Journal of Engineering Research and Reports

8(2): 1-16, 2019; Article no.JERR.52369
ISSN: 2582-2926

Comparative Studies and Optimization of the Process Factors for the Extraction of Beta-carotene from Palm Oil and Soybean Oil by Solvent Extraction

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Authors' contributions

This work was carried out in collaboration among all authors. Authors SOE and DCO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors COA and DCO managed the analyses of the study. Author COA managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JERR/2019/v8i216984
Editor(s):
(1) Dr. Djordje Cica, Associate Professor, Faculty of Mechanical Engineering, University of Banja Luka, Bosnia and Herzegovina.
Reviewers:
(1) Vera Lúcia Viana do Nascimento, Federal Institute of Piauí-Campus Teresina Central, Brazil.
(2) Julian Cruz-Olivares, Autonomous Mexico State University, Mexico.
Complete Peer review History: http://www.sdiarticle4.com/review-history/52369

Original Research Article

Received 27 August 2019
Accepted 01 November 2019
Published 11 November 2019

ABSTRACT

This research focuses on the extraction of beta-carotene from palm and soybean oils using solvents (Ethanol and Acetone), taking into account the effect of the extraction process factors such as time, temperature, dosage of the oil samples, solvent volume and solvent type. The extraction of beta-carotene from palm and soybean oils using acetone and ethanol was positively influenced by increase in temperature, time, solvent volume, dosage of the oil samples and solvent type. The effect of temperatures was carried out within the temperature ranges of 35ºC, 40ºC, 45ºC and 50ºC. It was observed that increase in temperature resulted in increase in concentration and 45ºC gave the highest concentration. From the study, it was observed that the extraction process for beta-carotene almost reached equilibrium after 50 mins for palm oil and 45 mins for soybean oil. Between acetone and ethanol used, ethanol was found to be the best solvent for the

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extraction of beta-carotene from palm oil and soybean oil. From the results, the concentration of the extracted beta-carotene increased as the volume of the solvent increased using both acetone and ethanol on both substrates. The concentration of extracted beta-carotene increased with increase in the dosage of the oil samples (palm and soybean). The increase in concentration could be attributed to the more active sites due to increase in the substrate volume.

Keywords: Extraction; palm oil; soybean oil; acetone; ethanol.

1. INTRODUCTION

Beta-carotene is one of the key products of food industry which has been largely utilized as nutrients and additives. It is a pigment found in plant that gives yellow and orange fruit and vegetables their colour [1]. Beta-carotene is a carotenoid that is a precursor for vitamin A and a powerful antioxidant that plays a critical role in maintaining vision, skin and neurological function. It is a natural occurring retinol precursor obtained from certain fruits and vegetable with potential antineoplastic and chemopreventive activities [2]. As antioxidants, beta carotene inhibits free radical damage to DNA. This agent also induces cell differentiation and apoptosis of some tumor cell types, particularly in early stages of tumorigenesis and enhances immune system activity by stimulating the release of natural killer cells; lymphocyte and monocyte. As a precursor of vitamin A, it is administered to reduce the severity of photosensitivity reactions in patients with erythropoetic protoporphyria [3].

However, recently there are rapid growing concerns about the source of the ingredients along with awareness about potential harmful synthetic compound which may result from synthetically produced beta-carotene. These rising consumer awareness and concerns have made the global demand for naturally produced carotene to increase appreciably. Hence, industries are continuously searching for the new sources as well as introducing different technologies to produce beta-carotene. Even though there has been numerous effort made to extract carotenoids either by removing chemically converted triglycerides through saponification or trans-esterification or by using observant materials still more effort are in progress towards efficient extraction of carotenoids [4]. Palm oil and soybean oil are interesting alternatives to the problem of natural sources of beta carotene. They are natural sources of carotene that are known to contain high concentration of natural carotenoids [2]. The main carotenoids of palm oil are alpha carotene and beta carotene. Together they make more than 80% of the total carotenoids in palm oil [5] with 36.4% alpha carotene and 54.4% beta-carotene [6]. It has greater carotenoids concentration than any other oil or fat [7] and these carotenes contribute to palm oil stability and nutritional value (Mustapha, et al. 2011). The concentration of carotenoids normally ranges between 400 and 3500ppm in palm oil, (Ahmed, Chan, et al. 2011). It is known that palm pressed fibre oil is enriched with natural carotenes [8].

Carotenoids are extracted by any of the following methods; atmospheric liquid extraction with maceration, soxhlet extraction, microwave-assisted extraction, supercritical fluid extraction, enzyme-assisted, liquid-liquid extraction (Ramesh, et al. 2018). Extraction using solvents is generally conducted at a temperature near the boiling point of the solvent, which reduces oil viscosity and improves its solubility in the solvent, ensuring the efficiency of the process (Gandlin, et al. 2003). According to Gandli, et al. ethanol is a biodegradable and nontoxic solvent, with great potential for oil extraction, potentially replacing n-hexane without yield losses. Acetone has also proven to be a solvent for oil extraction owing to its low polarity. Most authors have identified that yield/concentration of the extraction process using solvents is dependent on factors such as time, solvent volume, oil dosage, temperature, type of solvent used. Effective analysis and design of extraction units especially on an industrial scale, kinetic and thermodynamic data of the process are critical [9]. Carotenoids can be identified by a combination of absorption spectrum, High pressure Liquid Chromatography (HPLC) retention time, and mass spectrum (Buchecker and Noack 1995).Carotenoids handbook contains more than 700 types of carotenoids with their spectroscopic data and references, and it is useful to check the identity of any newly isolated carotenoids (Britten, et al. 2004).

Haven presented this information; this study is aimed at extracting beta-carotene from palm oil and soybean oil using solvent (acetone and ethanol). The effects of process factors were

considered as well as the kinetics study of extraction of beta-carotene from palm oil and soybean oil. Optimization of the process conditions was carried out using response surface methodology to develop mathematical models for the extraction.

2. MATERIALS AND METHODS

2.1 Materials

The freshly produced crude palm oil used for this study was procured from a local palm oil processing mill in Amator Amuzu in Aboh Mbaise L.G.A Imo State Nigeria. The soybean seeds were bought from a local market (Afia Abakpa) in Enugu State. The soybean seeds were carefully sorted out to remove contaminating particles such as pebbles of stone, seed shells and other impunities. The seeds were further crushed using mechanical milling machine and air-blown to remove the large quantities of the seed shell. The crushed mesocarps were hence ground to a smaller particle size to increase surface area and enable better extraction of the soybean seed oil by the solvent (n-hexane). The extracted soybean oil and the procured palm oil were then used for further experimental work in the laboratory.

