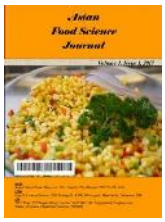

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Approximate Prediction of Chemical Changes in Peanut Oil during Intermittent Deep Frying Process Using UV-Visible Spectroscopy

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Aims: The study aimed at predicting some quality changes in peanut oil during intermittent frying of carbohydrate and protein-based foods, using visible spectroscopic and chemometric methods.
Study Design: Completely Randomized Design and Multivariate Linear Regression were used to achieve this study.
Place and Duration of Study: The study took place at the Department of Food Science and Technology, Federal University of Technology, Akure between February and September 2017.
Methodology: Equal weight of yam chips and marinated chicken [carbohydrate (CHO) and protein-based (PRO) foods, respectively] were fried at 170°C for 20, 40, 60, 80, 100, 120, and 140 min with oil samples taken and topped at every interval. Changes in quality parameters such as colour density, free fatty acid (FFA), acid value (AV), peroxide (PV and saponification values (SV), *K*-extinction coefficients (K_{232nm}, K_{266nm}, K_{270nm} and K_{274nm}), and ΔK, with time, were determined. UV-Visible spectra (350 – 800 nm) of the oil samples were taken, and the data were elaborated with Principal Component Analysis (PCA) and Partial Least Square (PLS) regression techniques.

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Results: Reduction in oxidative stability measured as increased values of FFA, PV, K-*extinction* values and *ΔK* were observed in all the samples and were particularly more pronounced (p = 0.05) in PRO-fried oils than those of CHO. Similarly, colour density increased linearly as frying time advanced in PRO-fried oil. PCA models of quality and spectra data revealed clear distinctions between PRO and CHO-fried oil samples. PLS regression coefficients showed that FFA (0.95), PV (0.92), SV (0.94), *ΔK* (0.98) and colour (0.95) were satisfactorily predicted; despite the relatively small sample size (15).
Conclusion: Non-destructive spectroscopic quality screening of vegetable oils during frying could facilitate rapid detection of degradation and the extent to which it can be reused. However, a large sample size is required to validate its reliability.

Keywords: Peanut oils; intermittent frying; quality parameters; UV-visible spectroscopy; PCA; PLS regression.

1. INTRODUCTION

Fried food has grown in popularity despite the low-fat/no-fat current health trend. Lipid oxidation is one of the major problems associated with this popular cooking process [1]. Several by-products are generated during continuous thermal treatment of oils, and these products include free fatty acids, alcohols, cyclic compounds, dimmers, polymers and some colourful derivatives [2]. The process is summarily an oil – food interaction at elevated temperatures that cause the food to become dehydrated, cooked with several physicochemical changes such as protein denaturation, starch gelatinisation, colour and flavour development [3]. There are extensive studies on the chemical changes that occurred in oil during repeated intermittent frying cycles [4,5,6]. Free fatty acids, peroxide values, total polar compounds and some other oxidative stability indices experience gradual but significant increase under frying conditions [2]. Frying fats and oils have a finite and relatively short lifespan. The sensory quality of the food diminishes gradually until the oil is discarded. Traditionally, there is a "dilemma to choose" when the oil has reached the end of its useful life because of diminished food quality and/or legal restrictions.

Most fast foods and restaurants reuse frying oil mainly for economic reasons [7]. In some cases, the unscrupulous act of adulteration of fresh oil with overused frying oils by food vendors is common, especially where legal regulations are less effective [8]. Frying oils are usually reused depending on the kind of oil, nature of food and how much of it is fried, how often the oil is filtered, how many hours oil is being used and at what temperature. In carbohydrate-based fried foods, caramelisation of sugar and non-enzymatic, high temperature-dependent browning reaction in frying foods, may lead to the hue colour of the oil and many other physicochemical changes [9]. In the case of protein-based foods, protein denaturation and subsequent release of monomeric and polymeric compounds into the oils may reduce the frying life and quality of the oils [10].

