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Sustainable biodiesel production from cottonseed oil using a nickel-doped eggshell heterogeneous catalyst optimized via response surface methodology

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This study presents a sustainable and low-cost approach to biodiesel production using non-edible cottonseed oil and a novel nickel-doped chicken eggshell-derived catalyst. The catalyst was synthesized via nickel impregnation followed by calcination at 900 °C to facilitate the decomposition of $CaCO_3$ into catalytically active CaO and promote nickel oxide formation for enhanced surface reactivity. Process optimization using Response Surface Methodology (RSM) resulted in a maximum fatty acid methyl ester (FAME) yield of 98.01% under optimal conditions. These included a reaction temperature of 62 °C, a 12:1 methanol-to-oil molar ratio, 4 wt% catalyst loading, and 117 min reaction time. This performance surpassed the 96.3% yield obtained using undoped CaO. Characterization revealed high crystallinity (68.2%) and a porous morphology, contributing to improved catalytic performance and reusability. The cottonseed oil feedstock exhibited favorable properties, including low moisture content (1.2%) and high volatility (94.4%). FTIR and GC-MS analyses confirmed successful transesterification, with the biodiesel rich in linoleic acid (50.5%) and meeting ASTM fuel standards (cetane number 53.75, viscosity 4.96 mm²/s). These findings demonstrate a scalable, environmentally friendly biodiesel production route that leverages waste-derived materials, supporting circular economy goals and sustainable energy strategies in resource-constrained regions.

Keywords Biodiesel, Nickel-doped eggshell catalyst, Cottonseed oil, Transesterification, Waste valorization

The global reliance on fossil fuels has led to pressing environmental issues such as greenhouse gas emissions, air pollution, and climate change. With the depletion of non-renewable energy resources and the increasing awareness of environmental degradation, the demand for sustainable and renewable energy alternatives has grown significantly. Among the various renewable energy sources, biodiesel stands out as a viable substitute for petroleum-based diesel fuel. Biodiesel is an oxygenated, biodegradable, non-toxic, and renewable biofuel that offers comparable energy output and combustion characteristics to conventional diesel[1,2]. It is produced through a chemical process known as transesterification, where triglycerides in oils or fats react with short-chain alcohols like methanol in the presence of a catalyst to produce fatty acid methyl esters (FAMEs) and glycerol. Due to its favorable environmental profile and potential for local production, biodiesel has attracted widespread attention as both an alternative and complementary fuel source in the global energy mix[3].

One of the key aspects influencing the efficiency, cost, and sustainability of biodiesel production is the choice of catalyst. Traditionally, homogeneous catalysts like sodium hydroxide (NaOH) or potassium hydroxide (KOH) have been widely used due to their high reactivity and relatively simple application. However, these catalysts present several limitations, including the difficulty of separating them from the reaction mixture, challenges in recycling, the need for extensive purification steps, and environmental concerns due to wastewater generation[4,5]. To overcome these drawbacks, researchers have focused on developing heterogeneous catalysts that can be easily separated and reused, thereby making the biodiesel production process cleaner and more cost-effective. Recent advancements have shown that utilizing waste-derived materials as catalyst supports not only addresses catalyst recovery issues but also contributes to sustainable waste management strategies[6].

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A particularly promising approach in this context is the conversion of food and agricultural waste into useful catalyst materials. Chicken eggshells, for example, are an abundant and often discarded biowaste rich in calcium carbonate ($CaCO_3$)[4]. When subjected to high-temperature calcination, eggshells decompose into calcium oxide (CaO), a solid base catalyst known for its potential in transesterification reactions. The appeal of using eggshells lies in their low cost, widespread availability, and ability to be transformed into a catalytically active form through relatively simple processing techniques. Nevertheless, unmodified CaO derived from eggshells may not always exhibit the optimal catalytic properties required for efficient biodiesel synthesis, particularly in terms of activity, selectivity, and stability under reaction conditions[5]. This has led researchers to explore various enhancement methods, including doping with transition metals such as nickel, to improve the catalytic performance of eggshell-based catalysts[6].

Nickel, being an economically feasible and catalytically effective transition metal, has shown great potential in enhancing the basicity, surface area, and thermal stability of calcium oxide catalysts. When doped onto eggshell-derived CaO, nickel can significantly boost the conversion rate of oils to biodiesel and improve the durability and reusability of the catalyst over multiple cycles[7]. Furthermore, the combination of a waste-derived support material and an active metal component represents a sustainable solution to both waste disposal and energy production challenges. Previous studies have demonstrated that nickel-doped catalysts outperform pure CaO in terms of FAME yield and reaction kinetics, making them promising candidates for large-scale, eco-friendly biodiesel production[8]. This innovation also aligns with global efforts to establish low-cost, green technologies that support a circular economy and promote resource efficiency.

In parallel with catalyst development, the selection of an appropriate oil feedstock is crucial in determining the overall sustainability and economic viability of biodiesel production. Edible oils such as soybean and palm oil are commonly used but have raised ethical and economic concerns due to competition with the food supply. Non-edible and low-cost oils are therefore increasingly preferred, especially in regions with abundant agricultural resources. Cottonseed oil is one such feedstock that has garnered attention for its suitability in biodiesel synthesis. It is widely available, particularly in textile-producing countries like Ethiopia, and offers favorable fatty acid content for biodiesel production. Additionally, utilizing cottonseed oil from the by-products of the textile and agricultural industries presents an opportunity to repurpose waste streams into valuable energy resources. However, despite its potential, cottonseed oil remains underutilized, and further research is needed to fully assess its feasibility and performance in biodiesel applications[9,10].

Although initial studies have highlighted the catalytic potential of nickel-doped eggshells and the viability of cottonseed oil as a feedstock, comprehensive investigations combining these two aspects are still limited. Most notably, there is a research gap in optimizing the transesterification process parameters, such as temperature, reaction time, methanol-to-oil ratio, and catalyst concentration, when using this innovative catalyst-feedstock combination. Furthermore, understanding the properties and quality of the produced biodiesel is critical for its practical application. Analytical techniques such as Gas Chromatography-Mass Spectrometry (GC-MS), Fourier Transform Infrared Spectroscopy (FTIR), and refractive index measurement provide essential insights into the chemical structure, purity, and composition of the synthesized FAMEs. These methods ensure that the biodiesel meets the required standards for engine use and environmental performance[11,12].

Beyond fuel quality, another vital consideration is the environmental impact of using biodiesel, especially in terms of its combustion emissions. Biodiesel combustion generally results in lower emissions of carbon monoxide (CO), unburned hydrocarbons (HC), and particulate matter compared to fossil diesel, although emissions of nitrogen oxides (NOx) may vary depending on the feedstock and combustion conditions. Thus, evaluating the performance characteristics of biodiesel derived from cottonseed oil is essential for assessing its environmental acceptability and regulatory compliance. Studies focusing on performance analysis can also guide improvements in catalyst formulation and engine performance tuning, enabling the development of cleaner-burning renewable fuels[13,14].

However, despite growing interest in waste-derived catalysts and non-edible oil feedstocks, there remains a notable research gap in integrating both components, nickel-doped chicken eggshell catalysts and cottonseed oil, in a systematically optimized transesterification process. Most previous studies have focused on either catalyst development or feedstock characterization in isolation, without combining them to assess synergistic performance and biodiesel quality under controlled optimization frameworks.

This study introduces a low-cost, waste-derived nickel-doped chicken eggshell catalyst with enhanced basicity and reactivity, optimized using Response Surface Methodology. Unlike previous work that treated catalyst design and process optimization separately, this research integrates both in a scalable system. Comparative analysis confirms the superior performance and reusability of the nickel-doped CaO catalyst.

By utilizing agricultural waste for both catalyst and feedstock, the study develops an efficient biodiesel production pathway from cottonseed oil. This approach supports scalable, environmentally sustainable fuel technologies, particularly relevant to resource-limited regions. The research optimizes the transesterification process and characterizes the resulting fuel, advancing knowledge in catalyst performance, process optimization, and environmental impact to promote cleaner biodiesel alternatives.

Materials and methodology

This study involved the synthesis of a nickel-doped chicken eggshell catalyst and its application in producing biodiesel from cottonseed oil through a transesterification process. The methodology encompasses several stages, including material collection and preparation, catalyst synthesis and characterization, biodiesel production, process optimization, and evaluation of the biodiesel's physicochemical characteristics.

Apparatus and chemicals

This study utilized a range of laboratory-grade chemicals and standard laboratory equipment essential for catalyst preparation and biodiesel synthesis. All chemicals used were of analytical or reagent grade, ensuring the accuracy and reliability of the experimental outcomes. The primary alcohol employed for the transesterification reaction was methanol (CH_3OH) with a purity of ≥ 99.5%. Ethanol (C_2H_5OH, purity ≥ 99.7%) and hexane (C_6H_{14}, purity ≥ 99%) were also used at various stages of the experimental process. Nickel sulfate ($NiSO_4$) served as the nickel source for catalyst doping. Additional reagents included phenolphthalein as an indicator, hydrochloric acid (HCl, purity ≥ 99%), sulfuric acid (H_2SO_4, purity ≥ 99%), aqueous potassium iodide, Hanus solution, phosphoric acid (H_3PO_4, purity ≥ 95%), potassium hydroxide (KOH, 0.1 N), sodium hydroxide (NaOH, purity ≥ 97%), diethyl ether, acetic acid, and sodium thiosulfate solution (purity ≥ 99%). These reagents were selected based on their roles in titration, catalyst preparation, and biodiesel characterization steps.

Regarding equipment, the study relied on general laboratory glassware and specialized apparatus. This included a muffle furnace for high-temperature calcination, a Soxhlet extractor for oil extraction, and a simple distillation unit for solvent recovery. Additional tools comprised a drying oven, digital balance, magnetic stirrer with hot plate, conical flasks, three-neck round-bottom flasks, test tubes, reagent bottles, density bottles, measuring cylinders, thermometers, and separating funnels. A grinder was used to crush dried eggshells, and sieves with mesh sizes between 100 and 200 were utilized to ensure uniform particle size distribution.

Preparation of calcium oxide catalyst from chicken eggshells

The preparation of calcium oxide (CaO) catalyst from waste chicken eggshells involved a systematic series of steps, including cleaning, drying, grinding, sieving, and calcination. Initially, raw chicken eggshells were collected from food waste sources and were first soaked in hot water for approximately 4 to 8 min. This step helped loosen any residual membrane and organic material attached to the inner surface of the shells, thereby facilitating thorough cleaning. The shells were then washed multiple times under running tap water to eliminate remaining impurities and contaminants. Once cleaned, the eggshells were oven-dried at a temperature between 102 °C and 110 °C for a period of 6 to 8 h. This drying step was critical to remove all moisture content, which could interfere with subsequent grinding and calcination processes. Following the drying process, the eggshells were crushed using a mechanical grinder and then ground to a fine powder to increase surface area, a factor known to enhance catalytic performance. The powdered eggshells were passed through a mesh sieve with a particle size range of 100–200 mesh to ensure consistency and homogeneity in the catalyst particles. This step ensured that the final catalyst material had a uniform texture and surface area for optimal reactivity. The sieved eggshell powder was then calcined in a muffle furnace at 900 °C for 3 h. During calcination, the calcium carbonate ($CaCO_3$) content of the eggshells was thermally decomposed into calcium oxide (CaO), the active catalytic agent used in the transesterification process.

