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# CaO-Based Heterogeneous Catalyst from Cow Bone for Direct Cracking of Triglycerides

A calcium oxide (CaO)-based heterogeneous catalyst derived from waste cow bone for the direct cracking of *Moringa oleifera* seed oil was investigated. Optimization was carried out using a five-level-four-factorial central composite design based on response surface methodology in 30 experimental runs. Under the optimal conditions of reaction temperature, catalyst concentration, reaction time, and agitation speed, 68 % biodiesel was achieved. A total of four major fatty acid methyl esters were identified in the synthesized biodiesel by the retention time and fragmentation pattern data of the gas chromatography-mass spectrometry analysis. The statistical model for predicting the biodiesel yield was developed for the effects and percentage contributions of the optimization variables.

**Keywords:** Biodiesel, CaO-based catalysts, *Moringa oleifera* seeds, Response surface methodology, Triglyceride cracking

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## 1 Introduction

The efforts for local content development, reduction of greenhouse gases, and economic gains have led to various methods of biodiesel production and use of biodiesel from both vegetable oils and animal fats [1]. The significance of biodiesel production and its subsequent usage as a renewable energy source ranges from the reduction of overdependence on crude oil resources that are finite in nature, to its combustion properties, biodegradability, ecological and environmental friendliness, renewability, and its sustainability development, thereby making it an alternative [2]. Biodiesel, as the name implies, is a mixture of fatty acid methyl esters (FAMES) that is derived from either vegetable oils (waste and virgin) and animal fats via transesterification processes involving acidic or basic catalysts. Moreover, advancements in biodiesel production technology have paved ways for biodiesel production using catalysts that are homogeneous in nature [3].

Usually, hydroxides of sodium and potassium are often employed as base-oriented homogeneous catalysts in the production of biodiesel [4]. However, the catalyst recovery and the quantum of water needed to dissolve and refine alkaline hydroxides have always been the disadvantages associated with homogeneous catalysis in biodiesel production [5]. On the other hand, the use of heterogeneous catalysts for biodiesel production solves both the economic and environmental problems posed by homogeneous catalysts. First, a cheaper and easier process is involved, thereby limiting the amount and generation of wastewater, and second, simplification of production processes, i.e., bypassing some routes. This leads

to energy and time savings, and cutting down the cost of production [6].

Mootabadi et al. [7] studied the production of biodiesel from palm oil using alkaline earth oxides, namely, CaO, SrO, and BaO calcined at 50 °C for 3 h. The activities of each of the three catalysts were sequentially CaO < SrO < BaO. At 0.03 and 9:1 catalyst-to-oil mass ratio and methanol-to-oil molar ratio, respectively, the CaO biodiesel yield was about 5 % after 1 h. The results demonstrated that the strengths of the heterogeneous catalysts developed were in the same sequence of CaO < SrO < BaO. Hence, basicity properties were established as the major determinants of the catalytic activities recorded among the heterogeneous catalysts used.

Chen et al. [8] studied the basicity of the alumina-supported alkaline earth metal oxides calcined at 550 °C for 6 h (CaO/Al<sub>2</sub>O<sub>3</sub> calcined between 300 °C and 750 °C). The authors found that the calcination temperature has a significant influence on the amounts of basic sites of alkaline earth oxides only, as it does not influence the strength of basic sites. At a calcination temperature of 550 °C, the optimal biodiesel yield was

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obtained, i.e., it serves as the temperature at which the optimum amount of basic sites of CaO was obtained.

In this work, kaolinite clay from Ukpok, Anambra State of Nigeria, and cow bone were synthesized and applied in triglyceride cracking for biodiesel production. These raw materials and the synthesized catalyst were characterized for XRF, FTIR, SEM, XRD, EDS, BET, XPS, and TGA to study their molecular structures, compounds of interest, porosity, grain sizes, elemental proportions, surface areas, IR spectra, weight losses, and morphologies. The synthesized catalyst was thereafter utilized for triglyceride cracking for biodiesel production from transesterification of *Moringa oleifera* oil.

## 2 Materials and Methods

### 2.1 Materials

*Moringa oleifera* seed oil was purchased from Dorayi, a local market in Kano State, Nigeria. The cow bone was obtained from Minna Abattoir, Niger State, Nigeria, and the Ukpok clay was collected from Ukpok in Anambra State, Nigeria. The chemicals and reagents used in this study were of analytical grade purchased from Steve Moore Chemicals, Zaria, Kaduna State, Nigeria, including analytical reagent-grade sulfuric acid ( $\text{H}_2\text{SO}_4$ ) solution (95 % concentration,  $1.83 \text{ g mL}^{-1}$ ), analytical standard ammonium sulfate (132.14 molecular weight), and ACS reagent-grade ethanol (98 % purity).

### 2.2 Methods

#### 2.2.1 Preparation of Heterogeneous Catalysts

De-alumination of Ukpok clay was carried out according to Eterigho et al. [9]. A known weight of uncalcined clay was mixed with a standard analytical-grade 36 N  $\text{H}_2\text{SO}_4$  solution at a molar ratio of 1:5. An aliquot of the mixture was placed in an open quartz crucible, which was heated in a furnace at  $500^\circ\text{C}$ , ramped at  $5^\circ\text{C min}^{-1}$  for 1 h. Once the reaction temperature was reached, the crucible was left in the furnace for another 6 h.

#### 2.2.2 Preparation of Cow Bone

The cow bones were washed thoroughly with distilled water to remove dirt. Thereafter, the samples were sun-dried for a maximum of five days to transform the calcium species to an active catalyst. After the attainment of a constant dry weight, the samples were calcined in a furnace. Each sample was calcined at  $1000^\circ\text{C}$  for 2 h. This is because calcination at high temperature reduces the carbon and oxygen content and produces metal oxides such as  $\text{K}_2\text{O}$ , CaO, and MgO as the major ingredients in the ash, which are the active base catalysts for biodiesel production [14]. The calcined samples were left to cool at room temperature before grinding each sample into powder using laboratory ball mills. Finally, the samples were sieved using  $75\text{-}\mu\text{m}$  sieve size and stored in air-tight containers [10].

#### 2.2.3 Modification of Sulfated Cow Bone

To activate the silica for the reactions involved in the study, there was the need to convert the kaolin into metakaolin by thermal treatment. The resultant material (metakaolin) was separately doped with cow bone before sulfating the sample with ammonium sulfate ( $(\text{NH}_4)_2\text{SO}_4$ ) using a molar ratio of 0.5:1:6 [9].

