

NATURAL ADDITIVES EFFECTS ON SOME QUALITY PARAMETERS OF UN-REFINED PEANUT OIL

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ABSTRACT

Edible oils are widely utilized in diets as a major source of fat and a heat transfer medium during frying. Improvement on nutritive value is a major concern in edible oil industry. Therefore, objectives of this work were to investigate the effects of sweet pepper (*Capsicum annum*) and tomato fruit (*Lycopersicon esculentum*.) as additives on some physical and functional properties, nutritive values and stability of peanut (*Arachis hypogaea*) oil. Response surface methodology was employed to study effect of natural additives on peanut oil. The variables were additives ratio (5:0, 3:2, 2.5:2.5, 2:3, 0:5) of sweet pepper: tomato and incubation time (1, 1.5, 2.5, 4, 6 hrs). While the responses were oil iodine value, peroxide value, free fatty acid, saponification value, colour, viscosity, beta-carotene, vitamin E and vitamin K. The calculated means for iodine value, peroxide value, free fatty acid, saponification value, colour, viscosity, β -carotene, vitamin E and vitamin K were 105.39 ± 10.38 Wijs, 9.89 ± 2.81 mEqkg⁻¹, $2.4 \pm 1.13\%$, 166.1 ± 20.53 mgKOH/g, 0.16 ± 0.2 A, 0.02 ± 0.00 Nsm⁻², 788.3 ± 69.50 μ g/100g, 12.7 ± 0.87 μ g/100g and 7.8 ± 0.59 μ g/100g, respectively. Coefficient of determination (R^2) of models for iodine value, peroxide value, free fatty acid, colour, viscosity, β -carotene and vitamin E were 0.76, 0.91, 0.76, 0.89, 0.93, 0.97 and 0.84, respectively. Functional properties of the oil including iodine value and saponification value reacted differently to treatment. Iodine value was significantly ($p < 0.05$) influenced while non-significant ($p > 0.05$) effect was recorded for the saponification value. The treatments significantly ($p < 0.05$) influence peroxide value and free fatty acid of the extracted oil. Physical properties of the peanut oil measured by colour and viscosity were significantly influenced by the treatment ($p < 0.05$). Nutritive values of the peanut oil which were beta-carotene and Vitamin E were significantly ($p < 0.05$) influenced by the treatments but Vitamin K was not significantly influenced.

Key words: Peanut, Additives, Oil, Stability, Properties

INTRODUCTION

Peanut (*Arachis hypogaea*) is a leguminous plant grown mainly for its seeds. It is the thirteenth most important food crop and fourth most important source of vegetable protein in the world [1]. The major peanut producers in the world are China, India, Nigeria, and Sudan [2]. Peanut is a concentrated source of energy with its seeds having nutritive values of fat (41-52%), easily digestible protein (21-30%), and carbohydrate (11-27%) [3]. The seed is valued for its high oil content [4]. The oil extracted from peanut is useful in the manufacture of margarine, mayonnaise, salad oils, soaps and lather shaving creams [5].

Fortification of edible oil to reduce deficiency of vitamin A is recommended [6]. Synthesized additives are commonly used to fortify edible oil. The availability and increased consumption of beta-carotene-rich foods in the daily diet is a preferred strategy for improving nutritional status than synthetic vitamin A dosage approaches [5]. Vegetables are highly nutritious sources of vitamins and small amounts of minerals [7]. Sweet pepper (*Capsicum annum*) and tomato fruit (*Lycopersicon esculentum*.) are vegetables widely cultivated in Nigeria. Sweet pepper it's known for its pungency, impartation of flavours to foods and richness in vitamins. Sweet pepper and Tomato being rich in carotenoids also will impart colour and increase the β -carotene content of the oil. In human medicine, β -carotene not only serves as valuable source of vitamin A, but also serves as a potent antioxidant, scavenging free radicals and quenching singlet oxygen; thereby it is capable of reducing the risk of development of certain types of cancer [8].

Improvement on nutritive value is a major concern in edible oil industry. Therefore, objectives of this work were to study the effects of sweet pepper and tomato fruit as additives on some physical and functional properties, nutritive value, and stability of peanut oil.

MATERIALS AND METHODS

Experimental design

The experimental design was based on the rotatable design of Response Surface Methodology for a two-variable case as described by Montgomery [9]. The two independent variables were the additives ratio (A) and incubation time (B). The combinations of the additives were 100:0, 60:40, 50:50, 40:60 and 0:100 corresponding to 5g of sweet pepper/0g of tomato, 3g of sweet pepper/2g of tomato, 2.5g of sweet pepper/2.5g of tomato, 2g of sweet pepper/3g of tomato, and 0g of sweet pepper/5g of tomato. The incubation time ranged from 1 to 6 hrs. The pH of the mixture was unmodified and the temperature of incubation was kept constant at 40 ± 2 °C for all the treatments. Total numbers of treatments were 13 (Table 1). All the experimental procedures were replicated three times; mean values were recorded.

Preparation of samples

Cultivars of sweet pepper, tomato and peanut used for the experiment were *OAB 99-7*, *Roma- VF* and *Boro light* respectively. The raw materials (sweet pepper, tomato fruits and peanut) were obtained from National Horticultural Research Institute, Ibadan, Nigeria. They were cleaned and ground using plate mill (model FE 145A, FOBA Engineering, Nigeria) separately. Their initial moisture contents were determined using ASABE [10], while sweet pepper and tomato pH determination were carried out as described using the glass electrode TOA pH meter [11]. Sweet pepper-tomato ratio (5 g) was prepared as additive. This mixture was added to 20 g of peanut and incubated at 40 ± 2 °C at varied time (Table 1) using a Gallenkamp (size two) incubator. Oil was extracted from incubated samples using Soxhlet apparatus using gravimetric method for crude fat determination [12]. Oil from un-treated sample of same oilseed was used as control.

