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An Optimization Approach to Oil Extraction from Chrysophyllum Albidium Oilseeds and Its Quality Characterization

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Adepoju, T. F.
Chemical Engineering Department, Landmark University, Omu-aran, P.M.B. 1001, Kwara State, Nigeria

Ojediran, J. O.
Agric & Biosystem Engineering Department, Landmark University, Omu-aran, P.M.B. 1001, Kwara State, Nigeria

Okunola A.A.
Agric & Biosystem Engineering Department, Landmark University, Omu-aran, P.M.B. 1001, Kwara State, Nigeria

Abstract: *This work focused on optimization of oil extraction from Chrysophyllum albidium oilseeds using Response Surface Methodology (RSM), it also to investigate the physicochemical properties and fatty acid profile of the oil. Based on Box-Behnken design, 17 experimental runs were conducted to investigate the effects of extraction time, solvent volume sample weight and their reciprocal interactions on the oil yield. A quadratic polynomial was obtained to predict the oil yield and the ANOVA test showed the model to be significant ($p < 0.05$). A statistical model predicted the highest oil yield to be 24.402% (w/w), at the optimal condition of sample weight of 59.99 g, solvent volume of 167.57 ml and extraction time of 60 min. Using these optimal factor values in three independent replicates, an average oil content of 24.35% (w/w) was achieved, which was well within the range predicted by the model.The physicochemical properties oil extracted showed the oil to be yellow liquid at room temperature, the refractive index was found to be, 1.4246; the moisture content, 0.015%; specific gravity, 0.884; viscosity, 1.24 P; FFA, 1.25; acid value, 2.50; saponification value, 190.50 (mg KOH/g oil); peroxide value, 1.96 (meq O_2/kg oil); iodine value, 102.50 (g I_2/100g oil); higher heating value, 40.08 (MJ/kg) and cetane no, 51.89. The physicochemical analysis of oil suggested the oil could have important food and industrial applications. The fatty acid profile of the seed oil revealed that the oil was highly unsaturated (72.15%). In addition, the quality of oil extracted under the optimal condition revealed that the oil is edible and could serve as feedstock for many industrial applications.*

***Keywords**: Optimization, Chrysophyllum albidium, response surface methodology, fatty acid, physicochemical properties.*

1. Introduction:

Oilseed crops are vital sources of oils of nutritional, pharmaceutical and industrial importance. The characteristics of oils from different sources depend mainly on their compositions and no oil from single source can be suitable for all purposes (Ramadan and Mörsel, 2003). Presently, the quest for traditional vegetable oils has increased immensely because of the ever-growing World population and their use for industrial purposes. Several oils such as moringa oil, sunflower oil, rapeseed oil, palm oil, soybean oil, corn oil and pumpkin oil have been used for industrial purposes (Alcantara *et al*., 2000; Dorado *et al*., 2004; Mitra *et al*., 2009). New low-cost oilseed crops are needed to produce inexpensive oils suitable for food, pharmaceutical and industrial applications. One of the possible alternative crops is *Chrysophyllum albidium*, also known as White Star Apple. It isa forest fruit tree described by the Scottish botanist George Don. They are commonly found throughout tropical Africa. It is closely related to the African star apple (*Chrysophyllum africanum*) which is also common throughout West Africa. Some schools of thought feel that they may just be a variety of the same species(The National Academies Press. 2008). The *Chrysophyllum albidium* seed oil is rich in both linoleic (36.0%) and oleic (37.6 %) fatty acids (Ajewole, 1991). Ugboge and Akukwe. (2000) reported that there is a potential to use oils from non-utilized oil seeds inmanagement of wounds.

Numerous methods exist in oil separation from oilseeds such as mechanical pressing, pressurized solvent extraction, Soxhlet extraction, and ultra-sonic extraction, Aqueous Enzymatic Oil Extraction (AEOE), among others. Mechanical pressing is the most widely used but the oils produced with this method usually have low value. With extraction method using supercritical fluid such as CO_2, the oil produced has very high purity but for the high operating and investment cost. Extraction with solvent has a number of advantages, which include higher yield and less turbidity as well as relatively low operating cost. Previous studies showed that extraction with organic solvents have been one of the major approaches employed. Some of the recent work on oil extraction using solvent extraction technique include oils from *Washingtonia filifera* (Nehdi, 2011), *Moringa oleifera* (Rashid *et al*., 2011), bitter seed, pumpkin (*Cucurbita pepo* L.), Kalahari melon seed, kenaf and Sorrel (Nyam et al., 2009).

