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Effect of Incorporation of Amaranth Leaf Flour on the Chemical, Functional and Sensory Properties of Yellow Maize/Soybean Based Extrudates

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Abstract: *This study evaluated the effect of incorporation of amaranth leaf flour on the chemical, functional and sensory properties of extrudates from flour blends of yellow maize and soybean. Flour samples of wheat, yellow maize, soybean and amaranth leaf were produced by cleaning, drying, milling and sieving through 2 mm mesh sieve. Amaranth leaf flour was incorporated into yellow maize and soybean (70:30) composite flour at 0, 5, 10, 15 and 20 % replacement levels and wheat flour (100 %) served as control. Baking ingredients were added to the composite flour blends and wheat flour, to produce extruded snack samples using a single screw extruder, before drying in an oven at 60 ^{o}C for 30 minutes. The proximate composition, minerals, vitamins and anti-nutritional factors of the extrudates were analysed. Incorporation of amaranth leaf and soybean flour significantly ($p < 0.05$) increased the protein content from 17.88 to 21.67 % and energy from 349.72 to 360.07 kcal/100g. Amaranth leaf flour incorporation significantly ($p < 0.05$) increased the mineral and vitamin contents of the extrudates. Addition of 5 % amaranth leaf flour significantly ($p < 0.05$) increased protein, mineral and vitamin contents of extruded snacks compared to the control. The extrusion cooking and drying treatments significantly ($p < 0.05$) reduced anti-nutritional factors. Addition of amaranth leaf flour significantly ($p < 0.05$) reduced expansion ratio, water absorption capacity and significantly ($p < 0.05$) increased the bulk density, pH and oil absorption capacity of the extrudates. The microbial load which was very low, ranged from 1.08 to 9.4 x 10^1 cfu/g for bacteria count, 0.00 to 3 x 10^1 cfu/g for mould count, while coliform was not detected in all extruded snack samples. The most acceptable snack was the wheat flour (control), while acceptability of other extrudates decreased with increasing addition of amaranth leaf flour. The extrudate with 10 % level of amaranth leaf flour incorporation was still acceptable.*

Keywords: *Amaranth leaf flour, Chemical composition, Extrudates, Functional properties, Sensory quality*

I. Introduction

Snack foods are commonly available foods that are eaten between main meals or ‘a light meal eaten between regular meals [1]. Basically, snack foods are prepared from natural ingredients or components according to predesigned plans to yield products with specified functional properties [2]. Snack food consumption is becoming popular and contributes to the total energy intake [3]. The proper consumption of healthy snacks, therefore, can help the supplementation of energy and nutrient requirements [4]. The most widely consumed snacks are cereal-based products, which generally are low in nutrient density [5]. However, incorporation of ingredients with high nutrient density into these snack products could increase their nutritional values [6]. The nutritional quality of yellow maize-soybean based snacks containing amaranth leaf flour has not been investigated. Therefore, combination of yellow maize (Zea mays L.), soybean (Glycine max L. Merrill) and amaranth leaf (*Amaranthus hybridus* L.) flours to produce snacks, would help to increase nutrient density of extruded snacks available in Rwanda. One of the most important technologies which have shown great potential for the development of new snack product is extrusion cooking [7]. Extrusion cooking is preferable to other food-processing techniques in terms of continuous process with high productivity and significant nutrient retention, owing to the high temperature and short time required [8]. The objective of this work was to incorporate amaranth leaf flour in yellow maize and soybean flour blends to develop extrudates and assess their quality characteristics.

II. Materials and Methods

2.1 Source of Raw Materials

The materials used in this study were amaranth leaf, yellow maize, soybean and wheat. Five kilograms (5 kg) of soybean, two kilograms (2 kg) of wheat, and ten kilograms (10 kg) of fresh amaranth leaves were bought from Kimironko market, Gasabo district, Kigali city, Rwanda. Eight kilograms (8 kg) of yellow maize as well as other ingredients (sugar, salt, baking fat, baking powder and vanilla) used for this work were obtained from Ogige market in Nsukka, Enugu State, Nigeria.