#### 2.2.4 Characterization of Doped Heterogeneous Catalyst

The surface morphology of the doped catalyst was analyzed by SEM (ZEISS SIGMA model). The elemental composition was determined by XRF. The crystalline phase was analyzed by XRD (PANalytical X'Pert<sup>3</sup> powder diffractometer) using  $\text{CuK}\alpha$  radiation at 5000 kV and 30 mA, in the scanning angle  $2\theta$  ranging between  $10$  and  $90^\circ$  with a step size of  $0.0130^\circ$ . The surface area of the catalyst was determined by Brunauer-Emmett-Teller (BET) NOVA STATION-D instrument. The FTIR spectrum was obtained by FTIR spectroscopy (Bruker, Germany) to analyze the functional groups present in the heterogeneous catalyst [9, 11].

#### 2.2.5 Optimization of the Transesterification Process

The transesterification process was optimized using response surface methodology (RSM). The effects of temperature, catalyst concentration, agitation speed, and time were evaluated. Design Expert Software (7.0) was used in carrying out the regression analysis where the level of interactions of the variable under consideration and their significance were ascertained. The interaction effects of the variables were used to fit the empirical model with the data obtained. The latter was correlated with the independent variables using a polynomial equation in addition to the ANOVA used in testing for variance. Five-level four-factorial central composite design (CCD) variables and the optimization experimental design matrix were used to produce biodiesel from cow bone catalyst. The coded and uncoded levels of the independent variables are presented in Tab. 1. Zero (0) represents the central point for the variables while  $-1$  and  $+1$  denote the lower and upper value of the four process variables;  $-\alpha$  and  $+\alpha$  are the lowest and highest value suggested by the model for every parameter studied within the range.

**Table 1.** Independent variables and levels used for CCD.

S/N	Variable(s)	Symbol	Levels				
1	Temperature [ $^\circ\text{C}$ ]	$X_1$	$-1$	0	$+1$	$-\alpha$	$+\alpha$
2	Catalyst conc. [wt %]	$X_2$	$-1$	0	$+1$	$-\alpha$	$+\alpha$
3	Time [h]	$X_3$	$-1$	0	$+1$	$-\alpha$	$+\alpha$
4	Agitation speed [rpm]	$X_4$	$-1$	0	$+1$	$-\alpha$	$+\alpha$

## 2.3 Composition Analysis of Produced Biodiesel Samples

### 2.3.1 FTIR

Functional groups present in the heterogeneous catalyst samples of biodiesel produced from NaOH, and cow bone catalysts were measured using FTIR. Sixteen background scans involving eight resolutions were taken and the samples were recorded in the range of 4000 to 650  $\text{cm}^{-1}$ .

### 2.3.2 GC-MS

The biodiesel samples produced from NaOH, and cow bone were analyzed by means of a PerkinElmer Clarus 500 gas chromatograph coupled to a Clarus 500 mass spectrometer with liquid automatic sampler using helium as the vector gas in a 30 m long and 0.25  $\mu\text{m}$  in diameter capillary column with 2  $\mu\text{L}$  sample injection rate. Initially, the temperature was held at 50  $^{\circ}\text{C}$  for 3 min, then it was raised to 280  $^{\circ}\text{C}$  at an increment rate of 10  $^{\circ}\text{C min}^{-1}$  for 3 min [12].

## 3 Results and Discussion

### 3.1 Composition Analysis

#### 3.1.1 XRF Analysis

XRF analysis was carried out on the locally abundant raw material (cow bone) showing the presence of elements and compounds of interest (Al, Si, and  $\text{SO}_3$ ) at a ratio of 1:5:4, respectively. Tab. 2 presents the compositions of cow bone before ( $C_1$ ) and after calcination ( $C_2$ ) at 1000  $^{\circ}\text{C}$  for 2 h, respectively. It can be seen that the approximate compositions of mineral elements of the fresh peels increases in the order  $\text{Si} > \text{P} > \text{Cl} > \text{Al} > \text{Na} > \text{Mg}$ , with Si being the main element. This has also been reported by similar observation from *Carica papaya* peels [13]. Al, Si, CaO, and  $\text{SO}_3$  increases of 36, 14, 34, and 60 %, respectively, were recorded for the cow bone before and after calcination of the sample. It is deduced that heating the sample (cow bone) at high temperature and below its melting point eliminates elements and/or compounds that can release water, increasing their crystallinities while at the same time improving their specific properties. Hence, there exist increments in the value of CaO after calcination.

Etim et al. [14] also reported that during calcination at high temperatures the carbon and oxygen content are reduced to produce metal oxides such as  $\text{K}_2\text{O}$ , CaO, and MgO as the major ingredients in the ash, which are the active base catalysts

for biodiesel production. It can be concluded that both time and calcination temperature are significant parameters [15].

#### 3.1.2 FTIR Analysis of Cow Bone CaO-Based Heterogeneous Catalyst

The FTIR spectra of the heterogeneous catalyst (cow bone) is presented in Fig. 1. It shows several adsorption peaks with highly significant broad bands seen at 3628  $\text{cm}^{-1}$  representing the hydroxyl groups (OH) stretching vibrations of the hydroxyl groups in the brucite-like sheets. This phenomenon is attributed to the total dehydration and calcination of the samples, thereby demonstrating lack of water quantities. The spectra bands in the range of 600  $\text{cm}^{-1}$  portrayed by cow bone heterogeneous catalysts are a summary representation of magnesium hydroxide (Mg-OH) stretch modes. The peak at 1098  $\text{cm}^{-1}$  observed in cow bone heterogeneous catalysts signifies the OH deformities modes. The absorption peak at approximately 3628  $\text{cm}^{-1}$  confirms the presence of many different bonded oxides of Fe, Mn, Cr, V, Ti, K, P, Al, Na, and S [16, 17].

#### 3.1.3 SEM Analysis

The SEM morphology of the CaO-based heterogeneous catalysts after calcination at 1000  $^{\circ}\text{C}$  for 2 h is presented in Fig. 2. The morphological features of the sample were studied at 5 KX magnification. The particle size was found to be approximately 5  $\mu\text{m}$ . The image is characterized by large micropores and particle agglomeration. This may be due to sintering of metal oxides as a result of calcination [14]. The structural porosity increases in natural materials (limestone and clay) and is significant for increasing the yield of biodiesel [18, 19].

The porous surface obtained after calcination could be due to the release of water and  $\text{CO}_2$  during decomposition of  $\text{CaCO}_3$  to CaO which act as porogens [20, 21]. The porosity on the surface of the catalyst increases the BET surface area that leads to an increase in the catalytic activity [20]. The biodiesel produced from this material will require high steps of washing which will ultimately affect the yield. A similar type of structure was reported by Laskar et al. [22] for calcined waste snail shells.