There are quite a number of conventional physical and chemical analyses; traditionally used to detect oil quality before and after frying procedure [11] such as conductivity testing [12] and gas chromatography coupled with compositional determination [13]. However, application of UV-visible spectroscopy to determine quality changes during continuous frying of oil has been relatively scarce in the literature. Gonçalves et al. [14] monitored tocopherol degradation and formation of oxidation products in thermally-treated edible oils using UV-visible spectroscopy and chemometrics. Similarly, accelerated oxidation stress was detected in olive oil using synchronous fluorescence spectroscopy [15]. This region of the electromagnetic spectrum is divided into the ultraviolet (200 - 400 nm) and visible (400 - 800 nm) regions. Food molecules especially lipids have a unique and specific fingerprint that can be rapidly revealed using spectroscopy [16,17]. A slight alteration of these patterns can be used as indicators of chemical or physical changes in foods. Harnessing this method to monitor changes in the quality of frying oil could be a quicker and more easily adaptable technique at the industrial level. Therefore, this study aims to compare the conventional classical chemical analysis with non-destructive spectroscopic methods in elucidating physicochemical changes in vegetable oils during continuous frying of both protein and carbohydrate-based foods. The study has the potential to serve as a time-serving means of

predicting appropriate lifespan of oil during constant frying process.

2. MATERIALS AND METHODS

2.1 Peanut Oil Frying and Sampling procedures

Wholesome mature white yams (*Dioscorea alata*) were obtained from Oba Market, Akure. Frozen chicken wings and fresh peanut oil were purchased from Shoprite grocery store in Akure Ondo State Nigeria. The yams were cleaned, peeled, washed and drained. Sliced yam (500 g) were cut into pieces (approximately 40 – 50 x 10 x 10 mm) and fried in a Eurosonic domestic deep fryer (model ES-388, China), initially filled with peanut oil (2 L). The oil was heated to 170°C, and the yam slices were added and fried for 20 mins. For the oil sampling, after each successive frying operation, the oil was allowed to cool to less than 50°C, and 30 g was taken into a dark bottle. The oil in the fryer was topped with fresh oil before repeating the frying procedure. Seven batches of yam were fried, and the samples taken were labelled as C20 to C140 (CHO-fried oils). Vegetable oils are not usually reused beyond 3 times [18]. The same procedures were repeated for marinated chicken wings using another 2 L peanut oil and the samples obtained were labelled as P20 to P140 (PRO-fried oils). Thus, the total frying period by the end of the experiment on each of the food materials was 2 hr, 20 mins, given rising to 14 samples. Fresh oil was taken as reference and all samples were kept in dark bottles, corked and stored in cool dry place prior to analysis.

2.2 Chemical Analysis

2.2.1 Free fatty acids, acid Value, peroxide value and K specific extinction coefficients determinations

Quality parameters and oxidative stability indices of the oil samples such as FFA, AV, PV and K values were measured using Official standard methods of American Oil Chemist' Society [19]. Free acidity (0.503AV) expressed as oleic acid (%) was determined by titrating ethanol: ethyl ether (1:1) solution of the oil against 0.1N KOH using phenolphthalein as an indicator. Peroxide value was determined by potassium iodide reaction with oil solution (chloroform, acetic acid and oil) in the absence of light and the iodine generated was titrated with 0.1N sodium thiosulfate solution using 1% starch solution as an indicator. The results were expressed as equivalents of oxygen per kg of oil ($meqO_2/kg$). *K*-extinction coefficients (K_{232nm}, K_{266nm}, K_{270nm} and K_{274nm}), and ΔK were measured spectrophotometrically as the absorption values of the isooctane-oil solution at 232, 266, 270 and 274 nm wavelengths respectively, using UV-vis spectrophotometer (Shimadzu UV-1800 Kyoto, Japan) with 1 cm path length.

2.2.2 Saponification value determination

The standard method of AOAC No. 920.160 [20] was used to determine the saponification value (SV) of the oil samples.

2.2.3 Colour Density

Colour density was determined spectroscopically according to the method described by Wroistad [21]. The sample colour was extracted by mixing 1 mL of oil sample (1 mL) with 10 mL methanol and stirred properly. The mixture was allowed to stand for 10 min and centrifuged. The absorbance of the supernatant was recorded as optical density (OD) at 420 and 520 nm wavelengths using UV-vis spectrophotometer (Shimadzu UV-1800 Kyoto: Japan). Sum of the absorbances of the wavelengths was recorded as colour density thus: $Colour\ Density = A_{420nm} + A_{520nm.}$

2.2.4 UV-visible spectra acquisition

UV-visible spectrophotometer (Shimadzu UV-1800 Kyoto: Japan) with the following operational parameters: deuterium-discharge lamp as an ultraviolet source, tungsten lamp for the visible and 2.0 nm resolution; was used to take the spectra of the oil samples. There were two rectangular cells, one for sample (1 mL peanut oil dissolved in 2 mL hexane) and the other for blank (pure n-hexane). Quartz cuvette of 10 mm path length was used for sample and blank holders. The UV-vis spectra of the samples taken between 200-800 nm with 2.0 nm equally spaced wavelength interval.

