OPTIMIZATION OF SCREW PRESS EXTRACTION OF *CITRILLUS LANATUS* SEED OIL AND PHYSICOCHEMICAL CHARACTERIZATION

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ABSTRACT: *The aim of this study was to optimize the extraction of C. lanatus seed oil using a screw press. Response surface methodology (RSM) was employed to describe the effects of pressing temperature and restriction diameter on oil yield and residual oil content using a Doehlert design. The seed oil extracted was characterized to determine its quality. Results showed that the experimental data were adequately fitted into the second-order polynomial model. The pressing temperature and restriction diameter had a significant effect on the screw press performance. The optimum conditions within the experimental domain were the pressing temperature of 92 °C and restriction diameter close to 4 mm. Under these conditions, the oil yield was 38.79%. The colour of clarified oil was light yellow. Its physical and chemical properties come up to the required standard for edible oil. Therefore, C. lanatus seed oil extracted by screw press could have important applications in human nutrition.*

KEYWORDS: Citrullus Lanatus, Seed Oil, Screw Press, Oil Yield, Optimization.

INTRODUCTION

Citrullus lanatus (Thunb.) Matsum. & Nakai is one of the most widespread and commercialized oleaginous cucurbits in Côte d'Ivoire (Zoro Bi *et al*., 2006). *C. lanatus* varieties, mainly grown for oilseeds, are generally found in West Africa (Guner and Wehner, 2004). They are mainly grown in savannah regions, in pre-forest regions, and are well adapted to low input production systems. In West Africa, *C. lanatus* seeds are usually transformed into a paste that is used only to make a sauce called "pistachio sauce" (Zoro Bi *et al*., 2006). However, some studies suggested that *C. lanatus* seeds are rich in oil which contains a large amount of linoleic acid, an essential fatty acid (Gbogouri *et al*., 2011; Olaofe *et al*., 2012; Raziq *et al*., 2012). Therefore, the *C. lanatus* seed is a source of high quality edible oil.

This polyunsaturated oil has a high sensitivity to oxidation (Loukou *et al*., 2013). It therefore requires special extraction conditions. The edible vegetable oil is conventionally extracted using a mechanical press or chemical solvent. Although the solvent extraction gives high oil yield, the oil is not free of solvent residues (Smassel, 2013). Moreover, the high temperature level alters the polyunsaturated oil quality (Liauw, 2008). Faced with the challenge of food safety, the mechanical pressing is a promising alternative to solvent extraction. The main reason is that mechanical pressing, carried out at a relatively low temperature and without solvent, contributes to preserving the oil quality (Soetaredjo *et al*., 2008; Uquiche *et al*., 2008). Currently, the screw press is mainly used in oil mills, the use of hydraulic presses is limited to a certain type of raw materials (cocoa, olives). For this reason, the screw press extraction was chosen as a reference method.

ISSN 2056-5798(Print), ISSN 2056-5801(online)

This mechanical pressing process must be optimized to increase the oil yield. According to Ferchau (2000), the screw press performance can be affected by many factors including pressing temperature and restriction diameter. According to Jacobsen and Backer (1986), pressing efficiency is mainly defined by pressing capacity, oil flow, oil yield and/or residual oil content in the oilcake. To optimize a process by locating the optimum set of experimental conditions, then one must resort to designs for second-order models (response surface designs), which employ more than two factor levels to allow fitting of a full quadratic polynomial. A very useful experimental design for second-order models is the uniform shell design proposed by Doehlert (1970). Doehlert designs are easily applied to optimize variables (Nechar *et al*., 1995, Massart *et al*., 2003) and offer advantages in relation to central composite (Box and Wilson, 1951) and Box-Behnken (Box and Benhken, 1960) designs. They need fewer experiments, which are more efficient and can move through the experimental domain. Multivariate designs, which allow the simultaneous study of several control variables, are faster to implement and more cost-effective than traditional univariate approaches (Montgomery, 1997; Neto *et al*., 2001).

Ultimately, *C. lanatus* seed oil is of high quality, but it has a high sensitivity to oxidation. The mechanical pressing can preserve the quality of this oil, but it often gives a low oil yield. So, how to optimize this process to maximize his oil yield? To date, no research has been reported on optimization of screw press extraction of *C. lanatus* seed oil. In this work, response surface methodology (RSM) is applied to investigate the effects of pressing temperature and restriction diameter on the residual oil content and *C. lanatus* seed oil yield using screw press extraction.