Response surface methodology (RSM) is a useful optimization tool, which has been applied in research to study the effect of individual variables and their interactions on response variables, Box and Wilson (1951). It has been used extensively on the optimization of extractions of edible and non-edible oils from different oil sources such as pumpkin, palm oil, silkworm pupae, *Vetiveria zizanioides*, locust bean, to mention but a few (Mitra et al., 2009; Danh et al., 2009; Tan et al., 2009; Akinoso and Raji, 2011). The major advantage of RSM is the ability to reduced number of experimental runs needed to arrive at optimized and statistically acceptable results (Akinoso and Raji, 2011). Thus, it saves time and less difficult compared with full-factorial design (Tan et al., 2009).

This study was aimed at oil separation from White star apple (*Chrysophyllum albidium*.) oilseeds via application of solvent extraction method. To optimize the extraction conditions for the process, RSM was applied to determine the effects of three-level-three factors and their reciprocal interactions on the oil extracted. In addition, the quality of the oil extracted was evaluated by carrying out physicochemical analysis with a view to determining its potential use.

2. Materials And Methods:

2.1. Materials:

Chrysophyllum albidium oilseed samples were collected from Omu-Aran market Area in Kwara State, Nigeria. The oilseeds had breakable pericarp, which were removed by breaking and the white star was then separated from the pericarp. The seeds were sun dried for 4-days. Separation of the chaffs from the oilseeds was carried out by winnowing. Finally, the cleaned oilseeds were milled into powder by grinding with a milling machine. All chemicals and reagents used for this work were of analytical grades.

2.2. Methods:

2.2.1. Experimental Design:

In this study, the Box-Behnken experimental design was employed in order to optimize the *Chrysophyllum albidium* oil extraction. The coded independent factors levels are presented in Table 1. Selected extraction parameters for the separation of oil from the *Chrysophyllum albidium* seeds were extraction time (X_1), solvent volume (X_2) and sample weight (X_3).

Factor	Symbol	Coded factor levels		
		-1	0	+1
Sample weight (g)	X_1	40	50	60

Solvent volume (ml)	X_2	150	200	250
Extraction time (min)	X_3	40	50	60

Table 1: Factors and their levels for Box-Behnken design.

A three-level-three-factors design was applied, which generated 17 experimental runs (Table 2). This included 6 factorial points, 6 axial points and 5 central points to provide information regarding the interior of the experimental region, making it possible to evaluate the curvature effect. Depicted in Table 2 also are the observed yields, the predicted yields and the residual values. The effects of unexplained variability in the observed response due to extraneous factors were minimized by randomizing the order of experiments.

2.2.2. Oil Extraction Procedure:

500-ml Soxhlet apparatus and n-hexane as solvent were used for this study. Initially, the apparatus was charged with a known weight (Table 2) of*Chrysophyllum albidium*oilseeds powder in a muslin cloth placed in a thimble of Soxhlet apparatus. A round bottom flask containing known volume of n-hexane was fixed to the end of the apparatus and a condenser was tightly fixed at the bottom end of the extractor. The whole set up was heated up in a heating mantle at temperature of 68 ^{0}C. The excess solvent in the oil was recycled by heating at temperature of 70 ^{o}C after the extraction. Quantity of oil extracted was determined gravimetrically. The oil yield was evaluated as the ratio of the weight of the extracted oil to the weight of the *Chrysophyllum albidium* oilseed powder sample (Eq.1).The oil obtained was stored appropriately for further processing.

