ORIGINAL ARTICLE



# Development of cookies made with cocoyam, fermented sorghum and germinated pigeon pea flour blends using response surface methodology

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Abstract Cookies were produced from blends of cocoyam, fermented sorghum and germinated pigeon pea flours. The study was carried out to evaluate the effects of varying the proportions of these components on the sensory and protein quality of the cookies. The sensory attributes studied were colour, taste, texture, crispness and general acceptability while the protein quality indices were biological value (BV) and net protein utilization (NPU). Mixture response surface methodology was used to model the sensory and protein quality with single, binary and ternary combinations of germinated pigeon pea, fermented sorghum and cocoyam flours. Results showed that BV and NPU of most of the cookies were above minimum acceptable levels. With the exception of cookies containing high levels of pigeon pea flour, cookies had acceptable sensory scores. Increase in pigeon pea flour resulted in increase in the BV and NPU. Regression equations suggested that the ternary blends produced the highest increase in all the sensory attributes (with the exception of colour).

Keywords Cocoyam flour  $\cdot$  Fermented sorghum flour  $\cdot$  Germinated pigeon pea flour  $\cdot$  Cookies

### Introduction

Cookies are widely consumed throughout the world. In fact, they represent the largest category of snack foods in most parts of the world (Lorenz 1983). Wheat flour, which is the flour of choice for producing cookies, is unavailable or

L. C. Okpala (⊠) · E. C. Okoli Department of Food Science and Technology, Ebonyi State University, Abakaliki, Ebonyi State, Nigeria e-mail: lcokpala@yahoo.com uneconomical in many regions of the world. Therefore to produce baked goods, regions with limited supplies of wheat flour must rely on imports or exclude wheat products from the diet (Holt et al. 1992). The consumption of cereal based foods like cookies require the development of an adequate substitute for wheat (Eneche 1999). The substitute should be one that is readily available, cheap and able to replace wheat flour in terms of functionality.

Composite flours produced from cereals and legumes have the advantage of improving overall nutrition (FAO 1995) while composite flours produced from legumes and tubers will have high protein content and high calorific value (Chinma et al. 2007). In selecting the components to be used in composite flour blends, the materials should preferably be readily available, culturally acceptable and provide increased nutritional potential (Akobundu et al. 1998).

Pigeon pea (*Cajanus cajan*) is a legume which has a relatively high amount of protein (Tiwari et al. 2008). Sorghum (*Sorghum bicolor*) is a cereal. Even though sorghum compares favourably with other cereals in terms of nutrition, world food consumption of the cereal has remained stagnant mainly because it is regarded in many countries as an inferior grain (FAO 1995). Cocoyam (*Xanthosoma sagittifolium*) is a tuber. It has fine granular starch which has been reported to improve binding and reduce breakage of snack products (Huang 2005). These three crops are grown in large quantities in the tropics but are underutilized.

Plant proteins have been reported to have limiting amino acids (Ihekoronye and Ngoddy 1985) and it is necessary to combine these plant proteins in proportions that will improve the protein intake of consumers. Many processes are available for improving the nutritional quality of plant foods. These methods include traditional processing methods like cooking, soaking, dewatering, fermentation, germination, smoking, salting, curing etc. (Teutonico and Knorr 1985). Germination has been reported to induce an increase in free limiting amino acids and available vitamins with modified functional properties of seed components (Hallén et al. 2004). It has also been shown to decrease antinutritional factors, increase the protein digestibility, crude fibre and protein contents of chick pea (El–Adawy 2002). Fermentation improves amino acid composition and vitamin content, increases protein and starch availabilities and lowers levels of antinutrients (Chavan and Kadam 1989). The germination of pigeon pea and fermentation of sorghum should therefore be beneficial to the nutritional quality of the seeds.

Mixture response surface methodology (MRSM) is a statistical technique that can be used to systematically determine the effects of multiple variables in a mixture on quality attributes while minimizing the number of experiments that must be conducted (Cornell 1979). It is a special response surface experiment in which the design factors are the components or ingredients of a mixture, and the response depends on the proportions of the ingredients that are present (Myers et al. 2009).

This work seeks to produce cookies from blends of cocoyam, fermented sorghum and germinated pigeon pea flours (hereafter known as components) and use MRSM to study the effects of varying the proportions of these components on the sensory and protein quality of the cookies.