MATERIALS AND METHODS

Materials and Chemicals

C. lanatus seeds were collected in May 2015 from farmers in the Dikodougou department (latitude 9°04'03.3"N, longitude 5°46'20.0"W) located in northern of Côte d'Ivoire. The seeds were sorted, soaked in lukewarm water for 15 min and then dehulled manually. The kernels were washed with tap water, dried in an oven (UFB 400, Memmert, Schwabach, Germany) at 60 °C for 2 h and then packed in black plastic bags. All chemicals and reagents used for this work were of analytical grades.

Experimental Design

The two-factor Doehlert (1970) design was used to optimize screw press extraction of *C. lanatus* seed oil. The independent variables selected were pressing temperature and restriction diameter. Factor levels were defined on the basis of preliminary experimental results as follows:

- pressing temperature: center point 60 °C, step of variation 20 °C;

- restriction diameter: center point 6 mm, step of variation 2 mm.

The coded and uncoded levels of the independent variables are listed in Table 1.

The number of experiments required (N) was given by $N = k^2 + k + 1$, where k is the number of variables. For two variables, seven (7) experiments were necessary. The experiments were performed in duplicate in order to evaluate the experimental error. All experiments were carried

ISSN 2056-5798(Print), ISSN 2056-5801(online)

out in randomized order to minimize the effect of unexplained variability in the observed responses due to extraneous factors. Table 2 shows the order of execution, variables conditions and observed responses.

Table 1. Variables and levels for the Doehlert design

Variable	Symbol	levels						
Coded value	X_i	-1	-0.866	-0.5	0	+0.5	+0.866	+1
Pressing Temperature	T	20		40	60	80		100
Restriction diameter	D		4		6		8	

Note. The black cells mean that there are no uncoded values corresponding to the coded values for the variable.

Screw Press Extraction

The oil extraction was carried out using a Komet screw press (CA 59 G, IBG Monforts Oekotec, Mönchengladbach, Germany) according to the method described by Ferchau (2000). The pressing temperature was between 20 and 100 °C, which were reached by attaching an electrical resistance-heating ring, coupled to a Störk-Tronic thermoregulator (Störk, Stuttgart, Germany), around the press barrel. The minimum and maximum restriction diameter used were 4 and 8 mm, respectively. The screw speed was set at 20 rpm. For all tests, extraction was carried out on kernels 500 ± 5 g. The extracted oils and residual oilcakes were suitably packaged and stored in the refrigerator at 4 °C for further processing and analysis.

Determination of experimental responses

The experimental responses were oil yield (OY) and residual oil content (RO). The oil yield was evaluated as the ratio of the weight of the extracted seed oil to the weight of the *C. lanatus* oilseed introduced into the press. The residual oil content was determined on the oilcake according to the AOCS (2009) method using a Soxhlet extractor (behrostest R104 T-SK, Behr Labor-Technik, Düsseldorf, Germany). Oil yield and residual oil content were expressed as percentage (g oil/100 g seed or oilcake).

Statistical Analysis of Data

Extraction experiments of *C. lanatus* seed oil were repeated twice. The mean values ± standard deviations were reported for all observed responses. A second-order polynomial regression model was used to express Y as a function of the independent variables as follows:

$$Y = \beta_0 + \sum \beta_i X_i + \sum \beta_{ii} X_i^2 + \sum \sum \beta_{ij} X_i X_j \quad (1)$$

where Y represents the response variables, β_0 is a constant, β_i, β_{ii} and β_{ij} are the linear, quadratic and interactive coefficients, respectively. X_i and X_j are the levels of the independent variables. Statistica software version 7.1 (Statsoft, 2005) was used for multiple regression analysis, analysis of variance (ANOVA), canonical analysis in the response surface regression (RSREG) procedure. The analysis includes the Fisher's test (overall model significance), its associated probability *P(F)* and determination coefficient R^2 which measures the fit goodness of regression model. It also includes the *t*-value for the estimated coefficients and associated probabilities. Response surface and contour plots were developed using the fitted quadratic

ISSN 2056-5798(Print), ISSN 2056-5801(online)

polynomial equation obtained from RSREG analysis. The test of statistical significance was based on the total error criteria with a confidence level of 95.0%.