2.2 Production of Flour

Each of the samples was cleaned to remove foreign materials, sorted, washed with distilled water, dried at 60 ^{o}C, milled, sieved through a 2.0 mm mesh sieve then packaged in polyethylene bags and kept at 10 ^{o}C in a refrigerator, until analyses, formulation, and preparation of extruded snacks.

2.3 Preparation of Flour Blends

Amaranth leaf flour was incorporated into yellow maize and soybean (70:30) composite flour at 0, 5, 10, 15 and 20 % replacement levels as well as wheat flour at 100 %, served as a control.

2.4 Samples Preparation for Extrusion

The flour (500 g), sugar (50 g), salt (5 g), baking fat (5 g) were mixed together manually for six minutes to get a creamy dough. The baking powder (5 g) and vanilla (7.5 g) were then added. Distilled water (103 ml) was gradually added while mixing until good textured, slightly firm dough was obtained. The dough was kneaded on a clean flat surface for three minutes.

2.5 Extrudates Preparation

Extrusion of samples was performed using locally fabricated FST 001 single-screw extruder available at the Department of Food Science and Technology, University of Nigeria, Nsukka, Enugu State, Nigeria. The output shaft of worm reduction gear was provided with a torque limiter coupling. The barrel of the extruder received the feed from a co-rotating variable speed feeder. The barrel was provided with three electric band heaters: entry barrel temperature (T_1), centre barrel temperature (T_2) and end barrel temperature (T_3) and their regulators (80, 100, and 160 ^{o}C), respectively. The die was required to be fixed on the face of barrel by a screw nut tightened by a special wrench provided. The single screw extruder was kept on for 30 min to stabilize the set temperatures, prior to feeding each sample through the hopper at 500 g/min. The die diameter was selected at 4 mm as provided by the model manufacturer. The product was collected at the die end and dried at 60 °C in an oven (Fulton, Model NYC-101 oven) for 30 min to remove extra moisture from the extrudates.

2.6 Analytical Methods

The moisture, fat, ash, crude fibre and crude protein contents were determined using the methods of Association of Official Analytical Chemists [9]. The total carbohydrate content was determined by difference method as described by [10]. The calorific value was computed by summing up the values obtained by multiplying the values with Atwater constants for carbohydrates, crude fat and crude protein with the factors 4, 9 and 4, respectively [11].

Lectin was determined by the spectrophotometric method of (9). The saponin contents of samples were determined following the (9) method. Oxalate was determined by using the method of [12]. The method of [13] was used to determine trypsin inhibitor, while phytate content was determined using spectrophotometric method described by [14].

The vitamin A, C, B_1 and B_2 contents were determined using the the methods of Association of Official Analytical Chemists [9], while the vitamin E content was determined using the method described by [14]. Iron and zinc were determined by the method described by [15]. Magnesium was determined using the method described by [16]. Calcium was determined using the method described by [14]. Phosphorus was determined according to [15] by the molybdate method using hydroquinone as a reducing agent, while potassium was determined by a procedure described by [17] using a flame photometer.

Water absorption capacity, fat absorption capacity, bulk density and pH were determined using the method described by [18], while the expansion ratio was determined using the method described by [19]. Sensory evaluation of the samples was conducted as described in [51]. The attributes evaluated were taste, aftertaste, mouthfeel, aroma, texture, crust colour, clumb colour and the overall acceptability, using a 9 point Hedonic scale.

2.7 Statistical Analysis

All experiments were performed in triplicates. The data were subjected to One-way Analysis of Variance (ANOVA) and Duncan's New Multiple Range Test (DNMRT) was used to determine the difference between means of the tested parameters. Differences were determined as significant or non-significant at a significance level of 0.05 in all cases.

