Applied Tropical Agriculture
Volume 23, No. 2, 82-89, 2018
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The Federal University of Technology, P.M.B. 704, Akure, Nigerian.

QUALITY AND *IN VITRO* ESTIMATED GLYCEMIC INDEX OF COOKIES FROM UNRIPE PLANTAIN-CRAYFISH-WHEAT COMPOSITE FLOUR

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Abstract
The behaviour of composite flour obtained from unripe plantain (PF), crayfish (CF) and wheat (WF) flours in the production of cookies was investigated. The physicochemical, estimated glycemic index and sensory parameters of the produced cookies were used to identify the potentiality of the plantain-crayfish-wheat composite flour. Proximate composition of the composite cookies ranged from 15-18% protein, ~2% fibre, 23-25% fat and ~50% carbohydrate while 100% WF-cookies had 12.60% protein, 3.28% fibre, 7.70% fat and 71% carbohydrate, respectively. On the contrary, all the experimental composite cookies had very high (≤86) estimated glycemic index as compared to the control (what's the GI of the control, should be placed in this parenthesis). Notably, potassium was the abundant mineral (2-52 mg/100 g) present in the composite cookies while calcium (6 mg/100 g) was abundant in the 100% WF-cookies. The physical properties (thickness, bulk density, breaking strength, diameter) and sensory parameters (colour, taste, crispiness, aroma) of the experimental cookies were comparable and significantly (p>0.05) similar to the commercial cookies. The improved physicochemical and nutritional properties of the resultant composite cookies may indicate the potential of the composite for production of acceptable cookies.

Keywords: *plantain, crayfish, composite flour, cookies, glycemic index*

Introduction
Research into the possible utilization of composite flours from indigenous crops had improved the value addition of these crops beyond their current basic use. For instance, indigenous crops have been utilized in producing flours with close-resemblance characteristics to the well- known wheat flour (Udeh & Malomo, 2018) for confectionaries (such as cookies) production. Cookies have been described as a low moisture content-based confectionery product when compared to biscuits, but with a larger and softer chewier texture (Okaka, 2009). It is consumed extensively all over the world as a snack food and on a large scale in developing countries with prevalent and exponentially higher protein and caloric malnutrition (Chinma & Gernah, 2007). A current trend in nutrition is the consumption of low-carbohydrate diets, including slowly-digestible food products, as well as an increased intake of functional foods (Aparicio-Saguilan et al., 2007; Hurs & Martin, 2005). For instance, there exists a recent WHO recommendation to reduce the overall consumption of sugars and foods that promote high glucose responses (WHO/FAO, 2003). This is as a result of increased advocacy on the consumption of functional foods due to different health problems related with food consumption such as celiac disease (life-long intolerance to wheat gluten, characterized by inflammation of the proximal small intestine), diabetes and heart diseases (Eke-Ejiofor & Kiin-Kabari, 2012).

Cookies are considered to be a concentrated food due to high contents of carbohydrates, fats and low moisture (Aparicio-Saguilan et al., 2007). As such, they are a substantial source of energy in which their quality can be enhanced by including a number of ingredients, like crayfish, in recipe formulations. The initiation of composite flour programmes to evaluate the feasibility of alternative locally available flours as a substitute for wheat flour in novel food production had been encouraged (Abdelghafor et al., 2011). For instance, the use of high yielding, native plant species as a means of growing economy and sources of protein supply for human nutrition have been reported in Malaysia (Hasmadi et al. 2014) and Nigeria (Adeloye, 2012). Therefore, cookies can serve as vehicle for delivery of important nutrients if made readily available to the population (Chinma & Gernah, 2007). This partly stimulated research into the production of cookies from non-wheat flour blends containing functional ingredients (principally those with high fiber and resistant starch) such as unripe plantain. Dietary fiber in human diets lowers serum cholesterol, reduces the risk of heart attack, colon cancer, obesity, blood pressure, appendicitis and many other disease (Rehinan et al., 2004).

Therefore, this study is aimed at producing plantain flour-based cookies enriched with crayfish protein concentrate in order to improve the nutritional quality of the cookies and increase the utilization of unripe plantain flour.

Materials and Methods

Materials

Matured freshly cut unripe green plantain was obtained from a farm in Oke-ogba Akure, Ondo State, Nigeria. Dried Crayfish was purchased at Sabo market and wheat flour was purchased at Oja-oba Market Akure, Ondo State, Nigeria. The basic formulation ingredients for the baking of cookies were all purchased from Mummy T. Supermarket, Akure, Ondo State, Nigeria.