#### 3.1.4 XRD Analysis

XRD is employed for estimation of the average crystallites or grain sizes of catalysts. The XRD analyses were carried out on the CaO-based heterogeneous catalysts developed from cow bone mainly to derive structural information of the catalysts

**Table 2.** Elemental composition (in %) of cow bone.

	Na	Mg	Al	Si	P	$\text{SO}_3$	Cl	$\text{K}_2\text{O}$	CaO	$\text{TiO}_2$	$\text{Cr}_2\text{O}_3$	$\text{Mn}_2\text{O}_3$	$\text{Fe}_2\text{O}_3$	ZnO	SrO	Total
$C_1$	1.51	0.99	2.01	13.44	4.63	60.48	2.10	1.84	4.22	1.19	1.03	0.99	3.01	0.87	1.03	100.00
$C_2$	1.96	0.54	3.13	15.54	3.51	68.33	0.06	0.01	6.41	0.27	0.02	0.01	0.33	0.00	0.01	

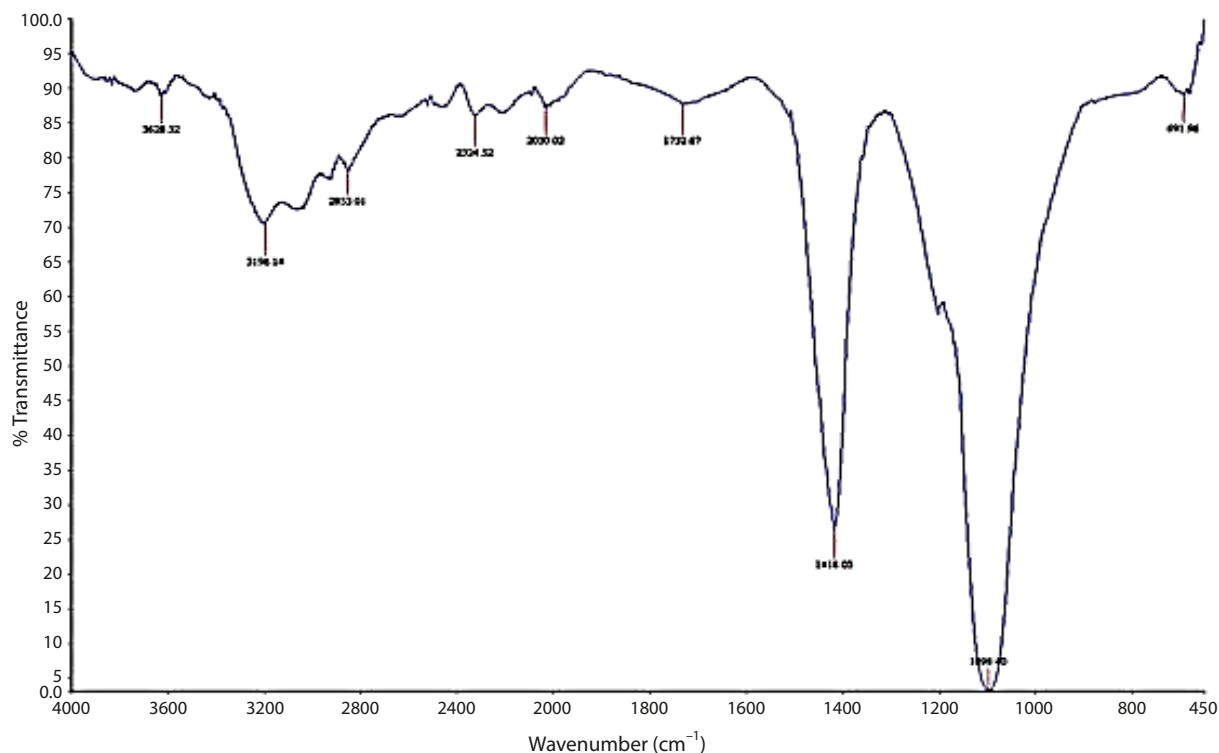


Figure 1. FTIR spectrum of the CaO-based heterogeneous cow bone catalyst.

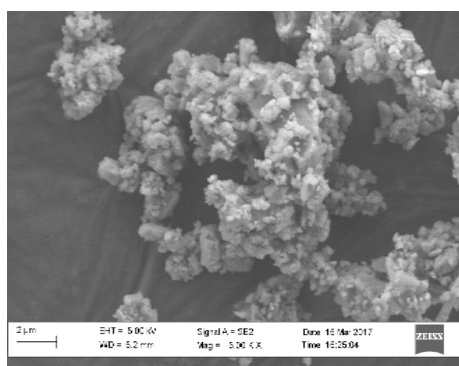


Figure 2. SEM image of the CaO-based heterogeneous cow bone catalyst.

produced. Fig. 3 depicts the XRD patterns of CaO-based heterogeneous catalysts developed from cow bone. Due to the fact that the samples are in the form of polycrystalline powders, the XRD analyses therefore aimed at the identification of specific lattice planes that produce peaks at their corresponding angular positions  $2\theta$ , as determined by Bragg's law [23]. Moreover, the interaction of cow bone powder indicates that hydroxyl groups of sodium (Na) have reacted with the support of  $\text{Al}_2\text{O}_3$  giving rise to the formation of alumina that originated from the stronger basic sites of the catalysts [24]. Hence, the XRD peaks obtained from the heterogeneous catalyst are intense and sharp because they have sufficient long-range order, thereby becoming broader for crystallite sizes below about 100 nm [25]. The disappearance of major peaks gave way to an amorphous

material with a very broad band within the low-range angles of  $15\text{--}35^\circ$  and such broad bands are said to be the amorphous phase of silica ( $\text{SiO}_2$ ).

### 3.1.5 BET Analysis

The BET analyses of the cow bone CaO-based heterogeneous catalysts were carried out using a NOVA STATION D instrument under the conditions of 0.25 g weight, 3 h out-gas time, nitrogen as analysis gas, 0.100 precision tolerance, 6054 s analysis time,  $250^\circ\text{C}$  out-gas temperature, and  $273^\circ\text{K}$  bath time. The surface area of  $392.740\text{ m}^2\text{g}^{-1}$  was recorded for the cow bone CaO-based heterogeneous catalyst. This phenomenon can be attributed to the collapse of the surface area during the second calcination due to fortification of the cow bone [26]. Tab. 3 presents the BET analysis results for the heterogeneous catalysts while Fig. 4 depicts the BET result of the cow bone CaO-based heterogeneous catalyst.