III. Results and Discussion

The proximate composition of extruded snacks presented in Table 1,showed significant ($p < 0.05$) differences in all measured parameters. The result revealed that incorporation of amaranth leaf flour in yellow maize-soybean blends increased protein content from 17.88 to 21.67 %, ash content from 3.17 to 4.60 %, fat content from 2.70 to 5.27 %, crude fiber content from 3.33 to 4.97 % and energy from 349.72 to 360.07 kcal/100g and reduced the carbohydrate content from 65.72 to 56.98 %. The extrudates showed higher amount of protein, ash, fat, crude fiber and carbohydrates than the control (I) which was produced from wheat flour only. The moisture content of the extruded snacks detected in this study were within the limit (10 %) specified by World Food Programme (WFP) as maximum moisture content for efficient storage of maize-soy snack blends. The protein contents detected in this study ranged from 14.82 to 21.67 % and were higher than the range (15.13-16.43 %) reported by [20] for extruded products from corn, millet and soybean blend. The protein content of extruded snack samples were significantly ($p < 0.05$) higher than that of the control snack sample (I) and FAO/WHO [21] recommended level (> 15 %).

Table 1: Proximate composition of the extrudates

Extruded Snack Samples	**Moisture (%)**	**Protein (%)**	**Ash (%)**	**Fat (%)**	**Crude Fibre (%)**	**Carbohydrate (%)**	**Energy (Kcal/100g)**
I(100:0:0)	$10.03^{a} \pm 0.55$	$14.82^{f} \pm 0.20$	$1.30^{d} \pm 0.10$	$5.37^{a} \pm 0.06$	$3.68^{d} \pm 0.02$	$64.80^{b} \pm 0.34$	$366.77^{a} \pm 2.19$
II(70:30:0)	$8.37^{c} \pm 0.15$	$17.88^{e} \pm 0.08$	$3.17^{c} \pm 0.29$	$2.70^{e} \pm 0.10$	$3.33^{e} \pm 0.03$	$65.72^{a} \pm 0.28$	$349.72^{c} \pm 0.72$
III(65:30:5)	$7.40^{d} \pm 0.10$	$18.73^{d} \pm 0.03$	$3.91^{b} \pm 0.08$	$3.58^{d} \pm 0.08$	$3.66^{d} \pm 0.05$	$62.73^{c} \pm 0.20$	$358.09^{b} \pm 0.23$
IV(60:30:10)	$8.9^{b} \pm 0.36$	$19.30^{c} \pm 0.27$	$3.80^{b} \pm 0.00$	$5.27^{a} \pm 0.21$	$4.00^{c} \pm 0.20$	$58.73^{d} \pm 0.26$	$359.53^{b} \pm 1.72$
V(55:30:15)	$9.23^{b} \pm 0.21$	$20.23^{b} \pm 0.25$	$4.52^{a} \pm 0.07$	$4.37^{c} \pm 0.15$	$4.68^{b} \pm 0.02$	$56.98^{e} \pm 0.50$	$348.17^{c} \pm 0.64$
VI(50:30:20)	$6.67^{e} \pm 0.15$	$21.67^{a} \pm 0.08$	$4.60^{a} \pm 0.00$	$5.00^{b} \pm 0.20$	$4.97^{a} \pm 0.15$	$57.09^{e} \pm 0.28$	$360.07^{b} \pm 1.33$