Oil Clarification and Amount of Matter in Crude Oil

The extracted oil was weighed, put into 50 ml tubes, and then centrifuged (Z 300 K, Hermle Labortechnik, Wehingen, Germany) at 6,000 rpm and 25 °C for 15 min. The supernatant clarified oil was carefully recovered and stored in the refrigerator at 4 °C for further physicochemical analyzes. The precipitated solids were washed with n-hexane, dried and weighed. Solid content was expressed as percentage (g solids/100 g extract (oil + solid)).

Physicochemical Analysis of Oil

The clarified oil was subjected to physicochemical characterization. The colour and state of the oil at 25 °C were noted by visual inspection, while the specific gravity was determined by the method of IUPAC (1979). The refractive index was determined at 25 °C using a refractometer (AR200, Leica Microsystems, New York, USA). The viscosity was determined at 40 °C using a viscosimetric tube (AFNOR, 1986). The acid, peroxide, iodine and saponification values were determined according to the official method (AOAC, 1997). The unsaponifiable matter content was determined following IUPAC (1979) protocol. The carotenoid and total phenol contents were determined according to the colorimetric methods described by Zoué *et al*. (2012) using a UV-Visible spectrophotometer (V-530, Jasco International, Tokyo, Japan). Each oil sample was analyzed in triplicate. The mean values ± standard deviations were reported for all results.

RESULTS AND DISCUSSION

Analysis of Experimental Results

The experimental responses and suspended solid contents obtained from all the experiments are listed in Table 2.

Table 2. Experimental design, experimental responses and suspended solids content

Test	Coded values		Uncoded values		Observed responses		SS (%)
	X_1	X_2	T (°C)	D (mm)	OY (%)	RO (%)	
1	+1	0	100	6	35.07 ± 0.20	16.60 ± 0.18	8.33 ± 0.02
2	+0.5	+0.866	80	8	33.98 ± 0.03	17.60 ± 0.03	8.20 ± 0.01
3	-0.5	+0.866	40	8	29.62 ± 0.35	21.37 ± 0.29	8.14 ± 0.18
4	-1	0	20	6	24.16 ± 0.77	25.62 ± 0.57	9.73 ± 0.25
5	-0.5	-0.866	40	4	32.47 ± 0.44	18.94 ± 0.39	6.47 ± 0.10
6	+0.5	-0.866	80	4	38.27 ± 0.07	13.52 ± 0.07	6.53 ± 0.12
7	0	0	60	6	32.63 ± 0.17	18.80 ± 0.15	8.37 ± 0.32

Note. All tests were performed in duplicate and the mean values ± standard deviations are reported. T: pressing temperature (°C); D: restriction diameter (mm); X_1 and X_2: coded values of T and D, respectively; OY: oil yield (%); RO: residual oil content (%), SS: suspended solids content (%).

ISSN 2056-5798(Print), ISSN 2056-5801(online)

The oil yield varied between 24.16 ± 0.77% and 38.27 ± 0.07%. These values were significantly better than those generally obtained by artisanal extraction, which rarely exceeds 3% of the weight of the fruit (Charrouf, 2002). Indeed, the artisanal extraction, which consists in grinding the seeds, mixing the paste obtained, with addition of water, and pressing it manually, causes a lot of losses of matter. However, these oil yields were significantly lower than those of solvent extraction, which is the most widely used method for extracting vegetable oils. In this extraction method, the apolar organic solvent, which has a very high affinity with the oil, removes almost all the oil contained in the matrix (Smassel, 2013).

Press extraction did not remove all the oil from the seeds. The residual oil content oscillated between 13.52 ± 0.07% and 25.62 ± 0.57%. These values were lower than those recorded in neem seed oilcakes (Azadirachta indica A.Juss [Meliaceae]), 27.8 and 28.7% (Nitièma-Yefanova *et al*., 2012). These authors obtained the oilcake using a hydraulic press. This difference in results could therefore be explained by the type of machine used, but also by the type of seed and the operating conditions.