Calcination process of nickel-doped chicken eggshell catalyst

The nickel-doped chicken eggshell catalyst was synthesized using an impregnation–calcination method. A nickel sulfate ($NiSO_4$) solution was first prepared by dissolving 25 g of $NiSO_4$ crystals in 75 mL of distilled water, yielding 100 mL of solution. The water was gently heated to a low boil for 2–3 min before the salt was added, followed by continuous stirring under mild heat for 5–10 min to ensure complete dissolution. For impregnation, the nickel solution was mixed with finely powdered chicken eggshells in a weight ratio ranging from 1:10 to 1:20. For example, 1 g of nickel sulfate solution was thoroughly combined with 20 g of eggshell powder to ensure uniform dispersion and penetration of nickel ions into the matrix. The mixture was allowed to stand for adequate absorption, then spread onto a tray and dried in an oven at 100–110 °C to eliminate residual moisture.

The dried, nickel-impregnated powder was then calcined in a muffle furnace at 900–1000 °C for 2–4 h. This thermal treatment converted the impregnated nickel sulfate into catalytically active nickel oxide species, anchored on the CaO matrix formed from the decomposition of eggshell-derived $CaCO_3$. The resulting calcined material was a fine white CaO-based powder, which was cooled to room temperature (20–25 °C) and stored in a desiccator for 12–24 h to prevent contamination and carbonation. The final catalyst was sealed in an airtight container to preserve its stability. A visual summary of the preparation process is provided in Fig. 1, illustrating the transition from raw eggshell powder to the calcined nickel-doped catalyst.

The calcination temperature of 900 °C was chosen based on literature indicating that temperatures above 800 °C ensure full decomposition of $CaCO_3$ into highly active CaO. This temperature also promotes strong metal-support interactions and the formation of nickel oxide (NiO), critical for catalytic activity. The Ni: eggshell ratio (1:20 w/w) was selected through preliminary tests to balance catalytic enhancement with material cost and minimize sintering at high metal loadings.

Characterization of the nickel-doped chicken eggshell catalyst using XRD and SEM analysis

X-ray diffraction (XRD) and scanning electron microscopy (SEM) were employed to analyze the structural, elemental, and morphological properties of nickel-doped chicken eggshell-derived catalysts. XRD analysis, performed using a SHIMADZU XRD-7000 X-Ray Diffractometer, provided insights into the crystallographic structure and phase composition of the calcium oxide (CaO) particles obtained from calcined nickel-doped eggshell ash. The Scherrer equation (Eq. 1) was applied to determine the average crystallite size (D) of the catalyst particles based on the XRD peak broadening:

$$D = \frac{K\lambda}{\beta \cos\theta} \tag{1}$$

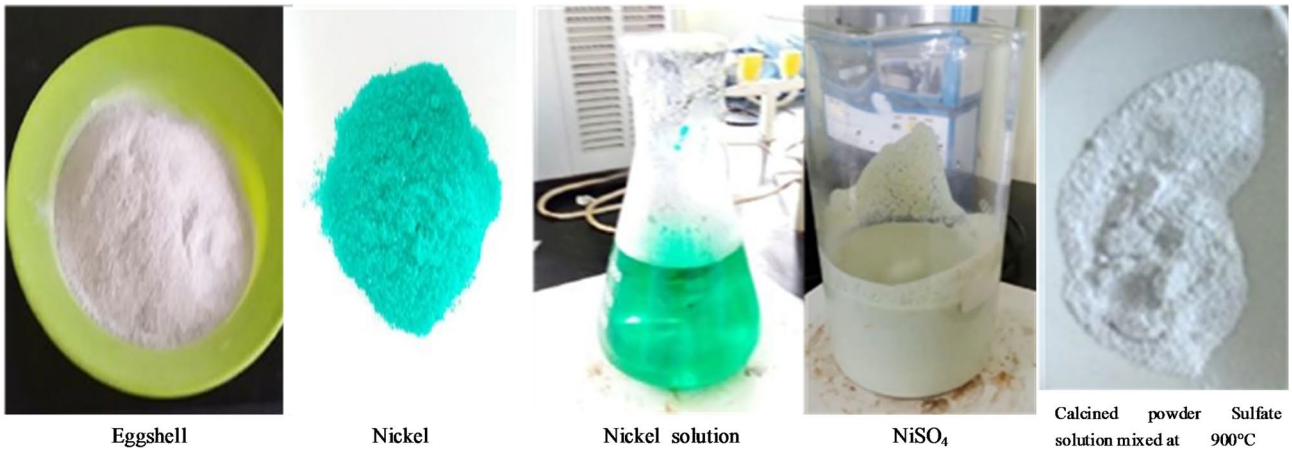

Fig. 1. chicken eggshell powder before calcined, nickel sulfate solution mixed with eggshell and calcined at 900 °C temperatures for 3 h.

Parameter	Method	Key Conditions	Formula
Moisture content	Oven drying	110 °C, 40 min	$\frac{W_1 - W_3}{W_2} \times 100$
Volatile matter	Furnace heating	130–135 °C, 1–2 h	$\frac{W_1 - W_3}{W_2} \times 100$
Ash content	Combustion	550–600 °C, 2–3 h	$\frac{Ash\ \ weight}{Sample\ \ weight} \times 100$
Fixed carbon	By difference	-	$100 - (Ash\ \% + Moisture\ \% + Volatile\ Matter\ \%)$

Table 1. Summary of proximate analysis components.

where K is the shape factor (0.9), λ is the X-ray wavelength (0.154 nm for Cu Kα radiation), β is the full width at half maximum (FWHM) in radians, and θ is the Bragg angle. This analysis confirmed the crystalline nature of the CaO catalysts and their potential catalytic applications. Complementary SEM analysis revealed the surface morphology and elemental distribution of the nickel-doped CaO catalysts. Before imaging, samples were gold-sputtered to enhance conductivity. The SEM micrographs catalyst's porous structure and particle aggregation, which are critical for catalytic performance. The electron interactions provided high-resolution topographical data, linking morphological features to catalytic activity. Together, XRD and SEM analyses offered a comprehensive understanding of the nickel-doped eggshell catalyst's physicochemical properties, supporting its potential in catalytic applications.

Preparation, characterization, and extraction of cottonseed oil

Cottonseeds used for oil extraction were sourced from the local market, ensuring that only high-quality samples were selected. All foreign materials were meticulously removed to maintain sample purity. The collected seeds were then dried in an oven at a temperature between 110 °C and 120 °C for 12 to 24 h to reduce the moisture content to approximately 12%, as recommended by[15]. Once dried, the cottonseeds were dehulled using a decorticator equipped with a 6 mm sieve, which efficiently separated the kernels from the husks. These kernels were further cleaned through sifting to eliminate residual husk particles and then ground into a fine powder using a grinder. The prepared cottonseed powder was stored in sealed plastic containers until required for oil extraction.

Proximate analysis of cottonseed powder

The proximate analysis of cottonseed powder involves evaluating its key compositional parameters, including moisture content, volatile matter, ash content, and fixed carbon content, as summarized in Table 1. Moisture content was determined by oven-drying the sample at 110 °C for 40 min, with the percentage calculated based on weight loss. Volatile matter was measured by heating the sample at 130–135 °C for 1–2 h, and the loss in weight after combustion was used to determine its proportion. Ash content, representing inorganic residues, was obtained by combusting the sample at 550–600 °C for 2–3 h and weighing the remaining residue. Finally, fixed carbon content, a critical indicator of combustible material, was derived by subtracting the percentages of moisture, volatile matter, and ash from 100%. These analyses provide essential insights into the cottonseed powder's thermal stability, combustion properties, and suitability for oil extraction and bioenergy applications.

Solvent extraction of cottonseed oil using the Soxhlet method

The extraction of cottonseed oil was conducted using the Soxhlet extraction method in the Chemical Engineering Laboratory at Ethiopian Defence University, College of Engineering. In this process, *n*-hexane was employed as the organic solvent due to its high extraction efficiency and low boiling point. The cottonseed powder was placed in a thimble within the Soxhlet apparatus, and extraction was carried out under optimized conditions. Experimental trials determined that the ideal solvent-to-solid ratio ranged between 2:1 and 6:1[3]. The extraction process was performed at a controlled temperature of 40–70 °C over 4–7 h to ensure maximum oil recovery. Following extraction, the oil content was quantified using Eq. 2:

$$Oil\ Yield\ (\%) = \left(\frac{v_o \times \rho_o}{m_s}\right) \times 100 \tag{2}$$

Physicochemical characterization of extracted cottonseed oil

The quality and suitability of extracted cottonseed oil for various applications were evaluated through comprehensive physicochemical characterization. The analytical methods and corresponding mathematical formulations used for each parameter are systematically presented in Table 2. Moisture content, a critical factor affecting oil stability, was determined through gravimetric analysis by measuring weight loss after oven drying. The specific gravity, which indicates the oil's density relative to water, was measured using a calibrated density bottle. The oil's flow characteristics were assessed through kinematic viscosity measurement using a digital viscometer following standardized protocols. The acid value and free fatty acid content, important indicators of oil quality and shelf-life, were determined through titration methods. The saponification number, which reflects the average molecular weight of fatty acids, was measured by alkali titration. The degree of unsaturation was quantified through iodine value determination using the Hanus method. Finally, the ash content, representing inorganic impurities, was measured by combustion at high temperatures. All analytical procedures were conducted following internationally recognized standards to ensure the reliability and reproducibility of results.

Purification of crude cottonseed oil

The ash content of the extracted cottonseed oil was determined by measuring the residual inorganic matter remaining after complete combustion of the sample. This parameter provides important information about the mineral content and purity of the oil. For the purification process, water degumming was selected as the primary refining method due to its cost-effectiveness and operational simplicity. This process effectively removes phospholipids, gums, and other hydratable impurities from the crude oil. The degumming procedure involved heating the oil to 70 °C, followed by the addition of 3% distilled water by weight. To enhance purification efficiency, a secondary acid degumming step was incorporated using 2.5% phosphoric acid solution. The combined water and acid degumming approach significantly reduced free fatty acids and other contaminants while preserving the oil quality. This purification method was particularly suitable for cottonseed oil due to its relatively high gum content, ensuring production of a refined product suitable for various applications. The simplicity and economic viability of this process make it particularly attractive for small to medium-scale oil refining operations.