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The least value of protein content (14.82 %) was detected from the 100% wheat sample, which served as a control but compared well with the value (15.83 %) reported by [22] for maize-pigeon pea biscuit. The ash content which ranged from 1.30 to 4.60 % showed significantly ($p < 0.05$) different variations. The ash contents of these extruded snacks were higher than the range (1.72 - 2.04 %) reported by [20] for extruded products from corn, millet and soybean blend, but compared well with the value (3.53 %) reported by [23] for extruded acha/soybean blends and compared favorably with 5.15 % reported by [24] for African breadfruit-soybean-corn snack. The fat content of the extruded snacks was high and ranged from 2.70 to 5.93 %. This could be due to the baking fat added as ingredient before extrusion, which acts as a placiticizer and lubricant in extrusion. The fat contents observed for the extruded snacks were below the value (8.33 %) reported by [23] for extruded acha/soybean blends, while 7.60 % fat was reported by [24] for African breadfruit-soybean-corn snack. However, fat contents of these extruded snacks were close to the range (2.32 - 4.58 %) reported by [20] for extruded products from corn, millet and soybean blend. The fibre content of the extruded snacks ranged from 3.33 to 5.93 %. In general, crude fibre of extruded snacks was low. Foods with low fiber content are very important in maintaining children health, considering their stomach capacity since they have to consume more to get satisfied and meet their daily energy requirement [25]. The crude fibre content of the samples, compared favourably with the value (4.5 %) reported by [24] for African breadfruit-soybean-corn snack. Fibre is promoted in diets due to its tendency to absorb water, acts as "bulking agent", facilitates faster transit of foods in the gastrointestinal tract, reduces the retention time of faeces in the colon, could prevent colon cancer and other bowel disorders by decreasing retention time of faeces in the colon[26]. It could bind bile salts, help in increasing the loss of cholesterol, act as a hypocholesterolemic agent and therefore useful in dietary management of cardiovascular diseases [26]. The carbohydrate contents of the snack samples were generally high, and ranged from 56.98 to 65.72 %. High level of carbohydrate in extruded snacks provides energy to the body for metabolic activities and work. A small amount of vegetable oil from 0.5 to 1 % protects against degradation of carbohydrates and further the process of pre-gelatinization of starch [27]. These values were high compared to the range (54.34 - 58.34 %) reported by [20] for extruded products from corn, millet and soybean blend. This is obviously due to the high accumulation of carbohydrates in cereals. The similar finding of 54.10 % of carbohydrate was also detected by [24] for African breadfruit-soybean-corn snack, while 54.86 % was reported by [22] for maize-pigeon pea biscuit. The energy content of the extruded snacks which ranged from 348.17 to 366.77 kcal/100g, were significantly ($p < 0.05$) different from each other. With regard to the energy value (332.17 kcal/100g) of corn snacks reported by [28], the values found in this study were higher, but conformed to the range (354.02 - 362.65 kcal/100g) reported by [29] for extruded snack products from lentil-corn blends, but lower than the value (390.19 kcal/100g) of recent work [30] for snack produced from the by-products of rice and soybean.

Amaranth leaf flour incorporation significantly ($p < 0.05$) increased the mineral contents (mg/100g) such as calcium (135.17 - 202.00), iron (5.30 - 5.80), zinc (5.51 - 6.81), magnesium (20.37 - 83.36), potassium (166.30 - 207.54) and phosphorus (51.72 - 58.97) of the extrudates (Table 2). The calcium content ranged from 117.50 to 202 mg/100g. The calcium content increased as the level of incorporation of amaranth leaf flour increased.

Table 2: Mineral composition (mg/100g) of the extruded snacks

Extruded Snack Samples	Calcium	Iron	Zinc	Magnesium	Potassium	Phosphorus
I(100:0:0)	$117.50^{f} \pm 0.68$	$3.51^{b} \pm 0.18$	$2.81^{e} \pm 0.22$	$12.56^{f} \pm 0.81$	$154.26^{f} \pm 0.70$	$49.15^{e} \pm 0.18$
II(70:30:0)	$135.17^{e} \pm 0.13$	$5.30^{a} \pm 0.58$	$5.51^{d} \pm 0.42$	$20.37^{e} \pm 0.36$	$166.30^{e} \pm 0.75$	$51.72^{d} \pm 0.49$
III(65:30:5)	$193.12^{d} \pm 0.37$	$5.45^{a} \pm 0.30$	$6.43^{c} \pm 0.98$	$73.52^{d} \pm 0.27$	$177.90^{d} \pm 0.55$	$52.32^{d} \pm 0.54$
IV(60:30:10)	$197.20^{c} \pm 0.48$	$5.58^{a} \pm 0.35$	$6.50^{c} \pm 0.21$	$76.68^{c} \pm 0.89$	$187.16^{c} \pm 0.82$	$54.33^{c} \pm 0.86$
V(55:30:15)	$198.33^{b} \pm 0.27$	$5.71^{a} \pm 0.08$	$6.70^{b} \pm 0.58$	$79.68^{b} \pm 0.71$	$195.73^{b} \pm 0.56$	$55.35^{b} \pm 0.79$
VI(50:30:20)	$202.00^{a} \pm 0.61$	$5.80^{a} \pm 0.11$	$6.81^{a} \pm 0.22$	$83.36^{a} \pm 0.67$	$207.54^{a} \pm 0.93$	$58.97^{a} \pm 0.30$