#### Materials and methods

#### Materials

The white variety of pigeon pea (*Cajanus cajan*), the white variety of sorghum (*Sorghum bicolor*) and the tannia variety of cocoyam (*Xanthosoma sagittifolium*) were purchased from a retail outlet in Abakaliki, Ebonyi State, Nigeria. Wheat flour and all other baking ingredients such as eggs, baking powder, fat, milk and flavourings were also obtained from the same source. Corn starch was purchased from a retail outlet in Enugu, Enugu State; Nigeria, casein was bought from a chemical store in Nsukka, Enugu State while vitamin and mineral premixes were bought from Bioorganics Nigeria Plc, Lagos, Nigeria.

Cocoyam flour (CF) Cocoyam flour was produced using the method described by Ijeoma (1983). The corms were washed, peeled, sliced and blanched at 80 °C for four minutes. They were dried and milled to pass through 100  $\mu$ m mesh sieve.

*Germinated Pigeon pea flour (GPF)* Pigeon peas were germinated using the modified method of Hallén et al (2004). Cleaned grains were soaked in 0.1 % sodium hypochlorite solution for 30 min to prevent mould growth. After that time, the grains were thoroughly washed and soaked in water (10 h). The hydrated seeds were spread on jute bags and allowed to germinate for 4 days after which they were dried at 50 °C. Thereafter, formed roots and testa were rubbed off before milling and sieving through 100  $\mu$ m mesh sieve.

Fermented Sorghum flour (FSF) The sorghum flour was subjected to natural lactic acid fermentation using the method of Hallén et al (2004). Washed and dried grains were ground into fine flour. The flour was mixed with water (1:5 wt/wt) to form slurry followed by the addition of 5 % sugar by weight of flour. The slurry was left to ferment in trays at room temperature until the pH of the slurry reached 5.5. The fermented slurry was dried at 50 °C and then ground through a 100  $\mu$ m mesh screen. Both of the flour samples were kept in airtight containers until needed for analysis.

*Experimental design* A three-component augmented simplex centroid design was used as described by Scheffé (1963). The three mixture components evaluated in this study were cocoyam flour  $(x_1)$ , fermented sorghum flour  $(x_2)$  and germinated pigeon pea flour  $(x_3)$ . The proportions for each ingredient were expressed as a fraction of the mixture and for each treatment combination, the sum of the component proportions was equal to one, where:

$$\sum X_i = x_1 + x_2 + x_3 = 1$$

In this design, the number of points (n) necessary to run a mixture experiment is

 $n = 2^q - 1$ ,

where q is equal to the number of components being studied (3). This design resulted in 7 flour mixtures. Three additional points were included to provide extra points within the mixture triangle. Four runs were replicated to give an internal estimate of error (Table 1).

Cookie preparation The ingredients used were: flour, 100.0 g; hydrogenated vegetable fat, 40.0 g; sugar (granulated cane), 25.0 g; egg (whole, fresh), 31.0 g; milk (filled, powdered); 7.8 g; nutmeg; 0.3 g, vanilla (liquid), 5.0 ml, salt, 1.0 g and baking powder, 1.0 g. Fat and sugar were mixed in a Hobart mixer until fluffy. Whole eggs and powdered milk were added while mixing and then mixed for a total of about 30 min. Vanilla flavour, nutmeg, flour, baking powder and salt were mixed thoroughly and added to the cream mixture where they were all mixed together to form a dough. The dough was rolled and cut into circular shapes of 5 cm diameter. Baking was carried out at 185 °C

Table 1 Experimental design used to produce flour blends

Blends	Cocoyam (CF) (%)	Germinated pigeon pea (GPF) (%)	Fermented sorghum (FSF) (%)		
B1	100	0	0		
B 2	0	100	0		
B 3	0	0	100		
B 4	50	0	50		
В 5	0	50	50		
B 6	50	50	0		
В 7	33.3	33.3	33.3		
B 8	16.7	66.6	16.7		
B 9	16.7	16.7	66.6		
B 10	66.6	16.7	16.7		
B 11	100	0	0		
B 12	0	100	0		
B 13	0	0	100		
B14	50	0	50		

Mixture design results in blends B 1–B7. Blends B 8–B10 are additional formulations selected as interior points of the design. They were derived from the relation: [(q + 1/2q, 1/q, 1/2q....1/2q), Cornell 1986]

Blends nos. B11–B14 = replicate runs as recommended by Design-Expert Software.

for 20–25 min. Cookies samples were cooled and stored in polyethylene bags until needed. Cookies were made from wheat to serve as a control.