2.2 Methods

2.2.1 Determination of the physical parameters of the oil samples

The specific gravities, viscosity, peroxide value, acid value, iodine value, saponification value, and free fatty acid (FFA) of the oil samples were determined as prescribed by AOAC (2005) and presented in Table 1.

2.2.2 Extraction of soybean oil from soybean seeds

Solvent extraction was performed by soaking the ground soybean seed sample with n-hexane in an air-tight container for three (3) days after which the mixture of soybean oil and n-hexane was recovered. The mixture was further separated through the process of distillation to recover the solvent and the extracted soybean oil sample was used for further analysis. The distillation of the mixture of extracted soybean oil and solvent (n-hexane) was done by adding the mixture into a single neck round bottom flask mounted on a heating mantle, with a counter-current flow soxhlet extraction tube connected to the flask with cold water entering and leaving the tube. The mixture was heated slightly above the boiling point of n-hexane 68-69, 72ºC which promoted the n-hexane to evaporate into the steam, hence condensed by the action of the cold water.

Table 1. Physiochemical properties of the oil samples

Parameters	Palm oil	Soybean oil
Specific gravity (mg/cm^3)	0.900	0.885
Viscosity (Cp_s at 25ºC)	0.056	0.035
Peroxide value (meq/cm^3)	4.9806	4.9806
Acid value ($mgKOH/cm^3$)	1.662	1.53
Iodine value (Wijs)	44.9136	45.3939
Saponification value (mg/cm^3)	210.22	212.9
FFA (%)	0.831	0.765

2.2.3 Effect of volume (Dosage) of the oil samples on the extraction of beta-carotene using acetone and ethanol

During the determination of effect of the dosage of the oil samples, time was kept constant at 20 mins, solvent volume was also kept constant at 30 cm^3, and temperature was at room temperature while the dosage of the oil samples varied from 5 cm^3, 10 cm^3, 15 cm^3 and 20 cm^3. This was done to determine the effect of dosage of the oil samples on the extraction of beta carotene from the samples under the sample operating conditions for the process factors.

2.2.4 Effect of temperature on the extraction of beta-carotene using acetone and ethanol from palm and soybean oil samples

The determination of the effect of temperature was done by using the same procedure as in 2.2.7 while keeping dosage of the oil samples constant at 10 cm^3, solvent volume at 30 cm^3, time at 20 mins, while the temperature was varied from 35, 40, 45, 50ºC. This was done to determine the effect temperature has on the extraction of beta carotene using acetone and ethanol from palm oil and soybean oil respectively.

2.2.5 Effect of time on the extraction of beta-carotene from palm oil and soybean oil using acetone and ethanol

The extent of extraction of beta carotene from palm oil and soybean oil using acetone and ethanol was studied as a function of time. The substrates volume was kept constant at 10 cm^3, solvents volume was kept constant at 30 cm^3. Time varied at the range of 10, 20, 30, 40, 50, 60 and 70 mins at different temperature of 35ºC, 40ºC, 45ºC and 50ºC.

2.2.6 Effect of solvent volume on the extraction of beta-carotene from palm oil and soybean oil using acetone and ethanol

During the determination of the effect of solvent volume on the extraction of beta carotene from the oil samples, the volume of the oil samples were kept constant at 10 cm^3 and also the time for extraction was kept constant at 20 mins while the volume of the solvents volume were varied from 20,40,60 and 80 cm^3. This was done in

Table 2. Design matrix with response

						Response	
Standard order	**Run order**	**Temperature (ºC)**	**Volume of oil (MLs)**	**Time (Mins)**	**Type of solvent**	**Palm oil (Mg/Ml)**	**SBo (Mg/Ml)**
8	1	45.00	20.00	50.00	Ethanol	0.88362	0.44181
16	2	40.00	15.00	40.00	Ethanol	0.86958	0.43479
15	3	40.00	15.00	40.00	Ethanol	0.86868	0.43434
35	4	40.00	15.00	40.00	Acetone	0.81639	0.408195
34	5	40.00	15.00	60.00	Acetone	0.83115	0.415575
25	6	35.00	10.00	50.00	Acetone	0.68103	0.340515
1	7	35.00	10.00	30.00	Ethanol	0.67815	0.339075
4	8	45.00	20.00	30.00	Ethanol	0.891	0.4455
31	9	40.00	5.00	40.00	Acetone	0.65655	0.328275
20	10	40.00	15.00	40.00	Ethanol	0.85914	0.42957
23	11	35.00	20.00	30.00	Acetone	0.79929	0.399645
28	12	45.00	20.00	50.00	Acetone	0.8748	0.4374
39	13	40.00	15.00	40.00	Acetone	0.85203	0.426015
5	14	35.00	10.00	50.00	Ethanol	0.69462	0.34731
29	15	30.00	15.00	40.00	Acetone	0.66888	0.33444
27	16	35.00	20.00	50.00	Acetone	0.81684	0.40842
24	17	45.00	20.00	30.00	Acetone	0.8838	0.4419
33	18	40.00	15.00	20.00	Acetone	0.80757	0.403785
13	19	40.00	15.00	20.00	Ethanol	0.82368	0.41184
7	20	35.00	20.00	50.00	Ethanol	0.83322	0.41661
32	21	40.00	25.00	40.00	Acetone	0.81945	0.409725
12	22	22.00	25.00	40.00	Ethanol	0.83583	0.417915
16	23	45.00	10.00	50.00	Acetone	0.80487	0.402435
21	24	35.00	10.00	30.00	Acetone	0.66483	0.332415
6	25	45.00	10.00	50.00	Ethanol	0.78426	0.39213
10	26	50.00	15.00	40.00	Ethanol	0.85275	0.426375
14	27	40.00	15.00	60.00	Ethanol	0.8478	0.4239
3	28	35.00	20.00	30.00	Ethanol	0.81531	0.407655
2	29	45.00	10.00	30.00	Ethanol	0.80082	0.40041
36	30	40.00	15.00	40.00	Acetone	0.85167	0.425835
9	31	30.00	15.00	40.00	Ethanol	0.68229	0.341145
18	32	40.00	15.00	40.00	Ethanol	0.86004	0.43002
30	33	50.00	15.00	40.00	Acetone	0.83574	0.41787
22	34	45.00	15.00	30.00	Acetone	0.75861	0.3793305
19	35	40.00	15.00	40.00	Ethanol	0.83268	0.41634
11	36	40.00	5.00	40.00	Ethanol	0.66969	0.334845
37	37	40.00	15.00	40.00	Acetone	0.84321	0.421605
17	38	40.00	15.00	40.00	Ethanol	0.86904	0.43452
40	39	40.00	15.00	40.00	Acetone	0.84231	0.421155
38	40	40.00	15.00	40.00	Acetone	0.85257	0.426285

effect to determine at the same operating conditions of time, dosage of the samples, temperature, and the best value of the solvents for the extraction of beta carotene from the oil samples.