Std. run	X_1	X_2	X_3	OOY% (w/w)	POY % (w/w)	RES.
1	-1	-1	0	20.50	20.36	0.14
2	1	-1	0	22.50	22.41	0.088
3	-1	1	0	22.00	22.09	-0.088
4	1	1	0	22.40	22.54	-0.14
5	-1	0	-1	21.00	21.04	-0.038
6	1	0	-1	20.60	20.59	0.013
7	-1	0	1	21.00	21.01	-0012
8	1	0	1	24.00	23.96	0.038
9	0	-1	-1	19.95	20.05	-0.10
10	0	1	-1	23.00	22.88	0.12
11	0	-1	1	23.50	23.62	-0.12
12	0	1	1	22.75	22.65	0.10
13	0	0	0	23.50	23.68	-0.18
14	0	0	0	23.00	23.68	-0.68
15	0	0	0	23.40	23.68	-0.28
16	0	0	0	24.00	23.68	0.32
17	0	0	0	24.50	23.68	0.82

**OOY = Observed oil Yield, POY = Predicted oil Yield, RES. = Residual

Table 2: Experimental design matrix by Box-Behnken for three-level-three-factors response surface study

$$\%\ Oil\ yield = \frac{weight\ in\ gram\ of\ oil\ extracted}{weight\ in\ gram\ of\ Chrysophyllum\ albidium\ \text{oilseeds powder}} \qquad (1)$$

2.2.3. Statistical Data Analysis:

The data obtained from the *Chrysophyllum albidium*oilseeds extraction experiments were analysed statistically using response surface methodology, so as to fit the second-order mathematical model generated by the Design-Expert software version 8.0.3.1 (Stat-Ease Inc., Minneapolis, USA). To correlate the response variable to the independent variables, multiple regressions was used to fit the coefficient of the polynomial model of the response. The quality of the fit of the model was evaluated using test of significance and analysis of variance (ANOVA). The fitted second-order mathematical model is described in Eq. 2.

$$Y = b_0 + \sum_{i=1}^{k} b_i X_i + \sum_{i=1}^{k} b_{ii} X_i^2 + \sum_{i<j}^{k} b_{ij} X_i X_j + e \qquad (2)$$

Where, Y is response factor (*Chrysophyllum albidium* oil yield), b_o is the intercept value, b_i (i = 1, 2, k) is the first order model coefficient, b_{ij} is the interaction effect, and b_{ii} represents the quadratic coefficients of X_i, and e is the random error.

2.2.4. Physicochemical Analysis Of The Extracted Chrysophyllum Albidium Seed Oil:

The evaluation of the following physicochemical properties of the extracted seed oil were determined by the AOAC methods: refractive index, moisture content, viscosity, acid value, saponification value, peroxide value, specific gravity, % FFA, the mean molecular mass was obtained by the method of Akintayo and Bayer (2002), whereas the higher heating value was determined using the method of Demirbas (1998) and iodine value was obtained by Wijs method.

2.2.5. Analysis Of Fatty Acid Compositions Of Chrysophyllum Albidium Seed Oil:

Fatty acid profile of the *Chrysophyllum albidium* seed oil was determined using gas chromatography (HP 6890 powered with HP ChemStation Rev. A 09.01 [1206] Software). Oil sample (50 mg) was esterified for five minute at 95 ^oC with 3.4 ml of the 0.5 M KOH in dry methanol. The mixture was neutralized using 0.7 M HCl and 3 ml of 14% boron triflouride in methanol was added. The mixture was heated for 5 min at the temperature of 90 ^oC to achieve complete methylation process. The fatty acids were thrice extracted from the mixture with redistilled n-hexane. The content was concentrated to 1 µl for gas chromatography analysis and 1 µl was injected into the injection port of GC.

3. Results And Discussion:

3.1. Optimization Of Chrysophyllum Albidium Seed Oil Extraction:

Table 2 shows the coded factors considered in this study with observed values, predicted valuesas well as the residual valuesobtained. Design Expert 8.0.3.1 software was employed to evaluate and determine the coefficients of the full regression model equation and their statistical significance. Table 3 described the results of test of significance for every regression coefficient.Considering the large F-values and low corresponding p-values, all the model terms have very strong effects on the oil yield except X_1X_2 with $p > 0.05$ (Table 3). However, second-order term X_1^2 with F-value of 31.14 and p- value of 0.0008, was the most significant model term. In order to minimize error, all the coefficients were considered in the design. The results of the second-order response surface model fitting in the form of ANOVA are presented in Table 4. The model F-value of 15.63 with low p-value (0.0008) implied a high significance for the regression model (Yuan *et al.*, 2008). The goodness of fit of the model was checked by the coefficient of determination (R^2).

