Ultrasound Assisted Extraction of Oil from Soybean

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A B S T R A C T

Box-Behnken Experimental design of Response Surface Methodology (RSM) was used in this research for designing experiments and selecting finest standards. Power (60,80 and 100 %), sonication time (20, 40 and 60 minutes), and solvent volume (75,100 and 125 ml) were selected as the independent variable, whereas sonication frequency (20Hz) sample particle size (1mm) sample weight (25g) were kept constant. The extraction was carried out using ultrasound assisted technique. This research work was basically done to study the optimized parameters on the basis of oil yield, which was the dependent parameter. The data from all the experiments were analyzed using Design Expert 9.0.6.2 and the response functions were developed using multiple regression analysis and second order model was fitted for each response. It was found that power, sonication time and solvent volume had highly noteworthy consequence on oil yield. With upsurge in level of all the three variables, oil yield augmented.

Introduction

Oils and fats are the substances that have a profound position in human life. Biochemically, oils are usually grouped together with fats as a class of composites under lipids, one of the four major groups of large polymeric biological molecules that structure living organisms; the others being proteins, polysaccharides and nucleic acids. Fats are chemically similar to oils but differ mainly in their physical state; oils are liquid at room temperature, while fats are solids (Nelson *et al.,* 2008). Vegetable oils are the utmost significant class of biological oils and comprise those that are edible. Oils of plant origin make up the enormous popular of these as a result of their diverse and enormous number of applications in food preparation, the most notable of which are: flavor, texture, shortening, and cooking. Few animal fats such as butter and ghee also share similar functions (Gunstone2005; Gunstone and Harwood 2007). Among vegetable oils, the leading market shares belong to those obtained from oil palm, soybeans, canola (rapeseed), sunflower, peanuts and cottonseed.

Traditionally the most preferred method of oil extraction involves the usage of an organic

solvent to selectively transfer lipids from their biological source material (Birch and Ian 2000; Wang 2004). Hexane is the industrial favorite solvent for oil extraction from soybeans. In the case of soybeans, these tedious steps have profound industrial implications; they include cleaning, dehulling, moisture-conditioning and flaking (Hammond, Johnson *et al.,* 2005). Hence, several advanced approaches have been suggested, that could be utilized to 'assist' the solvent extraction process. Among these, two methods have had significant success; ultrasound and microwaves and ultrasound (Lee *et al.,* 2010).

Microwave-assisted extraction (MAE) is a thermal process while ultrasound assisted solvent extraction (UASE) is a more mechanical process. Even though MAE has been proven to extract several valuable phytochemicals, the thermal character of the method makes it unsuitable for the extraction of PUFAs due to their high vulnerability to oxidation and degradation. UASE in contrast, has been successfully utilized for the extraction of oils from different biomaterials including soybeans to changing grades of success (Prabakaran and Ravindran, 2011).

Considering all the above proofs, the aim of this research is that hexane is an efficient solvent for extracting Soybean oil, and that ultrasound-assisted solvent extraction (UASE) with hexane as solvent could be used to obtain high-quality soybean oil with greater efficiency than conventional methods.

The overall objective of the research was to boost the effectiveness of oil extraction with respect to oil yield from ground soybeans. The specific objective was thus to study the consequence of process parameters like sonication power, solvent volume and sonication time on the oil yield from soybean using Ultrasound assisted extraction.

Materials and Methods

The main objective of this research work is to standardize the operating conditions for extraction of oil from soybeans and then optimize the parameters (showed in Table 2 and Table 3) on the basis of oil recovery. Materials used and procedure adopted in this research is described as:

Raw Material, Chemicals and glassware

White Soybean variety (PS-1477) was used for the extraction of Soybean oil. This variety was obtained from the Crop research Centre of G B Pant university of Agriculture and Technology. Hexane was utilized as the solvent for the extraction of soybean oil from soybean. Hexane was taken from the department lab. Glassware like beakers, conical flasks, measuring cylinders and funnels were collected from the department's store room.

Experimental setup

It consists of different equipment required to carry out experimental work which includes sonicator, hot plate Magnetic Stirrer, refrigerator, weighing balance, grinder, soxhlet apparatus etc.