2.2.7 Determination of beta-carotene

For each of the oil samples (palm & soybean oil), 10 $cm^3$was measured into a conical flask and 30 cm^3 of 95% ethanol was measured and added into the conical flask and maintained at room temperature in a water bath for 20 mins with periodic shaking. The supernatant was decanted and allowed to cool. The ethanol concentration of the mixture was brought to 85% by adding 5cm^3 distilled water and further cooled for about 5mins. The mixture was transferred into a separating funnel and 15 cm^3 of pet ether and 10 cm^3 of ethanol were added into it. It was allowed to stand until two layers were formed. The button layer was emptied into a beaker while the top layer was collected into a 250 cm^3 conical flask, the bottom layer was transferred into a funnel and re-extracted with 15 cm^3 of pet-ether ones to get a fairly yellow extract. The entire pet-ether was collected into 250 cm^3 conical flask and transferred into a separating funnel for another re-extraction with 15 cm^3 of 80% ethanol. The extract formed was measured and poured into a sample bottle for further analysis using a U-V-spectrophotometer at 390 Nm.

According to Beer Lambert's law which states that absorbance of a material sample is directly proportional to its thickness (path length).

$A \propto L$

$$A = \varepsilon LC \quad (2.1)$$

$$\text{Conc. (c)} = \frac{Absorbance \times volume\ of\ cuvette}{Molar\ Extinction\ Coefficient} \quad (2.2)$$

Where;

A = Absorbance
ε = Molar extinction coefficient 1.25 x 10^4
C = Concentration

Path Length → Volume of cuvette.

2.3 Optimization of Process Factors for the Extraction of Beta Carotene from Palm Oil and Soybean Oil

The experiment was designed using Design Expert version 8.1.0.1. Central Composite (CCD) was utilized in order to optimize the extraction of beta-carotene from palm oil and soybean oil. The experiment was designed on three (3) levels, four factors (three numeric and one categorical) that gave 40 experimental runs. The factors are temperature of extraction, extraction time, volume of the oil samples and type of solvent used. The design matrix is shown in Table 2.

3. RESULTS AND DISCUSSION

3.1 Effect of Volume (Dosage)

The extent of extraction of beta carotene from palm oil and soybean oil was studied as a function of dosage of the oil samples with regards to concentration. From Fig. 1(a), it can be see that, as the volume of the oil sample increased, the concentration also increased in the extraction of beta carotene from palm oil using acetone and ethanol. Time was kept constant at 20 mins, temperature at 35ºC and solvent volume at 30 cm^3. This increase in the concentration could be attributed to the increase in more active sites by increasing the substrate volume for the solvent to act on. This present study varied the volume of the oil samples from 5,10,15 and 20 cm^3.

Fig. 1(b) also showed the same trend of increase in the concentration of extracted beta-carotene from soybean oil using acetone and ethanol. This increase in concentration would continue until equilibrium between the volume of the oil sample and the solvent is [10].

3.2 Effect of Temperature

3.2.1 Effect of temperature on the extraction of beta carotene from palm oil and soybean oil using acetone and ethanol

The effect of temperature on the extraction of beta-carotene from palm oil and soybean oil using acetone and ethanol was evaluated based on the concentration at temperature range of 35ºC, 40ºC, 45ºC, and 50ºC, at a dosage of 10 cm^3, solvents volume of 30 cm^3 and constant time of 20 mins. It is clear from the result as shown in Fig. 2 (a&b), that as the temperature increased, the concentration of the extracted beta carotene increased. This is due to increased mass transfer rate with increase in temperature. [11] when temperature is increased, it facilitates the breakdown of cell walls and improve extraction with solvent. As reported by Oliveira, et al. (2010), that carotenoid degradation occurs

in temperature close to 40ºC. Therefore, temperature up to 45ºC resulted in a decrease in the yield of carotenoids content. Norshizila, et al. [10] reported that carotenoids concentration decreased at the temperature of 45ºC. This present study is in agreement with Norshazila, et al. [10] in the case of acetone whereby the concentration decreased after 45ºC as evident in Fig. 2 (a&b). Also, the increase in concentration as temperature increased shows that the process is an endothermic process [12].

3.3 Concentration of Beta-carotene at Different Temperature and Time

The concentration of the extracted beta carotene from palm oil and soybean oil using acetone and ethanol was studied as a function of time and temperature. The substrates volume was kept constant at 10 cm^3, solvents volume was kept constant at 30 cm^3. Time of extraction varied at the range of 10,20,30,40,50,60 and 70 mins, at different temperature of 35ºC, 40ºC, 45ºC and 50ºC.

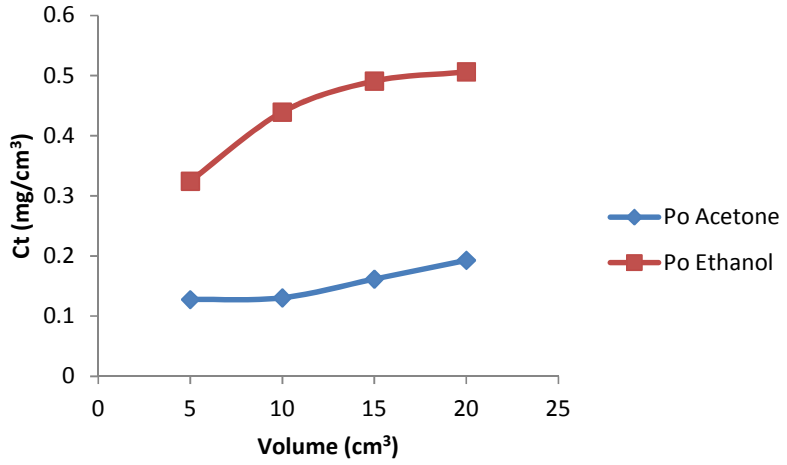

Fig. 1a. Effect of volume of palm oil on the extraction of beta carotene using acetone and ethanol