Source	Sum of squares	df	Mean Square	F-value	p-value
X_1	3.13	1	3.13	15.03	0.0061
X_2	1.71	1	1.71	8.23	0.0240
X_3	5.61	1	5.61	26.99	0.0013
X_1X_2	0.64	1	0.64	3.08	0.1228
X_1X_3	2.89	1	2.89	13.90	0.0074
X_2X_3	3.61	1	3.61	17.36	0.0042
X_1^2	6.47	1	6.47	31.14	0.0008
X_2^2	1.47	1	1.47	7.05	0.0327
X_3^2	2.63	1	2.63	12.64	0.0093

Table 3: Test of significance for all regression coefficient terms

Guan and Yao (2008) reported that an R^2 should be at least 0.80 for the good fit of a model. In this case, the R^2 value of 0.9526 indicated that the sample variation of 95.26% for the oil extraction is attributed to the independent factors (sample weight, solvent volume and extraction time) and only 4.74% of the total variations are not explained by the model. The value of the adjusted determination coefficient (Adj. R^2 of 0.8916) was also very high, supporting a high significance of the model (Akhnazarova and Kefarov, 1982; Khuri and Cornell, 1987) and all p-values were less than 0.05 except X_1X_2 (sample weight-solvent volume), implying that the model proved suitable for the adequate representation of the actual relationship among the selected factors. The lack-of-fit term of 0.11 was not significant relative to the pure error. In this case, a non-significant lack of fit is good. Hence, the model could be used in theoretical prediction of the oil extraction. The developed regression model describing the relationship between the oil yield (Y) and the coded values of independent factors of sample weight (X_1), solvent volume (X_2) and extraction time (X_3) and their respective interactions is described in Eq. (3).

$$Y = 23.68 + 0.63X_1 + 0.46X_2 + 0.84X_3 - 0.40X_1X_2 + 0.85X_1X_3 - 0.95X_2X_3 – 1.24X_1^2 - 0.59X_2^2 - 0.79X_3^2 \quad (3)$$

The model coefficients and probability values i.e. coded value are shown in Table 5. The low values of standard error observed in the intercept and all the model terms showed that the regression model fits the data well, and the prediction is good (Table 5). The variance inflation factor (VIF) obtained in this study showed that the centre points are orthogonal to all other factors in the model (Table 5). The model also proved suitable for the adequate representation of the real relationship among the selected independent factors.

Figure 1a shows the response surface plot representing the effect of solvent volume, sample weight and their reciprocal interaction on oil yield while keeping extraction time constant at zero level. The results revealed that lowsolvent volume and high sample weight favoured oil yield while decreasing in both variables led to low oil yield. Response surface plot describing the effect of extraction time, sample weight and their reciprocal interaction on oil yield while keeping solvent volume constant at zero level is depicted in Figure 1b. It was observed that high oil yield was observed at the high extraction time andhigh sample weight; the reverse resulted into only marginal decrease in the oil yield. The combination of high extraction time and high solvent volume did not significantlyincrease the oil yield. However, the high oil yield was observed at low solvent volume and high extraction time. The curvatures nature of the surface plots in Figure 1(a and b) indicate mutual interactions between sample weightand solvent volume and, between sample weight andextraction time, respectively (Rashid et al., 2011). Figure 1c shows the response surface plot of the effect of solvent volume, extraction time and their reciprocal interaction on oil yield while sample weight constant at zero level. The optimal values of the independent factors selected for the extraction process were obtained by solving the regression equation (Eq. 3) using the Design-Expert software package. The optimal condition was established as sample weight of 59.99 g, solvent volume of 167.57 ml and extraction time of 60 min.The predicted oil yield under the optimal condition was Y = 24.402% (w/w).