Determination of some physicochemical properties of raw materials

The physicochemical properties of sweet pepper, tomato and peanut were determined to establish their quality characteristics. These were determined using recommended analytical and sampling methods, Codex Stan 234-1999 as guide [13]. Methodologies adopted were AOAC method for apparent density for specific gravity [12]; ASABE S410.1 method for moisture content [10]; AOAC 981.12 method for pH; AOAC 920.39C method for fat; AOAC 942.15 method for titratable acidity; AOAC 950.49 method for ash; AOAC 962.10 method for fiber; AOAC 954.01 for crude protein determination [12] and carbohydrate by calculation as reported [14]. Unit mass was measured by electronic weigh balance of 0.01g accuracy (JD-3G Series, Shenyang Longteng Electronic Co. Ltd, Liaoning, China). The colour of the additives was determined with the aid of spectrophotometer (UVIKON XL, North Star Scientific, Leeds, UK) at 470 nm absorbance.

Determination of iodine value

The iodine value was determined using AOCS Method Cd 1-25 [15]. The procedure involved adding excess Wijs reagent to the oil sample and allowing the mixture to react for 30 min at 25 ± 5 °C. Excess reagent was treated with potassium iodide to convert it to equivalent iodine and titrating with thiosulfate reagent and a starch indicator until blue color disappears.

Determination of peroxide value

The peroxide value of the oil was determined by a modified American Oil Chemists' Society standard method Cd 8-53 [15]. Five milliliters of oil was placed on a test paper (United States Patent 4098575). Ten milliliters of water was added to oil soaked paper. The intensity of the blue colour developed on the test paper is proportional to the peroxide value of the oil. Colour of the paper was compared to a predetermined standard [16].

Determination of free fatty acid content (FFA)

Free fatty acid (FFA) was calculated using AOCS method Ca5a – 40 [15]. Mixture of 1.0g of oil, 25ml of diethyl ether, 25 ml of ethanol and 1ml of phenolphthalein solution was prepared. The mixture was titrated with aqueous 0.5N NaOH, which was shaken vigorously until a permanent faint pink colour appeared and persisted for 15 sec. The percentage of FFA in the sample was calculated using the equation 1 below.

$$\%FFA = \frac{\text{Titration (ml)} \times 0.141 \times 100}{\text{Wt of sample used}} \dots\dots\dots 1$$

Determination of saponification value (SV)

Saponification value of oil sample was determined according to the method reported by Pearson [17]. This was done by dissolving 1g of the oil in 12.5 ml of 0.5% ethanolic KOH. One milliliter of phenolphthalein was added to the mixture as indicator before titration with 0.5 N HCL. A blank determination was also carried out under the same condition. Saponification value was calculated using equation 2 below.

$$SV = 56.1 N \frac{(Q - Z)}{W} \dots\dots\dots 2$$

Where N is normality of HCL; Q is volume (ml) of H₂SO₄ for blank (ml); Z is volume (ml) of H₂SO₄ for sample (ml); W is Weight of oil (g) and 56.1 is constant which is equivalent to weight of potassium hydroxide.

Determination of colour

The color of the oil was determined using spectrophotometer colour method AOCS method Cc 13c -50 [15]. Mixture of oil and n-hexane was prepared in ratio 1 to 9 ml and the absorbance was read at 470 nm on UVIKON XL spectrophotometers (North Star Scientific, Leeds, UK).

Determination of oil viscosity

The oil viscosity was evaluated as described [18]. It was determined at 25 °C using Brookfield Synchroelectric Viscometer (Model RVT, Rookfield Engineering Laboratories, Stoughton, MA) and a helipath stand equipped with a T – B spindle operated at 2.5 rpm.

Determination of beta-carotene (β-carotene)

Beta-carotene was determined according to AOAC method No 970.64 [12]. A mixture of 2 g of oil sample, 10 ml of distilled water and 25ml alcoholic KOH solution was heated in boiling water bath (Techmel and Tecmel, Texas, USA) for 1 hour. The mixture was cooled to room temperature (29°C), after which 30 ml of water was added. After hydroxylation, the mixture was extracted using chloroform. Traces of water in the extract was removed by the addition of 2 g anhydrous Na₂SO₄. Standard solution of β-carotene

was obtained by dissolving 0.003 g of β-carotene in 100 ml of chloroform. Thereafter, a series of standard solutions of different concentration were obtained and their absorbance determined. Absorbance of sample and standards were read on the spectrophotometer at 470 nm wavelength (UVIKON XL spectrophotometers, North Star Scientific, Leeds, UK). Values of β-carotene were calculated using equation 3 below:

$$\beta - \text{carotene} \left(\frac{\mu\text{g}}{100\text{g}} \right) = \frac{\text{absorbance of sample X dilution factor X gradient}}{\text{weight of sample}} \dots\dots\dots 3$$

Determination of Vitamin E

Vitamin E was determined using AOAC 971.30 method [12]. A mixture of oil (1g), absolute alcohol (10 ml) and 1M alcoholic sulphuric acid (20ml) was refluxed for 45 minutes in a condenser and cooled for 15 minutes. Unsaponifiable matter in the mixture was extracted with dimethyl ether. The extracts evaporated at a low temperature and the residues obtained were dissolved in 10 ml absolute alcohol. Absolute alcohol (5 ml) and 1ml conc. HNO₃ was added to aliquots of the sample and standards (0.3-3.0 mg vitamin E). The mixture was evaporated in a water bath (Techmel and Tecmel, Texas, USA) at 90°C for 3 minutes from the time the alcohol started boiling. A series of standard solutions of known concentrations were determined with reference to their absorbance from which average was recorded. It absorbance was measured at 290 nm using a UVIKON XL spectrophotometer (North Star Scientific, Leeds, UK) against a blank containing 5 ml absolute alcohol and 1ml conc. HNO₃ and treated in a similar manner. Equation 4 was applied to determine Vitamin E.