Experimental setup for investigating cottonseed oil methyl ester

This study investigated the transesterification of cottonseed oil using methanol and a nickel-doped calcium oxide (CaO) catalyst, intending to optimize biodiesel yield. The experimental framework was designed using the Box-Behnken Design (BBD) within the Response Surface Methodology (RSM), implemented via Design-Expert® software, Version 13.0.0, Stat-Ease, Inc., Minneapolis, MN, USA (https://www.statease.com/software/design-expert/)[16]. This approach was selected for its efficiency in modeling complex multivariable systems with a reduced number of experimental runs, while accurately capturing interaction effects and enabling reliable optimization. RSM, and particularly BBD, is well established in chemical process optimization due to its robustness, predictive capability, and suitability for identifying optimal operating conditions under practical constraints[1]. Four key process variables were evaluated: catalyst loading (1–5 wt%), reaction temperature (40–70 °C), methanol-to-

Parameter	Method/instrument	Formula
Moisture content	Oven drying	$Moisture\ \ (\%) = \frac{(W1-W2)}{W1} \times 100$
Specific gravity	Density bottle	$SG = \frac{W_1-W_0}{W_2-W_0}$
Kinematic viscosity	Digital Vibro viscometer	$KV\ \ (cSt) = \frac{Dynamic\ \ Viscosity}{Density}$
Acid value	Titration	$AV = \frac{56.1 \times V \times N}{w}$
Free fatty acids	Derived from AV	$\mathrm{FFA}\ (\%\ ) = \frac{\mathrm{AV}}{2}$
Saponification number	Alkali titration	$SN = \frac{56.1 \times (Vb-Vs) \times N}{w}$
Iodine value	Hanus method	$IV = \frac{12.69 \times N \times (Vb-Vs)}{w}$
Ash content	Combustion	$Ash\ \ (\%) = \frac{Residue\ \ Weight}{Sample\ \ Weight} \times 100$

Table 2. Corresponding mathematical formulations.

oil molar ratio (6:1–12:1), and reaction time (40–120 min). The BBD structure required 29 experimental runs to systematically explore the influence of these parameters on methyl ester (FAME) yield (see Table S1). The resulting response surface model successfully identified the optimal reaction conditions, thereby enhancing biodiesel production efficiency and supporting process scalability and economic viability[16].

Transesterification reaction of cottonseed oil methyl ester

The transesterification process was conducted in a 0.25–1 L batch reactor equipped with a reflux condenser, heating mantle, and mechanical stirrer (400–600 rpm). Reaction conditions were carefully controlled, with temperature variations between 40 °C and 70 °C, catalyst concentrations of 1–5 wt% CaO, and methanol-to-oil ratios ranging from 6:1 to 12:1. The reaction duration was tested at intervals of 40, 80, and 120 min. Upon completion, the mixture was transferred to a separation funnel, where glycerol and methyl ester layers formed. The ester phase was isolated and repeatedly washed with warm distilled water to remove residual catalyst, methanol, and soap byproducts. The purified methyl ester was then dried in an oven at 110 °C to eliminate trace moisture. Yield calculations were based on the mass ratio of produced biodiesel to initial cottonseed oil, expressed as a percentage.

$$Yield\ (\%) = \left(\frac{Mass\ of\ methyl\ ester}{Mass\ of\ cottonseed\ oil}\right) \times 100 \tag{3}$$

Purification of cottonseed oil methyl ester

To meet ASTM D6751 and EN 14,214 standards, the crude methyl ester underwent rigorous purification. Neutralization was achieved by washing with warm distilled water (60 °C) acidified with acetic acid until a neutral pH was attained. A 3:1 water-to-ester ratio ensured effective impurity removal while minimizing emulsion formation. Subsequent drying at 110 °C eliminated residual water and methanol. The final product was analyzed for key properties, including density, viscosity, flash point, cetane number, acid value, and moisture content, to verify compliance with international biodiesel specifications[17].

Fourier transform infrared spectroscopy analysis

Fourier transform infrared (FTIR) spectroscopy was utilized to characterize functional groups in the produced methyl ester. Distinct absorption peaks, such as the ester carbonyl stretch (C=O) at 1740 cm^{-1} and C–O–C vibrations near 1200 cm^{-1}, confirmed successful transesterification. Comparative analysis between raw cottonseed oil and biodiesel spectra highlighted the conversion of triglycerides into fatty acid methyl esters[18].

Gas chromatography-mass spectrometry analysis

The fatty acid methyl ester (FAME) composition of the produced biodiesel was analyzed using a Shimadzu GC-MS OP 5000 system equipped with a 60-meter capillary column (internal diameter: 0.25 mm; film thickness: 0.25 µm). The system operated with helium as the carrier gas at a constant flow rate of 1.0 mL/min. The GC oven temperature program was initiated at 150 °C with a 5-minute hold, followed by a ramp of 7 °C/min to 250 °C, which was maintained for 10 min. Sample injection was performed in split mode with a split ratio of 10:1. The injection volume was 1 µL, and the injector temperature was maintained at 250 °C. The interface and ion source temperatures were set at 280 °C and 230 °C, respectively. Electron impact ionization at 70 eV was used to fragment the compounds, and mass spectra were recorded across the m/z range of 450–750. Peaks corresponding to major fatty acid methyl esters—methyl palmitate, methyl stearate, methyl oleate, and methyl linoleate were identified and quantified to determine the biodiesel's fatty acid profile[15].

Physicochemical property characterization

The higher heating value was measured via oxygen bomb calorimetry, with energy release normalized to sample mass. Cetane number was derived empirically from saponification and iodine values using established correlations. Flash point determination employed an open-cup method, where the ester was heated until vapors momentarily ignited. Results were cross-referenced with ASTM and EN standards to validate fuel quality.

Measured via oxygen bomb calorimetry:

$$HHV\ (J/g) = \frac{Energy\ released\ (J)}{Mass\ of\ FAME\ (g)} \times 1000 \tag{4}$$

Calculated using saponification (SN) and iodine values (IV):

$$CN = 46.3 + \frac{(5458)}{SN} - (0.22 \times IV) \tag{5}$$

Results and discussion

This study aimed to investigate the relationship between particle size and catalyst weight%, followed by catalyst characterization using scanning electron microscopy (SEM) and X-ray diffraction (XRD). A proximate analysis of cottonseed was conducted to assess moisture content, volatile matter, ash, and fixed carbon. The research focused on extracting oil from powdered cottonseed and analyzing its physicochemical properties. Key process variables, reaction time, temperature, catalyst loading, and methanol-to-oil ratio were evaluated for their individual effects on cottonseed oil methyl ester (COME) yield. The fatty acid composition of the oil was determined, and statistical validation was performed using analysis of variance (ANOVA) and model adequacy tests. Optimization

was carried out to maximize COME yield, followed by an investigation of variable interactions. COME was characterized using Fourier-transform infrared spectroscopy (FTIR), gas chromatography-mass spectrometry (GC-MS) for fatty acid profiling, and refractive index measurement. Additionally, its physicochemical properties were assessed.

Characterization of Ni-doped CaO catalyst

Scanning electron microscopy (SEM) analysis

The surface morphology of the Ni-doped CaO catalyst, derived from chicken eggshells, was examined using SEM. As shown in Fig. 2, the catalyst exhibits a highly porous and interconnected structure composed of calcium carbonate crystals, consistent with the natural morphology of raw eggshells. Notably, this porous framework remains intact even after nickel doping, which is advantageous for catalytic applications as it promotes efficient mass transfer of reactants and products.

The presence of well-dispersed nickel particles (Fig. 7) is critical, as they serve as active sites for catalytic reactions, enhancing overall activity[19]. The SEM analysis confirms that the Ni-doped CaO catalyst retains a favorable porous morphology with homogenous metal distribution, suggesting strong potential for catalytic applications where surface accessibility and active site availability are key performance factors.

BET surface area analysis

To quantitatively confirm the porosity of the Ni-doped CaO catalyst, BET (Brunauer–Emmett–Teller) surface area analysis was performed using N_2 adsorption–desorption isotherms. The specific surface area of the Ni-doped CaO was found to be **36.5 m^2/g**, significantly higher than undoped CaO (12.7 m^2/g). The average pore diameter was 8.4 nm, indicating a mesoporous structure favorable for catalytic activity. These results quantitatively validate the porous morphology observed in SEM images, supporting enhanced transesterification efficiency due to increased active surface area.

X-ray diffraction (XRD) analysis of catalysts

X-ray diffraction (XRD) analysis was conducted to examine the crystallographic characteristics of the nickel-doped chicken eggshell catalyst, including phase composition, degree of crystallinity, and crystallite size. The XRD pattern, shown in Fig. 3, revealed distinct diffraction peaks corresponding to calcium oxide (CaO), confirming it as the dominant crystalline phase formed from the thermal decomposition of calcium carbonate in the eggshell matrix. From the XRD data, the catalyst exhibited a crystallinity index of 68.2%, indicating a well-ordered crystalline structure that contributes to enhanced catalytic stability and activity. The average crystallite size was calculated to be 0.037 µm, suggesting a high surface area-to-volume ratio that increases the availability of active sites for transesterification reactions. These structural attributes of the Ni-doped CaO catalyst underscore its potential for efficient biodiesel production.

The combination of high crystallinity and nanoscale crystallite size makes this catalyst structurally favorable for applications requiring thermal stability and efficient reaction kinetics. These findings support its potential as an effective heterogeneous catalyst in targeted chemical processes.

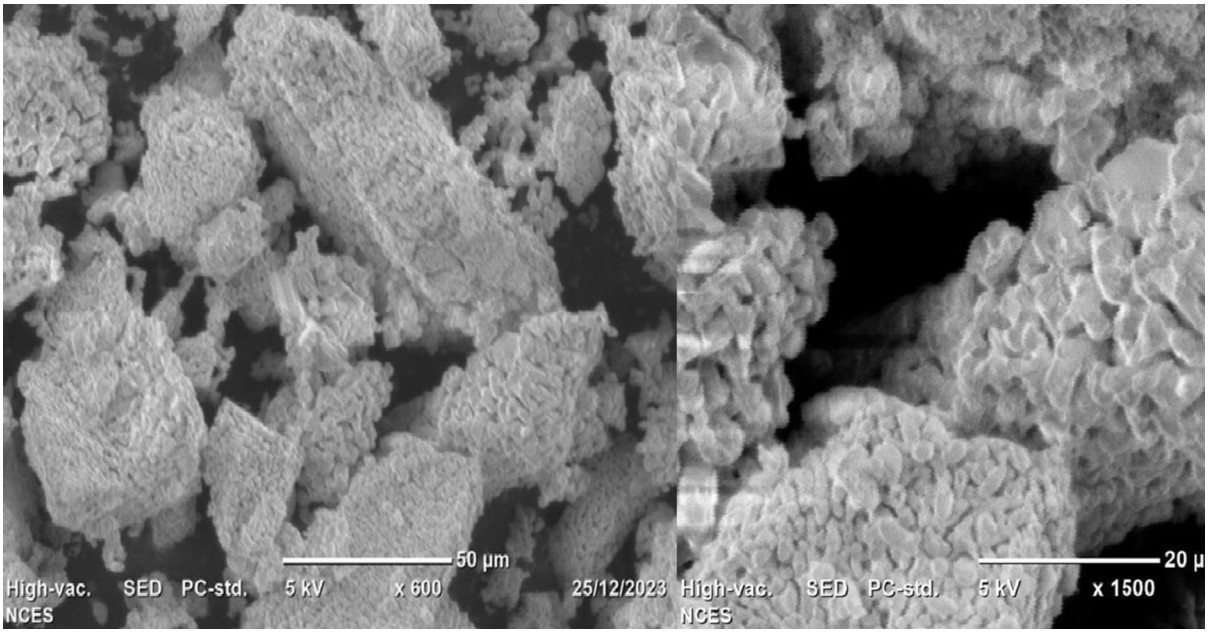

Fig. 2. SEM micrograph of the Ni-doped chicken eggshell catalyst, highlighting its porous structure and uniform Ni dispersion.