Rat feeding protocol The test diets were adjusted to provide 6, 8 and 10 % protein. The diets made from cookies with protein content above 10 % were adjusted to provide 10 % protein; test diets made from cookies with protein content ranging from 8.01-9.99 % were adjusted to provide 8 % protein while diets from cookies with protein content ranging from 6.01–7.99 % were adjusted to provide 6 % protein. There were three reference protein diets of casein at 6, 8 and 10 % protein levels for comparison of the protein quality of these test diets (Prabhavat et al. 1991). Other ingredients included 5 % vegetable oil, 0.25 % vitamins, 0.045 % minerals and the remainder, corn starch added to balance the diets (Ugwu and Ugwu 2009). The diets were thoroughly mixed, pelletized and stored in polyethylene bags labeled with designated names. The polyethylene bags were kept in airtight containers until ready for use.

Eighteen groups (fourteen assays + three casein control groups + group receiving a protein-free diet) of five male adult albino rats of the Wistar strain with average initial weight of 120-210 g were used. They were divided in such a way that all the groups of rats had the same average weight. Subsequently, they were housed in individual screened bottomed cages designed to separately collect faeces and urine. Experimental animals each received 20 g of

the corresponding group diets and water ad libitum. The temperature of the laboratory was 28±1 °C with alternate 12 h periods of light and dark. These animals were used to assess the BV and NPU of the diets based on casein. Following the method described by Al-Numair and Ahmed (2008), a 9-day balance study which included a four-day adjustment and five-day nitrogen (N) balance period was carried out. There was a preliminary feeding period of four days followed by a balance period of five days during which complete collection of faeces and urine was performed for each rat. Food intake was monitored daily and final body weights were recorded. Urine was collected in sample bottles, preserved in 0.1 N HCl to prevent loss of ammonia and stored in a refrigerator until analyzed for urinary nitrogen. Faeces of individual rats were pooled, dried at 85 °C for 4 h, weighed before being ground into fine powder and stored for faecal N determination. The concentration of nitrogen in the diet, faeces and urine was estimated by the Kjeldahl method (AOAC 2000).

Sensory evaluation Sensory evaluation was conducted on the cookies. A total of twenty semi-trained panelists were recruited from staff and students of the Ebonyi State University, Abakaliki. A randomized complete block design was used whereby each panelist evaluated all the samples prepared for each treatment. Evaluation of all the samples took place in one session. Criteria for selection of panelists were that panelists were regular consumers of cookies and were not allergic to any food. Panelists were instructed to evaluate colour, flavour, texture, crispness and general acceptability of the cookies. A 9-point hedonic scale was used with 1 = dislike extremely, 5 = neither like nor dislike and 9 = like extremely (Ihekoronye and Ngoddy 1985). Samples were identified with three digit code numbers and presented in a random sequence to panelists. The panelists were instructed to rinse their mouths with water after every sample and not to make comments during evaluation to prevent influencing other panelists. They were also asked to comment freely on samples on the questionnaires given to them.

*Statistical analysis* Analysis of variance (ANOVA) was carried out on data from sensory and protein quality tests followed by Duncan's Multiple Range test (SPSS v. 16.0 for windows, SPSS Inc, Illinois, USA). A p-value below 0.05 was considered significant.

The experimental data from sensory and protein quality tests were also evaluated using MRSM. The analysis was performed using Design-Expert (Version 8.0.3, State-Ease, Inc. Minneapolis, 2010) software. Model significance (p < 0.05), lack of fit and adjusted regression coefficients ( $R_{adj}^2$ ) which indicate the model fitness were determined from the analysis.