2.3 Data Matrices and Statistical Analysis

Analysis of Variance (ANOVA) was used to determine the significant changes in the quality characteristics of the oil samples after each frying cycle (Minitab 16.0, Minitab Inc., State College, USA). In the multivariate analysis, data matrices were created in two categories

(chemical and spectroscopic data) using SIMCA (v. 13, Umetrics, Umea, Sweden) thus:

a. Chemical data matrix (14 x 11) which involves of 14 peanut oil samples (n observations) and 11 measured variables (K variables) such as free fatty acids, colour density, acid and peroxide values, *K*-extinction coefficients (K_{232nm}, K_{266nm}, K_{270nm} and K_{274nm}), and ΔK values.
b. UV-visible spectral data matrix composed of 350 – 800 nm regions constituting the most significant segments; with high signal-to-noise ratio.

PCA (Principal Component Analysis) was applied to examine the natural clustering pattern in each of the data matrices, by factorizing the **X** matrix into two matrices: score (**T**) indicating the position of the oil samples relative to that of the predictors (variables) loading (**P**) and residual error (**E**) as shown in the equation below:

$$X = T*P + E$$

It is an unsupervised multivariate data elaboration technique with the objective of obtaining a linear transformed and a dimensionally reduced data output, that keeps a reasonable amount of variances in the original data [22]. The vectors of measured variances are called principal components, and they are arranged based on the amount of variance explained. PC 1 is the axis that lies in the direction containing most explained variation. The subsequent principal component (PC 2 and above) describes the lesser amount of variance, and the order continues.

PLS regression technique was applied to evaluate the predictive capacity of the spectra (UV-visible) over some important chemical parameters of the oil samples. Due to the limited sample size (15), only leave-one-out cross-validation was used to estimate the performance of the model. The selection of these variables was based on the previous knowledge of their importance to oil stability [17]. Details of the computed statistical outputs were reported regarding root to mean square error of calibration (RMSEE), cross-validation (RMSECV), regression coefficients for calibration, R^2_{cal}, cross-validation R^2cv and a significant number of PCs as a criterion that define the predictive ability of the model and its applicability to independent data set. Overfitting of the models was avoided by using automatic fitting embedded in SIMCA 13.30 software.