$$\text{Vitamin E} \left(\frac{\mu\text{g}}{100\text{g}} \right) = \frac{\text{absorbance of sample X dilution factor X gradient}}{\text{weight of sample}} \dots\dots\dots 4$$

Determination of vitamin K

Vitamin K was determined using AOAC method 992.27 [12]. Five grams of oil sample was weighed into a 250 ml beaker and 30ml of butyl alcohol was added. Magnetic stirrer was used to homogenise the solution. The resulting mixture was filtered through a Whatman No 42 filter paper. Three drops of 2, 4-dinitrophenyl hydrazine and 3 ml of alcoholic ammonia were added to 10 ml of the mix. Standard solutions of vitamin K from 0-20 μg/ml were prepared and treated as samples to obtain the slope of the standard curve. The absorbance of standards and sample were read using UVIKON XL spectrophotometers (North Star Scientific, Leeds, UK) at 325 nm wavelength. Vitamin K in μg/100g was calculated using equation 5.

$$\text{Vitamin K} \left(\frac{\mu\text{g}}{100\text{g}} \right) = \frac{\text{absorbance of sample X dilution factor X gradient}}{\text{weight of sample}} \dots\dots\dots 5$$

Data analysis

Mean values of three replicates recorded as obtained data were analyzed using Analysis of Variance (ANOVA) and regression. Test of significance was fixed at 95% confidence level. Second-order polynomial equations were developed to express the relationship between variables and responses using proprietary software (Design-Expert, Statsoft Inc., Minneapolis, USA).

RESULTS

Preliminary data

The physical and chemical properties of sweet pepper, tomato and peanut used for the experiment were presented as Table 2.

Iodine value

The obtained iodine values (IV) ranged from 100.07 to 116.9 Wijs (Table 3). Least value is slightly lower than control value of 103.38 Wijs. Analysis of variance of the data showed significant effect of treatments ($p < 0.05$) on iodine value of the oil. A second order model 2FI (Equation 6) was adequate to describe the variation in the iodine value of the peanut oil due to additive ratio (A) and incubation time (B). Additives ratio (A) and interaction between the responses were the significant model terms at $p < 0.05$. Visual illustration of the relationship is shown as Fig. 1. A curve linear trend was observed with additives showing higher effect on the response.

$$IV = +77.13 + 16.91A - 7.93B - 15.86AB \dots \dots \dots 6$$

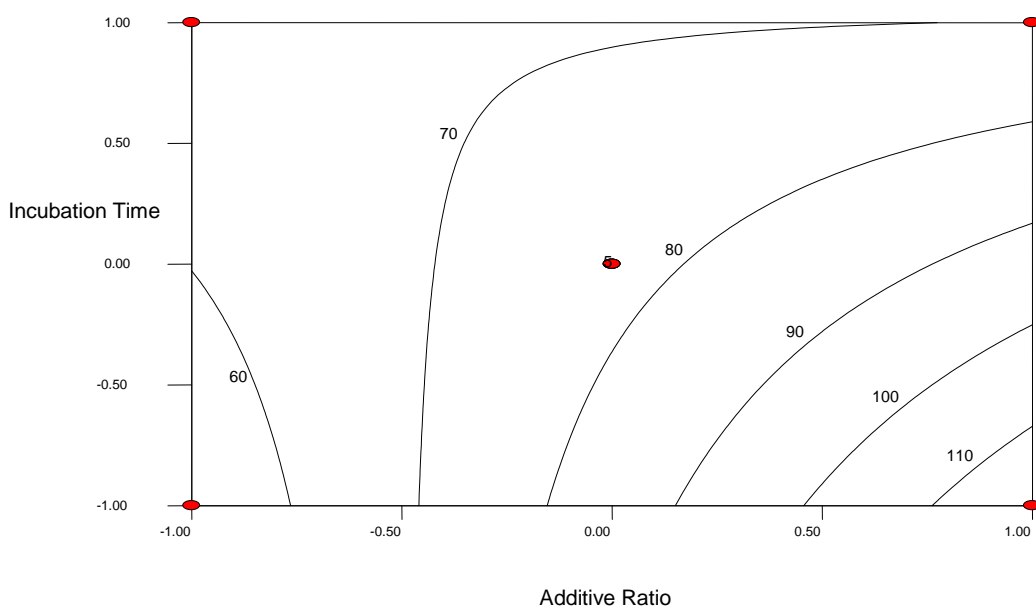


Figure 1: Contour plot of treatments effect on iodine value of oil

Peroxide value

Minimum and maximum peroxide values (PV) were 5.4 and 12.3 meqkg⁻¹, respectively (Table 3). Least peroxide value recorded from the experiment was higher than the control (3.6 meqkg⁻¹). Treatments significantly influenced (p<0.05) the peroxide value of the oil. Response surface cubic model was appropriate to illustrate the relationship (Equation 7). The contour plot of the relationship showed asymmetric shape (Fig. 2). Coefficient of the determination R² of the model was high 0.91.

$$PV = 10.70 + 1.5A + 1.2B - 2.2AB - 1.1A^2 + 0.6B^2 - 3.5A^2B + 0.8AB^2 \dots\dots\dots 7$$