Experimental design

RSM contains of a collection of statistical and mathematical methods that can be utilized to describe the relations between the independent and response variables. It has significant usage in the design, improvement and formulation of novel commodity, also in the enhancement of current product design. It only describes the result of the independent variables or in combination, on the procedures. The usage of Box-Behnken designs to prepare experimental design is seen in the current research work. Box-Behnken designs (BBD) are a class of

rotatable or nearly rotatable second-order designs built on three-level incomplete factorial designs. Box and Behnken recommends how to choose points from the three-level factorial plan, which permits the effectual assessment of the first- and second-order coefficients of the mathematical mode (Box and Behnken, 1960). In Box–Behnken designs (Ferreira *et al.,* 2007) the experimental points are situated on a hyper sphere equidistant from the central point. The number of experiments (N) required for the development of BBD is defined as

$$N = 2k\,(k-1) + Co \quad \ldots (1)$$

Where k is number of factors and Co is the number of central points.

For 3 factors at three levels BBD had 17 experiments with 5 experimental runs at central points, as presented in the Table 1.

Preparation of sample

Raw soybeans were washed to eradicate any foreign material for examples and, small stones and plant leaves that might exist after harvesting, drying, transportation and storage. Washed and conditioned soybeans were ground by means of a Cyclotec mill. A stainless steel screen with a mesh dimension of 1 mm was utilized to attain a constant particle dimension distribution of soybean flakes. Ground soybean flakes of size 1mm were then packaged in air-tight glass jar until used.

Ultrasound treatment

Ground soybean flakes (25 g) were mixed with the selected quantity of solvent (75,100,125 ml) in a 250 ml glass beaker. The soybean–solvent suspension was ultrasonicated for 20, 40 and 60 minutes using a 20 kHz ultrasonic Sonicator (Electrosonic

Industries, India) with a 2.0 cm probe that was immersed in the suspension. Ultrasonic wave amplitude used were 60, 80 and 100%. Suspensions were endlessly stimulated at a continuous stirring rate of 500 rpm by a magnetic stirrer to avert heating of suspensions under the effect of high-intensity ultrasound. After extraction, oil: solvent mixture (miscella) was filtered using Wattman filter paper 1. The filtrate from the filtration comprises of oil and solvent. The oil was then separated from this mixture by means of a countercurrent distillation arrangement with the heating mantle as the heat source and water as the coolant. The solvent used was also recovered. Quantity of oil extracted was determined by weighing the oil obtained after solvent removal which was kept in properly washed and sterilized test tubes.

Yield determination

The yield of oil was calculated using the following formula:
$$Yield\,(\%) = (W_e\,/\,W_t) \times 100 \ldots\ldots\ldots\ldots(2)$$

Where, W_e is the quantity of oil extracted from the sample (g) and W_t is the quantity of total oil in the sample (g). W_t was obtained by Soxhlet standard extraction mode.

Data analysis

The model development was done by means of response surface methodology through use of Design expert V 9.6.0.2.Complete second order model as given in Equation-1 was fitted to the data and the model adequacy was tested using R^2 (coefficient of multiple determination) and fisher's F-test. The parametric effect on various responses was done through the analysis of developed models. Regarding three independent variables a second order response function has the following general form

$$Y = \beta_0 + \sum_{i=1}^{4} \beta_i X_i + \sum_{i=1}^{3} \sum_{j=i+1}^{4} \beta_{ij} X_i X_j + \sum_{i=1}^{4} \beta_{ii} X_i^2$$

… (3)

Where, β_0, β_i, β_{ij} are constants X_i, X_j are variables (coded).

Multiple regression analysis was used to examine the experimental data so as to develop response functions and obtain variable parameters optimized corresponding to best outputs. The values of model coefficients and connected statistics in relations of lack of fit and P-value were obtained through the program. The value of P represents the possibility of significance. A high F-value is an indication of the point that the model had a significant lack of fit and therefore is inadequate. A model with lower values of P is considered better. The models having P-value lower than 0.1 (indicating the lack of fit is unimportant at 90% confidence level) were acknowledged.