### 3.1.6 XPS Analysis

Fig. 5 presents the XPS plot of the CaO-based heterogeneous catalyst developed from cow bone sample carried out at a take-off angle, pass energy, scan rate, analyses area, and pressure of  $45^\circ\text{C}$ ,  $178.95\text{ eV}$ ,  $2.5\text{ eVs}^{-1}$ ,  $3 \times 10\text{ mm}^2$ , and  $< 5 \times 10^{-9}\text{ Torr}$ , respectively. The effect of de-aluminating kaolin on the composition of the silica-to-alumina (Si/Al) ratio is also depicted by XPS analysis on the CaO-based heterogeneous catalyst. The

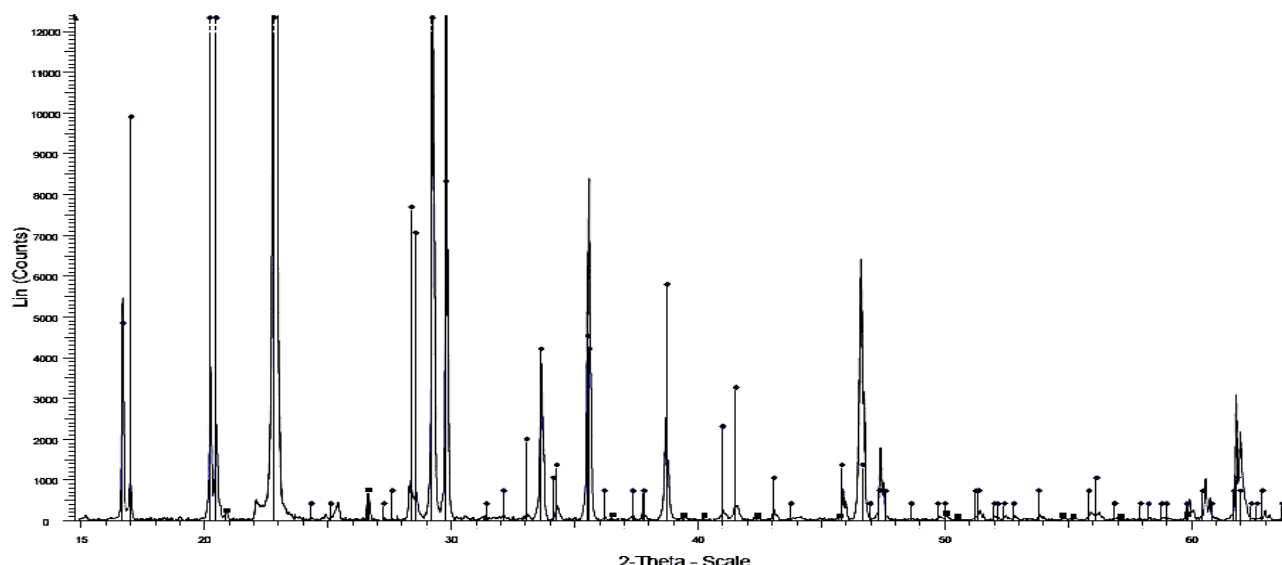


Figure 3. XRD pattern of the CaO-based heterogeneous cow bone catalyst.

Table 3. BET analysis of the CaO-based heterogeneous cow bone catalyst.

S/N	Properties	Values
1	Slope [-]	6.3960
2	Intercept [-]	2.4700
3	Correlation coefficient [-]	0.9969
4	Surface area [ $\text{m}^2\text{g}^{-1}$ ]	392.7400
5	Pore radius [ $\text{\AA}$ ]	9.2370

Al2p spectra for all the samples at the same binding energy correspond to aluminium(III) oxide. Also, the silicon (Si2p) spectrum of the sample is similar around 100 eV. Similarly, Eterigho et al. [9] reported 103.30 eV (the binding energy of silicon), which indicated that it was  $\text{Si}^{4+}$  in silicon oxide. This little variation in the pass energy is attributed to the doping of cow bone powder.

### 3.1.7 TGA Analysis

Fig. 6 shows the TGA profiles of the heterogeneous catalysts. The TGA curve features two major peaks in which the first one is endothermic, and the other is exothermic for the sample. The first endothermic peak appears at lower temperature (250 °C). This phenomenon (weight loss) can be attributed to the de-adsorption of humidity on the exterior surface of the sample. Residual water can be desorbed from the surface during heating the sample by the TG analyzer [27]. The second stage occurs at about 250–600 °C and an exothermic peak at about 2500 °C may be attributed to the crystallization of amorphous compounds therein [28]. According to Eterigho et al. [9] the only element responsible for the weight loss at the tempera-

ture of the thermal decomposition is sulfate decomposition. The TGA results of the CaO-based heterogeneous catalyst showed accelerated weight loss. Hence, the method is said to be more viable in terms of sulfate retention on the surface of the catalysts.

### 3.2 Optimization

The optimization of biodiesel produced from *Moringa oleifera* seed oil using CaO-based heterogeneous catalyst produced from cow bone was carried out to determine the effect of operating parameters (time, temperature, agitation speed, and catalyst concentration) on biodiesel yield. A five-level four-factorial experimental design was applied via central composite rotatable design matrix (CCRD) and response surface methodology (RMS). The results were analyzed by means of the design software Design Expert 7.0.0. The CCRD design matrix generated for the experimental range and levels is presented in Tab. 4.

Table 4. Experimental range and level of independent variables.

S/N	Variable	Symbol	Limits				
			-1	0	+1	$-\alpha$	$+\alpha$
1	Temperature [°C]	$X_1$	200	250	300	150	350
2	Catalyst conc. [wt %]	$X_2$	2	3	4	1	5
3	Time [min]	$X_3$	120	180	240	60	300
4	Agitation speed [rpm]	$X_4$	200	250	300	150	350