Source	Sum of squares	df	Mean square	F-value	p-value
Model	29.24	9	3.25	15.63	0.0008

Residual	1.46	7	0.21		
Lack of fit	0.11	3	0.036	0.11	0.9520
Pure error	1.35	4	0.036		
Cor total	30.70	16			

R^2 = 95.26%, R^2 (adj) = 89.16%

Table 4: Analysis of variance (ANOVA) of regression equation

Fact.	Coefficient estimate	df	Standard error	95% CI low	95% CI high	VIF
Intercept	23.68	1	0.20	23.20	24.16	-
X_1	0.63	1	0.16	0.24	1.01	1.00
X_2	0.46	1	0.16	0.081	0.84	1.00
X_3	0.84	1	0.16	0.46	1.22	1.00
X_1X_2	-0.40	1	0.23	0.31	1.39	1.00
X_1X_3	0.85	1	0.23	0.31	1.39	1.00
X_2X_3	-0.95	1	0.23	-1.49	-0.41	1.00
X_1^2	-1.24	1	0.22	-1.77	-0.71	1.01
X_2^2	-0.59	1	0.22	-1.12	-0.065	1.01
X_3^2	-0.79	1	0.22	-1.32	-0.26	1.01

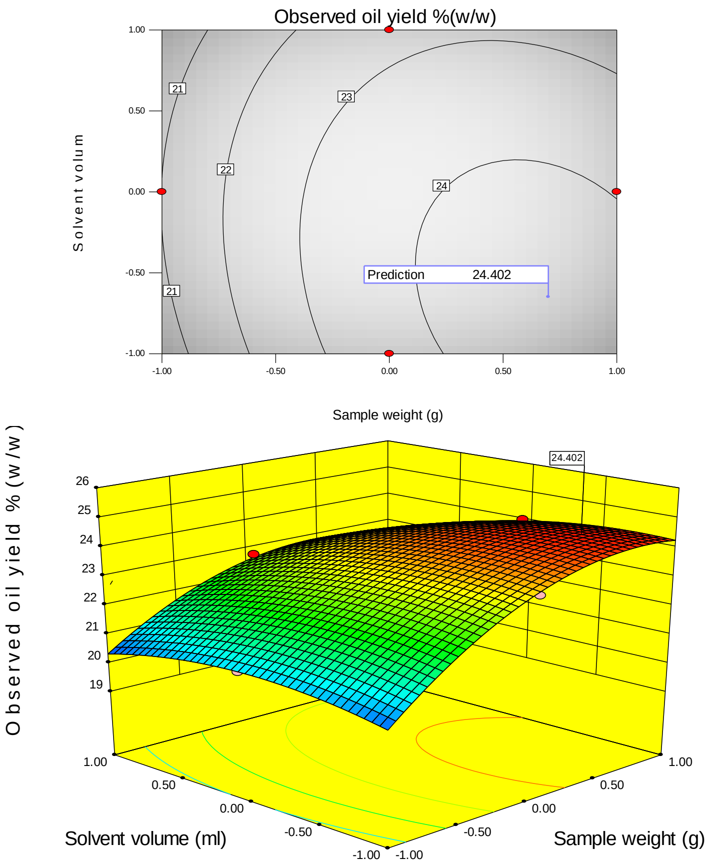
Table 5: Regression coefficients and significance of response surface quadratic

To verify the prediction of the model, three independent replicates experiment was carried out and the average oil yield obtained was 24.35% (w/w), which was well within the predictable value of the model equation. The results of this study demonstrate that RSM with appropriate experimental design can be effectively applied to the optimization of the process factors in oil extraction work.

3.2. Quality Characterization Of Chrysophyllum Albidium Seed Oil:

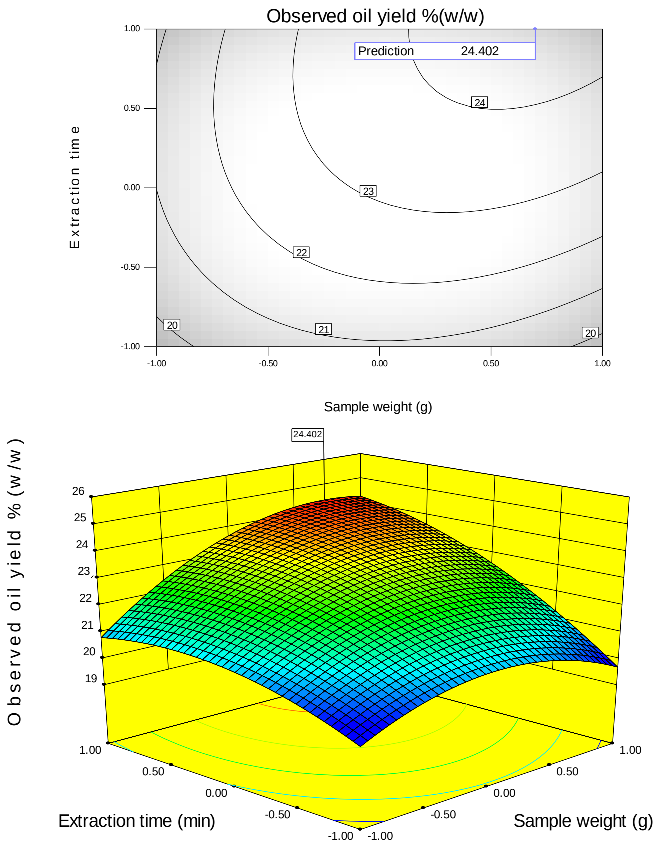
3.2.1. Physical Properties Of The Chrysophyllum Albidiumseed Oil:

To characterize the quality of the *Chrysophyllum albidium* seed oil extracted in this work, the oil was subjected to physicochemical analysis and the results obtained are presented in Table 6. At room temperature, the seed oilwas liquid yellowin colour with refractive index and moisture content of 1.4246 and 0.015% (wet.b), respectively. Observations on the colour and



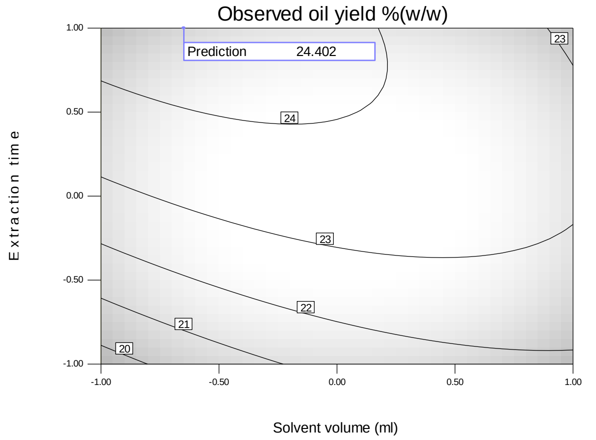
Observed oil yield %(w/w)
Solvent volum
Sample weight (g)
Prediction 24.402
Observed oil yield % (w/w)
Solvent volume (ml)
Sample weight (g)
24.402

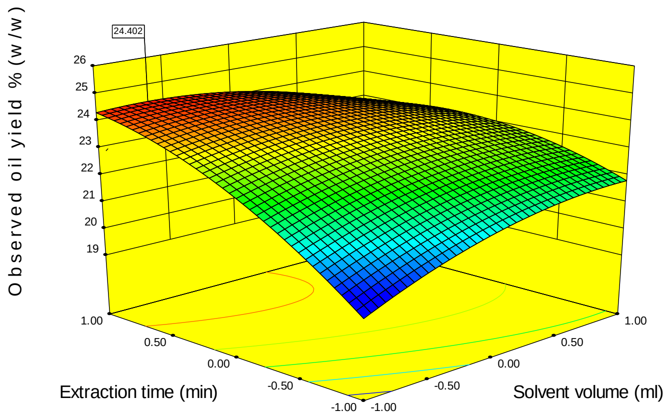
(a)



Observed oil yield %(w/w)
Extraction time
Sample weight (g)
Prediction 24.402
Observed oil yield % (w/w)
Extraction time (min)
Sample weight (g)
24.402

(b)





(c)

Figure 1 :(a-c): Surface plots for solvent extraction ofChrysophyllum albidium seed oil.

the refractive index of the oil agreed with previous published reports (Ajewole and Adeyeye 1991; Audu et al., 2013). The specific gravity of the seed oil was determined as 0.884 and the viscosity, which is a measure of the resistance of oil to shear, was 1.24 (P). These values are within the ranges reported for *Chrysophyllum albidium* seed oil (Audu et al., 2013).