The model search was started with the special cubic equation as shown in Eq. (1) below:

$$Y = \beta_1 x_1 + \beta_2 x_2 + \beta_3 x_3 + \beta_{12} x_1 x_2 + \beta_{13} x_1 x_3 + \beta_{23} x_2 x_3 + \beta_{123} x_1 x_2 x_3$$
(1)

where Y is the predicted response,  $\beta$ 's are the parameter estimates for each linear and cross product term for the prediction model,  $x_1$ ,  $x_2$ ,  $x_3$ ,  $x_1x_2$ ,  $x_1x_3$ ,  $x_2x_3$  and  $x_1x_2x_3$  are the linear terms of cocoyam, sorghum and pigeon pea and the cross product terms of cocoyam x sorghum, cocoyam x pigeon pea, sorghum x pigeon pea, cocoyam x sorghum x pigeon pea flours respectively. The model chosen was based on a significant model (p < 0.05), insignificant lack of fit and highest R<sup>2</sup> as recommended by Cornell (1986).

## **Results and discussion**

Protein quality evaluation The mean values of the BV and NPU of the cookies produced are shown in Table 2. Significant differences (p < 0.05) existed among the blends. The minimum requirement for modelling a surface is that variation among the assays is observed (Capitani et al. 2009).

The biological value (BV) of the cookies ranged from 73.86 to 95.46 %. Cookies made with 100 % cocoyam flour (100CF) had the lowest BV while those made with 50 % cocoyam flour and 50 % fermented sorghum (50CF:50FSF) had the highest BV. Biological value measures the efficiency of utilization of absorbed nitrogen (Hackler 1977). A value of 100 % denotes the highest quality protein (James et al. 2009). The Net protein utilization (NPU) values ranged from 65.09 % for cookies made with 100CF to 91.98 % for cookies made with 50CF:50FSF. Net protein utilization is a

Table 2 Protein quality						
of cookies made with cocoyam, fermented	Blends	Biological Value <sup>f</sup> (%)	Net protein utilization <sup>f</sup> (%)			
sorghum and germinat- ed pigeon pea flour	B1	78.16 <sup>de</sup>	70.08 <sup>de</sup>			
blends	В2	92.53 <sup>abc</sup>	89.74 <sup>abc</sup>			
	В 3	78.98 <sup>d</sup>	72.95 <sup>d</sup>			
	B 4	95.46 <sup>a</sup>	91.98 <sup>a</sup>			
	В 5	88.11 <sup>c</sup>	85.25 <sup>bc</sup>			
	B 6	90.64 <sup>abc</sup>	88.11 <sup>abc</sup>			
	В 7	86.64 <sup>c</sup>	84.60 <sup>c</sup>			
<sup>f</sup> Values are means	B 8	88.79 <sup>bc</sup>	86.68 <sup>abc</sup>			
obtained from 5 rats	В9	91.28 <sup>abc</sup>	89.05 <sup>abc</sup>			
Mean values within a	B 10	86.79 <sup>c</sup>	83.99 <sup>c</sup>			
column with the same superscript are not sig-	B 11	73.86 <sup>e</sup>	65.09 <sup>e</sup>			
nificantly different $(p>$	B 12	90.27 <sup>abc</sup>	87.59 <sup>abc</sup>			
0.05)	B 13	80.14 <sup>d</sup>	75.34 <sup>d</sup>			
B1–B14 refer to blends as shown in Table 1	B14	93.74 <sup>abc</sup>	88.45 <sup>abc</sup>			

measure of both digestibility and BV of the amino acid mixture absorbed from food (Ene-Obong and Obizoba 1995). Values obtained were above 70 % recommended for good food protein and dietary mixture (FNB 1974). The findings in this study agree with a report by FAO (1990), which stated that the amino acid content of roots and tubers (such as cocoyam), is not complemented by that of legumes as both are limiting in respect of the sulphur containing amino acids. It further suggested that in order to maximize their contribution to the diet, roots and tubers should be complemented by a wide variety of foods including cereals.

Table 3 shows the coefficients estimates, adjusted regression coefficients  $(R_{adj}^2)$ , the results of model significance as well as the lack of fit for BV and NPU for the cookies.

The multiple regression analysis showed that the special cubic model was significant in predicting the BV of the cookies (p=0.0005) and it could explain 89.55 % of all variance in the data. Furthermore, it did not present significant lack of fit (p=0.34). A non-significant lack of fit in a model makes it useful as a predictive model (Yağci and Gőğüş 2008). If a model has a significant lack of fit, it is not a good indicator of the response and should not be used for the prediction (Myers et al. 2009).