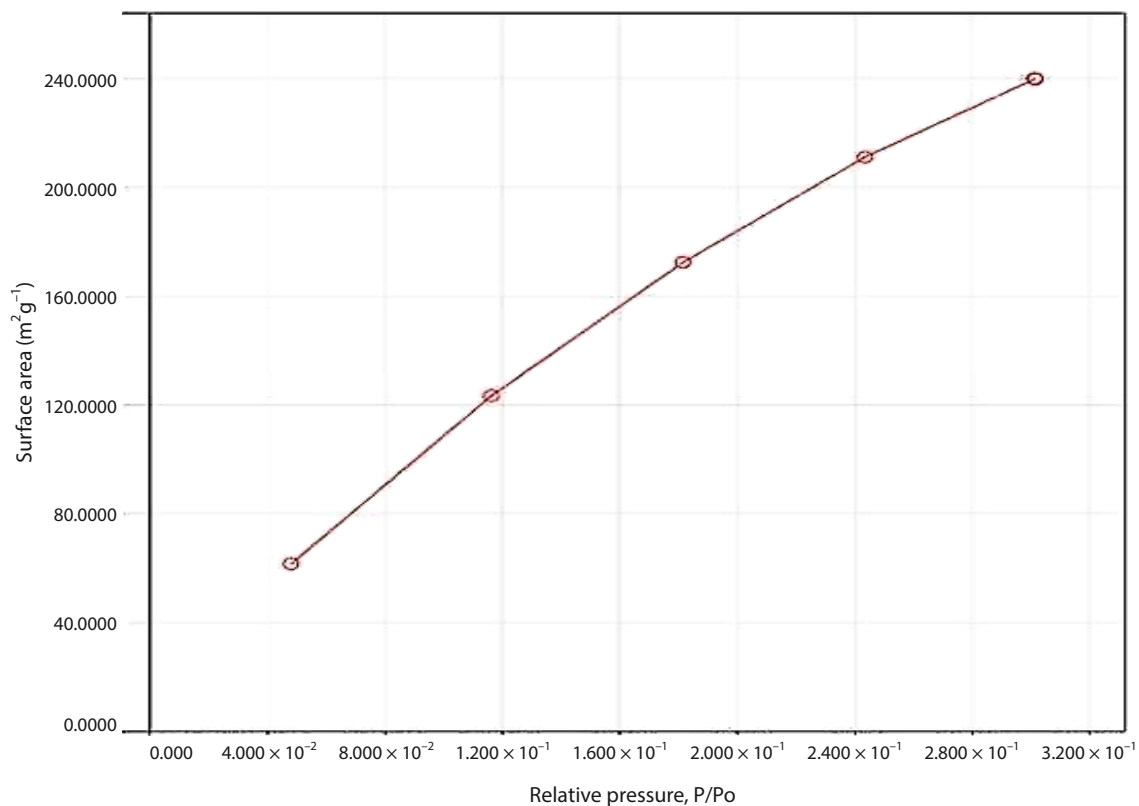


Figure 4. BET plot of the CaO-based heterogeneous cow bone catalyst.

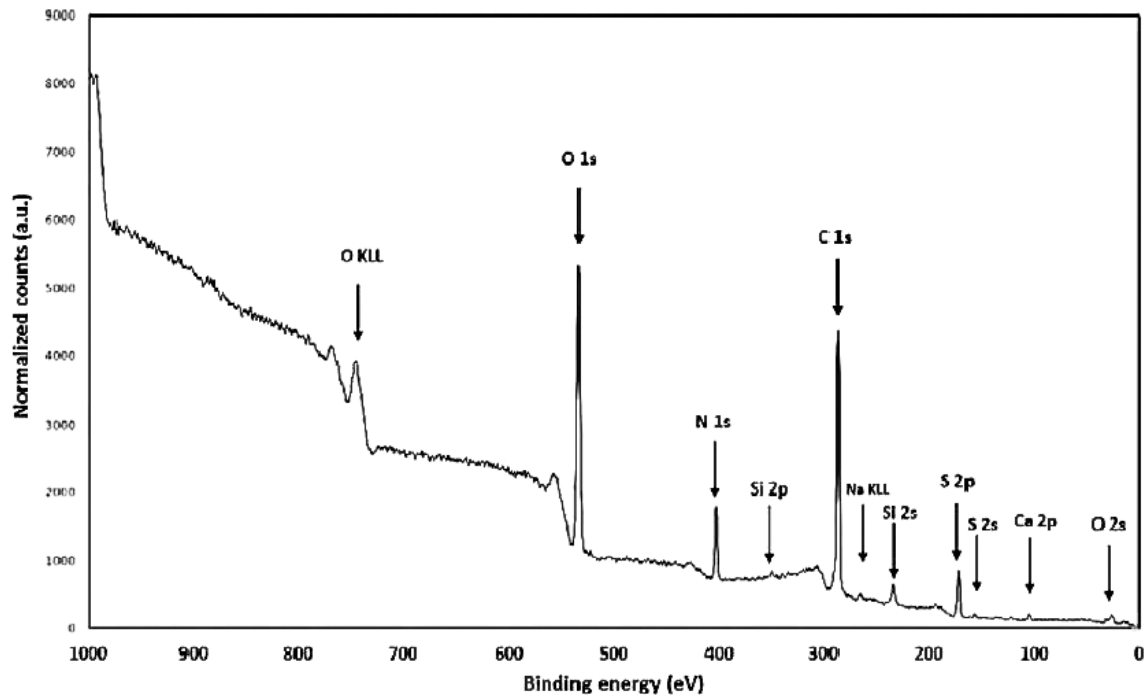
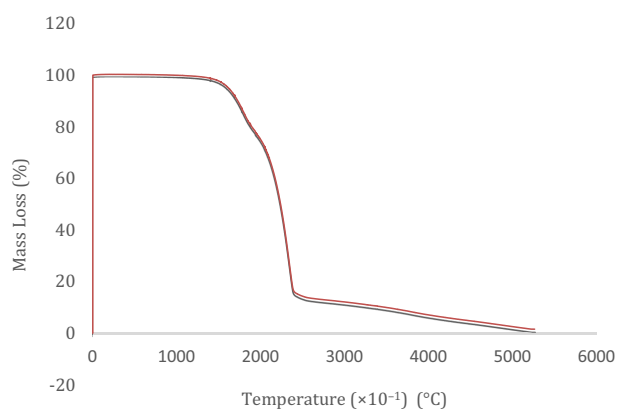


Figure 5. XPS spectra of the CaO-based heterogeneous cow bone catalyst.



**Figure 6.** TGA plot of the CaO-based heterogeneous cow bone catalyst.

### 3.2.1 Optimization Process

The results of the cow bone heterogeneous catalyst are presented in Tab. 5. The experimental percentage yields obtained

for all the experimental runs depicted close ranges with the predicted results. To this end, whenever the predicted percentages of yields are closer to the percentage experimental yields, it has become more than apparent that the statistical model development seeking to establish valid correlations between the operating parameters (time, temperature, agitation speed, and catalyst concentration) is said to be reliable [28]. However, it is deduced that operating parameters have an impact on the percentage yields of biodiesel produced from *Moringa oleifera* seed oil.

According to Sharma and Singh [29], optimized operating parameters increase the biodiesel yield. It is also observed that biodiesel yields from *Moringa oleifera* seed oil decrease with higher catalyst concentrations. This trend is attributed to the fact that the higher the amount of catalyst, the more triglycerides react with the catalyst thereby forming soap, subsequently leading to lower yields [30]. Additionally, the higher the amount of catalyst, the higher the pH of the methyl ester produced. This may lead to accumulation of deposited catalyst in diesel engines [31]. The ranges for percentage experimental yields recorded of the sixteen points factorized were 20.00–68.00%. At a combination of 180 min, 350 °C, 250 rpm, and

**Table 5.** CCRD experimental matrix depicting observed and predicted yields.

Run	Factor 1 ( $X_1$ )	Factor 2 ( $X_2$ )	Factor 3 ( $X_3$ )	Factor 4 ( $X_4$ )	Actual yield [%]	Predicted yield [%]	Difference [%]
1	-1	-1	-1	-1	48.90	47.92	0.98
2	+1	-1	-1	-1	55.00	56.83	1.83
3	-1	+1	-1	-1	56.00	57.33	1.33
4	+1	+1	-1	-1	59.00	58.25	0.75
5	-1	-1	+1	-1	30.40	33.33	2.93
6	+1	-1	+1	-1	51.70	52.25	0.55
7	-1	+1	+1	-1	50.00	50.25	0.25
8	+1	+1	+1	-1	63.00	61.17	1.83
9	-1	-1	-1	+1	33.00	37.50	4.5
10	+1	-1	-1	+1	57.00	55.92	1.08
11	-1	+1	-1	+1	55.00	52.92	2.08
12	+1	+1	-1	+1	64.00	63.33	0.67
13	-1	-1	+1	+1	20.00	19.92	0.08
14	+1	-1	+1	+1	47.00	48.33	1.33
15	-1	+1	+1	+1	42.00	42.83	0.83
16	+1	+1	+1	+1	64.00	63.25	0.75
17	$-\alpha$	0	0	0	37.00	33.92	3.08
18	$+\alpha$	0	0	0	62.00	63.25	1.25
19	0	$-\alpha$	0	0	51.00	46.42	4.58
20	0	$+\alpha$	0	0	68.00	70.75	2.75
21	0	0	$-\alpha$	0	61.00	60.42	0.58
22	0	0	$+\alpha$	0	47.00	45.75	1.25

Table 5. Continued.