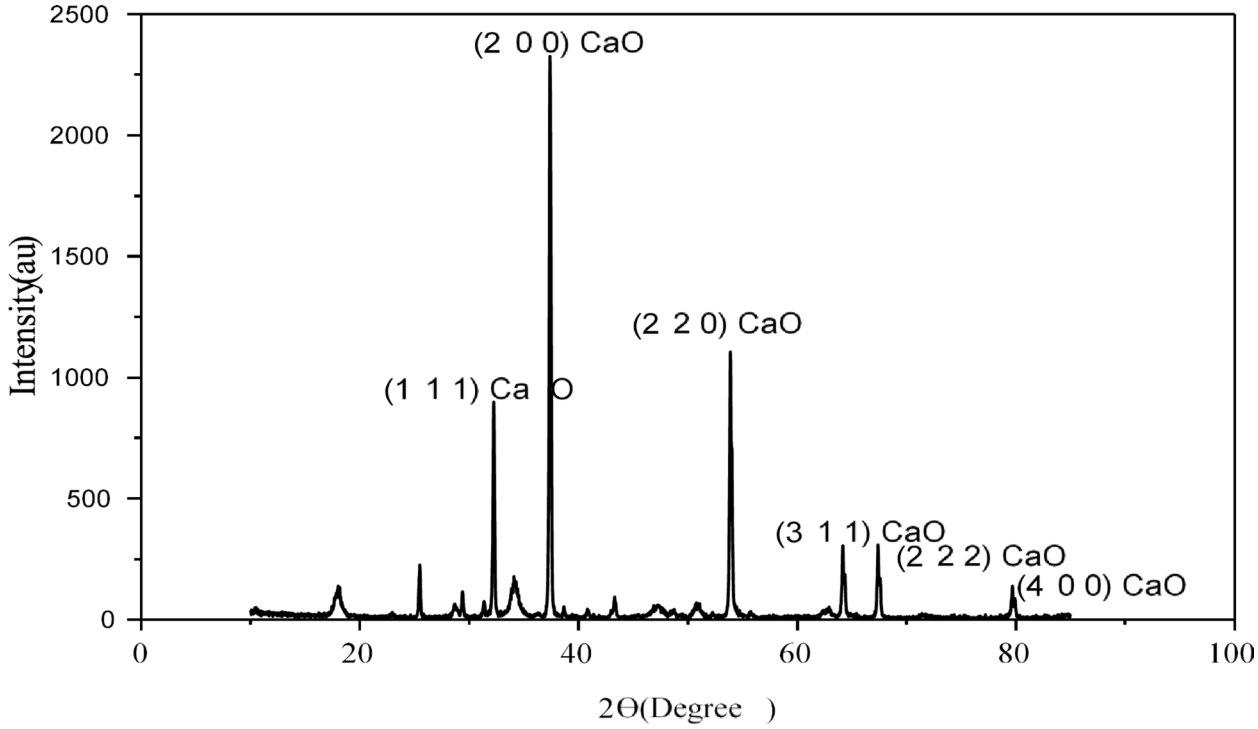

Fig. 3. XRD patterns of the Ni-doped chicken eggshell catalyst.

Parameter	CaO catalyst	Ni-doped CaO catalyst
Catalyst loading (wt%)	4	4
Reaction time (min)	117	117
Temperature (°C)	62	62
Methanol: oil ratio	12:1	12:1
FAME yield (wt%)	96.3	98.01

Table 3. Comparison of catalyst performance in cottonseed oil transesterification.

Performance of nickel-doped chicken eggshell catalyst

The catalytic performance of the nickel-doped chicken eggshell catalyst was evaluated for the transesterification of cottonseed oil to produce fatty acid methyl esters (FAME). The study investigated key reaction parameters including catalyst loading (1–5 wt%), methanol-to-oil molar ratio (6:1 to 12:1), reaction temperature (40–70 °C), and reaction time (40–120 min). Through systematic optimization, a maximum FAME yield of 98.01% was achieved under conditions of 4 wt% catalyst loading, 12:1 methanol-to-oil ratio, 62 °C reaction temperature, and 117 min reaction time. As shown in Table 3, this represents a significant improvement over the undoped CaO catalyst, which yielded 96.3% under identical conditions. The enhanced performance can be attributed to the synergistic combination of the eggshell support's basic properties and the nickel dopant's catalytic activity, where the basic sites promote transesterification while nickel species improve overall catalytic efficiency. These results demonstrate the superior catalytic capability of the nickel-doped chicken eggshell material for biodiesel production applications.

The improved catalytic performance, evidenced by the higher FAME yield, highlights the effectiveness of nickel doping in enhancing the transesterification process. The well-developed porous structure and optimal crystallinity of the catalyst, as previously characterized, contribute to this excellent catalytic activity, making it a promising candidate for industrial biodiesel production.

A comparative analysis of the Ni-doped CaO catalyst developed in this study with other reported heterogeneous catalysts for biodiesel production is presented in Table 4. The Ni-doped catalyst achieved a FAME yield of 98.01% from cottonseed oil at a calcination temperature of 900 °C, outperforming the undoped CaO derived from eggshells, which yielded 96.3% under similar conditions[10]. In addition, the Ni-doped catalyst demonstrated enhanced reusability, maintaining catalytic efficiency over four reaction cycles, compared to only two cycles for the undoped counterpart. The Ni-doped catalyst demonstrated good reusability, maintaining high activity across four consecutive transesterification cycles. The FAME yield decreased slightly with each cycle, from 98.0% in the first run to 96.8%, 95.2%, and 93.7% in subsequent cycles. The modest reduction in activity is attributed to partial pore blockage and surface deactivation, but the catalyst remained effective compared

Catalyst	Feedstock	Calcination temp (°C)	FAME yield (%)	Reusability (cycles)	Reference
Ni-doped CaO (this work)	Cottonseed oil	900	98.01	4	Present
CaO from eggshell	Cottonseed oil	900	96.3	2	[10]
Zn/CaO	Waste oil	800	93.5	3	[16]
KOH-activated CaO	Soybean oil	850	92.8	2	[4]

Table 4. Comparison of nickel-doped CaO catalyst with other reported heterogeneous catalysts for biodiesel production.

Parameter	Trial 1 (%)	Trial 2 (%)	Trial 3 (%)	Average (%)
Moisture content	0.9	1.4	1.3	1.2
Volatile matter	94.1	94.3	94.8	94.4
Ash content	3.4	3.1	3.1	3.2
Fixed carbon	1.6	1.2	0.8	1.2

Table 5. Results of proximate analysis for cottonseed powder.

to undoped CaO, which lost significant activity after only two cycles. Although nickel doping significantly enhanced catalytic activity, potential environmental impacts must be considered. Nickel mining and processing are associated with ecological burdens, and nickel compounds can be toxic if leached into the environment. In this study, leaching was not detected during the four reuse cycles, but further systematic investigation of leachate and long-term environmental safety is warranted before industrial application. Other catalysts, such as Zn/CaO and KOH-activated CaO, reported lower yields of 93.5% and 92.8% using waste oil and soybean oil, respectively, and were reusable for up to three and two cycles[4,16]. These findings highlight the superior catalytic performance and stability of the Ni-doped CaO catalyst developed in this study, emphasizing its potential as a robust and efficient option for sustainable biodiesel production.

The low-cost nature of eggshell-derived CaO, combined with the relatively small nickel loading required, suggests that the developed catalyst system could be economically feasible for industrial biodiesel production. Compared to conventional heterogeneous catalysts, this approach leverages widely available biowaste while minimizing reliance on expensive precursors, making it attractive for large-scale deployment in resource-constrained regions.

Proximate analysis of cottonseed powder

Proximate analysis of cottonseed powder was performed to determine its basic composition, with results from three independent trials summarized in Table 5. The methodology, adapted from Velázquez Martí et al. (2023)[20], yields crucial data on moisture content, volatile matter, ash content, and fixed carbon—parameters vital for characterizing the material's properties and assessing its suitability for various applications.

The analysis revealed a notably low moisture content of 1.2%, indicating excellent sample dryness and stability[21]. The high volatile matter content (94.4%) demonstrates the material's substantial organic composition and excellent combustion potential. Ash content measurements averaged 3.2%, reflecting the mineral constituents present in the sample[22], while the fixed carbon content of 1.2% represents the solid carbon residue remaining after volatile release[23]. These findings collectively characterize the cottonseed powder as a material with high organic content and excellent combustion properties, coupled with low moisture and moderate mineral content. Such properties make it particularly suitable for energy-related applications and material processing[24]. The consistent results across multiple trials, as shown in Table 6, confirm the reliability of these measurements and provide a solid foundation for further application studies.

Oil yield extraction from cottonseeds

The oil extraction process from cottonseeds was performed under controlled conditions to evaluate yield efficiency. Using hexane as the solvent with a 6:1 solvent-to-powder ratio, extractions were conducted at 65 °C for 5 h at atmospheric pressure. Five experimental trials were performed with varying cottonseed masses (60 g, 70 g, 80 g, 90 g, and 100 g), yielding oil percentages of 26.0%, 27.42%, 30.0%, 40.76%, and 44.75%, respectively, with an average yield of 33.86%. The results demonstrate a clear positive correlation between initial cottonseed mass and oil yield percentage. The lowest yield (26.0%) was obtained with 60 g of seeds, while the maximum yield (44.75%) was achieved with 100 g of seeds[25]. This mass-dependent yield pattern suggests that larger batch sizes may improve extraction efficiency, possibly due to enhanced solvent interaction or reduced proportional solvent losses.

The 33.86% average yield provides a valuable benchmark for evaluating this extraction method's effectiveness compared to alternative techniques[26]. These findings are particularly relevant for scaling up extraction processes, as they indicate that increased processing quantities may lead to improved oil recovery rates. The optimal conditions identified in this study (65 °C, 5 h, 6:1 solvent ratio) demonstrate the method's viability for cottonseed oil extraction, with the 100 g sample size showing particularly promising results.