Adequacy of model

Equation is solved by means of a statistical method called technique of least square (MLS) which is a multiple regression method utilized to fit a mathematical model to a set of experiment data producing the lowest residual possible. The results of regression analysis are obtained in terms of ANOVA, regression coefficient and associated statistics, standard deviation, coefficient of determination (R^2), lack of fit, etc. These determine adequacy of predictive model and effect of independent parameters on the response (Table 5).

Results and Discussion

The current study was thus taken with the aim of studying the outcome of process factors on oil yield from soybean using Ultrasound assisted extraction and the optimization of process parameters for oil on the basis of its yield. The experimental design used in the current study was Box-Benhken design having three variables Sonicator Power, Extraction Time and Solvent Volume with three levels of each (60,80 and 100 %), (20, 40 and 60 minutes) and (75,100 and 125 ml) respectively. Response surface methodology was utilized to improve the process parameters. A complete second order mathematical model was fitted in the response. The competence of the model was tested using coefficient of determination (R^2) and Fisher's test. The results obtained on numerous characteristics of the study are given below.

Effect of independent parameters on oil yield

Soybean oil was extracted from ground Soybean using ultrasound assisted extraction while taking hexane as the solvent. To enhance the extraction process, parameters studies were Sonicator power (60, 80 and 100%), sonication time (20, 40 and 60 minutes), solvent volume (75, 100 and 125 ml). The quantity of oil obtained during extraction is showed in table. The oil yield ranged from 48 % to 84 % from sample amount of 25g of sample over entire experimental conditions.

The experimental data was studied to observe the noteworthy consequence of numerous process variables on the oil yield. The outcomes of study of variance for oil yield are given in Table 4. F-value was used to investigate the significance of linear, quadratic and interactive terms. F_{cal} value for model was superior to F_{tab}, which implies that model was significant (p < 0.01). The outcome of independent variables on oil yield at linear level was found highly significant at 1% level of significance ($F_{cal} > F_{tab}$), for quadratic and interactive terms it was also observed significant.

Multiple regression analysis was performed to fit the response parameter, oil yield. The regression coefficients for the second order model polynomial equations for the linear, quadratic and interaction terms are shown in Table 5. The statistical analysis shows that the projected model was sufficient, having significant fit and with very reasonable values of R^2 for drying time. The R^2 value for the oil yield was 99 %, which implies that the model could account for 99.00% data. Closer the value of R^2 to unity, the improved the empirical model fits the actual data. Insignificant value of lack of fit showed that the developed model is valid. The second order polynomial equation was developed which represents response, oil yield as functions of Sonicator Power, Solvent Volume and Extraction Time. An empirical relationship among the responses and input variables in coded form can be expressed by the following equation:

$$Y=70.83+12.78A+2.51B+1.35C + 0.38AB + 0.55AC + 0.12BC + 0.092A^2 - 0.92B^2 - 0.8C^2 \quad \dots (4)$$

Where, Y is oil yield, A, B and C are the coded values of Sonication Power, Solvent Volume and Sonication Time respectively.

Table.1 Experimental design table

Expt no.	Coded levels			Actual levels		
	A	**B**	**C**	**Power (A) (%)**	**Solvent volume(B) (Hexane) (ml)**	**Sonication time (C) (min)**
1	0	1	-1	80	100	40
2	0	-1	1	100	100	60
3	-1	-1	0	60	100	20
4	0	-1	-1	60	75	40
5	1	1	0	80	75	60
6	0	0	0	80	125	20
7	1	-1	0	80	100	40
8	0	0	0	60	125	40
9	0	0	0	80	100	40
10	1	0	-1	100	75	40
11	1	0	1	60	100	60
12	-1	0	1	80	75	20
13	-1	1	0	100	125	40
14	0	0	0	80	100	40
15	-1	0	-1	80	100	40
16	0	0	0	80	125	60
17	0	1	1	100	100	20

Table2 Values of levels of independent parameters

S No.	Independent parameter	Coded value	Value of levels		
			-1	0	+1
1	Power (%)	A	60	80	100
2	Solvent volume (ml)	B	75	100	125
3	Sonication time(mins)	C	20	40	60

Table.3 Values of constant parameters

S No	Parameter	Value
1	Sonication frequency	20 Hz
2	Sample particle Size	1 mm
3	Sample weight	25g