Run	Factor 1 ( $X_1$ )	Factor 2 ( $X_2$ )	Factor 3 ( $X_3$ )	Factor 4 ( $X_4$ )	Actual yield [%]	Predicted yield [%]	Difference [%]
23	0	0	0	$-\alpha$	46.00	44.25	1.75
24	0	0	0	$+\alpha$	36.00	35.92	0.08
25	0	0	0	0	54.00	46.00	8
26	0	0	0	0	47.00	46.00	1
27	0	0	0	0	38.00	46.00	8
28	0	0	0	0	46.00	46.00	0
29	0	0	0	0	40.00	46.00	6
30	0	0	0	0	51.00	46.00	5

3 wt %, maximum yield was obtained. Also, temperature and time have a positive influence on the percentage yield of biodiesel produced, contrary to the findings of Alhassan et al. [32]. This variation in the effect of operating parameters is attributed to the use of different seed oils. Hence, optimum methyl ester yields at elevated temperatures are mainly due to the degradation of *Moringa oleifera* seed oil viscosity, thereby increasing the rate of the reaction while at the same time improving the yield [33].

Eterigho et al. [9] developed and applied heterogeneous catalysts for direct cracking of triglycerides for biodiesel production using the same clay material and reported 59.00 % conversion of methyl ester for a solvent-free catalyst. The difference recorded in the optimum yield of methyl ester (9.00 %) can be attributed to the fact that the latter only utilized temperature and time whereas this study involved time, temperature, agitation speed, and catalyst concentration in addition to modifying the heterogeneous catalyst produced with cow bone for optimizing the methyl ester yield. The use of a heterogeneous catalyst developed from cow bone ultimately led to an increase in the biodiesel yield.

### 3.2.2 Analysis of Variance

Analysis of variance (ANOVA) on the optimization of biodiesel production from *Moringa oleifera* seed oil using cow bone oriented heterogeneous catalyst was employed mainly for estimating the effects of the major reaction parameters relative to the methyl ester yield. Tab. 6 depicts the response surface reduced quadratic equation based on partial statistical sum of squares – type III. According to Noordin

et al. [34], the partial statistical sum of squares is the sum of squares relative and corresponding to each and every effect adjusted for each and every other effect in the statistical equation selected after a reproducibility test of the equation often suggested by the software used (Design Expert 7.0.0) and relatively compared with the results obtained. The terms that appear insignificant in the equation are often corrected automatically via backward(s) cum stepwise(s) correction equation.

From the results obtained, it is statistically clear that the four reaction variables under study (time, temperature, agitation speed, and catalyst concentration) cum the statistical equation appear to be statistically significant  $p < 0.01$ . In statistics, a  $p$ -value less than 0.05 is a clear indication that the model terms

Table 6. Analysis of variance (ANOVA).

Source	Sum of squares	Df	Mean square	F-value	p-value	Remark
Model	3408.20	14	243.44	12.78	< 0.0001	Significant
A Time	1290.67	1	1290.67	67.77	< 0.0001	
B Temp.	888.17	1	888.17	46.64	< 0.0001	
C Agitation	322.67	1	322.67	16.94	0.0009	
D Catalyst conc.	104.17	1	104.17	5.47	0.0336	
AB	64.00	1	64.00	3.36	0.0867	
AC	100.00	1	100.00	5.25	0.0368	
AD	90.25	1	90.25	4.74	0.0459	
BC	56.25	1	56.25	2.95	0.1062	
BD	36.00	1	36.00	1.89	0.1894	
CD	9.00	1	9.00	0.47	0.5023	
A <sup>2</sup>	11.44	1	11.44	0.60	0.4503	
B <sup>2</sup>	271.44	1	271.44	14.25	0.0018	
C <sup>2</sup>	86.01	1	86.01	4.52	0.0506	
D <sup>2</sup>	60.01	1	60.01	3.15	0.0962	
Residual	285.67	15	19.04			
Lack-of-fit	95.67	10	9.57	9.57	0.9697	Not significant



are most significant, and the interaction between all the operating parameters is found to be more important [33]. The major effects of the variables (time, temperature, agitation speed, and catalyst concentration) depict significances of  $p < 0.01$ . Hence, the relationship of the variables under study in the prediction of the linearity of independent factors and their interactions in surface response is represented by a second-order polynomial equation (Eq. 1) [35].

$$Y = \alpha_0 + \sum_{i=1}^3 \alpha_i x_i + \sum_{i=1}^3 \alpha_{ii} x_i^2 + \sum_{i < j=1}^3 \alpha_{ij} x_i x_j \quad (1)$$

where  $Y$  is the biodiesel yield,  $\alpha_0$  is the intercept,  $x_i$  and  $x_j$  are uncoded independent variables, and  $\alpha_i$ ,  $\alpha_{ii}$ , and  $\alpha_{ij}$  denote linear, quadratic, and interaction constant coefficients, respectively.

### 3.2.3 Effect of Operating Parameters on Biodiesel Yield

From the ANOVA presented in Tab. 7, it is deduced that time, temperature, agitation speed, and catalyst concentration are significant independent variables. Also, the interaction of reaction time and temperature ( $AB$ ), reaction time and agitation speed ( $AC$ ), reaction time and catalyst concentration ( $AD$ ), temperature and agitation speed ( $BC$ ), temperature and catalyst concentration ( $BD$ ), and agitation speed and catalyst concentration ( $CD$ ) are significant for the first-order polynomial while reaction time, reaction temperature, agitation speed, and catalyst concentration are significant for the second-order polynomial. It has become more than apparent that all variables have an effect on biodiesel yield. For a variable to be significant in first-order and second-order polynomials, it does not statistically signify ascending or descending effects on the yield of methyl ester, instead it only signifies that whenever such a variable is intensified and vice versa, a corresponding increase or decrease is expected in the outcome. It is conveniently deduced from the analysis of statistical results that the process variables and their interaction effects have varying impacts on the yield biodiesel produced from *Moringa oleifera* seed oil using a cow bone oriented heterogeneous catalyst.