The crude oil was more or less brown, turbid, and contained suspended solids. The suspended solid contents ranged from 6.47 ± 0.10% to 9.73 ± 0.25%. Charrouf (2002) presented similar values (5% and 8%) for crude argan oil. This oil had also been extracted by a screw press of German origin. These results revealed that the press used in this study gave a crude oil with little suspended solid and easy to clarify. This press seems to be adequate for experimental work in the laboratory.

Analysis of the Model

Variance analysis of the factors studied for the response surface model is given in Table 3. Table 4 summarizes the multiple regression coefficients gained by a least squares technique to predict a second-order polynomial model for each response variable.

Table 3. Analysis of variance for response surface models

Variable	df	Oil yield				Residual oil content			
		Ss	Ms	*F*	*p*	Ss	Ms	*F*	*p*
Model	5	119.69	23.93	251.98	0.047	87.15	17.43	3999.02	0.012
T	1	85.24	85.24	897.22	0.021	61.75	61.75	14167.60	0.005
D	1	12.75	12.75	134.24	0.054	10.56	10.56	2423.63	0.012
TD	1	0.51	0.51	5.40	0.258	0.67	0.67	155.47	0.051
T^2	1	6.08	6.08	64.00	0.079	3.55	3.55	816.45	0.022
D^2	1	3.46	3.46	36.51	0.104	2.75	2.75	631.53	0.025
Residues	1	0.095	0.095	-	-	0.004	0.004	-	-

Note. T: pressing temperature (°C); D: restriction diameter (mm); df: degree of freedom; Ss: sum of squares; Ms: mean of squares; Fisher F test set at $p \leq 0.05$.

Table 4. Effects of independent variables on the response variables and coefficients of the 2nd degree model

Term	Oil yield				Residual oil content			
	Coefficient	Standard	*t*	*p*	Coefficient	Standard	*t*	*p*
β_0	32.63	0.30	105.8	0.006	18.80	0.06	284.80	0.002

ISSN 2056-5798(Print), ISSN 2056-5801(online)

T (β_1)	5.33	0.17	29.95	0.021	-4.53	0.03	-	0.005
D (β_2)	-2.06	0.17	-11.58	0.054	1.87	0.03	49.23	0.012
T*D (β_{12})	-0.82	0.35	-2.32	0.258	0.95	0.07	12.46	0.050
T^2 (β_{11})	-3.02	0.37	-8.00	0.079	2.31	0.08	28.57	0.022
D^2 (β_{22})	2.28	0.37	6.04	0.104	-2.03	0.08	-25.13	0.025
R^2	99.96 %				99.99 %			
adjusted	99.52 %				99.97 %			

Note. T: pressing temperature; D: restriction diameter; OY: oil yield; RO: residual oil content; R^2: coefficient of determination; student *t* test set at $p \leq 0.05$.

The statistical analysis showed that the regression models for the response variables were significant (P <0.05). The pressing temperature had significant effect (P <0.05) on the oil yield and residual oil content. The effect of restriction diameter was significant (P <0.05) only on residual oil content.

For the oil yield, examination of these coefficients with the *t*-test indicated that linear term of pressing temperature was the only significant term (P <0.05). For residual oil content, linear and quadratic terms of pressing temperature and restriction diameter were significant (P <0.05). Moreover, for the two response variables, interaction was not significant (P > 0.05) within the experimental domain. Therefore, these results suggest that linear effect of the pressing temperature may be the primary determining factor affecting the oil yield. However, the linear and quadratic effects of the independent variables were the main factors affecting the residual oil content.

For each response variable, the second-order polynomial equation in Eq. (1) can be written as follows with coefficient:

$$\mathrm{OY} = 32.63 + 5.33 \times \mathrm{T} - 2.06 \times \mathrm{D} - 0.82 \times \mathrm{T} \times \mathrm{D} - 3.02 \times \mathrm{T}^2 + 2.28 \times \mathrm{D}^2 \tag{2}$$

$$\mathrm{RO} = 18.80 - 4.53 \times \mathrm{T} + 1.87 \times \mathrm{D} + 0.95 \times \mathrm{T} \times \mathrm{D} + 2.31 \times \mathrm{T}^2 - 2.03 \times \mathrm{D}^2 \tag{3}$$

where T and D represent the coded values of pressing temperature and restriction diameter, respectively; OY and RO are the oil yield (%) and residual oil content (%), respectively. For the good fit of a model, the coefficient of determination (R^2) should be at least 80% (Guan and Yao, 2008). In this case, the adjusted R^2 were 99.52% and 99.97% for oil yield and residual oil content, respectively. This means that the regression models for the response variables were satisfactory, which adequately fits with the experimental results.