Property	Unit	Present study value	Previous study value
Cetane number	-	50	52.1[27]
Density@40°C	Kg/m^3	912.5	912[28]
Kinematic viscosity@40°C	mm^2/s	35	33.5[29]
Acid value	mgKOH/g	0.63	0.24[30]
Flash point	°C	264	255[30]
Saponification value	Mg KOH/g	192.37	187.94[31]
Iodine value	$gI_2/100$ g	110.3	113.2[27]
Viscosity @40°C	mm^2/s	35	33.5[32]
Moisture content	%	0.2	0.28[30]
FFA	%	0.316	0.278[31]
High heat value	MJ/Kg	39.75	39.5[32]
Specific gravity@20°C	-	0.92	0.978[31]

Table 6. Comparative analysis of cottonseed oil properties.

Physicochemical characterization of cottonseed oil

The comprehensive analysis of cottonseed oil revealed key physicochemical properties that determine its suitability for fuel applications. As presented in Table 6, the oil exhibits a cetane number of 50, indicating good ignition quality for diesel engines. The measured density of 912.5 kg/m^3 at 40 °C and kinematic viscosity of 35 cSt.significantly influence fuel injection and lubrication characteristics[25]. The acid value (0.63 mg KOH/g) and free fatty acid content (0.316%) suggest moderate acidity levels, while the exceptionally high flash point (264 °C) demonstrates superior safety for handling and storage.

The oil's saponification value (192.37 mg KOH/g) and iodine value (110.3 g I_2/100 g) reflect its fatty acid composition and degree of unsaturation[33]. With a high heating value of 39.75 MJ/kg and specific gravity of 0.92 at 20 °C, the oil demonstrates excellent energy content and physical characteristics[34]. The comparative data in Table 6 show close alignment with literature values, validating the experimental measurements while highlighting slight variations that may stem from differences in feedstock or processing conditions. These comprehensive characterization results provide essential data for evaluating the oil's performance in energy applications and establish a reliable basis for comparison with alternative vegetable oils. The combination of favorable ignition quality, appropriate viscosity, and high energy content positions cottonseed oil as a promising feedstock for biofuel production.

Transesterification of cottonseed oil: effect of reaction parameters on biodiesel yield

The transesterification of cottonseed oil was investigated using a nickel-doped chicken eggshell catalyst to synthesize cottonseed oil methyl ester (COME). The incorporation of nickel enhanced the catalytic activity of the eggshell-derived catalyst by improving basic site density and strength, leading to faster reaction kinetics and higher COME yields. The catalyst was prepared by mixing nickel sulfate ($NiSO_4$) solution with chicken eggshells, followed by calcination at 900 °C for 3 h to convert $CaCO_3$ into CaO. Optimal transesterification conditions using this catalyst included a reaction temperature of 62 °C, a methanol-to-oil molar ratio of 12:1, a catalyst loading of 4 wt%, and a reaction time of 117 min. Under these conditions, COME yields exceeded 98.01%, with the catalyst demonstrating good reusability and minimal activity loss over multiple cycles. The synthesized COME met key biodiesel standards, including viscosity, density, flash point, and cetane number.

The yield of cottonseed oil methyl ester (COME) increased with reaction time, reaching an optimum at 120 min. Beyond this point, further extension of reaction time did not enhance the yield and, in some cases, led to a slight decline. This reduction is likely due to the reversible nature of the transesterification reaction or the occurrence of side reactions. Reaction temperature also had a significant impact on COME synthesis. The yield steadily improved with increasing temperature up to 62 °C, as higher temperatures enhanced the solubility of reactants and accelerated reaction kinetics. However, temperatures exceeding 62 °C led to a decrease in yield, likely due to saponification and other thermal degradation effects. Thus, the optimal temperature range was determined to be 60–65 °C, where maximum conversion was achieved without excessive side-product formation. Catalyst loading influenced biodiesel yield in a similar trend. Increasing the catalyst concentration enhanced the reaction rate, with an optimal yield observed at 4 wt%. Beyond this point, further increases in catalyst loading resulted in reduced yields. This decline is attributed to increased soap and emulsion formation, which disrupted the separation of biodiesel from glycerol and reduced the accessibility of reactants to active catalytic sites. A saponification test confirmed this observation: soap content increased from 3.5 mg KOH/g at 4 wt% to 7.8 mg KOH/g at 5 wt%, validating the adverse effects of excessive catalyst on phase separation and product purity.

Likewise, the methanol-to-oil molar ratio played a critical role in driving the transesterification reaction toward completion. Although the stoichiometric requirement is 3:1, a higher ratio was necessary to shift the equilibrium in favor of methyl ester formation. The optimal yield was obtained at a 12:1 ratio, beyond which the yield declined due to dilution effects and reduced interaction between triglycerides and the catalyst. These combined effects of reaction time, temperature, catalyst loading, and methanol-to-oil ratio on biodiesel yield are summarized in Fig. 4, demonstrating their interdependent roles in optimizing the transesterification process.

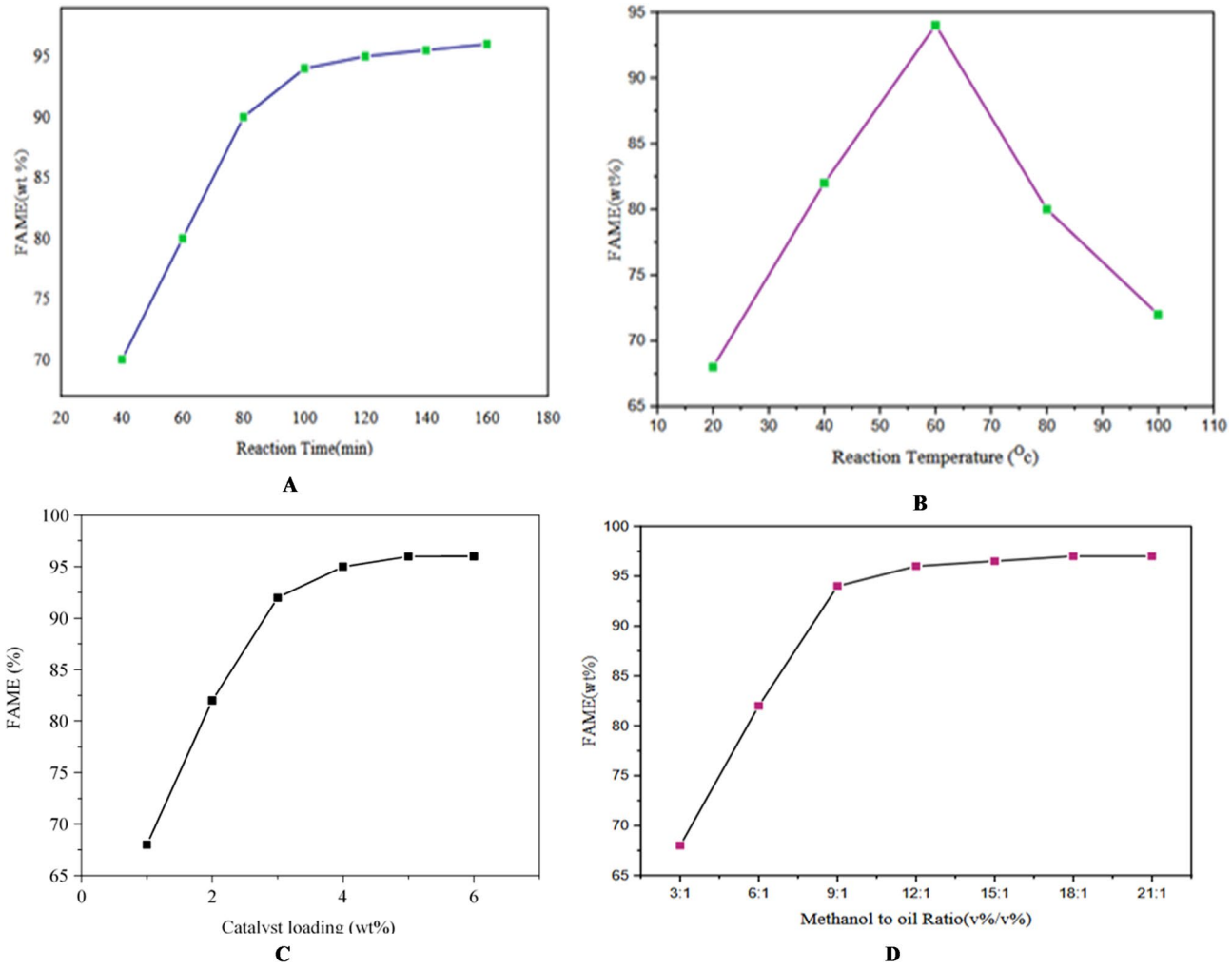

Fig. 4. Influence of Temperature, Time, Catalyst Loading, and Methanol-to-Oil Ratio.

Statistical analysis of cottonseed oil Methyl ester yield

Analysis of variance (ANOVA) was performed to evaluate the effects of key process variables on cottonseed oil methyl ester (COME) yield, based on 29 experimental runs. The results, summarized in Table 7, indicate that the quadratic model is highly significant, with an F-value of 213.29 and a p-value less than 0.0001, meaning there is only a 0.01% probability that this result is due to random chance. All main effects temperature (A), reaction time (B), catalyst loading (C), and methanol-to-oil molar ratio (D), along with their interaction and quadratic terms, were statistically significant ($p < 0.05$), confirming their strong influence on biodiesel yield. Furthermore, the lack-of-fit test was non-significant (F-value = 1.84, $p = 0.2919$), indicating that the model fits the experimental data well, with a 29.19% likelihood that any deviation is due to noise. These results demonstrate the model's robustness and suitability for accurately predicting and optimizing COME yield.

Model coefficients and predictive equations

The regression coefficients, as presented in Table S2 (supplementary), elucidate the individual and interactive effects of the process variables on COME yield. All variance inflation factor (VIF) values were equal to 1, indicating complete orthogonality among the variables and the absence of multicollinearity, which enhances the reliability of the model estimates. Based on the regression analysis, the final predictive equations were formulated in both coded form (Eq. 6) and actual process variables (Eq. 7), allowing for accurate yield prediction and process optimization within the studied parameter ranges.