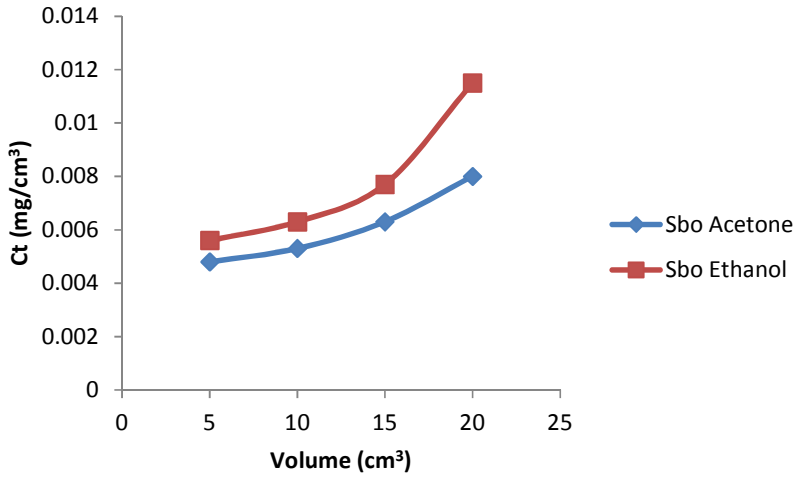

Fig. 1b. Effect of volume of soybean oil on the extraction of beta carotene using acetone and ethanol

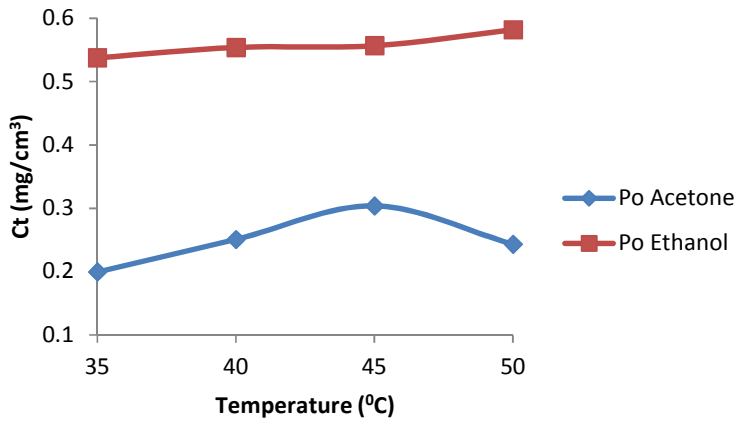

Fig. 2a. Effect of temperature on the extraction of beta carotene from palm oil

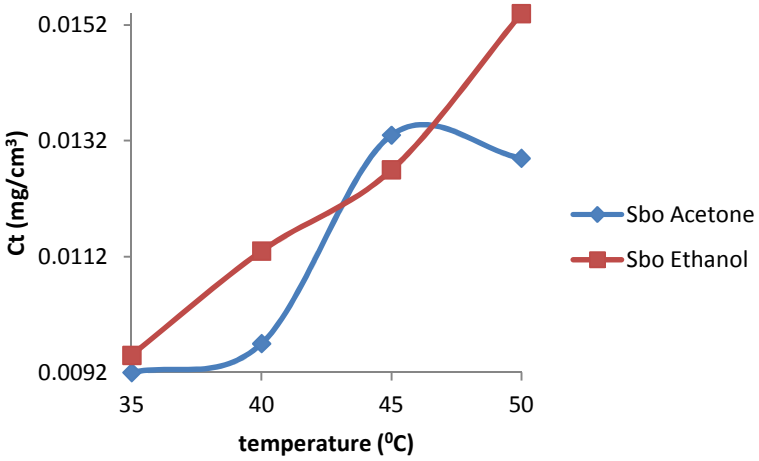

Fig. 2b. Effect of temperature on the extraction of beta-carotene from soybean oil

Fig. 3 (a&b) shows the effect of extraction time on the extraction capacity of acetone and ethanol at various temperatures. It was observed that the concentration increased as time and temperature increased, for both palm oil and soybean oil on both solvents. Fig. 3 (a) shows clearly that concentration was highest at temperature 45ºC. At temperature 50ºC, concentration was higher than that at 40ºC, but as time progressed, precisely after 50 mins, concentration at 40ºC became higher than that of 50ºC. This suggests that, the extraction of beta-carotene from palm oil using acetone can best be achieved between the temperature of 40ºC and 45ºC. Fig. 4(b) also shows a very close competition in the concentration of extracted beta-carotene at the four temperatures with 50ºC slightly higher and followed by 45ºC. This also suggests that the extraction of beta carotene from palm oil using ethanol can be best between the temperature of 45ºC and 50ºC. The increase in the concentration of the extracted beta-carotene with increase in temperature was also observed to go along with increase in time.

Fig. 3 (c&d) shows the concentration of the extracted beta carotene at different temperature and time from soybean oil using acetone and ethanol. From Fig. 4(c) it could be seen that the concentration was higher at the temperature of 45ºC with a good margin as time increased and this suggests that the extraction of beta-carotene

from soybean oil using acetone can best be done at 45ºC with increase in time of extraction. Fig. 3(d) shows the concentration of beta-carotene extracted from soybean oil using ethanol at different temperature and time. From the graph, temperature 50ºC showed highest concentration of beta-carotene and followed by temperature 45ºC. This also suggests that the extraction of beta–carotene from soybean oil using ethanol can be best achieved between temperature 45ºC and 50ºC with increase in time. As explained in section (2.2.7), increase in the concentration with increase in the temperature is due to increase in mass transfer rate with temperature. Increase in concentration with increase in time was as a result of longer time of shaking (agitation) which allowed for adequate mixing.

3.4 Effect of Time

The degree of extraction of beta-carotene from palm oil and soybean oil using acetone and ethanol was studied as a function of time separately. The volumes (dosages) of the oil samples were kept constant at 10 cm^3 for both acetone and ethanol. Solvent volume was kept constant at 30 cm^3 separately for palm oil and soybean oil and temperature was kept constant at 35ºC in the study for both substrates. Fig. 4(a) shows the concentration of beta-carotene with time using acetone and ethanol. It was observed that as the time of extraction increased, the concentration of beta-carotene from palm oil increased. Fig. 4(b) also shows the concentration of beta-carotene with time using acetone and ethanol. Fig. 4(b) shows that increase in time resulted in increase in the concentration of beta-carotene extracted from soybean oil until equilibrium was approached. The increase in concentration with increase in time observed in both Fig. 4 (a&b) could be attributed to the fact that as the time of extraction is increased it allows for longer time of shaking (agitation) which allows for adequate mixing of the substrates and solvents [13].