Analysis of the Stationary Point and Determination of the Optimum Conditions

To determine the nature of the stationary points, canonical analysis was performed on the second-order polynomial models after calculation of the stationary points coordinates. The stationary points coordinates, their corresponding experimental values and the canonical equations coefficients are presented in Table 5.

European Journal of Food Science and Technology
Vol.5, No.4, pp.35-46, September 2017
Published by European Centre for Research Training and Development UK (www.eajournals.org)

Table 5. Results of the canonical analysis

Stationary points			Corresponding values			Coefficients obtained		
Coordinate	OY	RO	Factor	OY	RO		OY	RO
Xs_1	0.80	0.84	T (°C)	92.03	93.85	λ_1	9.25	2.36
Xs_2	0.59	0.66	D (mm)	7.38	7.52	λ_2	-12.21	-2.08
Ds	0.99	1.07	Responses	34.15	17.50			

Note. Ds: distance from the stationary point to the center of the domain; Xs1 and Xs2: coordinates of the stationary point; T: pressing temperature; D: restriction diameter; OY: oil yield; RO: residual oil content; λ_1 and λ_2: coefficients of the canonical equation.

The coded coordinate of the stationary point was (0.8, 0.59) for the oil yield and (0.84; 0.66) for the residual oil content. It was converted to non-coded coordinate (92.03, 7.38) for the oil yield and (93.85; 7.52) for the residual oil content. At the stationary point, the predicted values of oil yield and residual oil were 34.15% and 17.50%, respectively. Results showed that all the stationary points were within the experimental domain because their distances to the domain center were all less than or equal to 1. The canonical forms of the equations demonstrating the nature of the response surfaces were:

$$Y_{OY} = 34.15 + 9.25 \times \omega_1^2 - 12.21 \times \omega_2^2 \quad (4)$$

$$Y_{RO} = 17.50 + 2.36 \times \omega_1^2 - 2.08 \times \omega_2^2 \quad (5)$$

where Y_{OY} and Y_{RO} represent canonical equations of oil yield and residual oil content, respectively; ω_1 and ω_2 are the axes of the response surface. The algebraic signs of the canonical equations coefficients are opposite; the stationary points are, in fact, saddle points. A saddle point is defined as the stationary point of a response surface which presents the maximum response for the levels of some variables and simultaneously the minimum response for the levels of other variables of the analytical system studied (Ferreira *et al*., 2004).

A three-dimensional response surface graph and a two-dimensional contour plot were generated for each response variable using the linear, quadratic and interaction terms in the second order polynomial model. The use of response surface graph and contour plot made possible to visualize and determine the optimum conditions of the screw press extraction. It was possible to follow the evolution of the independent variables and their influence on the response variables.

ISSN 2056-5798(Print), ISSN 2056-5801(online)