Methods

Processing of Unripe Plantain:

Matured unripe plantains (*Musa paradisiaca* known in Yoruba, Nigeria as *Agbagba*) cultivar was washed along with the skin to reduce contaminates and were peeled manually with the aid of a sharp stainless steel kitchen knife, the pulp was cut into uniform slices with thickness of about 1.5 mm, and soaked in hot water containing 1.25% sodium metabisulphite solution for 20 min and drained. The samples were dried at 65° C for 6 h using a cabinet dryer, milled using a hammer mill (at Ezedoc Foods Limited at Block MF Industrial Park, formerly Onyearugbulem Market Ilesha Owo expressway Akure, Ondo State, Nigeria) and sieved to pass a 0.25 mm sieve.

Processing of Crayfish into Flour

Dried crayfish (*Procombarus clarkia)* was cleaned and winnowed manually by sorting out foreign matters like stones and removing crayfish tails and heads before processing into flour using a manual hand engine milling machine to produce crayfish flour.

Preparation of Flour Blends

Graded levels of unripe plantain flour, crayfish and wheat composite flours were mixed at different proportions to obtain five (5) flour blends as shown in Table 1:

Table 1- Blend formulation of unripe plantain, crayfish and wheat flours for composite flour

Samples (%)	PF	CF	WF
A	80	10	10
B	70	10	20
C	60	10	30
D	50	10	40
E (control)	0	0	100

PF = Plantain flour; CF= Crayfish flour; WF= Wheat flour

Preparation of Cookies

Cookies were prepared according to the method of AACC (2000) with some modifications in the recipe: 100g flour, beet (2 g), margarine 5 g, baking powder 1g, one egg, salt 0.1 g and water. The dry ingredients (flour, beet, salt, milk and baking powder) were thoroughly mixed in a bowl by hand for 3 min, margarine was added and mixed until uniform. Egg was then added and the mixture kneaded. The batter was rolled on a flat rolling board sprinkled with flour for a uniform thickness, and cut with a 50 mm diameter cookie cutter. The cookies were placed on baking trays leaving a 25 mm space in between and were baked at 180 °C for 10 min the baking oven. After baking, the cookies were cooled at ambient temperature, packaged in polyethylene bags and stored at 23° C prior to subsequent analysis.

Determination of Proximate Composition

Proximate composition of the samples (moisture content, crude protein , fat, total ash and crude fibre) was determined using AOAC methods (AOAC, 2000). Total carbohydrate was quantified by calculating the difference.

Determination of Mineral Elements Composition

Mineral compositions were determined using AOAC method (1995). The ash was digested with 3 cm^3 of 3 ml HCL and made up to the mark in a 100 cm^3 standard flask with 0.36 M HCL before the mineral elements (calcium, zinc, magnesium, iron and potassium) were determined by atomic absorption spectrophotometer.

Physical Analysis of Cookies

Cookie diameter (*D*) and thickness (*T*) were determined using a Vernier caliper. Cookie spread was determined from the diameter and thickness, using the following previously described (Mir et al. 2015) formula:

$$SF = (D/T \times CF) \times 10$$

Where, CF is the correction factor, at constant atmospheric pressure. Bulk volume of cookies was determined with slight modifications to the method described by Mir et al. (2015). Bulk density of cookies was determined from the bulk volume and weight of cookie *viz*:

$$\text{Bulk density} = \text{Weight of cookie} / \text{Bulk volume}$$

Sensory Evaluation of Cookies

Sensory properties of cookies was carried out using a semi-trained fifty-member panelist, consisting of students and staff members from the Department of Food Science and Technology, Federal University of Technology, Akure, Nigeria. Cookie samples prepared from each flour blend were presented in coded white plastic plates. The order of presentation of samples to the panelists was randomized. The panelists were instructed to evaluate the coded samples for aroma, taste, crispiness, colour, texture and overall acceptability. Each sensory attribute was rated on a 9-point Hedonic scale ranging from 1-(dislike extremely) to 9-(like extremely).