3. RESULTS AND DISCUSSION

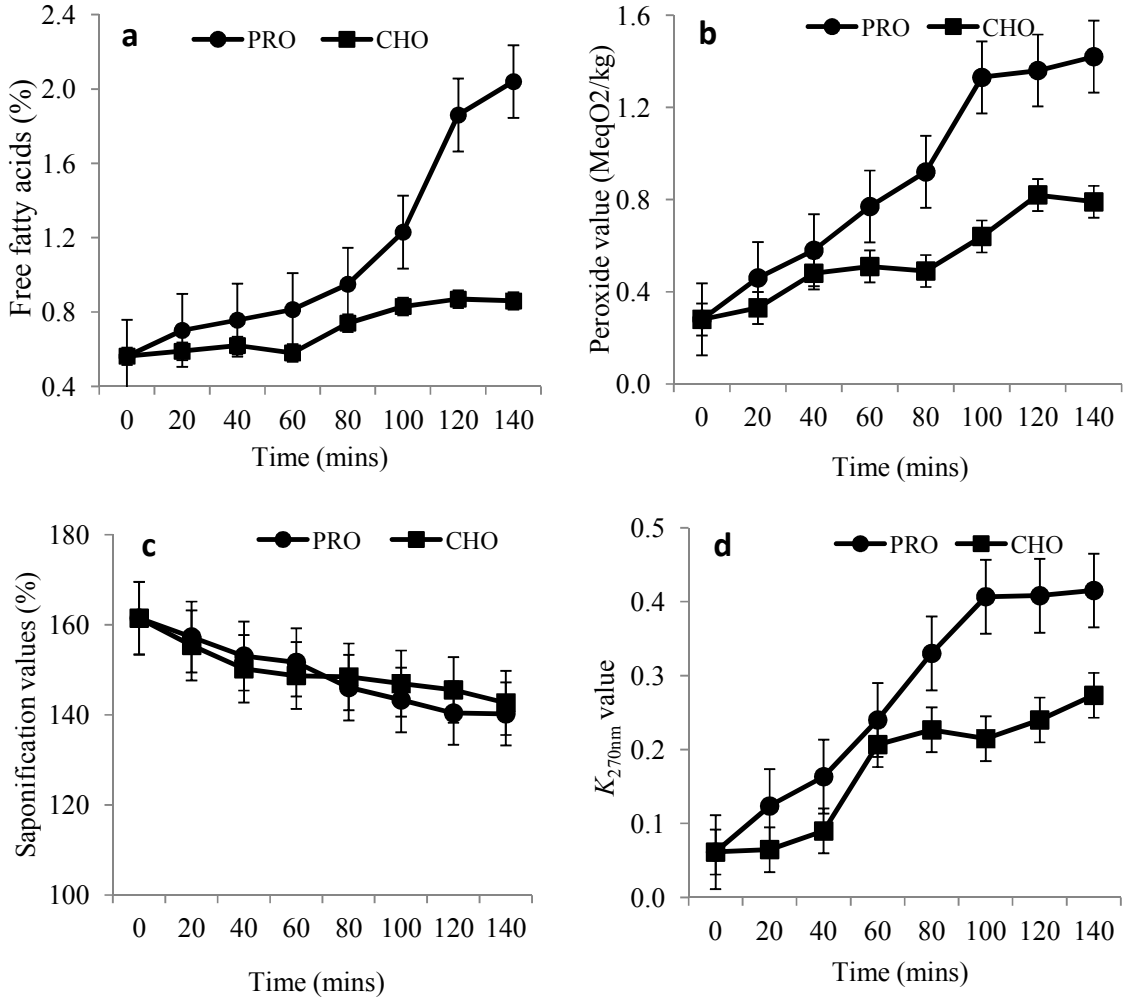
3.1 Changes in Oxidative and Quality Parameters

Fig. 1 shows the changes in oxidative and quality parameters of the oils concerning frying time and nature of fried food. FFA and PV increased with frying time, especially in protein-based food. Being a refined peanut oil, the FFA was already above the expected value (0.3 mgKOH/g) [23] even after 20 minutes of frying in both food groups. Food of high moisture content facilitates oil hydrolysis [10] leading to more FFA generation as observed in PRO-fried oil as compared to the CHO. The use of FFA as a monitoring tool for the quality of frying oil has been reported earlier [24]. PV is an indication of a primary oxidative product called "hydroperoxide" that could later be converted to secondary products responsible for the actual deterioration of frying fats [25]. Therefore, it is a useful biomarker that indicates the initial stage of oxidation. PV increased significantly as the frying time advanced and was more pronounced in PRO-fried oils. Conversely, SV decreased slightly with frying time. Conjugated dienes and trienes compounds generated during thermal treatment of oils can be qualitatively indicated by specific UV absorptions at 232 nm (K_{232}) and 270 nm (K_{270}), respectively [26]. The extinction values are a useful tool for quick quality comparison of oils, but they do not provide information on the actual polyunsaturated fatty acids responsible for the diene and triene compounds. These parameters showed a significant increase as frying time progressed. K_{270} linearly increased in PRO-fried oil up until 100 mins and levelled out; indicating accumulation of secondary oxidative products in oils with continuous oil reuse. The most remarkable change (over 50% increase) in K_{270} in CHO-fried oil was observed between 40 – 60 mins. A slightly different trend was observed in ΔK extinction value. However, ΔK of PRO-fried oil was at every point within the frying cycles; higher than that of CHO oil. Increase in K_{270} occurred in both groups. This indicates the presence of carbonylic compounds. Poiana [25] reported that α-diketones and unsaturated α-ketones absorb strongly at 270 nm. Colour change in oil during continuous frying is one of the physical changes due to the decomposition of nonvolatile components of the frying food. Nonvolatile monomeric and polymeric compounds are more in protein-based food than

carbohydrate, which may explain the difference in colour intensities of the oils from both food categories (Fig. 1f).