Coded Factors:

$$\begin{aligned} COME\ \ Yield\ \ (wt\%) &= 75.10 + 3.17A - 3.64B - 5.36C - 0.75D + 6.50AB + 2.75AC \\ &-2.25AD + 5.13BC + 7.20BD + 1.96CD + 2.65A^2 + 3.94B^2 + 5.07C^2 + 3.23D^2 \end{aligned} \quad (6)$$

Actual Factors:

Source	Sum of Squares	Df	Mean Square	F-value	*P*-value	Remark
Model	1433.90	14	102.42	213.29	< 0.0001	Significant
A-reaction temperature	120.33	1	120.33	250.60	< 0.0001	
B-reaction time	159.14	1	159.14	331.41	< 0.0001	
C-catalyst loading	344.90	1	344.90	718.26	< 0.0001	
D-methanol to oil molar ratio	6.80	1	6.80	14.16	< 0.0021	
AB	169.00	1	169.00	351.95	< 0.0001	
AC	30.25	1	30.25	63.00	< 0.0001	
AD	20.25	1	20.25	42.17	< 0.0001	
BC	105.06	1	105.06	218.79	< 0.0001	
BD	207.36	1	207.36	431.85	< 0.0001	
CD	15.35	1	15.34	31.95	< 0.0001	
A2	45.65	1	45.65	95.06	< 0.0001	
B2	100.71	1	100.71	209.73	< 0.0001	
C2	166.70	1	166.70	347.15	< 0.0001	
D2	67.75	1	67.75	141.10	< 0.0001	
Residual	6.72	14	0.4802			
Lack of Fit	5.52	10	0.5523	1.84	0.2919	Not significant

Table 7. Response surface quadratic model analysis of variance (ANOVA).

$$COME\ Yield\ \ (wt\%) = 278.42 - 1.78T - 1.81t - 23.39C - 9.74M + 0.01T \cdot t + 0.09T \cdot C - 0.05T \cdot M + 0.06t \cdot C + 0.06t \cdot M + 0.33C \cdot M + 0.01T2 + 0.002t^2 + 1.27C^2 + 0.36M^2 \quad (7)$$

Where T (reaction temperature), t (time), C (catalyst loading), and M (methanol ratio) are in their original units.

Model validation and optimization

Table 8 presents a comparison between the experimental and predicted yields of cottonseed oil methyl esters (COME), showing strong agreement across all experimental runs. For example, Run 1 yielded an experimental value of 98.00%, closely aligning with the predicted value of 98.24%, which underscores the reliability of the developed model. The model's robustness is further supported by key statistical indicators, including a high coefficient of determination ($R^2 = 0.9953$), adjusted R^2 (0.9907), and predicted R^2 (0.9766), as illustrated in Fig. 5. Additionally, the model's adequate precision value of 59.90 indicates a strong signal-to-noise ratio, confirming its effectiveness in exploring the design space and its suitability for accurate prediction and process optimization.

Through response surface methodology, the optimal conditions for maximizing biodiesel yield were identified as a reaction temperature of 70 °C, reaction time of 117–120 min, catalyst loading of 4.6–4.9 wt%, and a methanol-to-oil molar ratio of 11.5–11.8. Under these optimized conditions, a maximum experimental yield of 98.5% was achieved. Although the model predicted a theoretical yield of 100.5%, this value was pragmatically adjusted to a maximum of 100%, acknowledging the stoichiometric and thermodynamic limitations of the reaction. The slight overestimation is attributed to statistical extrapolation by the response surface model and does not represent a physically achievable outcome. The close alignment between predicted and experimental yields, as depicted in Fig. 5, further validates the model's predictive accuracy and practical reliability.

The ANOVA and regression analysis validated the model's reliability for COME synthesis optimization. The nickel-doped eggshell catalyst system demonstrated high efficiency under statistically validated conditions, aligning with biodiesel standards.

Interaction effects between process variables on cottonseed oil methyl ester (COME) yield

The interaction effects among key transesterification variables, methanol-to-oil molar ratio, catalyst loading, reaction temperature, and reaction time, had a significant impact on the yield of cottonseed oil methyl ester (COME). To visualize and interpret these interactions, two-dimensional (2D) contour plots and three-dimensional (3D) response surface plots were generated based on the fitted regression model. The 2D plots, provided in Figure S1 (Supplementary), offer a top-down view of the response landscape by illustrating yield variations to two variables at a time while holding others constant. In contrast, the 3D response surfaces, shown in Fig. 6 (A–F), provide a more comprehensive depiction of the interactive behavior between variables, revealing curvature and peak regions indicative of optimal operating conditions. As illustrated in Fig. 6A, the interaction between reaction temperature and reaction time demonstrated that yield increased with rising temperature, particularly at shorter reaction durations. The varying gradients on the surface confirm a significant temperature–time interaction, supported by close agreement between predicted and experimental yields within the 95% confidence interval. Figure 6B shows the effect of catalyst loading and reaction time. Yield improved with both parameters, but the rate of improvement was greater at lower catalyst concentrations and longer reaction times. At elevated catalyst levels, excessive soap formation hindered phase separation, thereby reducing yield. In Fig. 6C, the interaction between methanol-to-oil molar ratio and reaction time indicated that

Run	Reaction temperature (°C)	Reaction time (min)	Catalyst loading (wt% %)	Methanol to oil molar ratio	Actual value	Predicted value	Residual
1	40	40	3	9	98.00	98.24	-0.2375
2	40	55	5	9	88.00	88.60	-0.600
3	120	55	3	12	78.00	77.96	0.0389
4	80	55	3	9	80.00	80.23	-0.2319
5	40	55	1	9	88.00	87.77	0.2333
6	120	55	1	9	89.00	88.67	0.3319
7	80	55	5	6	77.50	76.83	0.6653
8	80	70	1	9	74.50	75.10	-0.6000
9	80	55	3	9	84.80	85.08	-0.2778
10	120	40	3	9	92.23	91.47	0.8597
11	80	70	3	12	75.00	75.10	-0.1000
12	40	55	3	6	80.50	80.70	-0.2042
13	80	40	1	9	94.00	93.87	0.1333
14	80	55	3	9	87.00	87.15	-0.1542
15	40	70	3	9	75.00	75.10	-0.1000
16	120	70	3	9	86.00	86.05	-0.0514
17	80	40	3	6	69.00	68.38	0.6153
18	80	55	3	9	82.00	81.15	0.8514
19	40	55	3	12	77.00	77.27	-0.2653
20	80	55	1	12	88.00	87.72	0.2819
21	80	40	3	12	79.00	79.25	-0.2458
22	80	55	3	9	72.00	71.54	0.4556
23	80	70	5	9	75.00	76.32	-1.32
24	120	55	5	9	72.00	72.18	-0.1833
25	80	40	5	9	75.00	75.10	-0.1000
26	80	55	5	12	76.00	75.10	0.9000
27	80	70	3	6	82.00	82.00	-0.0014
28	80	55	1	6	83.00	83.38	-0.3778
29	120	55	3	6	79.00	79.32	-0.3153

Table 8. Comparison of actual and predicted values for cottonseed oil methyl ester.

yield increased up to approximately 90 min and a 12:1 molar ratio. Beyond these values, further increases led to yield reduction, likely due to enhanced glycerol solubility affecting product separation.

The interaction of reaction temperature and catalyst loading is shown in Fig. 6D, where temperature positively influenced yield, while higher catalyst loadings had a negative effect, primarily due to saponification. The optimal yield (98.04%) was achieved at 70 °C and the lowest catalyst concentration, with predictions falling within the 95% confidence bounds. Figure 6E presents the combined effects of reaction temperature and methanol-to-oil molar ratio. A decline in yield was observed with simultaneous increases in both variables, suggesting that excessive methanol and high temperature negatively influence transesterification efficiency. Finally, Fig. 6F demonstrates the interaction between catalyst loading and methanol-to-oil molar ratio. An inverted U-shaped relationship was observed, with maximum yield occurring at approximately 3.5 wt% catalyst loading. Beyond this point, increased soap formation and methanol-induced glycerol solubility led to yield suppression. These findings reinforce the need for careful optimization of both variables to avoid side reactions and maximize yield. Across all cases, the close fit between model predictions and experimental data within the 95% confidence interval confirms the robustness and predictive capability of the model.

In general, the response surface methodology effectively elucidated the complex interactions governing COME yield. Optimal conditions were identified by balancing the methanol-to-oil ratio, catalyst concentration, reaction temperature, and time. Exceeding these thresholds often led to yield reduction due to side reactions (saponification) or solubility effects. These findings provide critical insights for process optimization in biodiesel production.

Characterization of cottonseed oil methyl ester (COME)

FTIR analysis of cottonseed oil methyl ester

FTIR spectroscopy was utilized to identify functional groups in cottonseed powder (CS), cottonseed oil (CSO), and cottonseed oil methyl ester (COME), as illustrated in Fig. 7. The FTIR spectrum of CS exhibited a strong absorption at 3637.15 cm^{-1}, attributed to O–H stretching vibrations, likely arising from hydroxyl groups in cellulose and other plant constituents[35]. Additional peaks at 1409.33 cm^{-1} (C–H bending) and 588.00 cm^{-1} (C–C bending) confirmed the presence of aliphatic hydrocarbon structures. In the CSO spectrum, a prominent peak at 1743.36 cm^{-1} was observed, corresponding to the carbonyl (C = O) stretch of ester groups, typical of triglyceride

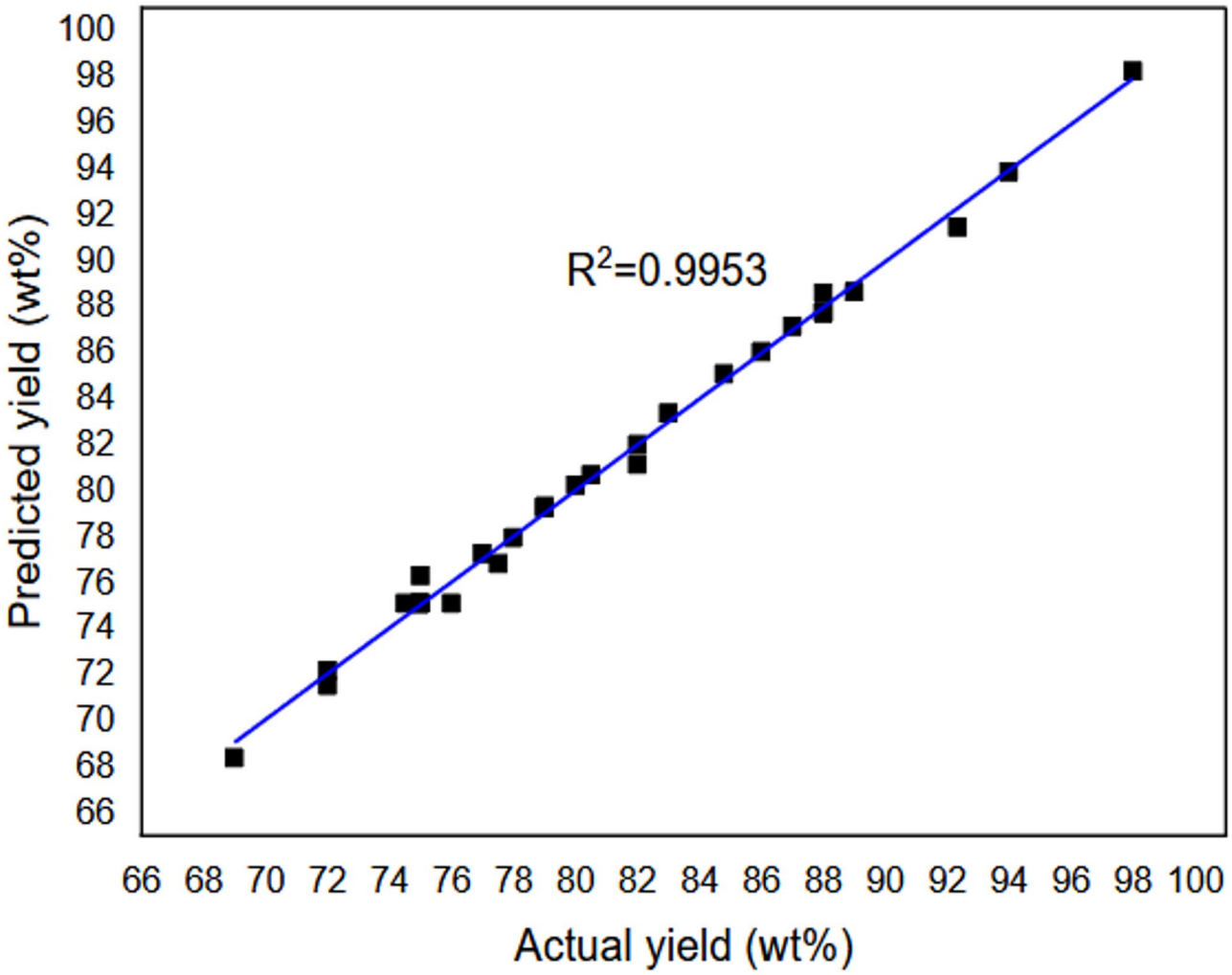

Fig. 5. Actual vs. Predicted.

structures (S. N. Shah, 2017). Other characteristic bands included 2933.25 cm^{-1} (C–H stretching), 1174.46 cm^{-1} (C–O stretching), and 705.83 cm^{-1} (=C–H bending), representing methylene, ester, and unsaturated moieties, respectively.