The regression model obtained for the BV of cookies was:

$$Y = 75.85x_1 + 80.08x_2 + 91.17x_3 + 68.33x_1x_2 + 26.09x_1x_2 + 12.22x_2x_2 - 170.03x_1x_2x_2$$
(2)

**Table 3** Coefficient estimates, model significance, adjusted regression coefficient (adj  $R^2$ ) and lack of fit values for biological value and net protein utilization of cookies produced from germinated pigeon pea, fermented sorghum and cocoyam flours

Variables	Biological value	Net protein utilization
X <sub>1</sub>	75.85	67.78
$X_2$	80.08	74.89
$X_3$	91.17	88.50
$X_1 X_2$	68.33	73.67
$X_1 X_3$	26.09	28.92
$X_2 X_3$	12.22	7.64
$X_1 X_2 X_3$	-170.03	n.s
Model (Prob>F)	$0.0005^{a}$	0.0003 <sup>a</sup>
Adj R <sup>2</sup>	0.8955	0.8690
Lack of fit	0.3384	0.2491

<sup>a</sup> significant at the 5 % level (p < 0.05); *n.s* non-significant

 $X_I$  cocoyam flour

 $X_2$  fermented sorghum flour

X<sub>3</sub> germinated pigeon pea flour

The positive (+) sign in the equations means that the response value increased with increase of the variables, whether it is linear, binary or ternary combinations while the negative (-) sign means that the response value decreased with increase of the variables thereby producing an antagonistic effect. From the equation, it was observed that increasing the linear and binary blends resulted in higher BV while increasing the ternary blends resulted in lower BV. Among the linear blends, germinated pigeon pea flour produced the highest increase in BV followed by fermented sorghum flour and cocoyam flour. Among the binary blends, the blend of cocoyam and fermented sorghum flour resulted in the highest BV.

The MRSM application on NPU data showed that quadratic model was significant (p=0.0003), no lack of fit was obtained (p=0.25) and it could explain 86.90 % of all variance in the data. All the components significantly increased the NPU as seen in the equation below:

$$Y = 67.78x_1 + 74.89x_2 + 88.50x_3 + 73.67x_1x_2 + 28.92x_1x_3 + 7.64x_2x_3$$
(3)

As was seen with BV, pigeon pea flour produced the highest increase in the NPU.

Sensory quality evaluation Mean values for consumer acceptance scores for the attributes tested for cookies produced are shown in Table 4. It was observed that while the addition of cocoyam flour improved ratings for the cookies, the addition of germinated pigeon pea flour had the opposite effect. Almost all the cookies containing at least 50 % GPF received low scores ( $\leq 6$ ) for texture, taste, crispness and general acceptability. Panelists described cookies containing

high levels of GPF as having a bitter aftertaste and lacking the characteristic crispness and texture associated with cookies. Except for those blends containing above 50 % GPF, all the cookies were at least slightly liked for the parameters studied. Cookies made with 100 % CF did not differ significantly (p>0.05) from the control (cookies made with 100 % wheat flour) for all the attributes tested. The most acceptable of the binary combinations was the one produced with 50CF:50FSF. Among the ternary blends, 66.6CF:16.7FSF:16.7GPF was the one with the highest acceptability and it did not differ significantly from cookies made with 50CF:50FSF and 100CF (p>0.05).

Results of the regression analyses listing the coefficients estimates, adjusted regression coefficients  $(R_{adj}^2)$  and the results of model significance and lack of fit for all the sensory attributes evaluated for cookies made with blends of germinated pigeon pea, fermented sorghum and blanched cocoyam flours are presented in Table 5.