3.5 Effect of Solvent Volume

The extraction of beta-carotene was also studied as a function of solvent volume. The volume of the oil samples was each kept constant at 10 cm^3, time at 20 mins, temperature at 35ºC and solvent volume varied from 20,40,60 and 80 cm^3.

Fig. 5(a) shows that he concentration of the extracted beta carotene increased as the volume of the solvent increased using both acetone and ethanol from palm oil. Fig. 5(b) on the other hand shows the concentration of beta carotene extracted from soybean oil using acetone and ethanol. The result equally shows increase in concentration with increase in solvent volume. This increase in concentration could be attributed to the increase in partitioning of the analytes (oil samples) into the solvent layer [13].

3.6 Effect of Solvent

The ideal solvent for oil extraction must possess several features that are impossible to find in any

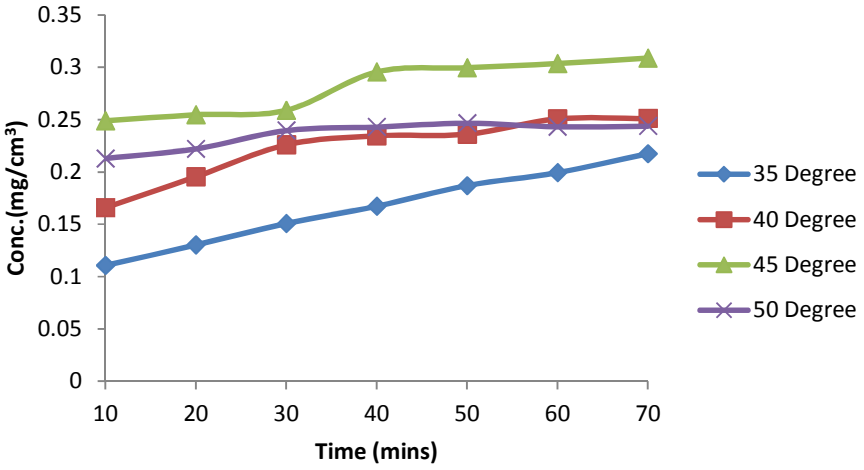

Fig. 3a. Concentration of beta carotene extracted from palm oil at different temperatures and time using acetone

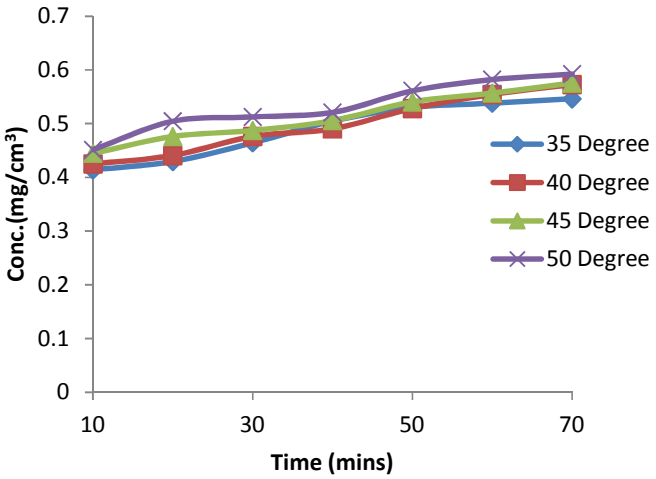

Fig. 3b. Concentration of beta carotene extracted from palm oil at different temperatures and time using ethanol

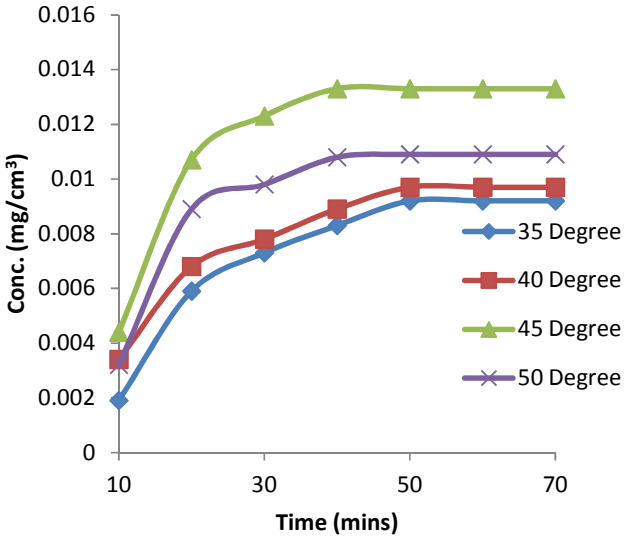

Fig. 3c. Concentration of beta carotene extracted from soybean oil at different temperatures and time using acetone

one solvent. One important factor regarding solvent selection is the lack of miscibility between them, such that they will form two distinct phases [13].

From the results in Fig. 6, it could be seen that the type of solvent had significant influence on the concentration of beta carotene extracted. This is to say that ethanol was able to extract higher concentration of beta carotene than acetone for both palm oil and soybean oil. This could be attributed to ethanol's immiscibility property with both palm oil and soybean oil.