In vitro Estimation of Glycemic Index of the Composite Cookies

The *in vitro* estimation of glycemic index of the composite cookies was done according to the previously described method (Dodd et al., 2011). Briefly, the carbohydrate content in each of the cookies was determined through the proximate analysis carried out. The total grams of carbohydrates were divided by the number of grams contributed by the individual component so as to find the proportion of carbohydrate for the component. This is followed by multiplying the proportions for the sample components by the predetermined GI of that component. GIs of individual foods can be found in an online GI database or in the table published by "*The American Journal of Clinical Nutrition*." The results were added together to determine the GI value of each of the biscuit samples.

Statistical Analysis

All determination reported in this study were carried out in triplicates. In each case, a mean value and standard deviation were calculated. One – way ANOVA was also performed using SPSS version 17.0 and separation of mean values using Duncan Multiple Range Test.

Results and Discussion

Proximate Compositions of the Cookies Produced from Wheat-plantain-Crayfish Composite Flours

Moisture content (MC) of the samples ranged from 7.52 to 9.47% (Table 2). The result showed that the 100% WF-cookies (control sample E) might have longer shelf live when compared to the composite cookies A-D of higher MC. The findings of Sanni et al. (2006) suggested longer shelf live of products with low MC. The current values in this study (7.52-9.47%) fall within the range (<10%) reported to have no adverse effect on the quality attribute of the food products (Abdelghafor et al., 2011) and are lesser than the 15% reported (Adebowale et al., 2012) for sorghum-wheat biscuit. Basically, the differences in the MC might be due to differences in raw materials (plantain-crayfish and sorghum) and processing methods involved (Agiriga, 2014). The 70%PF+10%CF+20%WF (sample B) cookies had the highest MC (9.47%) compared to other samples A, C, D and E. Hence, besides 100% WF cookies (sample E with 7.52%MC), the 80%PF+10%CF+10%WF (sample A with 8.08%MC) cookies might keep better than sample B.

The crude protein contents ranged from 12.60 to 19.83% (Table 2) with samples A and E having the highest (19.83%) and lowest (12.60%) values, respectively. Previous study (Vittadini & Vodovotz, 2003) reported the successful utilization of animal proteins in complementary products to obtain a protein-enriched product of cheap and moderate prices, which is found good for infants.. The inclusion of crayfish (animal protein) to sample A might have contributed to its high protein content (19.83%) when compared to that (12.60%) of the control (100% WF-cookies) sample, thus sample A might be found useful as potential agent in the management of chronic diseases (Oboh & Omotosho, 2005). Meanwhile about 18.15% protein content was reported (Odebode et al., 2017) for 70% unripe plantain-based flour, which suggested plantain to not be a good source of protein.

Total ash ranged from 3.89 to 5.32% with sample E having the highest value. The ash content of a food material could be used as an index of mineral constituents of the food (Sanni et al., 2008). Comparatively, the high ash content in sample A as against those in samples B, C and D could be attributed to high proportion of plantain (80%), and its mineral elements especially sodium, potassium and iron (Table 3). Crude fibre contents of the cookies (2.05-3.28%) showed that they could aid the digestion in the colon and reduce constipation often associated with confectionery products (Jideani & Onwubali, 2009). Previous study (Slavin, 2005) had confirmed a significant role of crude fibre in the prevention of several diseases such as cardiovascular diseases, diverticulosis, constipation and irritable colon.

The fat contents ranged from 7.70 to 27.27% with control sample E and D having the lowest (7.7%) and highest (27.27%) values, respectively. The high fat contents recorded for cookies from composite flours might be as a result of crayfish (animal origin) inclusion in the composite flour, which is not present in 100% WF-cookies. High fat content plays a significant role in the shelf life of food products by promoting rancidity in foods, leading to development of unpleasant and odorous compounds (Ikujenlola, 2014). Hence, food products with high fat content should be consumed immediately to prevent rancidity in the products (Ikujenlola, 2014). Carbohydrate contents of the cookies ranged from 48.10 to 71.11%. The carbohydrate content (71.11%) in the control sample (100% WF-cookies) is significantly ($p<0.05$) higher than sample B (52.43%), which might be as result of presence of non-digestible sugar in sample E compared to the composite cookies (Fig 1). The substitution of plantain flour had been reported to enrich the carbohydrate, fibre and minerals of an ideal food for diabetic, children and pregnant women (Eke-Ejiofor & Kiin-Kabari, 2012). The frequent consumption of plantain flour has aided the reduction in blood sugar (USDA Nutrient Database, 2010). Thus, consumption of sample A (having 80% plantain) with estimated low glycemic index (79) when compared to 87 recorded (Fig 1) for 100% wheat flour-cookies might efficiently regulate the blood sugar.

