 1.26^b	6.17 ± 0.29^c	88.17 ± 5.35^a	-
Viscosity (mm^2/s, 40°C)	34.45 ± 1.15^a	36.12 ± 3.11^a	5.07 ± 2.12^b	6.92 ± 0.93^b	2.3 – 6.0
Density (g/mL)	0.9112 ± 0.02^a	0.9269 ± 0.03^a	0.8712 ± 0.03^a	0.8920 ± 0.03^a	0.85 – 0.89

Note: values are expressed as mean ± standard deviation (n = 3). Means with different letters in the same row were significantly different at the $p < 0.05$ level.

After oxidation with $FeCl_3$, both chicken fat and its biodiesel exhibited an increase in the acid value. Desbruslais & Wealleans (2022) and Zamuz et al. (2022) demonstrated that prolonged exposure to oxygen and heat accelerates the release of free fatty acids via autoxidation processes in animal fats, particularly poultry fat. In biodiesel, an elevated acid value indicates the partial degradation of ester to short-chain fatty acids. This phenomenon is consistent with the findings of Longanesi et al. (2022) and de Menezes et al. (2022), who explained that biodiesel oxidation primarily involves the formation and decomposition of hydroperoxides during storage. This oxidative pathway oxidizes methyl ester components to form secondary products, such as saturated fatty acids, aldehydes, and ketones.

The iodine value, which indicates the degree of unsaturation of fats, decreased significantly in both chicken fat and its biodiesel after oxidation. This decline signifies that a considerable number of carbon-carbon double bonds (C=C) in unsaturated fatty acids, such as oleic and linoleic acids, underwent oxidation, resulting in a reduction in unsaturated fatty acid content. Similar findings were reported by de Menezes et al. (2022) and Yusri et al. (2020) for plant oil-based biodiesels rich in unsaturated fatty acids, in which prolonged exposure to oxygen and light triggers the oxidation of unsaturated carbon chains, substantially lowering the iodine value. As described by Machado et al. (2022), the low stability of double bonds in fatty acids is due to the vulnerability of pi bonds to oxygen attack; thus, the longer the oil is exposed to oxygen, the greater the decrease in the iodine value.

After oxidation, an increase in the peroxide value was observed in chicken fat, but a significantly higher increase was observed in chicken fat biodiesel. This indicates a much more intense formation of hydroperoxides in the ester phase. This pattern aligns with Kumar's (2017) report, which emphasized that biodiesel oxidation elevates peroxide values and other physical properties by forming primary and secondary lipid oxidation products. The greater susceptibility of biodiesel can be attributed to the higher content of carbon-carbon double bonds in fatty acid methyl esters (FAME), which are more susceptible to radical attack than the original triglycerides. Additionally, the natural antioxidants present in chicken fat are lost during transesterification. Fe^{3+} ions specifically accelerate the decomposition of hydroperoxides (ROOH) into free radicals via the Fe^{3+}/Fe^{2} redox cycle, thereby shortening the induction period and hastening peroxide accumulation (Sui et al., 2021; Singh et al., 2020).

The viscosities of both chicken fat and its biodiesel increased after oxidation. This increase is likely due to the formation of higher molecular weight polymeric compounds. Song et al. (2023) demonstrated that biodiesel interacting with iron ions can form soluble carboxylate complexes, thereby increasing fuel viscosity through the formation of stable metal-ester polymeric networks. Additionally, de Menezes et al. (2022) reported that the biodiesel viscosity increased with oxidation time, reaching a maximum at the same time as the acid values. In chicken fat–based biodiesel stored at elevated temperatures, prolonged oxidation leads to increased viscosity due to the accumulation of high-molecular-weight compounds.

The increase in density observed in chicken fat and its biodiesel after $FeCl_3$-induced oxidation is associated with the formation of intermediate compounds, such as peroxides, aldehydes, and polymers resulting from oxidative degradation, which leads to the accumulation of medium- and high-molecular-weight species in the liquid system (Silva et al., 2025). de Menezes et al. (2022) reported that oxidation of chicken fat-based biodiesel leads to the formation of hydroperoxides and polymers, which, with prolonged oxidative exposure, increase both density and viscosity. Furthermore, Masudi et al. (2022) noted that the presence of transition metals, such as iron, not only accelerates oxidation but also promotes polycondensation, thereby increasing the fuel density owing to a higher average molecular weight. Spacino et al. (2023) found that Fe^{3+} ions exhibited the highest prooxidant activity among the metals (Co^{2+} and Cr^{3+}), promoting the degradation of methyl esters into carboxylates and heavy resinous compounds.