The special cubic model was significant in predicting the texture of the cookies produced. Apart from the binary combination of CF and GPF, all the mixtures significantly increased the texture of the cookies as can be seen in the equation below:

$$Y = 7.81x_1 + 6.12x_2 + 5.62x_3 + 0.66x_1x_2 - 2.83x_1x_3 + 0.022x_2x_3 + 27.76x_1x_2x_3$$
(4)

The model could explain 88.74 % of the observed variations and it did not present a significant lack of fit (p>0.05). The equation suggests that the ternary blend produced the highest increase in sensory scores for texture.

consumer accep- erved for cook-	Blends	Texture	Taste	Colour	Crispness	General Acceptability
m germinated nented sorghum ours	B1	7.6 <sup>a</sup>	7.8 <sup>ab</sup>	7.6 <sup>ab</sup>	7.5 <sup>a</sup>	7.4 <sup>ab</sup>
	В 2	6.3 <sup>cd</sup>	6.5 <sup>bcd</sup>	6.5 <sup>bcd</sup>	6.2 <sup>cde</sup>	7.1 <sup>b</sup>
	В 3	5.9 <sup>d</sup>	5.0 <sup>e</sup>	5.9 <sup>d</sup>	5.3 <sup>e</sup>	5.0 <sup>d</sup>
	B 4	7.1 <sup>abc</sup>	7.1 <sup>abc</sup>	7.0 <sup>abcd</sup>	6.9 <sup>abcd</sup>	7.1 <sup>b</sup>
	В 5	5.9 <sup>d</sup>	6.0 <sup>cde</sup>	6.4 <sup>cd</sup>	5.1 <sup>e</sup>	5.0 <sup>d</sup>
	B 6	5.9 <sup>d</sup>	5.9 <sup>cde</sup>	6.7 <sup>bcd</sup>	5.4 <sup>e</sup>	5.5 <sup>cd</sup>
l on a 9-point	В 7	7.1 <sup>abc</sup>	6.9 <sup>abcd</sup>	6.9 <sup>abcd</sup>	6.8 <sup>abcd</sup>	6.8 <sup>bc</sup>
ith 1, dislike ex-	B 8	6.6 <sup>cd</sup>	5.6 <sup>de</sup>	6.8 <sup>bcd</sup>	6.0 <sup>de</sup>	5.8 <sup>cd</sup>
er like nor dis-	В9	6.7 <sup>bcd</sup>	6.7 <sup>bcd</sup>	6.9 <sup>abcd</sup>	6.5 <sup>bcd</sup>	6.5 <sup>bcd</sup>
extremely;	B 10	7.5 <sup>ab</sup>	7.5 <sup>ab</sup>	7.6 <sup>ab</sup>	7.1 <sup>abc</sup>	7.2 <sup>b</sup>
ists (n)=20	B 11	$8.0^{\mathrm{a}}$	$8.0^{\mathrm{a}}$	7.3 <sup>ab</sup>	7.7 <sup>a</sup>	8.3 <sup>a</sup>
vithin a column superscript are	B 12	5.3 <sup>e</sup>	5.3 <sup>e</sup>	5.8 <sup>d</sup>	5.3 <sup>e</sup>	5.3 <sup>d</sup>
y different $(p>$	B 13	6.0 <sup>cd</sup>	6.7 <sup>bcd</sup>	6.3 <sup>d</sup>	6.4 <sup>bcd</sup>	7.0 <sup>b</sup>
	B14	7.3 <sup>ab</sup>	7.4 <sup>ab</sup>	7.2 <sup>ab</sup>	6.4 <sup>bcd</sup>	7.1 <sup>b</sup>
to blends as	Control	8.0 <sup>a</sup>	8.2 <sup>a</sup>	$8.0^{\mathrm{a}}$	7.7 <sup>a</sup>	8.4 <sup>a</sup>

**Table 4** Mean consumer acceptance scores observed for cookies produced from germinatedpigeon pea, fermented sorghumand cocoyam flours

Scores are based on a 9-point hedonic scale with 1, dislike extremely; 5, neither like nor dislike; and 9, like extremely; number of panelists (n)=20

Mean values within a column with the same superscript are not significantly different (p > 0.05)