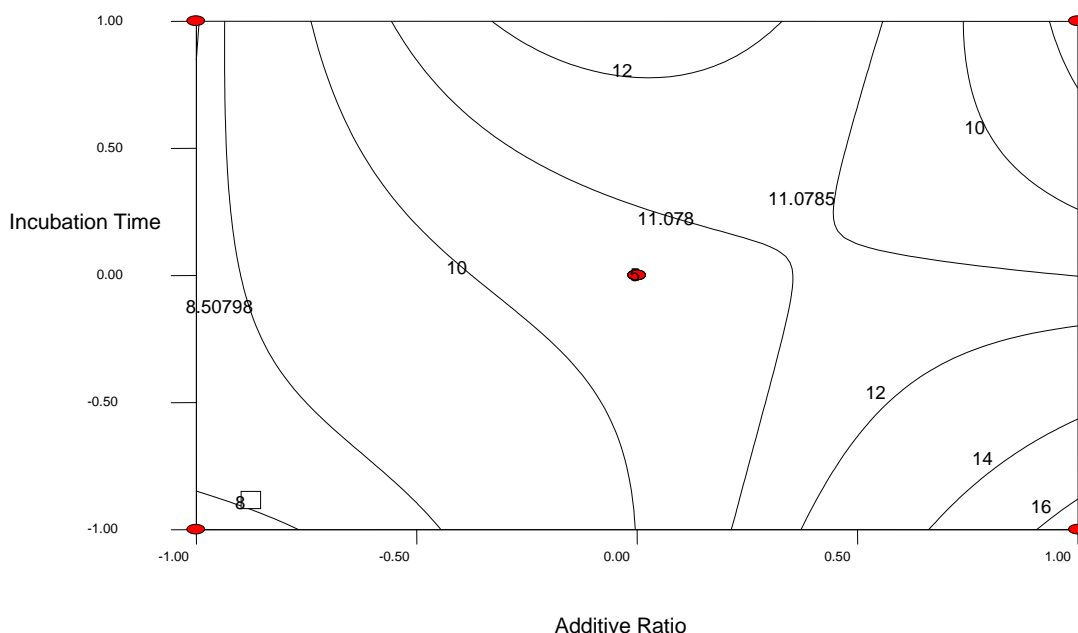


Figure 2: Contour plot of treatments effect on peroxide value of oil

Free fatty acid

Treatment of the oil by addition of tomato and pepper significantly (p < 0.05) affects its free fatty acid content (FFA). Obtained FFA ranged from 0.84 to 4.82 % (Table 3). Minimum value was higher than 0.56 % of untreated peanut oil used as control. Regression analysis results also revealed that linear and quadratic terms (A and A²) of the additives were significant model terms at p<0.05 (Equation 8). Figure 3, visually illustrates the interaction between additives, incubation time and free fatty acid content of the oil. Trend of the plot showed increase in free fatty acid with increase incubation time.

$$FFA = +3.14 + 0.63A - 0.32B - 0.85AB - 0.69A^2 - 0.54B^2 \dots\dots\dots 8$$

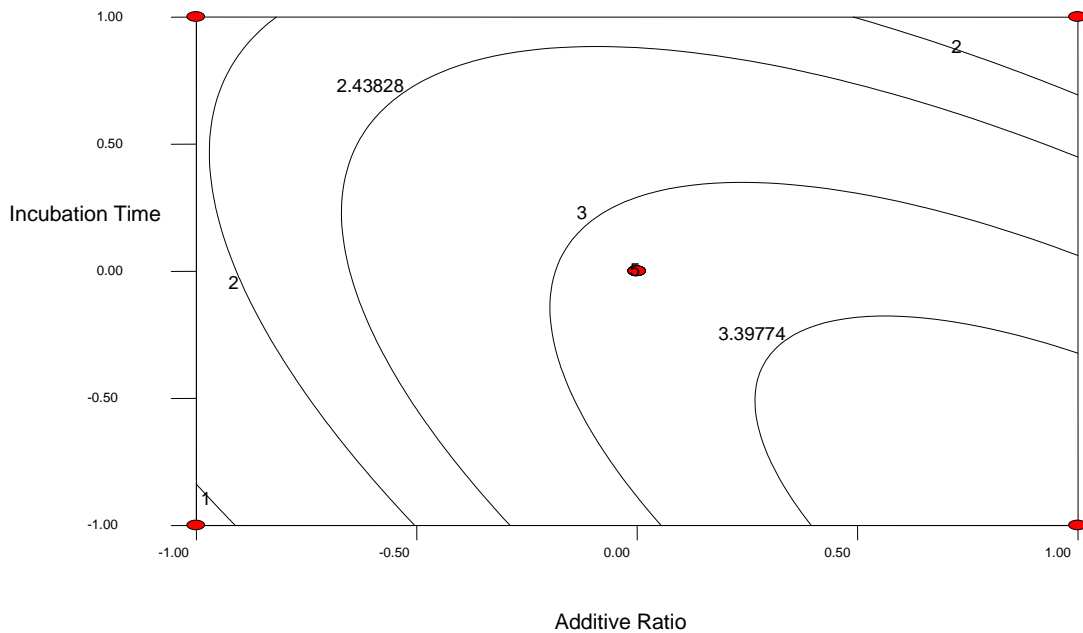


Figure 3: Contour plot of treatments effect on free fatty acid of oil

Saponification value

The mean saponification value (SV) of the oil was 166.14 ± 20.55 mg/100g. The contour plot (Fig. 4) demonstrates that linear relationship exists between the response and the variable. However, non significant effect ($p > 0.05$) of treatments on SV was observed. Generated regression model was not fit.