Table 1 indicates that chicken fat biodiesel after oxidation with $FeCl_3$ no longer meets the biodiesel quality standards. The acid value of the oxidized biodiesel (0.89 mg KOH/g) exceeded the limits set by ASTM D6751 and SNI 7182:2015 (≤ 0.50 mg KOH/g), while the viscosity (6.92 mm^2/s) surpassed the maximum allowable value of 6.0 mm^2/s specified by both standards (Adamu et al., 2025; Rachmadona et al., 2023). Although the peroxide value is not a criterion in ASTM D6751 and SNI 7182:2015 (where oxidative stability is evaluated by the induction period), it can serve as an early indicator of biodiesel quality deterioration. This finding suggests that exposure to iron significantly affects biodiesel quality by facilitating oxidative reactions.

All measured parameters exhibited shifts indicative of oxidative progression, with PV and AV displaying the most pronounced increases, especially in CF-BD samples. In contrast, IV declined, while viscosity and density experienced modest rises. These observed trends are consistent with recent literature regarding metal-catalyzed biodiesel aging, thereby reinforcing the current results (Fazal et al., 2022; Sui et al., 2021; Song et al., 2023). Most importantly, the findings directly address the study objective by confirming that Fe^{3+} ions function as an effective pro-oxidant, enabling rapid and practical screening of biodiesel degradation under accelerated oxidative conditions.

The appearance of chicken fat before oxidation (CF) and after oxidation (CFO), as well as chicken fat biodiesel before oxidation (CF-BD) and after oxidation (CF-BD-Ox), is shown in **Figure 1**.

Figure 1 illustrates the distinct visual changes observed in chicken fat and its transesterified biodiesel before and after the oxidation process.

Figure 1. The appearance of chicken fat before oxidation (CF) and after oxidation (CFO), as well as chicken fat biodiesel before oxidation (CF-BD) and after oxidation (CF-BD-Ox)

The chicken fat before oxidation (CF) was bright yellow. In contrast, after oxidation (CFO), the color became more turbid and slightly pale, indicative of the formation of oxidation products, such as peroxides and aldehydes. Biodiesel derived from chicken fat before oxidation (CF-BD) appeared clear with a yellowish-green hue; however, following oxidation (CF-BD-Ox), the color darkened to a brownish shade, and sediment formed at the bottom of the bottle. This suggests the formation of polymeric compounds via advanced oxidation and methyl ester degradation. Such color darkening and turbidity serve as visual indicators of oxidative reactions and quality deterioration in both fat and biodiesel, consistent with the findings of Lopresto et al. (2024) and Matbouei et al. (2020), who reported that oxidation induces a change from yellow to brown, accompanied by sediment due to heavy compound and polymer derivative formation. The infrared spectra of chicken fat and its biodiesel before and after oxidation with $FeCl_3$ are shown in **Figures 2** and **3**, respectively.

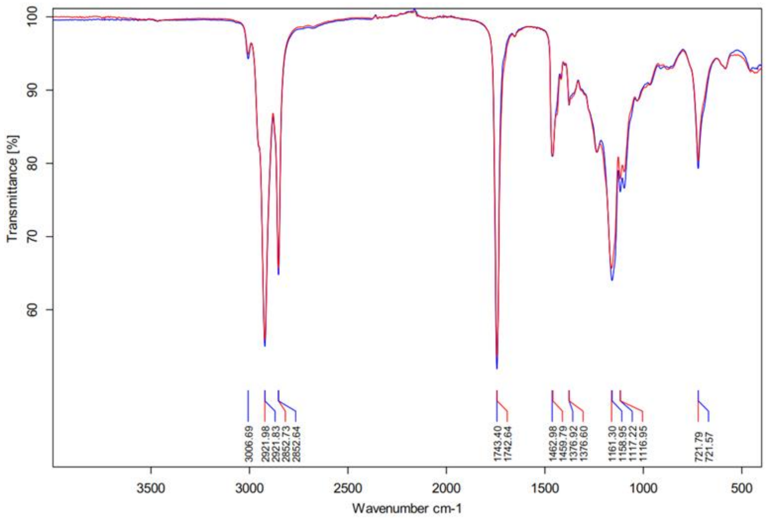

Figure 2. The infrared spectra of chicken fat before (CF, blue) and after oxidation with FeCl3 (CFO, red)