B1–B14 refer to blends as shown in Table 1

Table 5Coefficient estimates, model significance, adjusted re- gression coefficient (adj $R^2$ ) and lack of fit values for sensory attributes of cookies produced from germinated pigeon pea, fermented sorghum and coco- yam flours	Variables	Texture	Taste	Colour	Crispness	General Acceptability
	X <sub>1</sub>	7.81	8.16	7.70	7.58	7.82
	$X_2$	6.12	6.58	6.55	6.24	6.97
	$X_3$	5.62	5.10	5.99	5.31	5.11
	$X_1 X_2$	0.66	0.054	n.s	-1.07	-1.41
	$X_1 X_3$	-2.83	-2.57	n.s	-4.08	-3.58
	$X_2 X_3$	0.022	0.040	n.s	-2.54	-4.11
<sup>a</sup> significant at the 5 % level ( <i>p</i> < 0.05); <i>n.s</i> non-significant	$X_1 X_2 X_3$	25.76	14.84	n.s	37.06	31.40
	Model (Prob>F)	$0.0006^{a}$	<0.0001 <sup>a</sup>	< 0.0001 <sup>a</sup>	<0.0001 <sup>a</sup>	<0.0001 <sup>a</sup>
$X_1$ cocoyam flour	Adj R <sup>2</sup>	0.8874	0.9654	0.8056	0.9485	0.9786
$X_2$ fermented sorghum flour	Lack of fit	0.6570	0.4257	0.4308	0.5586	0.2241

 $X_3$  germinated pigeon pea flour

The regression model obtained for the colour of the cookies was:

$$Y = 7.70x_1 + 6.55x_2 + 5.99x_3 \tag{5}$$

There was significant influence of the linear terms on the colour of the cookies. The equation suggests that the cocovam flour produced the highest increase in sensory scores for colour. This was followed by sorghum and pigeon pea flours.

The model could explain 80.56 % of the variations in the colour observed. Thus, 19.44 % of the variation was attributed to factors not included in the model. The lack of fit test showed that model error and replicate error were small, meaning that there was no lack of fit (p=0.43) which further validated the suitability of the model for prediction purposes.

With regards the taste of the cookies, the equation obtained from the data is as follows:

$$Y = 8.16x_1 + 6.58x_2 + 5.10x_3 + 0.05x_1x_2 - 2.57x_1x_3 + 0.04x_2x_3 + 14.84x_1x_2x_3$$
(6)

The equation suggests that the combination of CF and GPF  $(x_1 x_3)$  contributed to an antagonistic effect on the taste of the cookies. According to the multiple regression analysis, the special cubic model for taste data was significant statistically (p < 0.0001) and could explain 96.54 % of all variance of the hedonic results. No lack of fit was obtained from the data (p=0.43). The equation suggests that the ternary blend produced the highest increase in sensory scores for taste.

The scores obtained for crispness differed statistically and it was observed that all of the binary combinations resulted in lower hedonic scores. Cookies made with 100CF did not differ statistically (p < 0.05) from the control. The MRSM application on crispness scores showed that the special cubic model was significant (p < 0.0001), no lack of fit was obtained (p=0.56) and it could explain 94.85 % of all variance data. Thus, about 5.15 % of the variation was due J Food Sci Technol (October 2014) 51(10):2671-2677

to other factors not included in the model. The model obtained for the crispness of the cookies was:

$$Y = 7.58x_1 + 6.24x_2 + 5.31x_3 - 1.07x_1x_2 - 4.08x_1x_3$$
$$- 2.54x_2x_3 + 37.06x_1x_2x_3$$
(7)

As was observed with texture and taste, it appeared that the crispness of cookies was increased mostly by the ternary blend.

The regression model obtained for the general acceptability of the cookies was:

$$Y = 7.82x_1 + 6.97x_2 + 5.11x_3 - 1.41x_1x_2 - 3.58x_1x_3$$
$$- 4.11x_2x_3 + 31.40x_1x_2x_3.$$
(8)

Statistical analysis indicated that the special cubic model was significant in predicting the general acceptability of the cookies (p < 0.0001). The model could explain about 97.86 % of the observed variations and did not present significant lack of fit (p=0.22). The equation shows that all of the mixtures except for the binary blends increased the general acceptability of the cookies. The equation also suggests that the ternary blend produced the highest increase in sensory scores for general acceptability.

#### Conclusion

Mixture response surface methodology could be used to determine the effect of variation in the levels of cocoyam, sorghum and pigeon pea flours on sensory scores and protein quality of cookie formulations. This research has also shown that the modelling of experimental data allowed the generation of useful equations which may be used to predict effects of various blends of germinated pigeon pea, fermented sorghum and cocoyam flours on the sensory and protein quality of cookies without preparing samples. The technique employed in this work can be used to develop novel foods.

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