Table.4 Oil yield

Std	Run	Power (%)	Solvent volume(ml)	Time (min)	Oil Yield (%)
5	1	80	100	40	67.6501
13	2	100	100	60	84.6316
11	3	60	100	20	48.6632
8	4	60	75	40	53.7241
2	5	80	75	60	67.4344
9	6	80	125	20	61.1463
15	7	80	100	40	68.9395
3	8	60	125	40	57.0819
10	9	80	100	40	68.9395
14	10	100	75	40	77.8611
4	11	60	100	60	58.3958
17	12	80	75	20	56.1954
6	13	100	125	40	82.7274
12	14	80	100	40	68.9395
16	15	80	100	40	68.9395
1	16	80	125	60	73.3756
7	17	100	100	20	70.5263

Table.5 ANOVA table for oil yield

| Analysis of variance table [Partial sum of squares - Type III]					
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob> F
Model	1577.37	9	175.26	473.45	< 0.0001
A-Power	871.32	1	871.32	2353.74	< 0.0001
B-Solvent Volume	33.69	1	33.69	91.00	< 0.0001
C-Time	20.02	1	20.02	54.09	0.0002
AB	0.57	1	0.57	1.54	0.2550
AC	4.78	1	4.78	12.91	0.0088
BC	0.25	1	0.25	0.66	0.4426
A^2	0.035	1	0.035	0.096	0.7662
B^2	3.60	1	3.60	9.72	0.0169
C^2	43.63	1	43.63	117.86	< 0.0001
Residual	2.59	7	0.37		
Lack of Fit	1.26	3	0.42	1.26	0.3991
Pure Error	1.33	4	0.33		
Cor Total	1579.96	16			

Fig.1 Effect of solvent volume (ml) and power (%) on oil yield
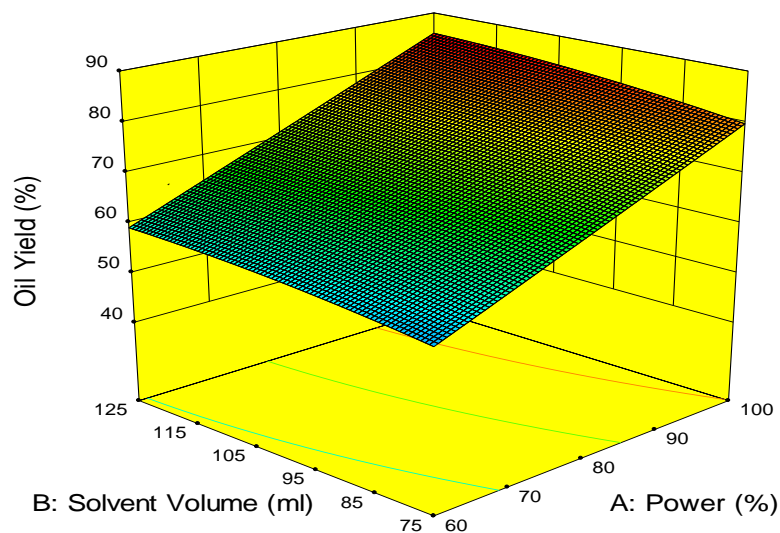
Fig.2 Effect of sonication time (min) and power (%) on oil yield
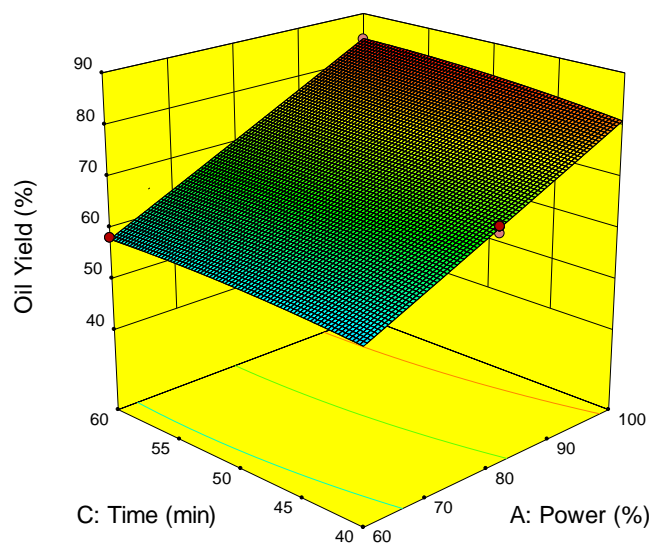
Fig.3 Effect of time (min) and solvent volume (ml) on oil yield
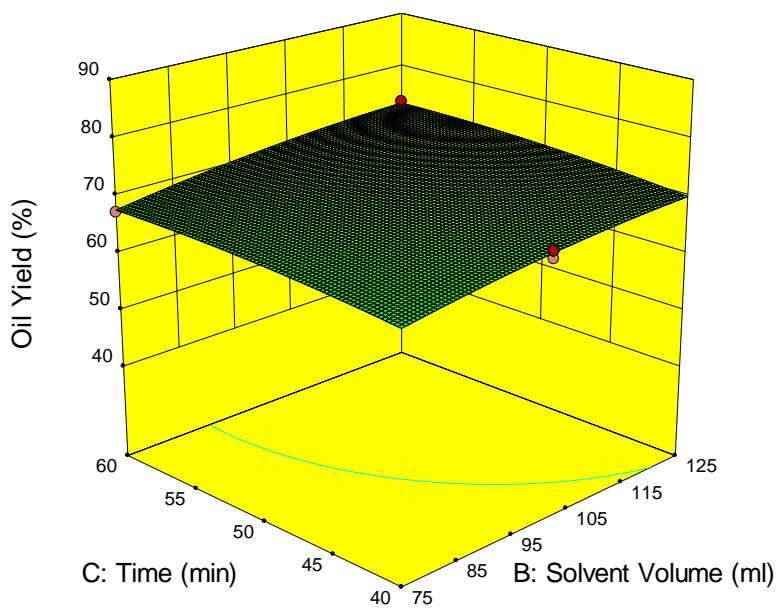
Graphical analysis