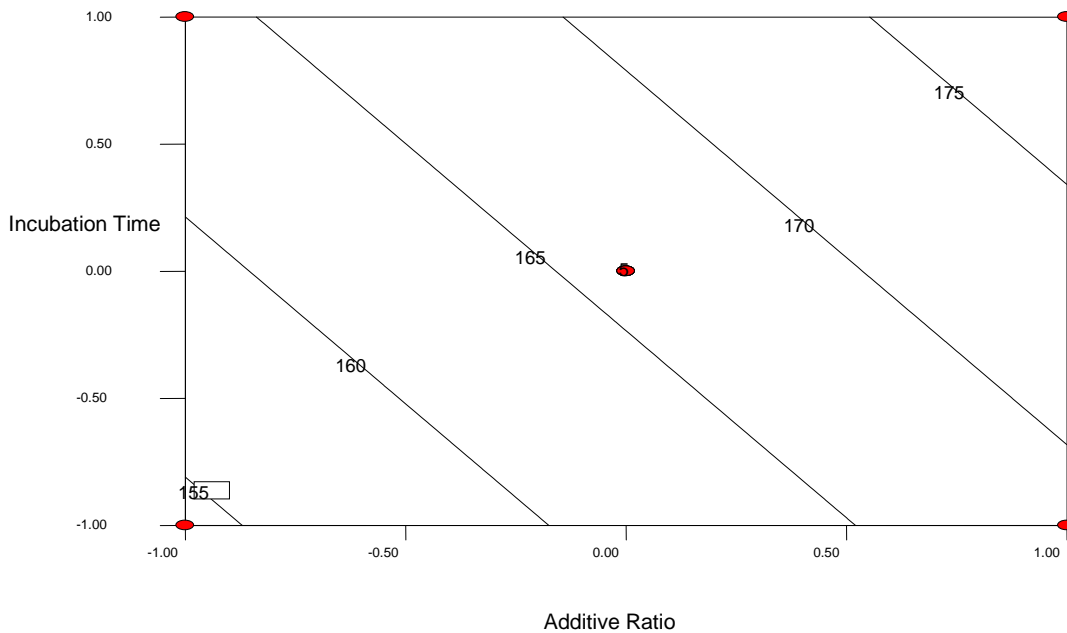


Figure 4: Contour plot of treatment effect on saponification value of oil

Colour

The colour (CL) readings ranged from 0.13 to 0.20 A (Table 3). It was observed that treatments with high incubation time produced high colour concentration. Effect of treatment was significant ($p < 0.05$) on oil colour. Both additives and incubation time were significant terms of the regression model (Equation 9). Figure 5 showed the contour plot of the interaction between treatment and oil colour. The plot revealed parabolic shape.

$$CL = +14.4 - 1.42A + 0.91B + 0.33AB + 1.24A^2 + 2.09B^2 \dots \dots \dots 9$$

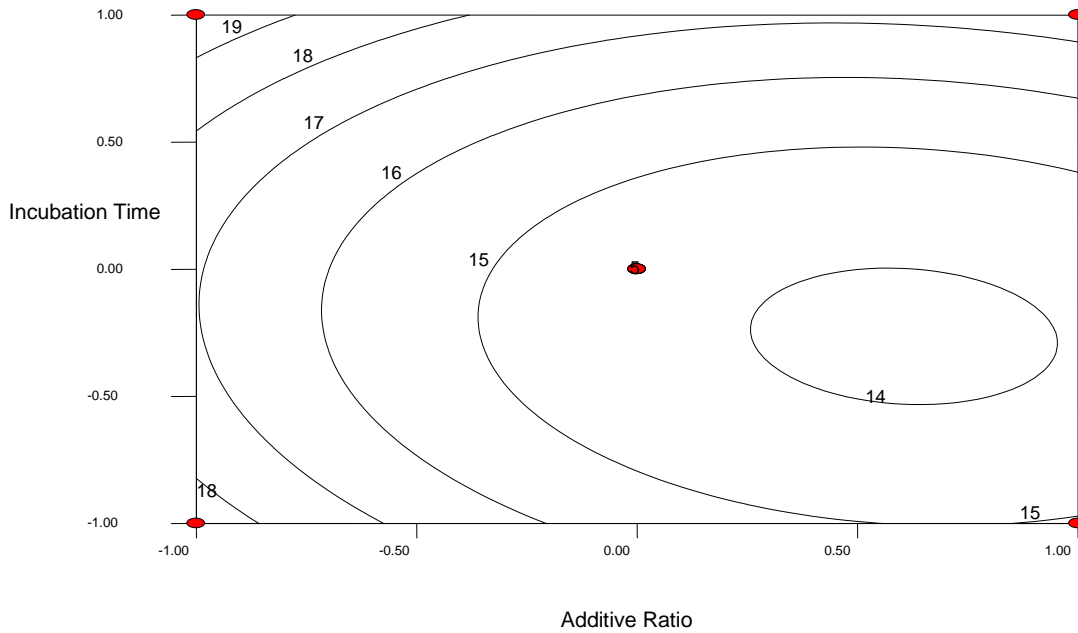


Figure 5: Contour plot of treatments effect colour of oil

Oil viscosity

Minimum and maximum viscosities (V_s) of treated oil were 0.01 and 0.02 Nsm^{-2} respectively, whereas that of the control was 0.01 Nsm^{-2} (Table 3). Effect of additives and incubation time were significant ($p < 0.05$) on oil viscosity. The contour plot (Fig. 6) revealed pronounced independent influence of the variables on the oil viscosity. Equation 10, mathematically expresses the relationship. Coefficient of the determination R^2 of the model was high 0.93.

$$V_s = 1.5 - 0.2A + 0.09B - 0.11AB + 0.18A^2 + 0.12B^2 - 0.18A^2B + 0.40AB^2 \dots \dots 10$$