3.2 Effects of Frying on UV-visible Spectra of Oils

The oxidative changes in the oil samples at different frying times and foods were monitored by UV-visible spectroscopy and the spectral differences (350 – 800 nm) were presented in Fig. 2. The spectral appeared similar to that observed by Zhang et al. [27]. A close observation of the spectra revealed a shift from left to right as wavelength increased. This is typical in the case of temperature variations during thermal treatment of oils [14]. The absorption at far ultraviolet and visible regions by an organic molecule is limited to certain functional groups called chromophores that contain valence electrons of low excitation energy state [28]. Different compounds present in the oil absorb differently causing a shift in their peaks toward longer wavelengths. The most significant absorption intensities for CHO-fried oils were between 400 – 800 nm, while that of PRO-fried oils were 400 – 600 nm. Compounds responsible for these absorbances may be products of thermal degradation. The left-to-right continuous shift in the spectra of the oil samples was more noticeable in PRO-fried oils than the CHO-fried. According to Koplik [29], the probable absorbing species include; poly-unsaturated fatty acids (oxidised linoleic acid), conjugated dienes and trienes, aromatic compounds. After 500 nm, the spectra maintained largely the same absorbance with a slight variation. A similar observation was reported for corn oil when used as adulterant [30].



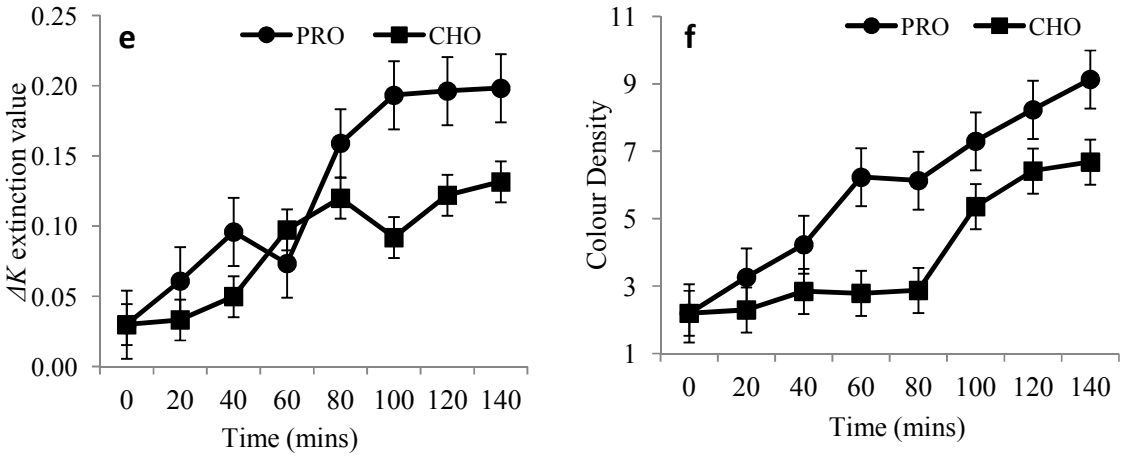


Fig 1. Changes in the quality parameters: (a) Free fatty acid, (b) Peroxide value, (c) Saponification value, (d) K_{270nm}, (e) ΔK extinction values and (f) Colour Density of peanut oil with time during intermittent frying of protein and carbohydrate-based food products