The effect of ultrasonic power on the yield of soybean oil is shown in Figure 1. It can be seen that the yield of soybean oil increases almost linearly from 48 to 84 % with increasing ultrasonic power from 60 to 100 %. Since the temperature and pressure were extremely great inside the bubbles and the breakdown of bubbles followed over very petite time, the violent shock wave and high-speed jet were produced which could boost the permeation of the solvent into the cell tissues and speed up the intracellular product discharge into the solvent by distracting the cell walls. Figure 2 shows the effect of experimental duration for extraction on the yield of soybean oil. It was also observed that the yield of oil increases from 48 to 84% with the upsurge in extraction time from 20 to 60 minutes. This process shows that the consequence of ultrasound is more effective at 60 minutes. It may be that ultrasonic wave possibly will disrupt the cell walls, so greater interaction region between solvent and material was generated and additional oil was released.

As shown in Figure 3, the solvent volume had significant result on the yield of soybean oil. The yield of oil extracted was found to upsurge with the rise in solvent volume from 75ml to 125 ml. The bigger liquid (solvent) to solid ratio signifies a larger concentration difference which aids mass transfer.

In conclusion, the use of ultrasound for soybean oil extraction showed a progressive consequence on the oil yield. The subsequent inferences were drawn from this research:

- Ultrasound had a noteworthy effect on soybean oil extraction.
- Sonication power intensity level affected the final soybean oil yield.
- Application of ultrasound increased oil yield irrespective of soybean variety.
- The selection of solvent influenced the proficiency of the ultrasound to improve yield.
- Application of ultrasound-assistance in oil extraction had a insignificant effect on oil quality, i.e., a high oil quality was still obtained.
- Choosing an ideal combination of treatment time and amplitude resulted in high oil yields.

The results obtained in this research have important implications for future usage in the soybean oil industry. This study revealed that ultrasound has the potential to be used in oil extraction method to improve efficiency and lower the processing cost. Hexane in amalgamation with other solvents can potentially be applied in soybean oil extraction because of its excellent oil extraction performance: less cost and improved safety features. Future studies will concentrate on finding the optimum combination of process parameters (ultrasound intensity, temperature, solvent, etc.) that can boost the effectiveness of oil extraction.

Acknowledgement

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