Table 2 - Proximate compositions of plantain-based composite cookies (%dry weight)

Samples	M.C.	Protein	Fat	Fibre	Ash	Carbohydrate
A	8.08 ± 0.03^{c}	19.83 ± 0.34^{a}	25.80 ± 0.37^{b}	2.12 ± 0.01^{b}	4.19 ± 0.06^{b}	48.10 ± 0.18^{e}
B	9.47 ± 0.21^{a}	16.65 ± 0.05^{c}	24.91 ± 0.69^{c}	2.13 ± 0.01^{b}	3.89 ± 0.46^{c}	52.43 ± 0.54^{b}
C	8.22 ± 0.27^{bc}	17.03 ± 0.16^{b}	25.58 ± 0.85^{b}	2.07 ± 0.01^{bc}	4.04 ± 0.25^{bc}	51.28 ± 0.92^{c}
D	8.32 ± 0.02^{b}	17.08 ± 0.25^{b}	27.27 ± 0.46^{a}	2.05 ± 0.00^{c}	3.96 ± 0.23^{c}	49.64 ± 0.56^{d}
E	7.52 ± 0.07^{d}	12.60 ± 0.09^{d}	7.70 ± 0.30^{d}	3.28 ± 0.01^{a}	5.32 ± 0.17^{a}	71.11 ± 0.14^{a}

Means with different letters are significantly different in the same column at the $p\leq0.05$. **A**- 80% Plantain Flour, 10% Crayfish Flour, 10% Wheat Flour; **B**- 70% Plantain Flour, 10% Crayfish Flour, 20% Wheat Flour; **C**- 60% Plantain Flour, 10% Crayfish Flour, 30% Wheat Flour; **D**-50% Plantain Flour, 10% Crayfish Flour, 40% Wheat Flour; **E**- 100% Wheat Flour

Mineral Compositions of the Composite Flour-produced Cookies

Table 3 showed that potassium content of the composite cookies was the highest (15.67-52.20 mg/100 g) followed by sodium (6.59-10.61 mg/100 g) while the iron content was 55.05-5.72 mg/100 g when compared to control sample (2.34, 1.44 and 1.36 mg/ 100 g), respectively.

Calcium (2.66-4.15 mg/100 g) and phosphorus (0.13-0.31 mg/100 g) are the least abundant minerals in the composite cookies but contrarily higher (6.06 and 0.74 mg/100 g) in control sample, respectively.

Generally, proper reduction in high blood pressure and hypertension pathogenesis had been linked (Zhang et al., 2013) with high potassium and low sodium intake, hence sample A could serve as potential snacks for the hypertensive patients. Interestingly, Lead (Pb) was not detected in any of the composite cookies and control, thus indicating their safe consumption and freed from critical health issues implicated with heavy metals.

Table 3- Mineral contents of plantain-based composite cookies (mg/100 g)

Samples	Na	K	Ca	P	Fe	Pb
A	10.61±0.02[a]	52.20±0.01[a]	4.15±0.06[b]	0.31±0.01[b]	5.72±0.03[a]	ND
B	6.59±0.01[d]	17.32±0.09[c]	2.66±0.02[e]	0.14±0.01[c]	5.05±0.01[d]	ND
C	7.66±0.01[b]	24.51±0.01[b]	2.86±0.02[d]	0.15±0.01[c]	5.20±0.01[c]	ND
D	7.08±0.02[c]	15.67±0.12[d]	2.99±0.04[c]	0.13±0.01[c]	5.42±0.01[b]	ND
E	1.44±0.04[e]	2.34±0.07[e]	6.06±0.02[a]	0.74±0.02[a]	1.36±0.05[e]	ND

Means with different letters are significantly different in the same column at the $p \leq 0.05$. **A**- 80% Plantain Flour, 10% Crayfish Flour, 10% Wheat Flour; **B**- 70% Plantain Flour, 10% Crayfish Flour, 20% Wheat Flour; **C**- 60% Plantain Flour, 10% Crayfish Flour, 30% Wheat Flour; **D**-50% Plantain Flour, 10% Crayfish Flour, 40% Wheat Flour; **E**- 100% Wheat Flour; **ND** = Not Detected