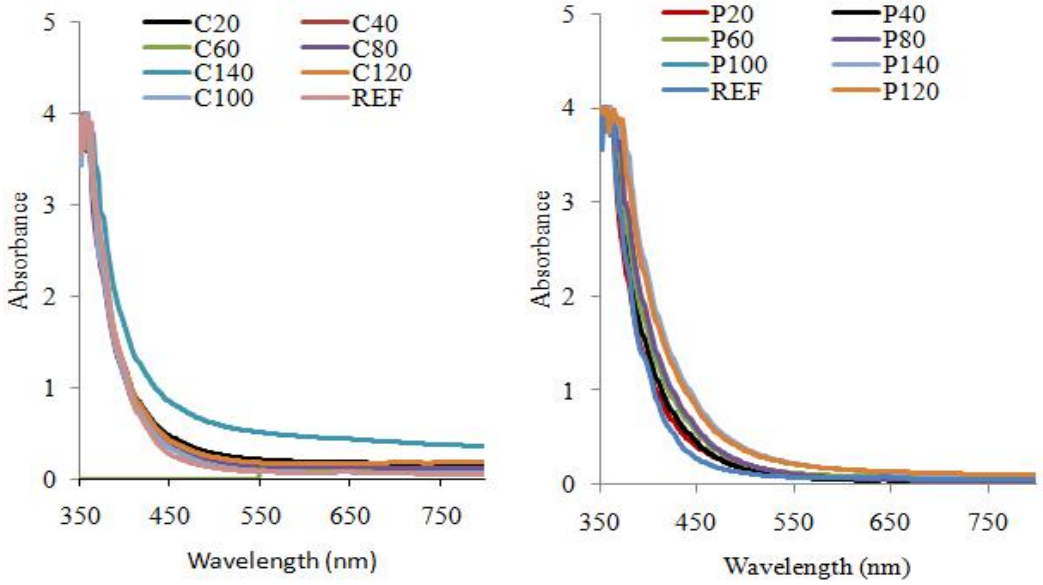


Fig. 2. UV-Visible spectra of peanut oil recorded at 20 min interval during intermittent deep frying of carbohydrate (C) and protein-based (P) foods

3.3 Multivariate Data Elaboration

3.3.1 Principal component analysis (PCA)

The oil quality parameters and spectral data were modelled separately with PCA to visualise the intrinsic pattern of change in the properties of the oil with frying time. PCA is an unsupervised multivariate statistical model [31] capable of revealing the clustering pattern of multiple observations (oil samples) relative to their responses (quality parameters and spectral wavelengths). PCA model of the quality parameters data with 2 PC and 94% explained variance, produced clusters of samples mainly based on the frying time. The upper left of the score plot (Fig. 3a) consists of short-time fried oils (20 – 40 mins), indicating no remarkable difference between PRO and CHO-fried oils within that time. The variable most responsible for this cluster is SV. The mid-section of the ellipse is predominantly occupied with the mixture of mid, long time CHO and mid-time PRO-fried oils. These oils had a minimum level

of all the quality parameters considered based on the central position in the score plot. However, the bulk of the variables such as high AV, FFA, K – extinction coefficients, PV and colour density were the characteristics of PRO-fried oils at 100, 120 and 140 mins. As the number of frying cycle increased, PRO-fried oils decreased in oxidative stability and quality properties than CHO-fried. This is supported by Peng et al. [32] who observed more accumulation of aldehydes in pork loin than potato during deep frying. However, PCA model of spectral data with lower percent explained variance (80%) and 2 PC produced a visually different score and loading plots (Fig. 3c & 3d). The upper part of the score plot is occupied by PRO-fried oils while CHO-fried oils occupied the lower part, indicating a more distinctive difference in the spectral information of the oil samples. High absorbance values of 366 – 506 nm range were responsible for the separation of PRO-fried oils at 100, 120 and 140 mins. Similarly, 552 – 800 nm spectra range was the most descriptive band separating 20 and 120 mins CHO-fried oils despite the low resolution of this region. There was a slight similarity between the information conveyed by the PCA models of chemical and spectral data of the oils, especially in separating 100 – 140 mins fried oils.

3.3.2 UV-visible spectra prediction of quality parameters

Partial Least Square (PLS) regression was used to relate the UV-visible spectral data (predictors) and the results of quality parameters (variables) of both PRO and CHO-fried oils. Models were built separately on each of the quality, and the performances of the models were evaluated using cross-validation (R^2cv) usually suitable for a low to medium-size data matrix [33]. Other model parameters used to verify fitness include: determination coefficient of calibration (R^2_{cal}), Root mean square error of calibration (RMSEE) and Root mean square error of cross-validation (RMSECV). UV-visible spectra predictive capacities over FFA, PV, SV, and ΔK were high (Fig. 4a) with a coefficient of determination above 0.90 in each parameter. However, SV had the highest R^2cv (0.90) among the variables