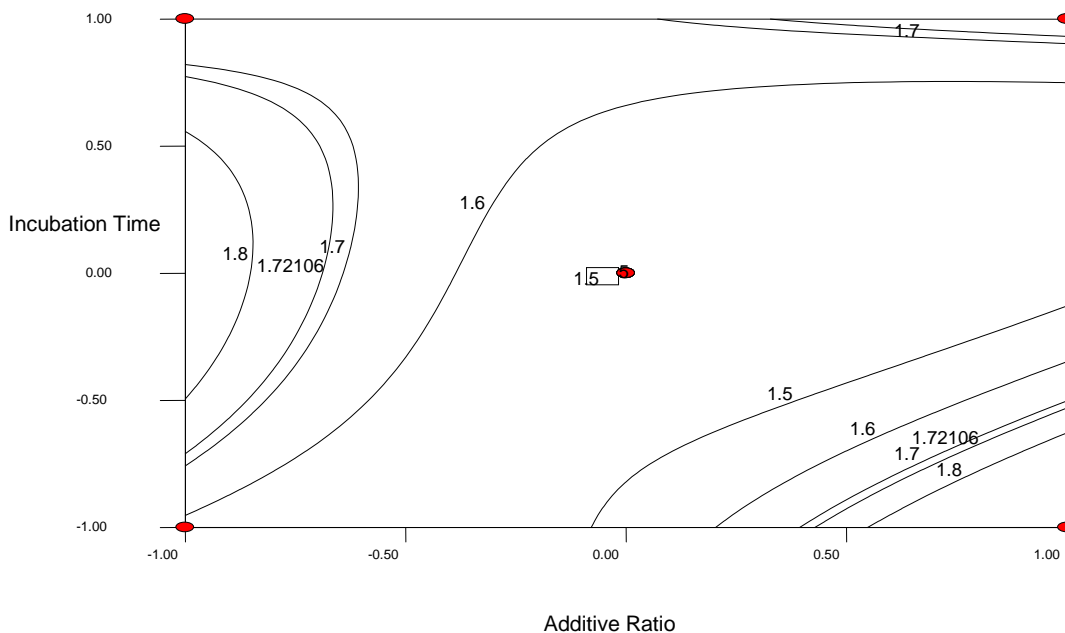


Figure 6: Contour plot of treatments effect on viscosity of oil

Beta-carotene (β -carotene)

Average beta-carotene of the treated oil was $788.3 \pm 69.5 \mu\text{g}/100\text{g}$. Least beta carotene recorded from the treated samples was $670 \mu\text{g}/100\text{g}$ while $866 \mu\text{g}/100\text{g}$ was the highest value (Table 3). Difference in control ($525 \mu\text{g}/100\text{g}$) and least value was $165 \mu\text{g}/100\text{g}$ (31.4%). Analysis of variance showed significant effect of treatments at $p < 0.05$ on beta-carotene. Response surface quadratic model (Equation 11) generated to predict the relationship revealed higher effect of additives (A) than incubation time (B). Coefficient of determination of the model was 0.94. Contour plot of the relationship is shown as Figure 7.

$$\beta - \text{carotene} = 866 - 17.61A + 3.30B + 18.5AB - 48.38A^2 - 77.89B^2 \dots\dots\dots 11$$

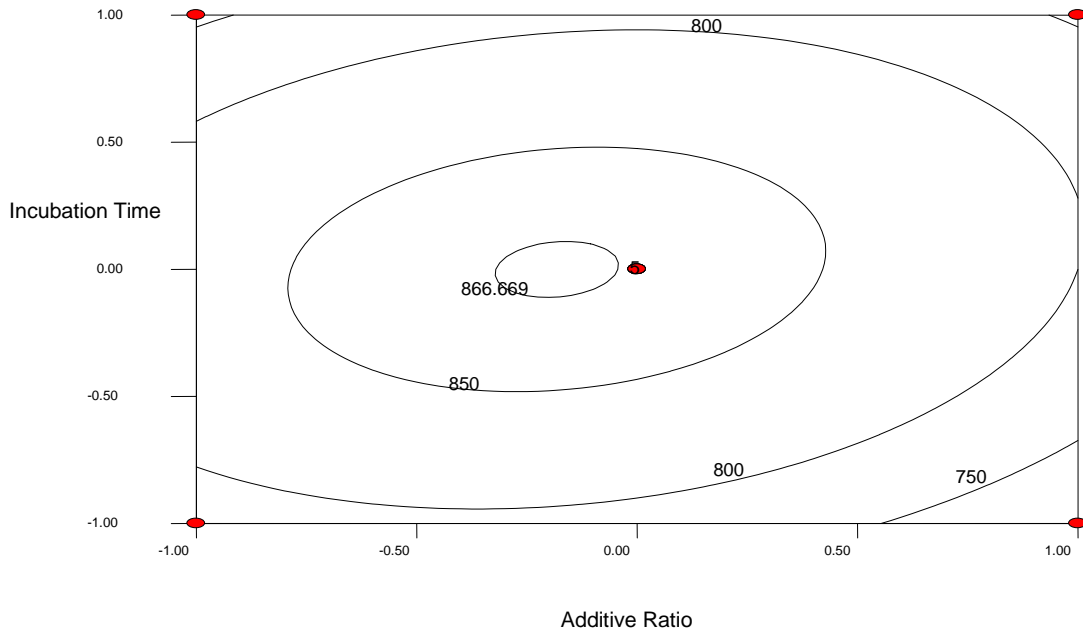


Figure 7: Contour plot of treatments effect on beta-carotene of oil

Vitamin E

The treatments significantly ($p < 0.05$) increased vitamin E (Vit E) content of the oil. About 50% increment was recorded between least and control values. From the contour plot (Fig. 8) and regression model (Equation 12) effect of incubation time (B) was more pronounced.