Physical Properties of Composite Flour-Produced Cookies

The physical characteristics of the composite cookie samples are presented in Table 4. The diameter of cookie samples ranged from 4.44 (sample D) to 4.79 cm (sample A) compared to the control sample (4.50 cm). However, thickness (5.10 mm) and spread ratio (12.24 mm) of the control sample (100% WF-cookies) were significantly ($p<0.05$) higher than those (0.51-0.86 and 5.22-9.38 mm) obtained for the composite cookies, respectively. The thickness of the composite cookies observed to increase gradually with decrease in the level of plantain and increase in wheat flour (samples A and D) in Table 4. The breaking strength and bulk density range from 3.47-6.03 g and 0.78-0.87 g/cm³ comparable to control sample (5.22 g and 0.48 g/cm³), respectively, whereas no significant difference ($p>0.05$) was observed for bulk densities of samples B, C and D. Meanwhile, the highest breaking strength (6.03 g) and bulk density (0.87 g/cm³) of sample C out of all the composite cookies could be as a result of its blend (60%PF and 30%WF) ratio.

Therefore, cookies prepared from 60%PF and 30%WF is favorable with better thickness and bulk density. The current observation from this blend (60%PF and 30%WF) ratio is similar to the previous findings on cookies from wheat-fluted pumpkin seed protein concentrates (Giami & Barber, 2004) and defatted sesame-unripe plantain (Chinma et al., 2012) composite flours.

Table 4- Physical properties of plantain-based composite cookies

Samples	Diameter (cm)	Thickness (mm)	Spread Ratio (mm)	Breaking Strength (g)	Bulk Density (g/cm³
A	4.79±0.06[a]	0.51±0.04[c]	9.38±0.77[b]	3.47±0.06[d]	0.78±0.02[b]
B	4.68±0.18[b]	0.59±0.08[c]	7.99±1.26[c]	3.90±0.10[c]	0.86±0.00[a]
C	4.70±0.06[ab]	0.53±0.06[c]	9.00±0.97[b]	6.03±0.15[a]	0.87±0.01[a]
D	4.44±0.06[c]	0.86±0.12[b]	5.22±0.83[b]	3.83±0.12[c]	0.84±0.01[a]
E	4.50±0.58[c]	5.10 ± 0.01[a]	12.24±0.04[a]	5.22±0.18[b]	0.48±0.02[c]

Means with different letters are significantly different in the same column at the $p \leq 0.05$. **A**- 80% Plantain Flour, 10% Crayfish Flour, 10% Wheat Flour; **B**- 70% Plantain Flour, 10% Crayfish Flour, 20% Wheat Flour; **C**- 60% Plantain Flour, 10% Crayfish Flour, 30% Wheat Flour; **D**-50% Plantain Flour, 10% Crayfish Flour, 40% Wheat Flour; **E**- 100% Wheat Flour

Glycemic Index of the Composite Flour

The results presented in Fig 1 showed that the cookies were having very high glycemic index (GI) (79.55-87.22), that is GI>70. It has been well documented (Foster-Powell et al., 2002) that when GI of any food or food product is >70, between 55-69 or <55, such food is classified as high, medium or low GI food, respectively. Although the present results are unexpected since plantain being the main ingredient has been reported (Mendosa, 2009) to have low GI. Thus, the high GI obtained in this study may be due to wheat inclusion, which has been reported (Aparicio-Saguilan et al., 2007) to possess high GI contents. This is further corroborated by the observed increasing GI as wheat content increased in the composite blends (Fig 1).