The COME spectrum showed a slight shift in the C=O peak to 1741.43 cm^{-1}, indicating the successful transesterification of triglycerides into fatty acid methyl esters (FAMEs). Peaks at 2913.92 cm^{-1} (C–H stretching), 1159.09 cm^{-1} (C–O–C stretching), and 705.83 cm^{-1} (=C–H bending) further confirmed the formation of methyl esters. These spectral variations collectively validate the chemical transformation during oil extraction and biodiesel synthesis, as summarized in Table 9.

GC-MS analysis of COME's fatty acid composition

The fatty acid profile of COME was analyzed using Gas Chromatography–Mass Spectrometry (GC-MS). As illustrated in Fig. 8 and detailed in Table 10, linoleic acid (C18:2) was the most abundant component at 50.5%, followed by palmitic acid (C16:0, 24.25%), oleic acid (C18:1, 20.25%), stearic acid (C18:0, 3.5%), and myristic acid (C14:0, 1.5%) (Jayakumar et al., 2019). The calculated molecular weight of the cottonseed oil based on its fatty acid distribution was 282.7 g/mol. This composition is consistent with reported profiles of cottonseed-derived biodiesel, confirming its potential for use as a renewable fuel[25].

Physicochemical properties of cottonseed oil methyl ester

The physicochemical properties of cottonseed oil methyl ester (COME) were evaluated to determine its suitability as a biodiesel fuel. As summarized in Table 11, the cetane number of 53.75 indicates excellent ignition quality, surpassing the minimum requirement of 47 set by biodiesel standards such as ASTM D6751 and EN 14,214. This higher cetane number ensures better combustion efficiency and smoother engine operation. The density of COME at 40 °C was measured at 865.7 kg/m^3, which falls within the acceptable biodiesel range (860–900 kg/m^3), ensuring compatibility with standard fuel injection systems. The kinematic viscosity, measured at 4.96 cSt. at 40 °C, also meets the standard range (1.9–6.0 mm^2/s), ensuring proper atomization and fuel flow within the engine. These parameters are crucial for maintaining consistent engine performance. The acid value was low at

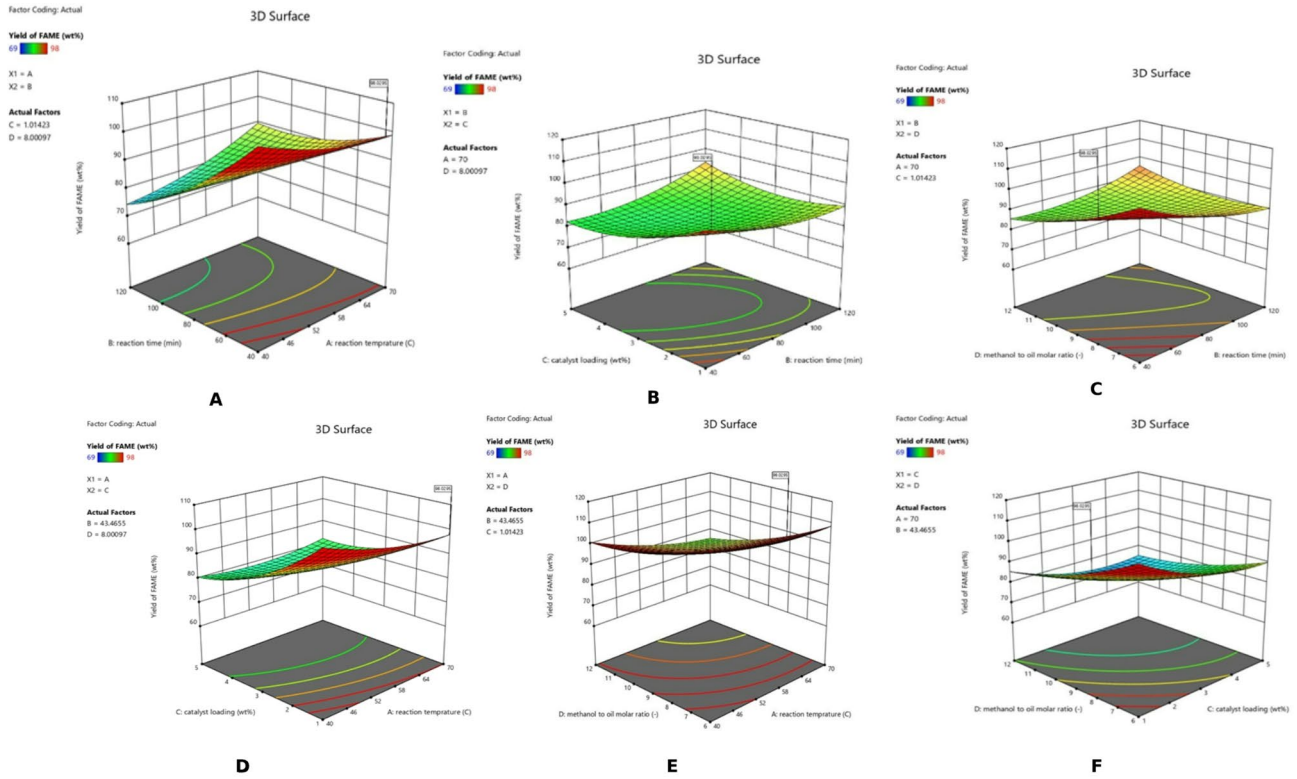

Fig. 6. (**A–F**) Three-dimensional response surface plots illustrating the interaction effects between process variables on cottonseed oil methyl ester (COME) yield.

0.18 mg KOH/g, indicating minimal free fatty acid (FFA) content and reducing the risk of corrosion and deposit formation in fuel systems. This low acidity highlights the effectiveness of the transesterification process and the overall fuel quality.

The flash point of COME was recorded at 162.4 °C, significantly above the minimum safety threshold (> 120 °C), which affirms its safe handling and storage properties. A higher flash point is desirable to reduce the risk of accidental ignition during transportation or storage. The saponification value was 193 mg KOH/g, reflecting the average molecular weight and ester content of the biodiesel, while the iodine value of 108.18 g I_2/100 g indicates a moderate level of unsaturation. These parameters influence oxidative stability and cold flow properties. The observed iodine value is consistent with linoleic acid-rich methyl esters and falls within the EN 14,214 maximum limit of 120 g I_2/100 g. COME exhibited a higher heating value (HHV) of 39.45 MJ/kg, which is comparable to petroleum diesel (around 42–45 MJ/kg), confirming its competitiveness in energy output. Small deviations in moisture content (0.5%) and FFA (0.09%) suggest good fuel stability, although further refinement may be pursued to enhance long-term storage quality.

The oxidative stability of COME was assessed using the Rancimat method (EN 14112). The induction period was measured at 6.2 h, slightly above the EN 14,214 requirement of 6 h. While this indicates that COME meets the minimum oxidative stability standard, the high linoleic acid content (50.5%) may compromise long-term shelf life. Future improvements could include the incorporation of natural or synthetic antioxidants to delay oxidation and preserve fuel integrity during storage.

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Conclusion

This study demonstrates an integrated and sustainable approach to biodiesel production using non-edible cottonseed oil and an innovative nickel-doped calcium oxide (CaO) catalyst derived from waste chicken eggshells. By addressing key challenges in feedstock selection, catalyst development, and process efficiency, the research highlights a viable pathway for the synthesis of resource-efficient biofuels. The nickel-doped CaO catalyst exhibited excellent catalytic activity, achieving a high fatty acid methyl ester (FAME) yield of 98.01% under optimized conditions (62 °C, 12:1 methanol-to-oil molar ratio, 4 wt% catalyst loading, and 117 min reaction time). The enhanced reactivity is attributed to its high crystallinity (68.2%), porous morphology, and the