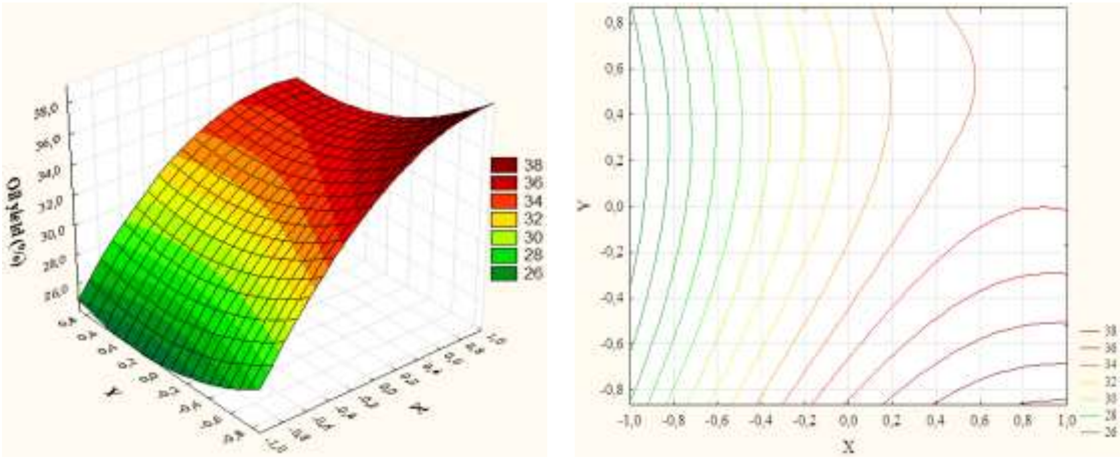

Fig. 1. Response surface and contour plots for the effect of pressing temperature and restriction diameter on the oil yield.

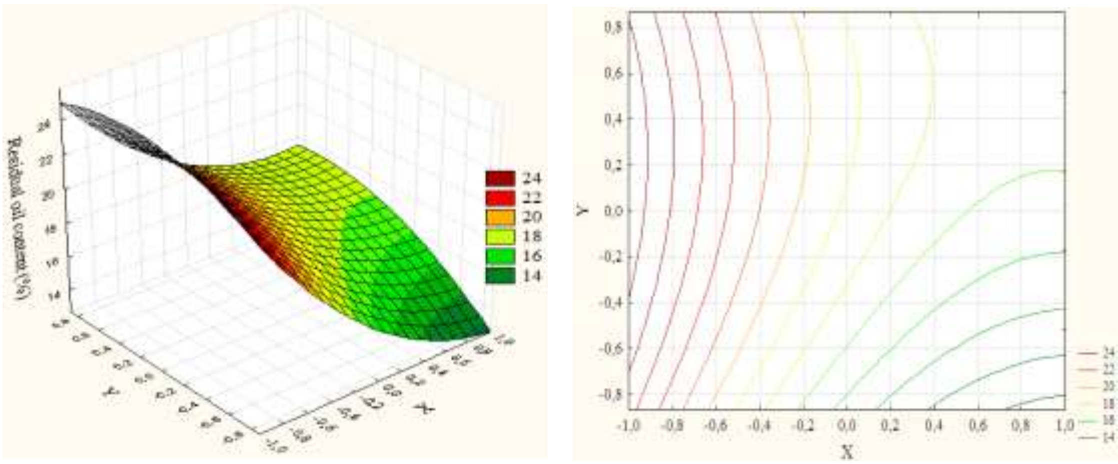

Fig. 2. Response surface and contour plots for the effect of pressing temperature and restriction diameter on the residual oil content.

Fig. 1 is the response surface and contour plots showing the effect of pressing temperature and restriction diameter on the oil yield. The oil yield increased with the increment of the pressing temperature. The possible reason for this is that increasing the temperature improves the fluidity of the oil within the paste, which facilitates the flow of oil from the paste during pressing and thus increases the oil yield. Reducing the restriction increased the compression exerted on the seeds and the dwell time of the material inside of the press, with a consequent increase of oil extracted.

The effect of pressing temperature and restriction diameter on the residual oil content can be seen in Fig. 2. The residual oil content decreased with the rise of pressing temperature and the reduction of restriction diameter, most likely due to the increase of oil extracted.

ISSN 2056-5798(Print), ISSN 2056-5801(online)

The screw press extraction conditions would be considered optimum if the oil yield reached maximum values. The canonical analysis showed that the stationary point of the response surface relative to the oil yield is a saddle point. In this case, the stationary point is not the optimum point. Then, optimization was carried out by visualizing the contour plots for the oil yield (Fig. 1). The optimum point represents the combination of extraction parameters that would give the maximum oil yield. According to these plots, the coded coordinate of maximum point for the oil yield was (0.8, -0.83) and corresponded to the uncoded coordinate (92 °C, 4.0 mm) according to the transforming equation of coded value. At the optimum point, the maximum predicted value of the oil yield was 38.79%. That is to say, when pressing temperature was 92 °C and restriction diameter was 4.0 mm, the predicted value of oil yield reached maximum value 38.79%. These results are distinctly different from those of Martínez *et al*. (2012) who studied the optimization of the chia seeds pressing (Salvia hispanica L.) using the same type of press. This difference in results would be due to the type of seed material, type of experimental design used or the parameters taken into account.