3.7 Optimization Result for the Extraction of Beta-carotene from Palm Oil and Soybean Oil

The response of the optimization design for the extraction of beta carotene is presented in Table 2. The varying response figures are indication that the extraction parameters considerably affect the concentration of the beta-carotene

extracted. From the responses, it was observed that the highest predicted value of concentration of beta-carotene from palm oil using ethanol was 0.89448 mg/cm^3. This value corresponded at extraction time of 44.12 mins, volume of oil of 18.9 cm^3 and at extraction temperature of 44.13ºC. Furthermore, the highest concentration of beta-carotene predicted to be extracted from palm oil using acetone was 0.880082 mg/cm^3 which corresponded at extraction temperature of 43.77ºC, extraction time of 44.33 mins and oil volume of 18.91 cm^3. Hence, these predicted optimum conditions for solvent extraction of beta-carotene from palm oil were further used in the laboratory which gave 0.89448 mg/cm^3 from ethanol and 0.88008 mg/cm^3 from acetone. On the other hand, the same conditions were predicted for maximum extraction of beta-carotene from soybean oil in both ethanol and acetone. The predicted concentration of beta-carotene from soybean oil using ethanol was 0.445569 mg/cm^3 and 0.440041 mg/cm^3 using acetone. Hence the predicted optimum conditions were further used in the laboratory which gave 0.44557 mg/cm^3 from ethanol and 0.440043 from acetone. From the predicted and

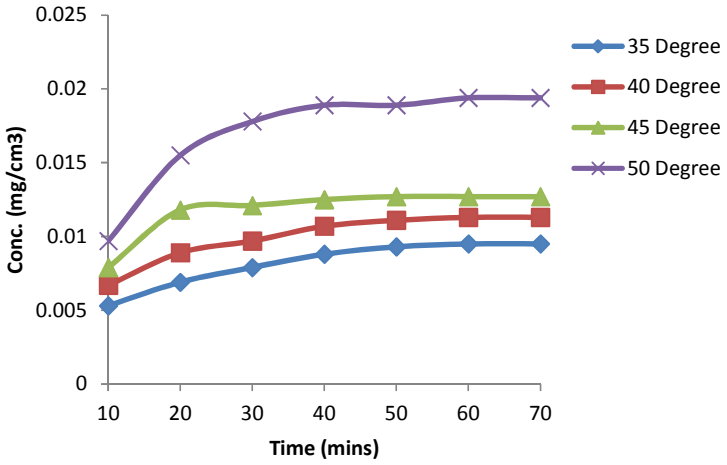

Fig. 3d. Concentration of beta carotene extracted from soybean oil at different temperatures and time using ethanol

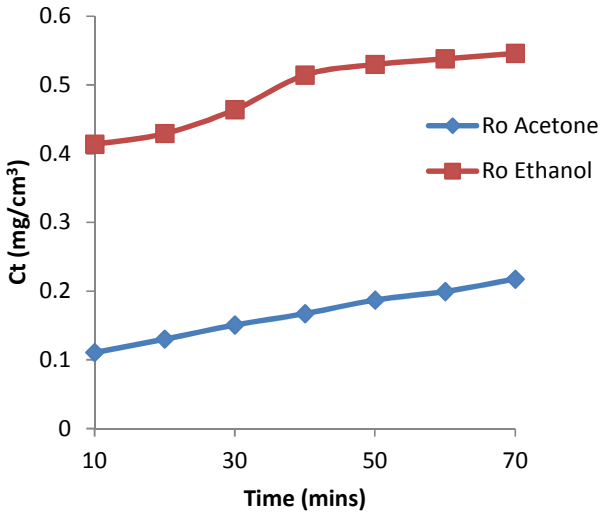

Fig. 4a. Effect of time on the extraction of beta carotene from palm oil using acetone and ethanol

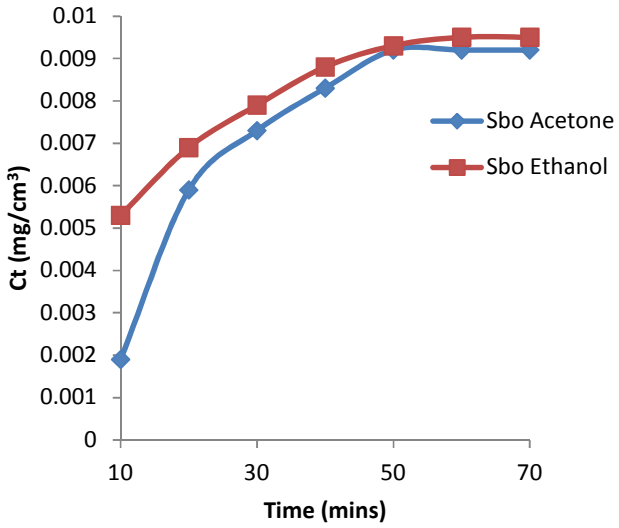

Fig. 4b. Effect of time on the extraction of beta carotene from soybean oil using acetone and ethanol

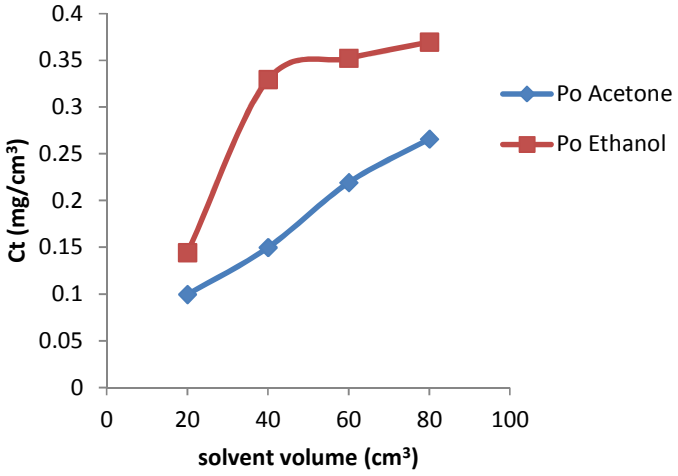

Fig. 5a. Effect of solvent volume on the extraction of beta carotene from palm oil using acetone and ethanol

experimental results, it was observed that there were negligible differences of 0.0001 using ethanol from palm oil 0.0002 using acetone from palm oil and 0.0002 using ethanol from soybean oil and 0.0005 using acetone from soybean oil. It can therefore be said that the model is a good model since it is capable of predicting the response. The values of concentration of beta-carotene extracted from palm oil using ethanol and acetone are close to value reported by Gob et al, 1985 to be between 500–700ppm (0.5 – 0.7mg/cm^3). The slight difference in concentration could be from the quality of the oil sample and the types of solvent/extraction method used.

Analysis of Variance (ANOVA) shows the model of the design; in this case, it is QUADRATIC MODEL.