3.2.2. Chemical Properties Of Chrysophyllum Albidium Seed Oil:

Among the most important characteristics used to determine the present condition and quality of oil samples are their chemical properties. Table 6 contains results obtained for the chemical properties of the *Chrysophyllum albidium*seed oil. Low FFA content (1.25%) of seed oil obtained in this study is indicative of the good resistance of this oil to hydrolysis. Ajewole and Adeyeye (1991) observed 0.85% FFA for the seed oil in their work while Audu et al., 2013 reported an FFA of 1.79% for the same oil. The low acid value (1.25 mg KOH/g oil) of this oil showed that it is not only edible but could also have a long shelf life. Ajewole and Adeyeye (1991) reported seed oil acidity of 1.7% while Audu *et al*. (2013) observed a very high acid value of 2.87 mg KOH/g oil for the same oil. These variations may be due to different cultivars used. A high saponification value of 190.50 (mg of KOH/g of oil) was obtained for the *Chrysophyllum albidium*seed oil, indicating high concentration of triglycerides. This value is closed to the result (193.7 mg KOH/g oil) reported by Audu *et al*. (2013). The iodine value of the seed oil was high (102.50 g of I_2/100 g of oil), showing that the oil contained a substantial level of unsaturation. The peroxide and p-anisidine values measure hydroperoxides and secondary oxidation products, i.e. aldehydes, of oils, respectively (Bockisch, 1998). The peroxide value obtained for the seed oil in this study was 2.60 milli-equivalent of peroxide/kg of oil, which is a low value. A peroxide value range of 1.96 milli-equivalent of peroxide/kg of oil has been earlier reported for seed oil (Audu *et al*., 2013). A p-anisidine value of 5.20 of the seed oil suggests the presence of significant amounts of secondary oxidation products in the seed oil. The combination of high iodine value and low peroxide value suggested the oil could be stored for a long period without deterioration. These also demonstrated the oil possessed the desirable qualities of edible oils. The Higher Heating Value (HHV) and cetane number determined for the seed oil are40.08 MJ/kg and 51.89, respectively. The value of HHV was within the range earlier reported by Demirbas (1998) for vegetable oils (37.47 – 40.62 MJ/kg). Hence, the physicochemical characteristics of the oil showed that the *Chrysophyllum albidium* seed oil is a good candidate for use as edible oil and as an industrial feedstock.

3.2.3. Fatty Acid Profile Ofchrysophyllum Albidiumseed Oil:

Gas chromatography analysis of fatty acids present in the seed oil is shown in Table 7. The results indicated that the oil was highly unsaturated. The major fatty acids present in the seed oil were linoleic, $C_{18:2}$; (38.09%), oleic, $C_{18:1}$; (34.06%), palmitic, $C_{16:0}$; (19.24%); stearic, $C_{18:2}$; (6.70%) and other trace fatty acids (1.91%). The total unsaturated fatty acid composition of the oil was 72.15%. Although this result followed the trend of reported fatty acid compositions for *Chrysophyllum albidium*seed oil, it has been observed that the quantity of each acid present in this seed oil varies considerably among the different cultivars studied (Ajewole and Adeyeye 1991; Audu *et al*., 2013).

Parameters	Mean values
Physical properties	
Physical state at 28 ^{o}C	Yellowin colour
Refractive index at 25 ^{o}C	1.4246
Moisture content (%. wet.b)	0.015
Specific gravity	0.884
Viscosity (P) at 40 ^{o}C	1.24
Chemical properties	
%FFA (as oleic acid)	1.25
Acid value (mg KOH/g oil)	2.50
Saponification value (mg KOH/g oil)	190.50
Iodine value (g I_2/100g oil)	102.50
Peroxide value (meq O_2/kg oil)	1.96
Higher heating value (MJ/kg)	40.082
Other properties	
Cetane number	51.89

Table 6: Physicochemical and other characteristics of seed oil

Parameters	Compositions %
Palmitic acids (C16:0)	19.24
Stearic acids (C18:0)	6.70
Oleic acids (C18:1)	34.06
Linoleic acids (C18:2)	38.09
Others	1.91

Table 7: Fatty acids compositions of the seed oil

4. Conclusions:

From the Box-Behnken design, a statistical model predicted the highest oil yield to be 24.402% (w/w), at the optimal condition of sample weight of 59.99 g, solvent volume of 167.57 ml and extraction time of 60 min. Using these optimal factor values in three independent replicates, an average oil content of 24.35% (w/w) was achieved, which was well within the range predicted by the model. The fatty acid profile of the seed oil revealed that the oil was highly unsaturated (72.15%). In addition, the quality of oil extracted under the optimal condition revealed that the oil is edible and could serve as feedstock for many industrial applications.

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