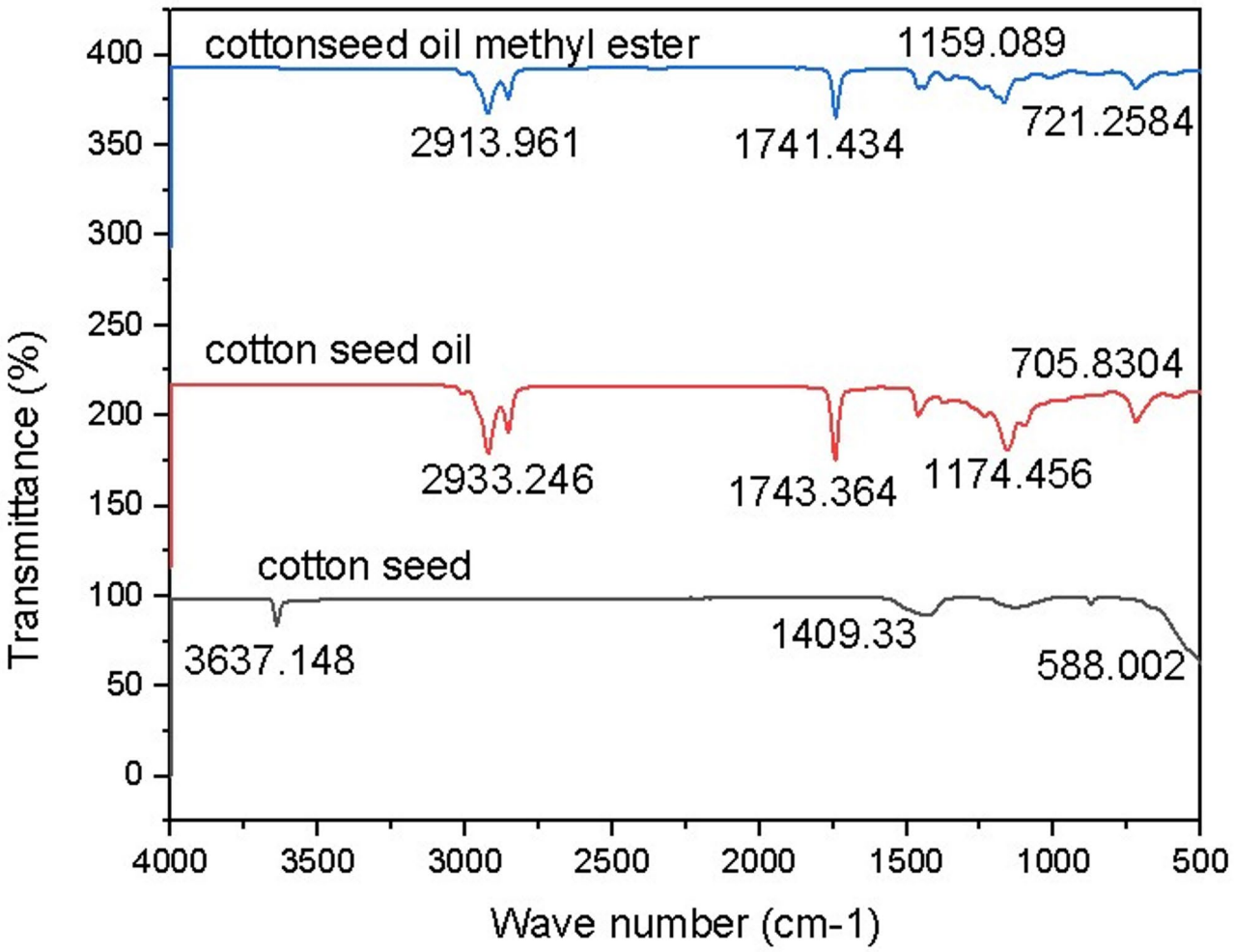

Fig. 7. FTIR spectra of CS, CSO, and COME.

Wave Number (cm^{-1})	Vibration type	Functional group	Sample identified in
3637.148	O–H stretch	–OH	Cottonseed powder (CS)
1409.33	C–H bending	$–CH_3$, $–CH_2–$	Cottonseed powder (CS)
588.002	C–C bending	–C–C–	Cottonseed Powder (CS)
2933.246	C–H stretch	$–CH_3$, $–CH_2–$	Cottonseed oil (CSO)
1743.364	C = O stretch	–C(= O)–O– (Ester)	Cottonseed oil (CSO)
1174.456	C–O stretch	–C–O–C–	Cottonseed oil (CSO)
705.83	=C–H bending	> C = CH_2, –CH = CH–	Cottonseed oil (CSO)
2913.916	C–H stretch	$–CH_3$, $–CH_2–$	Cottonseed oil methyl ester (COME)
1741.434	C = O stretch	–C(= O)–O– (Ester)	Cottonseed oil methyl ester (COME)
1159.089	C–O–C stretch	–C–O–C–	Cottonseed oil methyl ester (COME)
705.83	=C–H bending	> C = CH_2, –CH = CH–	Cottonseed oil methyl ester (COME)

Table 9. FTIR analysis of cottonseeds, oil, and Methyl ester (COME) showing functional group changes during transesterification.

incorporation of nickel, which collectively improved surface basicity and stability. Cottonseed oil was selected as a sustainable feedstock due to its non-edible nature, availability, and favorable characteristics, including low moisture content (1.2%) and high volatility (94.4%). The produced biodiesel displayed physicochemical properties within international standards (e.g., ASTM D6751), such as a cetane number of 53.75, kinematic viscosity of 4.96 cSt at 40 °C, and a flash point of 162.4 °C, indicating high quality and operational safety.

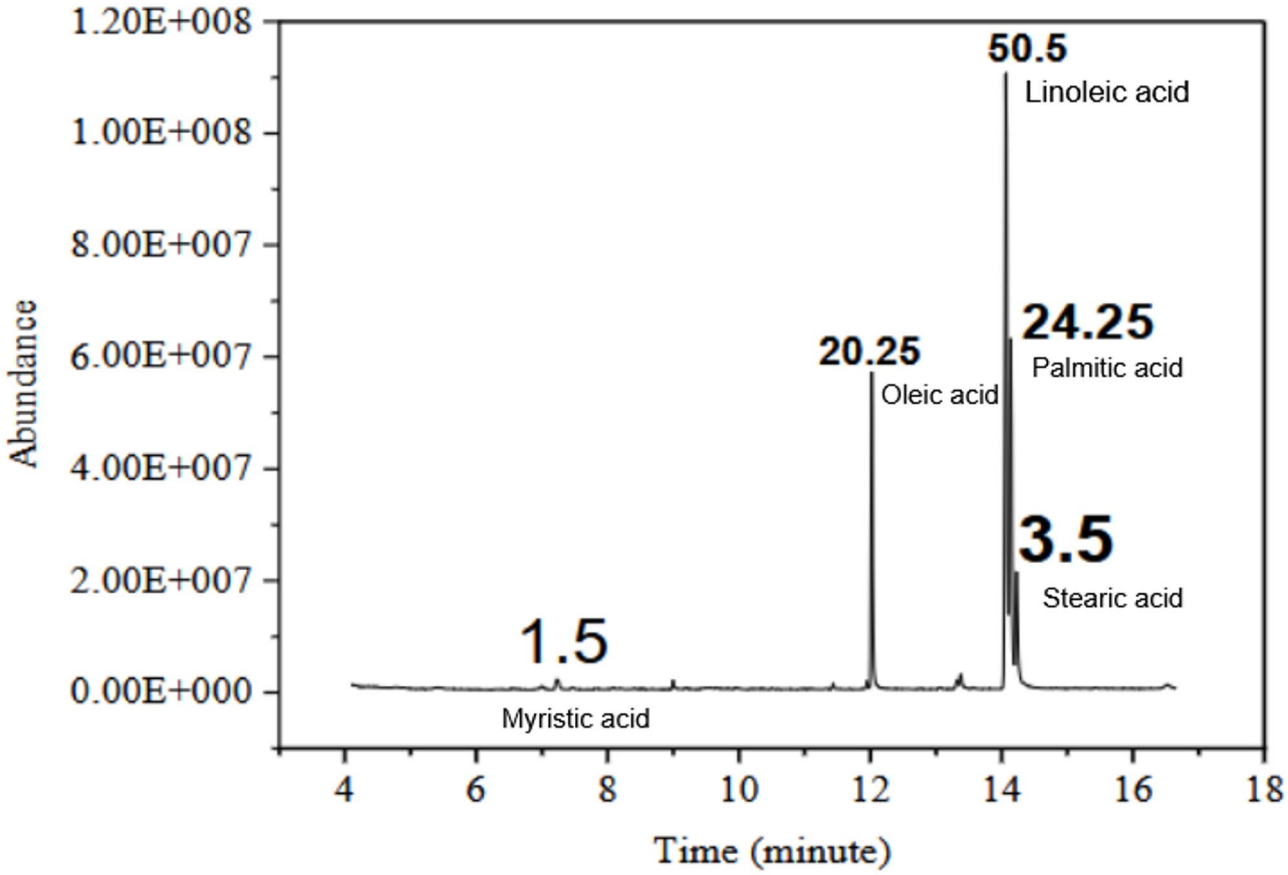

Fig. 8. Cottonseed oil methyl ester's fatty acid content as determined by GC-MS.

Fatty acid type	Structure	Formula	Area (%)	Molecular weight
Linoleic acid	C18:2	C18H32O2	50.5	280.45
Palmitic acid	C16:0	$C_{16}H_{32}O_2$	24.25	256.42
Oleic acid	C18:1	C18H34O2	20.25	282.46
Myristic acid	C14:0	C14H28O2	1.5	228.37
Stearic acid	C18:0	C18H36O2	3.5	284.48

Table 10. Cottonseed oil fatty acid Composition.

Oil properties	Unit	Current study values	Previous study value
Cetane number	-	53.75	53.1[36]
Density@40°C	Kg m^{-3}	865.7	877.5[37]
Kinematic viscosity@40°C	mm^2/s	4.96	4.63[38]
Acid value	mgKOH/g	0.18	0.1[36]
Flash point	°C	162.4	154[36]
Saponification value	mgKOH/G	193	208.411
Iodine value	$gI_2/100$ g	108.18	104.7[37]
viscosity@40°C	mm^2/s	4.4	4.81[36]
Moisture	%	0.5	0.02[39]
HHV	MJ/kg	39.45	38.6[38]
FFA	(%)	0.09	0.089[40]

Table 11. Comparison of COME properties between the present study and other Work.

Beyond its technical merits, this work exemplifies circular economy principles by valorizing agricultural and food waste into valuable materials for clean energy applications. The low-cost catalyst and accessible feedstock make this approach especially relevant for developing regions, where such resources are abundant and underutilized. The methodology supports decentralized, scalable biodiesel production strategies, contributing to more resilient and self-reliant energy systems. While the outcomes are promising, further investigation is warranted in several areas. These include evaluating long-term catalyst performance under continuous operation, assessing the economic feasibility of industrial-scale deployment, and exploring hybrid catalyst systems to enhance yield and reusability further. Moreover, expanding this approach to include other waste-derived feedstocks and alternative transesterification strategies could broaden its applicability.

In general, this study provides a practical and sustainable framework for biodiesel production by combining catalytic innovation, process optimization, and waste material utilization. Despite these promising outcomes, certain limitations should be acknowledged. The use of nickel salts, while effective for catalytic enhancement, may introduce supply or cost constraints depending on market availability. Additionally, the calcination temperature of 900 °C, although necessary to ensure complete $CaCO_3$ decomposition and nickel incorporation, contributes to high energy consumption. These factors represent potential trade-offs that must be considered when evaluating the full sustainability of the process. These findings contribute to the development of greener energy solutions and offer valuable insights for researchers, practitioners, and policymakers seeking to promote sustainable resource management and technological advancement in the biofuel sector.

Data availability

The datasets generated and analyzed during the current study are available from the corresponding author, Kumlachew Yeneneh (kumynnh2023@gmail.com), upon reasonable request. All relevant data supporting this study's findings are included in the manuscript.

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Author contributions

Kumlachew Yeneneh conceived and designed the research framework, performed the catalyst synthesis and characterization, conducted the transesterification experiments, analyzed the physicochemical and emission data, and prepared Figs. 1–18. Kumlachew Yeneneh also carried out the statistical modeling, optimization using Response Surface Methodology (RSM), and interpretation of GC-MS and FTIR results. He wrote the initial draft and integrated all data and figures into the manuscript. Gadisa Sufe provided critical guidance on experimental methodology, supported data interpretation, contributed to refining the statistical analysis, and reviewed and revised the manuscript for intellectual content and clarity. All authors reviewed and approved the final manuscript.

Declarations

Competing interests

The authors declare no competing interests.

Additional information

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