The Model F-value of 126.18 implies the model is significant. There is only a 0.01% chance that a

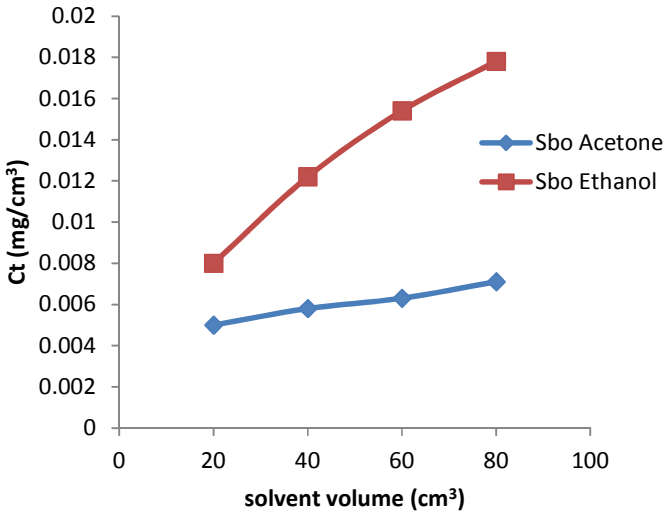

Fig. 5b. Effect of solvent volume on the extraction of beta carotene from soybean oil using acetone and ethanol

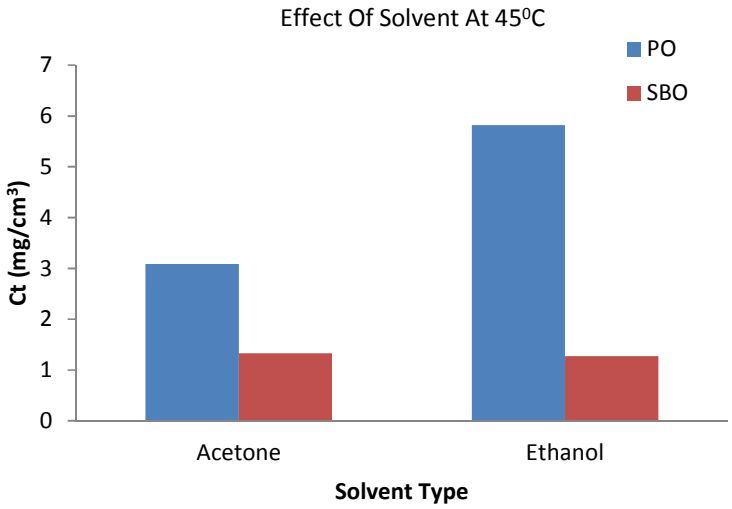

Fig. 6. Effect of solvent on the extraction of beta carotene from palm oil and soybean oil

"Model F-Value" this large could occur due to noise.Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case A, B, C, D, AB, A^2, B^2, C^2 are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. The "Lack of Fit F-value" of 0.99 implied the Lack of Fit was not significant relative to the pure error. There was a 53.47% chance that a "Lack of Fit F-value" this large could occur due to noise.

3.8 Model Equation (Palm Oil)

Final equation in terms of coded factors:

Beta Carotene from palm oil (Mg/cm3) = +0.85 + 0.043A + 0.05B + 5.527E–003C -7.265E–003D – 0.010AB – $0.023A^2$ – $0.027B^2$ – $6.383E–003C^2$ (3.1)

Final equation in terms of actual factors:

Types of Solvent; Ethanol

Beta carotene from palm oil (mg/cm^3) = -1.73366 + 0.089168Temp + 0.058378volume of oil + 5.65934E^{-003}Time – 4.03425E^{-004}Temp x volume of oil – 9.31684E^{-004}(Temp)^2 – 1.07703E^{-003} (Volume of oil)^2 – 6.38335E^{-005}(Time)^2 (3.2)

Type of solvent; Acetone

Beta carotene from palm oil (mg/cm^3) = 1.74819 + 0.089168Temp + 0.058378Volume of Oil + 5.65934E-003Time – 4.03425E-004Temp x volume of oil – 9.31684E-004(Temp)^2 – 1.07703E-003(Volume of oil)^2 – 6.3835E-005(Time)^2 (3.3)

3.9 Model Equation (Soybean Oil)

Model Equation (Soybean Oil)

Final equation in terms of coded factors:

Beta carotene from soybean oil (mg/cm3) = + 0.43 + 0.021A + 0.025B + 2.763E-003 C – 3.633E-003D – 5.043E-003AB – 0.012(A)^2 – 0.013(B)^2 – 3.192E-003 (C)^2 (3.4)

Final Equation in terms of actual factors:

Types of Solvent; Ethanol

Beta carotene from soybean oil (mg/cm^3) = -0.86683 + 0.44584Temp + 0.029189Volume of oil + 2.82967E-003Time – 2.01712E-004Temp x Volume of oil – 4.654842E-004(Temp)^2 – 5.38517E-004(Volume of oil)^2 – 3.19168-005(Time)^2 (3.5)

3.10 Model Graphs

Fig. 7 shows the one factor at a time plot of concentration of beta carotene extracted from palm oil using acetone and ethanol. From the plot (Fig. 7) can be seen that the concentration of beta-carotene extracted from palm oil using ethanol is higher than the concentration of beta-carotene extracted from palm oil using acetone. Therefore, ethanol can be said to be a better solvent for the extraction of beta carotene from palm oil than acetone. Similarly, the same result was gotten from soybean oil.

Fig. 8 show the 3-D plot of temperature and volume of the oil samples against beta carotene concentration from palm oil. Time was set constant at 40 mins and ethanol was used as the best solvent as identified by one factor plot. From Figure, it can be seen that temperature and volume of the oil samples had very good interaction which is evident from the positive effect of the increase in temperature of the extraction process with increase in the volume of the oil samples which resulted in increase in the concentration of the beta-carotene extracted from palm oil. In other words, increasing the temperature along with increasing the volume of the oil samples resulted in increase in the concentration of the extracted beta-carotene. In the same vein, the same result was gotten for soybean oil.

The Model F-value of 126.18 implies the model is significant. There is only a 0.01% chance that a "Model F-Value" this large could occur due to noise. Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case A, B, C, D, AB, A^2, B^2, C^2 are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. The "Lack of Fit F-value" of 0.99 implies the Lack of Fit is not significant relative to the pure error. There is a 53.47% chance that a "Lack of Fit F-value" this large could occur due to noise.