$$\text{Vit E} = 12 - 0.34A + 0.53B + 0.1AB + 0.73A^2 + 0.41B^2 \dots \dots \dots 12$$

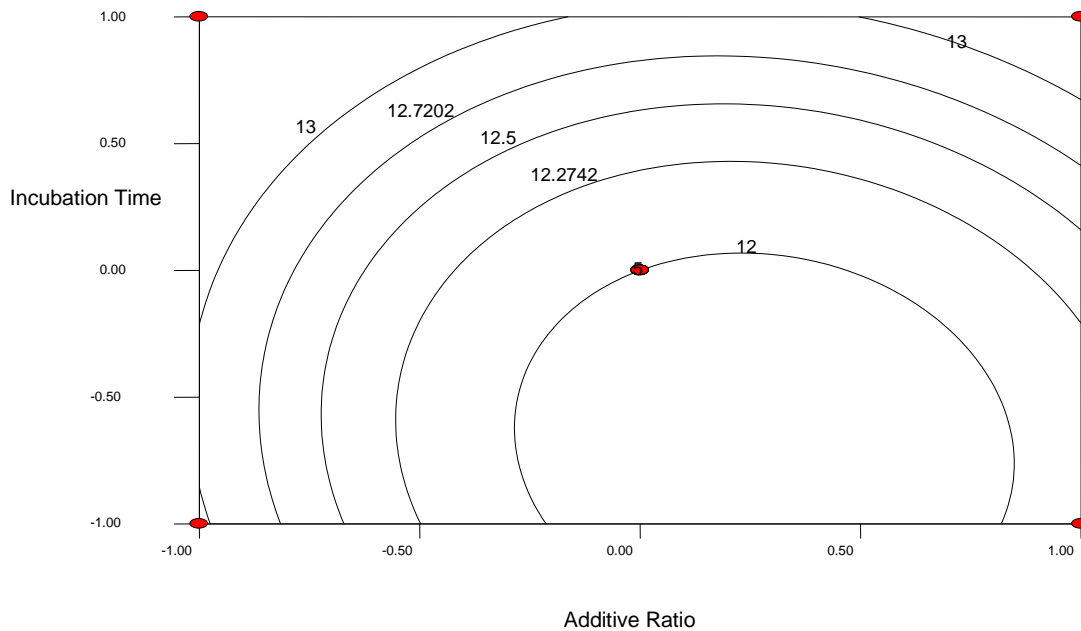


Figure 8: Contour plots of treatments effect on Vitamin E of oil

Vitamin K

Recorded Vitamin K (Vit K) ranged from 6.75 to 8.61 $\mu\text{g}/100\text{g}$ with mean value of $7.75 \pm 0.058 \mu\text{g}/100\text{g}$. The treatments had no significant effect ($p > 0.05$) on the Vitamin K content of the extracted oil. Attempt to use model equation to predict the relationship also showed poor fitness. The model did not satisfy lack of fit test.

DISCUSSION

Preliminary data

Fruits are graded by their physical characteristics (size, weight, colour and shape), internal attributes (soluble solids content, moisture content) and rating quality (proximate composition) [19]. Based on these quality parameters, pepper and tomato used for the study can be classified as matured-ripe fruits [20]. The general specification project for pickled products established by the processed fruit and vegetable committee of the Codex Alimentarius Commission FAO/OMS, recommend a maximum pH specification of 4.6 for pickled products [21]. The pH of the peanut and tomato fruit were in agreement with reported natural values for the crops [4, 7]. The information is required because enzymatic activities are pH dependent.

Iodine value

Iodine value is a measure of reactive groups like the double bound in the mixture. Natural enzymes that are present in tomato fruit might have acted as catalyst for the reaction [22].

Similar observation was reported on iodine value of olive fruit oil treated with pectinase, an enzyme [23]. Increase in iodine value denotes increase in the degree of un-saturation of the oil [24]. Coefficient of determination R^2 of the model was 0.76 which showed that model can only explain 76.18% variability in the response.

Peroxide value

This observed trend of peroxide values in this study was contrary to reports that enzymatic treatments reduced peroxide value of some other vegetable oils [23, 25]. Peroxides are among the oxidation products formed when the double bonds of unsaturated fats become oxidised and an increase in peroxide value could have been caused by the peroxidase activity present in tomato and pepper reference. Peroxide value is used as indicator of deterioration of oils. Recommended maximum peroxide value in edible oil is 10 meq kg⁻¹ [6]. High PV of the extracted oil may be traced to high moisture contents of pepper and tomato. Moisture content naturally accelerates rate of oxidative reaction [26]. High coefficient of determination (R^2) of the model 0.91 is an indication of model fitness. The irregular shape of the contour plot (Fig. 2) may justify why the cubic quadratic response model was appropriate.

Free fatty acid

Other authors reported an increase in free fatty acid of olive oil as result of addition of enzymes [25]. Thus, natural enzymes in pepper and tomato might have caused the observed FFA rise in the treated peanut oil. Free fatty acid content is one of the major quality parameters of edible oils. Permissible level of free fatty acid in un-refined vegetable oil is 2% [6].

Saponification value

Saponification values of domestic vegetable oils range from 180 to 200 mgKOH/g [27]. The higher the saponification number, the higher the number of fatty acids of low molecular weight. It is obvious from the contour plot (Fig. 4) that a linear relationship exists between the response and the variable. That is rate of change of saponification value is directly proportional combined effect of additive ratio and incubation time.

Colour

The observation that treatments with high incubation time produced high colour concentration is suggesting that incubation encourages colour formation reaction. The red colour of pepper and tomato is due to carotenoids [25]. The presence of appreciable quantity of carotenoids, a colour pigment in sweet pepper and tomato might have caused the noticeable difference in appearance. The parabolic shape of Fig. 5 is an indication that the saddle point was not centered.