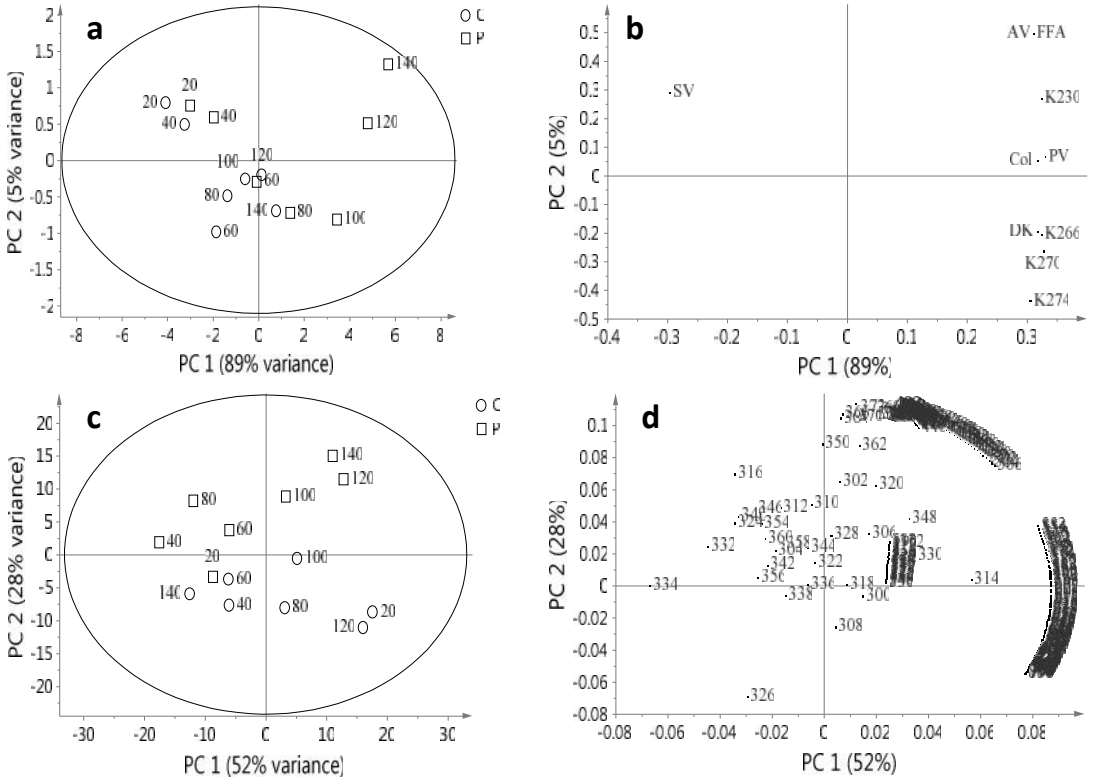


Fig. 3. Results of PCA multivariate models: (a) Score plot of quality parameters (b) loading plot of quality parameters, (c) Score plot of visible spectra and (b) loading plot of visible spectra of intermittently deep-fried peanut oils using carbohydrate (C) and protein-based (P) foods

considered (Table 1). The closer the R^2 value to 1.0 the better the reliability of the model. In the literature, UV-visible has been used to evaluate the vegetable oils under thermal oxidative stress [34]. Changes in PV during frying were reasonably predicted when compared to what is obtainable in the literature [35]. Similarly, colour density, K_{232nm} and K_{270nm} were significantly less predictable compared to other parameters. However, RMSEE and RMSECV values were very close in all the variables, and they are comparatively low compared to the mean values.