**Table 7.** Summary of the developed type III polynomial model.

Standard deviation	Mean	C. V. %	PRESS
4.36	49.27	8.86	824.64
$R^2$	Adj. $R^2$	Pred. $R^2$	Adeq. precision
0.9227	0.8505	0.7768	16.473

For ascertaining the independent and interactive contributions of the variables under study (time, temperature, agitation speed, and catalyst concentration), the linear statistical response model is used for both values (coded and actual) of the methyl ester yield. Eqs. (2) and (3) depict the response equation in terms of coded and actual factors, respectively. The linear statistical models also indicate the coefficients of all the variables in the linear regression equations, their statistical

significances, and their corresponding impacts on the yield of biodiesel from *Moringa oleifera* seed oil. According to Razali et al. [36], the positive and/or negative signs attached to individual terms and their interactions are indications of synergistic and antagonistic effects on the methyl ester yield.

$$\begin{aligned} \text{Yield} = & 46.00 + 7.33A + 6.08B - 3.67C - 2.08D - 2.00AB \\ & + 2.50AC + 2.38AD + 1.88BC + 1.50BD - 0.75CD + 0.65A^2 \\ & + 3.15B^2 + 1.77C^2 - 1.48D^2 \end{aligned} \quad (2)$$

$$\begin{aligned} \text{Yield} = & +220.58333 - 0.10278t - 0.66500T - 0.72000A \\ & + 4.08333CC - 6.66667 \times 10^{-4}tT + 8.3333 \times 10^{-4}tA + 0.039583tCC \\ & + 7.0000 \times 10^{-4}TA + 0.030000TCC - 0.015000ACC \\ & + 1.79398 \times 10^{-4}t^2 + 1.25833 \times 10^{-3}T^2 + 7.08333 \times 10^{-4}A^2 \\ & - 1.47917CC^2 \end{aligned} \quad (3)$$

where  $t$  is the time,  $T$  is the temperature,  $A$  stands for agitation, and  $CC$  is the catalyst concentration.

### 3.2.4 Reliability of the Statistical Model for the Optimization

To determine the reliability of the statistical model, the coefficient of determination, otherwise known as  $R^2$  value, is paramount because it indicates the fitness of the linear regression model developed [32]. As a rule of thumb, the closer the  $R^2$  value to unity, the higher the precision of the model [35]. It is deduced from the  $R^2$  value that 92.27 % of the methyl ester produced has been accounted for by the linear regression equation generated while 7.73 % of the entire variations experienced on the response of the biodiesel produced could not be explained by the linear regression equation generated. Additionally, the reliability of the linear regression model is ascertained by the values of standard deviation (4.36), the mean (49.27), and the adjusted  $R^2$  value (0.8505).

All the statistical information gathered was employed for predicting the optimum response of the biodiesel produced. In sum, the outcomes have shown that the optimal response of biodiesel obtained for the experimental result was  $98.00 \pm 2.00$  % at a confidence level of 95.00 %. The predicted  $R^2$  of 0.7768 (Tab. 7) is in reasonable agreement with the adjusted  $R^2$  of 0.8505. Also, adequate precision statistically measures the signal-to-noise ratio ( $S/N$ ). A ratio of 4.00 is desirable but an adequate precision of 16.473 is an indication of an adequate signal. Hence, the polynomial model developed can be employed in navigating the design space therein.

## 3.3 Characterization of the Developed CaO-Based Heterogeneous Catalyst

### 3.3.1 FTIR Analysis

The FTIR spectra of the produced biodiesel are displayed in Fig. 7. This is mainly to classify the unknown compounds

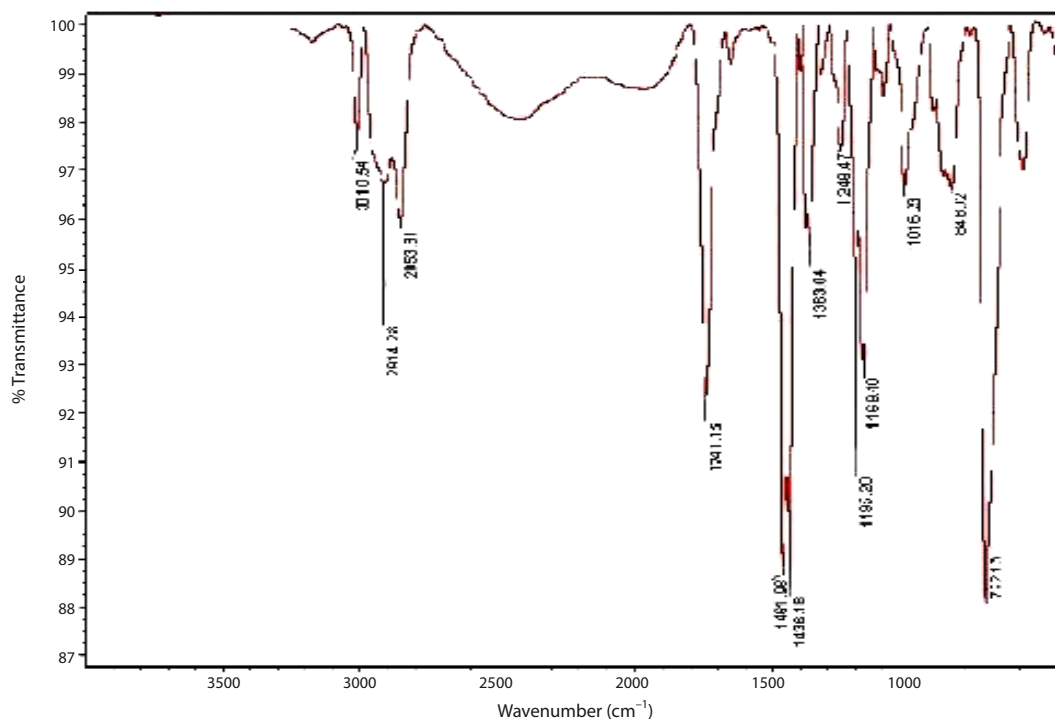


Figure 7. FTIR spectra of *Moringa oleifera* biodiesel using the CaO-based heterogeneous catalyst.

through identifying their functional groups. The IR spectrum appeared to lie between approximately 4000 and 500  $\text{cm}^{-1}$ . The IR spectrum obtained is segmented into two. The first is the left segment, i.e., between 2000  $\text{cm}^{-1}$  and above, which is often made up of peaks that are usually and relatively few in numbers with significant analytical information. The second segment is made up of numerous peaks in the range of 2000 to 500  $\text{cm}^{-1}$ . However, the peaks identified in both segments were ascertained via comparisons with established spectrums. Fig. 7 also demonstrates the

existence of C-CH<sub>3</sub>, CH<sub>2</sub>, CH<sub>3</sub>, C-O, CH=CH *trans*, and CH=CH *cis*, respectively.