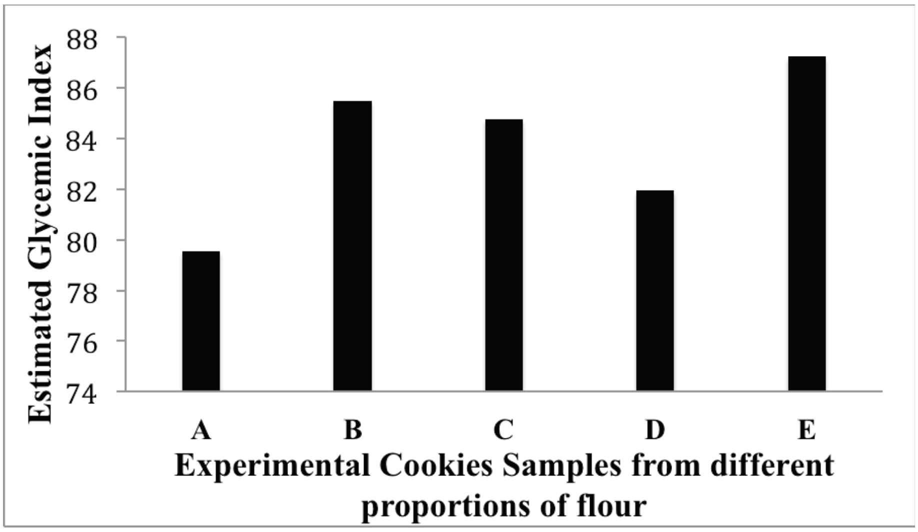

Fig 1- Estimated Glycemic Index of composite flour-produced cookies

A- 80% Plantain Flour, 10% Crayfish Flour, 10% Wheat Flour; **B**- 70% Plantain Flour, 10% Crayfish Flour, 20% Wheat Flour; **C**- 60% Plantain Flour, 10% Crayfish Flour, 30% Wheat Flour; **D**-50% Plantain Flour, 10% Crayfish Flour, 40% Wheat Flour; **E**- 100% Wheat Flour. **Note**: When GI value is: Less than 55 = low GI value; 55 – 69 = medium GI; Greater than 70 = high GI (Foster-Powell et al., 2002)

Sensory Properties of the Composite Flour-Produced Cookies

The result of sensory analysis of the plantain cookies is shown in Table 5. The results showed that a significant ($p<0.05$) difference exist among the experimental samples. This showed the samples may not be differentiated based on their physical appearances. It can be inferred from the appearance that samples A and E are moderately liked, sample B slightly liked and sample C dislike or not disliked. Among the composite flour blends, rating of the crispiness showed sample D having the highest and sample C having the least when compared to sample E (100% WF-cookies). This may be due to high presence of plantain flour, which is a good source of fibre (Chinma et al., 2012). There was no significant difference between the tastes of the samples A, B, C and D except for the control (E). The result (Table 5) further showed that the texture of sample D is very slightly liked compared to controls (E and F). The tastes of the control samples (E and F) are rated highest among them all, which might be as a result of their sweetened nature. The result shows no significant differences in the overall acceptability of the composite flour blends between samples A, C, D, but these samples were significantly ($p<0.05$) different from the control samples (E and F) according to the consumers' preference. This current observation corresponded with the previous findings earlier reported (Famakin et al., 2016) on functional dough meals.

Table 5- Sensory Evaluation of the Plantain cookies and Commercial cookies

Sample	Colour	Taste	Texture	Crispiness	Appearance	Aroma	Overall acceptability
A	7.00[c]	5.50[c]	7.00[b]	6.20[b]	5.75[c]	5.30[b]	6.70[b]
B	6.65[d]	5.70[c]	7.05[b]	6.20[b]	6.00[b]	4.80[c]	6.35[c]
C	6.75[d]	5.80[bc]	7.10[b]	6.15[b]	6.10[b]	5.55[b]	6.70[b]
D	6.65[d]	6.25[b]	6.30[c]	6.25[b]	6.30[b]	4.95[c]	6.70[b]
E	8.90[b]	8.80[a]	8.85[a]	8.90[a]	8.65[a]	8.70[a]	9.00[a]
F	9.00[a]	9.00[a]	9.00[a]	9.00[a]	9.00[a]	9.00[a]	9.00[a]

Means with different letters are significantly different in the same column at the p≤0.05. **A**- 80% Plantain Flour, 10% Crayfish Flour, 10% Wheat Flour; **B**- 70% Plantain Flour, 10% Crayfish Flour, 20% Wheat Flour; **C**- 60% Plantain Flour, 10% Crayfish Flour, 30% Wheat Flour; **D**-50% Plantain Flour, 10% Crayfish Flour, 40% Wheat Flour; **E**- 100% Wheat Flour; **F**- Control (Commercial Cookies)

Conclusion

The cookie made with flour blend of 80% plantain, 10% crayfish and 10% wheat gave the best overall acceptability and the best nutritional qualities in terms of the crude protein (18.23%), physical properties (diameter and spread ratio), mineral elements (calcium and potassium) and manageable (79) estimated GI. This would be of nutritional importance for coeliac disease and diabetic patients in most developing countries such as Nigeria, where people can hardly afford high proteinous foods from animals because of its expensive purchasing costs. The cookies produced had good sensory quality, which could compete with commercial cookies in terms of appearance and texture in the open market. Substitution of flours with different raw materials (plantain and crayfish flours) had greatly improved the protein, quality and nutritional benefits of cookies.

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