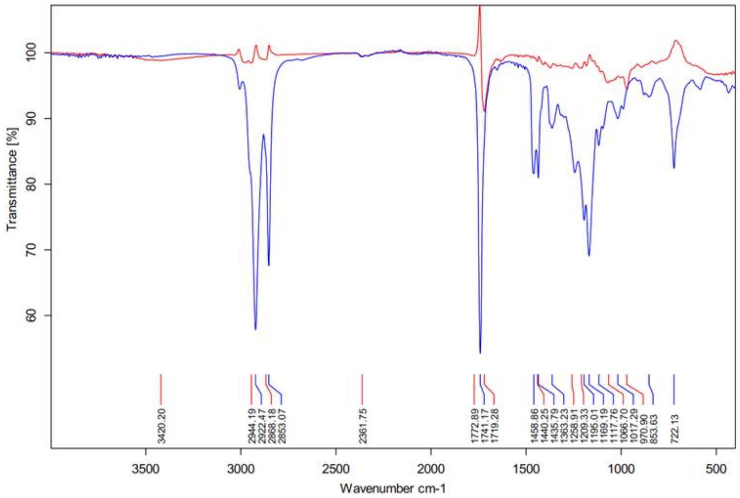

Figure 3. The appearance of chicken fat biodiesel before oxidation (CF-BD, blue) and after oxidation (CF-BD-Ox, red)

Spectra FTIR from **Figure 2** show that the peaks for chicken fat before (CF, blue) and after (CFO, red) oxidation both exhibit strong signals in the range ≈3006–2852 cm^{-1}, which are associated with the stretching vibrations of =C–H (cis) and aliphatic C–H bonds. The prominent peak at ≈1743 cm^{-1} corresponds to the carbonyl (C=O) stretching of esters, while the band around 1160–1118 cm^{-1} indicates C–O stretching in the ester groups (Narvaez et al., 2023). After oxidation, only slight decreases in intensity were observed in these regions, implying the formation of oxidation products.

Figure 3 shows the FTIR spectra of CF-BD (blue) and CF-BD-Ox (red), which reveal the characteristic ester peaks and modifications due to oxidation. The aliphatic C–H stretching peaks at ≈2922 and ≈2853 cm^{-1} show a marked decrease in intensity upon oxidation in CF-BD-Ox, indicating the consumption of saturated and unsaturated C–H bonds, respectively. The peak at ≈3006 cm^{-1}, attributed to =C–H (cis), also diminishes, signifying the oxidation of the double bonds. The sharpest ester carbonyl peak at ≈1740–1744 cm^{-1} reduces in intensity and shifts to ≈1719 cm^{-1} in CF-BD-Ox, consistent with the oxidation-induced alterations in ester functionalities. Furthermore, the peaks at ≈1465 and ≈1378 cm^{-1}, associated with CH_2/CH_3 bending vibrations, and the C–O–C stretching at ≈1240–1160 cm^{-1}, are significantly diminished, reflecting the formation of oxidized species, such as peroxides, alcohols, and carbonyl compounds.

A weak peak at ≈3400 cm^{-1} suggests the presence of O–H groups from hydroperoxides, while a peak at ≈722 cm^{-1} indicates aliphatic chain rocking vibrations (Chakarova et al., 2022; Abdiyev et al., 2025). Overall, the substantial reductions and shifts in specific IR bands support the idea that Fe^{3+} converts unsaturated bonds into primary (ROOH) and secondary oxidation products, such as aldehydes and ketones. These observations align with the findings of de Menezes et al. (2022) and Lau et al. (2024), who noted that shifts in carbonyl peaks and decreases in C–H intensities serve as early indicators of oxidation in animal fats and biodiesel.

From a practical standpoint, the rapid and dramatic deterioration of chicken fat biodiesel quality under $FeCl_3$ oxidation underscores the critical importance of metal-free handling and storage protocols in biodiesel supply chains. Iron contamination, from processing equipment, storage tanks, or fuel distribution systems, can severely compromise fuel stability and engine performance within days rather than months. The $FeCl_3$ method presented here offers a cost-effective, rapid alternative to traditional Rancimat testing for evaluating antioxidant protection and predicting storage life under realistic contamination scenarios.

Conclusions

This study demonstrates that $FeCl_3$-catalyzed oxidation significantly deteriorates the quality of biodiesel from chicken fat by increasing acid and peroxide values, reducing the iodine value, and raising viscosity and density through the formation of polymeric compounds and oxidation byproducts. Spectroscopic analysis confirmed the degradation of ester and aliphatic functional groups. Critically, oxidised CF-BD failed to meet ASTM D6751 and SNI 7182:2015 standards for acid value and viscosity, highlighting the adverse impact of metal contamination. These results validate $FeCl_3$ oxidation as a practical accelerated-degradation model for rapid stability screening. However, limitations include the use of a single iron source and relatively short oxidation duration; future work should examine multiple metal contaminants (copper, cobalt), explore synergistic effects with oxygen and light, and evaluate antioxidant protective efficacy under $FeCl_3$ challenge across broader temperature ranges. These investigations will strengthen biodiesel formulation strategies and support the development of improved storage and distribution protocols for animal-fat–based fuels.

Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this article. This research was conducted independently, without any commercial or financial relationships that could be perceived as potential conflicts of interest.

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