Physical and chemical properties of oil extracted by screw press

Table 6. Physical and chemical properties of C. lanatus seed oil extracted under conditions of optimum point.

Parameters	Mean values
Physical properties	
Colour/State à 25 °C	Light
Refractive index at 25 °C	1.47 ± 0.00
Specific gravity at 25 °C	0.92 ± 0.01
Viscosity at 40 °C (mPas)	53.63 ± 0.16
Chemical properties	
Acid value (mg KOH/g oil)	1.10 ± 0.08
Saponification value (mg KOH/g	198.46 ± 0.95
Iodine value (g I_2/100 g)	123.38 ± 1.38
Peroxide value (meq O_2/Kg oil)	0.60 ± 0.01
Total phenol (mg/100 g oil)	6.66 ± 0.30
Unsaponifiable matter (%)	0.80 ± 0.01
Carotenoid (mg/100 g oil)	94.61 ± 5.57

Note. All tests were performed in triplicate and the mean values ± standard deviations are reported.

Physical and chemical properties of the *C. lanatus* seed oil extracted under conditions of optimum point are shown in Table 6. At 25 °C, the *C. lanatus* seed oil was liquid, light yellow in colour with refractive index of 1.47 ± 00. Observations on the colour and the refractive index of the oil agreed with previous published report for seed oil extracted using hexane (N'guetta *et al*., 2015). The specific gravity was 0.92 ± 0.01 and the viscosity, which is a measure of the resistance of oil to shear, was 53.63 ± 0.16 mPas. These values are within the range earlier reported for most conventional oilseeds (Codex Alimentarius, 1999).

The low acid value (1.10 ± 0.08 mg KOH/g oil) showed that *C. lanatus* seed oil is not only edible but could also have a good resistance to hydrolysis. A high saponification value of 198.46 ± 0.95 (mg KOH/g oil) was obtained for the oil, indicating high concentration of triglycerides. The iodine value was high (123.38 ± 1.38 g I_2/100 g oil), which showed the oil contained a substantial level of unsaturation. The peroxide value was 0.60 ± 0.01 meq O_2/Kg oil. The value is lower relatively because oxidization could be avoided during the process of screw press extraction. The combination of high iodine value and low peroxide value indicated the *C. lanatus* seed oil could be stored for a long period without deterioration. These also demonstrated the oil possessed the desirable qualities of edible oils. The total phenol content was 6.66 ± 0.30 mg/100 g oil. The *C. lanatus* seed oil is a less important source of phenols than olive oil with a mean content of 20 mg/100 g oil (Gunstone, 2002). However, this compound with an antioxidant value was more abundant in *C. lanatus* seed oil than in pumpkin seed oil (3.9 ± 2.6 mg/100 g oil) (Andjelkovic *et al*., 2010). The unsaponifiable content was 0.80 ± 0.01%. This value was higher than those reported for other edible oils such as cotton (0.52%), peanut (0.33%) and palm kernel (0.22%) oils (Kapseu and Parmentier, 1997). This lipid fraction is a good source of minor compounds with bio-functional properties. The carotenoid content was 94.61 ± 5.57 mg/100 g oil. Carotenoids are substances of the unsaponifiable fraction with antioxidant properties. From the above results, the *C. lanatus* seed oil extracted using screw press can be used as edible oil and as an industrial feedstock.

CONCLUSION

Results showed that the second-order polynomial model is sufficient to describe and predict the response variables of oil yield and residual oil content using the pressing temperature and the restriction diameter as independent variables. The linear term of the pressing temperature greatly affects the oil yield. The linear and quadratic terms of the pressing temperature and the restriction diameter have a significant effect on the residual oil content. However, the interaction is not significant within the experimental domain. The method of graphic optimization, adopted to find the best extraction condition, predicts that the optimum extraction parameters within the experimental domain are: pressing temperature of 92 ° C and restriction diameter of 4 mm. Under such conditions, the oil yield is 38.79%. The colour of the *C. lanatus* seed oil extracted by screw press is light yellow. Its physical and chemical properties come up to the required standard for edible oil. Therefore, this seed oil would have a good prospect of upgrading in human food.

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ISSN 2056-5798(Print), ISSN 2056-5801(online)