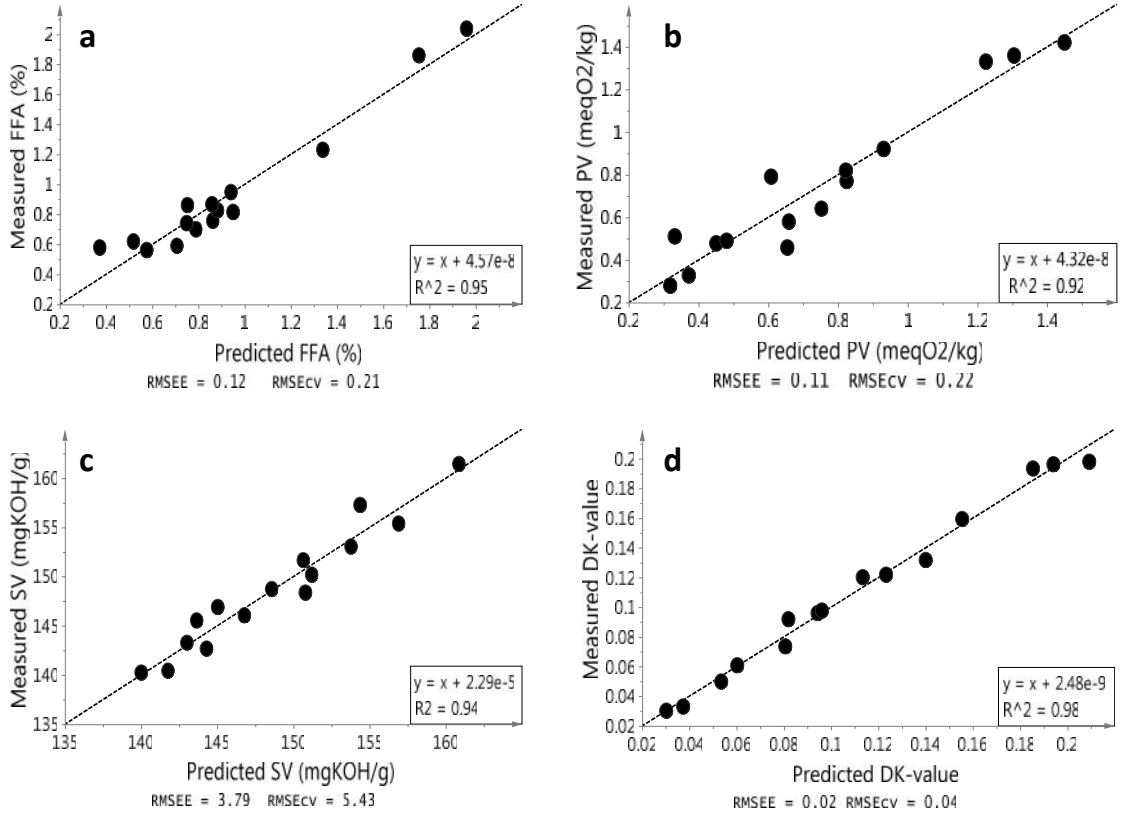


Fig. 4. PLS prediction of oxidative parameters: (a) Free fatty acid, (b) Peroxide value, (c) Saponification value, and (d) *ΔK* extinction coefficient of peanut oil during intermittent frying using UV-visible spectra spectroscopy data

Table 1. PLS regression models output for the prediction of quality parameters of deep-fried peanut oil using UV-visible spectra data

Parameters	Mean	Range	PC	R^2_{cal}	R^2_{cv}	RMSEE	RMSECV	Reg. Equation
FFA (%)	0.93	0.56 - 2.04	3	0.95	0.76	0.12	0.21	$y = x + 4.57*10^{-8}$
AV (%)	1.86	1.12 - 4.08	2	0.94	0.76	0.23	0.42	$y = x - 1.44*10^{-8}$
PV ($meqO_2$/kg)	0.75	0.28 - 1.42	3	0.92	0.60	0.11	0.22	$y = x + 4.32*10^{-8}$
SV (mgKOH/g)	148.77	140.22-161.48	4	0.94	0.90	3.79	5.43	$y = x + 2.29*10^{-5}$
Colour density	5.07	2.20 - 9.13	4	0.95	0.88	0.63	1.59	$y = x - 1.01*10^{-7}$
K_{232nm}	0.26	0.12 - 0.42	2	0.80	0.70	0.05	0.06	$y = x + 4.48*10^{-8}$
K_{270nm}	0.23	0.06 - 0.42	2	0.71	0.53	0.07	0.08	$y = x - 6.13*10^{-9}$
ΔK	0.11	0.03 - 0.20	5	0.98	0.89	0.07	0.04	$y = x + 2.48*10^{-9}$

PC: Principal components; R^2_{cal}: determination coefficient of calibration; R^2cv: determination coefficient of leave-one-out cross-validation; RMSEE: Root mean square error of calibration, RMSECV: Root mean square error of cross-validation

4. CONCLUSION

The changes in quality characteristics of peanut oil during intermittent and continuous frying using chicken (PRO-based) and yam (CHO-based) were evaluated using conventional chemical analysis and rapid spectroscopic technique. Results from the study suggested that the rate of quality deterioration of oil during deep-frying were dependent on the nature of the food being fried and a number of frying cycles. Quality parameters such as FFA, PV K-values, and Colour intensity increased with frying cycles and were more significant in PRO-fried oil. According to the PCA model, there was no remarkable difference in SV of PRO and CHO-fried oils within the first 40 mins of frying. However, reduced oxidative stabilities were the characteristics of the oil samples obtained after 100, 120 and 140 mins of frying. These oils were also distinguished by high UV-visible absorbance values at 366 – 506 nm range. Spectra data regression model showed high predictive potentials over most of the quality parameters of the fried oil samples. Therefore, better control and effective monitoring of the frying process can be rapidly achieved using UV-visible spectroscopy. However, it is recommended that a study that proposes appropriate stage and method of oil disposal that conforms to the international norm (i.e. less than 2% impurities for recycled oils used for animal feed), should be carried out.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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