Oil viscosity

Dependence of oil viscosity on treatment may be traced to the mixture of additives (sweet pepper & tomato) which possess high viscosity. High coefficient of determination R^2 (0.93), is an indication that the model can be used to navigate the design space.

Beta-carotene

Coefficient of determination of the model was 0.94. This suggests good fitness. β - carotene accounts for more than 90% of total carotenoids in vegetables [28]. Noticeable difference between treated and un-treated oil beta-carotene may be traced to migration of beta-carotene from the additives (tomato & sweet pepper) to the peanut during incubation. Symmetric shape of Fig. 7 is an indication of consistency.

Vitamin E

Vitamin E is a fat-soluble vitamin naturally occurring in peanuts with α -tocopherol on the greater side compared to other tocopherols [29]. Significant effect of pepper and tomato mixture on vitamin E content be associated with enzymatic reaction during the incubation period.

Vitamin K

Vitamin K is not originally present in peanut and its occurrence in vegetables is low [29]. This may account for the obtained result.

CONCLUSION

The extraction of peanut oil with the use of sweet pepper and tomato fruit as additives in a controlled environment at $40 \pm 2^\circ\text{C}$ revealed that functional properties of the oil, namely, iodine value and saponification value reacted differently to treatment. Iodine value was significantly influenced while no significant effect was recorded for the saponification value. Tests of rancidity of the oil showed that peroxide value and free fatty acid were significantly influenced by the treatments. Physical properties of the peanut oil measured by colour and viscosity were significantly influenced by the treatment. Nutritive values of the peanut oil viz beta-carotene and Vitamin E were significantly influenced by the treatments but Vitamin K was not significantly influenced. This information is of benefit to peanut oil producers and consumers.

Table 1: Design matrix of the experiment, additive ratio and incubation time interaction

S/N	Coded A pepper:tomato	Actual A pepper:tomato	Coded B Incubation time (hr)	Actual B Incubation time (hr)
1	-1	3:2	-1	1.5
2	-1	3:2	1	4.5
3	1	2:3	-1	1.5
4	1	2:3	1	4.5
5	1.414	0:5	0	3
6	-1.414	5:0	0	3
7	0	2.5: 2.5	1.414	6
8	0	2.5: 2.5	-1.414	1
9	0	2.5: 2.5	0	3
10	0	2.5: 2.5	0	3
11	0	2.5: 2.5	0	3
12	0	2.5: 2.5	0	3
13	0	2.5: 2.5	0	3

Table 2: Some physical and chemical properties of sweet pepper, tomato and peanut¹

Properties	Pepper	Tomato	Peanut
Unit Mass (g)	9.06 ± 1.02	42.45 ± 2.15	1.0 ± 0.00
Colour (A)	0.68 ± 0.01	0.40 ± 0.01	0.31 ± 0.01
Specific Gravity	0.73 ± 0.01	0.97 ± 0.00	0.86 ± 0.01
Moisture Content (%wb)	88.50 ± 4.20	91.61 ± 3.40	7.75 ± 1.04
pH	4.85 ± 0.74	4.60 ± 0.91	5.80 ± 1.04
Fat (%)	0.20 ± 0.00	0.23 ± 0.00	44.70 ± 7.10
Titrateable Acidity (%)	0.51 ± 0.00	0.68 ± 0.01	2.10 ± 1.01
Fiber (%)	1.42 ± 0.01	0.71 ± 0.00	1.14 ± 0.02
Protein (%)	1.16 ± 0.01	0.83 ± 0.01	22.07 ± 2.46
Carbohydrate (%)	7.56 ± 1.77	5.71 ± 1.93	24.34 ± 3.33

¹All the values are mean of three determinations.

Table 3: Recorded values of responses as influenced by variables²

S/N	IV (Wijs)	PV meqkg ⁻¹	FFA (%)	SV mgKOH/g	CL (A)	Vs Nsm ⁻²	β-caro μg/100g	Vit E μg/100g	Vit K μg/100g
1	101.45	9.07	1.41	140.25	0.19	0.01	755.00	12.30	8.61
2	101.37	9.10	1.41	140.25	0.20	0.02	751.00	13.60	8.40
3	116.90	11.90	4.82	180.50	0.13	0.02	690.00	12.00	7.54
4	101.41	9.01	1.41	140.25	0.16	0.02	760.00	13.70	7.10
5	108.83	9.53	1.97	196.35	0.16	0.02	740.00	12.80	6.90
6	100.07	5.45	0.85	184.15	0.18	0.02	800.00	14.60	6.75
7	108.83	12.50	1.97	196.35	0.20	0.19	697.00	13.50	7.40
8	108.83	9.00	1.41	140.25	0.17	0.01	725.00	12.60	7.60
9	101.14	10.70	3.14	168.30	0.14	0.01	857.00	11.90	8.60
10	103.21	11.07	3.14	168.10	0.13	0.02	861.00	12.10	7.90
11	107.69	10.03	2.94	169.00	0.13	0.01	867.00	12.60	8.50
12	105.57	11.16	3.18	167.70	0.13	0.02	859.00	11.80	8.20
13	104.79	10.41	3.01	166.30	0.14	0.01	860.00	12.30	8.10
C	103.38	3.66	0.56	56.10	0.13	0.01	525.00	6.70	5.30

Where C is without any treatment (control), IV is iodine value, PV is peroxide value, FFA is free fatty acid, SV is saponification value, CL is colour, β-caro is beta carotene Vit E is vitamin E and Vit K is vitamin K.

²All the values are mean of three replicates.

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