### 3.3.2 GC-MS Analysis

GC-MS analysis was primarily employed to determine the methyl ester groups present in the biodiesel produced using a CaO-based heterogeneous catalyst developed from cow bone. Fig. 8 depicts the GC-MS analysis of biodiesel produced from

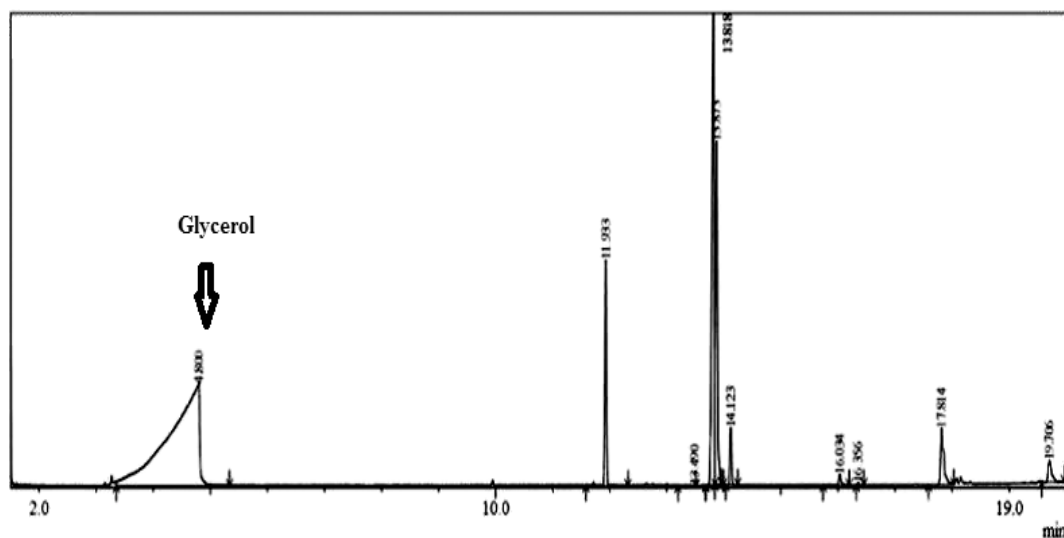


Figure 8. GC-MS analysis of *Moringa oleifera* biodiesel using the CaO-based heterogeneous catalyst.

*Moringa oleifera* seed oil. Four characteristic peaks for major fatty acid methyl esters (FAMES) that appear by the retention times and fragmentation pattern data of GC-MS analysis were observed. The FAME peaks identified are C16:1, C18:0, C18:1, and C18:2, respectively, i.e., palmitoleic acid methyl ester, stearic acid methyl ester, oleic acid methyl ester, and linoleic acid methyl ester.

The physicochemical properties of the synthesized biodiesels were evaluated to meet ASTM standards defined by international standards for the compatibility to diesel engines. The important properties presented in Tab. 8 were investigated to determine the quality of the synthesized *Moringa oleifera* seed oil and compared with the ASTM 6751 and other values reported in literature. The acid value of the biodiesel produced is an important parameter that determines the quality of the biodiesel obtained from *Moringa oleifera* seed oil using a cow bone oriented heterogeneous catalyst. The decrease in acid value recorded from the virgin oil stood at 98.50 %  $\text{mg}_{\text{KOH}}\text{g}^{-1}$ . This indicates a good transesterification process by the catalyst developed. The main properties to mitigate the fuel atomization difficulties are density and viscosity that were determined and found within acceptable limits.

## 4 Conclusion

A cow bone-derived catalyst was developed and applied as active heterogeneous catalyst for biodiesel production from *Moringa oleifera* seed oil. XRF analysis showed the presence of elements and compounds of interest (Al, Si, and  $\text{SO}_3$ ) at a ratio of 1:5:4, respectively, with approximate compositions of the elements increasing in the order  $\text{Si} > \text{P} > \text{Cl} > \text{Al} > \text{Na} > \text{Mg}$ , with Si being the main element. The FTIR spectra of the heterogeneous catalyst contained several adsorption peaks with a highly sig-

nificant broad band at  $3628\text{ cm}^{-1}$  representing the OH stretching vibrations of the hydroxyl groups in the brucite-like sheets, with the absorption peak at approximately  $3628\text{ cm}^{-1}$  confirming the presence of the availability of many different bonded oxides of Fe, Mn, Cr, V, Ti, K, P, Al, Na, and S.

The SEM image of the heterogeneous catalyst was characterized by large micropores and particle agglomeration. The BET of the cow bone CaO-based heterogeneous catalyst recorded a surface area of  $392.740\text{ m}^2\text{g}^{-1}$ . The catalyst was capable of converting up to a maximum of 68.00 wt % of biodiesel yield, under the parametric optimized conditions of 180 min,  $350\text{ }^\circ\text{C}$ , 250 rpm, and 3 wt %, which by prediction attained 70.75 wt %. Therefore, it can be concluded that this abundant raw material (cow bone) is useful in the development of catalysts for biodiesel production.

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**Table 8.** Physicochemical properties of the produced biodiesel.

S/N	Properties	Exp.	ASTM	Diesel	Singh [28]	Ten [37]	Amenaghawon [38]
1	Density at $15\text{ }^\circ\text{C}$ [ $\text{g cm}^{-3}$ ]	0.790	0.900 <sub>max</sub>	0.800	0.862	0.858	0.866
2	Total glycerine [%]	3.010	0.240 <sub>max</sub>	–	0.12		
3	Viscosity at $40\text{ }^\circ\text{C}$ [ $\text{mm}^2\text{s}^{-1}$ ]	4.400	6.00 <sub>max</sub>	16.50 <sub>max</sub>		5.1	5.25
4	Acid value [% $\text{mg}_{\text{KOH}}\text{g}^{-1}$ ]	0.380	0.500	–			0.12
5	Iodine value [g of I per 100 g oil]	96.00	–	–			84.21
6	Sulfated ash [wt %]	0.030	0.050 <sub>max</sub>	0.300 <sub>max</sub>	0.03	0.03	
7	Cetane number [–]	58.60	47.00 <sub>min</sub>	51.00	48	51	55.49
8	Cloud point [ $^\circ\text{C}$ ]	10.00	–3 to 12	–14.50	3.0	18	
9	Flash point [ $^\circ\text{C}$ ]	176.0	130 <sub>min</sub>	166 <sub>max</sub>	140	171	158
	Pour point [ $^\circ\text{C}$ ]	–7.00	–15 to 10	–			
10	Saponification value [ $\text{mg}_{\text{KOH}}\text{g}^{-1}$ ]	104.00	–	–	6.0	5	
11	Calorific value [MJ $\text{kg}^{-1}$ ]	44.00	–	–	39.01	38.1	39.17
12	Total glycerine [%]	3.010	0.240 <sub>max</sub>	–	0.12		

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