Table 3. Analysis of variance for extraction of beta-carotene from palm oil

Source	Sum of squares	df	Mean square	F Value	P-Value Prob> F
Model	0.20	8	0.024	126.18	<0.0001
=A-temp (ºC)	0.059	1	0.059	304.24	<0.0001
B-volume of oil (cm3)	0.079	1	0.079	407.30	<0.0001
C-Time(mins)	9.774E-0.04	1	9.774E-004	5.05	0.0319
D-Type of solvent	2.111E-003	1	2.111E-003	10.90	0.0024
AB	1.628E-003	1	1.628E-003	8.40	0.0068
A2	0.027	1	0.027	140.87	<0.0001
B2	0.036	1	0.036	188.25	<0.0001
C2	2.049E-003	1	2.049E-003	10.58	0.0028
Residual	6.004E-003	31	1.937E-004		
Lack of fit	4.051E-003	21	1.929E-004	0.99	0.5347
Pure Error	1.953E-003	10	1.953E-004		
Cor Total	0.20	39			

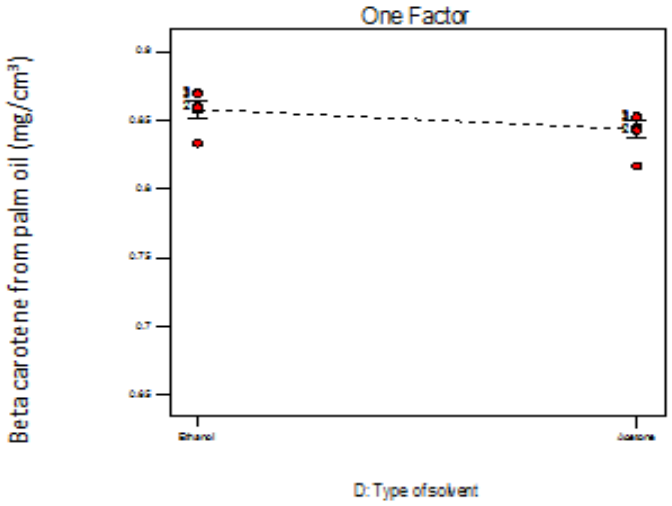

Fig. 7. One factor plot

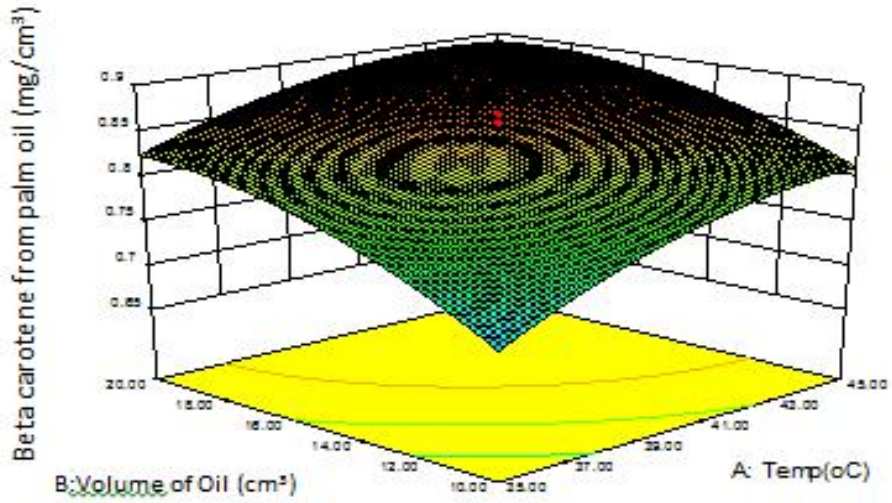

Fig. 8. 3-D plot of temp and volume of oil vs beta carotene conc. for palm oil

Table 4. Analysis of variance for extraction of beta-carotene from palm oil

Source	Sum of squares	Df	Mean square	F value	P-value (Prob> F)
Model	0.049	8	6.109E-003	126.18	<0.0001
A-temp (ºC)	0.015	1	0.015	304.24	<0.0001
B-volume of oil (cm3)	0.020	1	0.020	407.30	<0.0001
C-Time(mins)	2.443E-0.04	1	2.443E-004	5.05	0.0319
D-Type of solvent	5.278E-004	1	5.278E-004	10.90	0.0024
AB	4.969E-004	1	4.069E-004	8.40	0.0068
A2	6.820E-003	1	6.820E-003	140.87	<0.0001
B2	9.114E-003	1	9.114E-003	188.25	<0.0001
C2	5.123E-004	1	5.123E-004	10.58	0.0028
Residual	1.501E-003	31	4.842E-005		
Lack of fit	1.013E-003	21	4.823E-005	0.99	0.5347
Pure Error	4.882E-004	10	4.882E005		
Cor Total	0.050	39			

Table 5. Validation of model equation (Palm oil)

Volume of oil (ml)	Temperature (^oC)	Time (Mins)	Type of Solvent	Beta carotene (mg/ml)		Error (%)
				Predicted	Experimental	
18.9	44.13	44.12	Ethanol	0.894481	0.89448	0.0001
18.91	43.77	44.33	Acetone	0.880082	0.88008	0.0002

Table 6. Validation of model equation (Soybean oil)

Volume of oil (ml)	Temperature (^oC)	Time (Mins)	Type of solvent	Beta carotene (mg/ml)		Error (%)
				Predicted	Experimental	
18.9	44.13	44.12	Ethanol	0.445569	0.44557	0.0002
18.91	43.77	44.33	Acetone	0.440041	0.440043	0.0005

3.11 Optimum Conditions

The optimum conditions for the extraction of beta carotene from palm oil are as shown in Table 5 while the optimum conditions for the extraction of beta carotene from soya bean oil are as shown in Table 6.

4. CONCLUSION

Extraction of beta-carotene from palm oil and soybean was carried out by solvent extraction method using acetone and ethanol. Increasing the temperature, solvent volume, volume of the oil samples, time, all positively influenced the extraction of beta-carotene from palm oil and soybean oil. Among ethanol and acetone used for the extraction in both substrates, ethanol proved to be the best solvent for the extraction of beta-carotene from palm oil and soybean oil. Optimization of the process factors studied was done and the optimum conditions for the extraction of beta-carotene were 18.9 cm^3 of palm oil sample, 44.13ºC, 44.12 mins for ethanol and 18.91 cm^3 of palm oil, 44.33 mins, 43.77ºC, for acetone. The same conditions were generated by the model for the extraction of beta-carotene from soybean oil. Six kinetics models were used to test the experimental data and pseudo-second order model proved to fit the experimental data better with the largest correlation coefficient at all the temperatures studied. Therefore, it can be concluded that the extraction of beta-carotene from palm oil and soybean oil is possible provided the degradation temperature of beta-carotene (which is 45ºC) is not exceeded.

ACKNOWLEDGEMENT

The authors sincerely thank Pymotech research center and laboratories Enugu, Enugu State Nigeria for their facilities used.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/52369