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The values obtained in this study were higher than the values 51 and 71 mg/100g reported by [31] for weaning food product with or without fishmeal A and B, respectively prepared from fermented maize, rice, soybean and fishmeal blends. The iron content of the extruded snacks which ranged from 3.51 to 5.80 mg/100g were not significantly ($p > 0.05$) different from each other except the control (I). The result revealed increase in iron content with increasing addition of amaranth leaf flour. The values observed in this study were higher than the range (1.20 - 1.94 µg/ml) recorded by [20] for extruded products from corn, millet and soybean blend and compared well with the range (0.5 - 5 mg/100g) reported by [32] for extruded rice snacks. The recent work of [33] revealed comparable levels of iron (4.14, 4.25, 3.79 mg/100g) for fermented popcorn-African locust bean blend; fermented popcorn-bambara groundnut blend and fermented popcorn-African locust-bambara groundnut blend, respectively. The zinc content ranged from 2.81 to 6.81 mg/100g. These values were higher than the values (2.89, 3.16 and 3.83 mg/100g) from recent study conducted by [33] for fermented popcorn-African locust bean blend; fermented popcorn-bambara groundnut blend and fermented popcorn-African locust-bambara groundnut blend, respectively but compared well with the value (4.35 mg/100g) detected by [34] for extruded blends of pigeon pea and unripe plantain flours. The magnesium content showed significant ($p < 0.05$) differences, and ranged from 12.56 to 83.36 mg/100g. The values obtained in this study are in agreement with the value (81.40 mg/100g) of earlier work [23] for extruded acha-soybean blends. The potassium content of the formulated snacks ranged from 154.26 to 207.54 mg/100g. The values were within the range (88 - 233 mg/100g) reported by [32] for extruded rice snacks. The study conducted by [31] showed that high amount of potassium up to 263 and 244 mg/100g for weaning food product with or without fishmeal A and B, respectively can be produced from fermented maize, rice, soybean and fishmeal blend. The phosphorous content ranged from 49.15 to 58.97 mg/100g and fell within the range (42 - 148 mg/100g) reported by [32] for extruded rice snacks and compared also with the values (78.95, 88.90, and 96.90 mg/100g) reported by [33] for fermented popcorn-African locust bean blend; fermented popcorn-bambara groundnut blend and fermented popcorn-African locust-bambara groundnut blend, respectively.

The vitamin composition of the extruded snacks is shown in Table 3. There were significant ($p < 0.05$) differences between the extruded snack samples. Amaranth leaf flour incorporation significantly ($p < 0.05$) increased the vitamin contents such as β-carotene content (200.81- 3157.6 IU), thiamine (0.23 - 0.44 mg/100g), riboflavin (0.85 - 4.46 mg/100g) and vitamin E (0.56 - 0.86 mg/100g) of the extrudates. The β-carotene content ranged from 102.87 to 3157.6 IU. The β-carotene content of other samples increased with increasing levels of amaranth leaf flour addition. The values detected in this study were higher than the range (1.27-4.70 µg/ml) reported by [20] for extruded products from corn, millet and soybean blend but lower thanthe value (5.25 mg) obtained from a study by [35] for fresh leaves and (13.46 mg/100 g) from a study by [36]. Vitamin B_1 (Thiamine) contents of the extruded snacks ranged from 0.21 to 0.44 mg/100g. The thiamine content was higher than the value (0.06 mg/100g) detected by [24] for African breadfruit-soybean-corn snack. Vitamin B_2 (riboflavin) content of the samples revealed significant ($p < 0.05$) differences, and ranged from 0.28 to 4.46 mg/100g. Extruded snack sample with incorporation of different levels of amaranth leaf flour had high riboflavin contents with increasing incorporation levels (5, 10, 15 and 20 %). The values were higher than the level (0.40 mg/100g) detected in the previous study [34] for extruded blends of pigeon pea and unripe plantain flours

Table 3: Vitamin composition of the extruded snacks

Extruded Snack Samples	β-carotene (IU)	Vitamin B_1 (mg/100g)	Vitamin B_2 (mg/100g)	Vitamin C (mg/100g)	Vitamin E (mg/100g)
I(100:0:0)	$102.87^{f} \pm 0.60$	$0.21^{e} \pm 0.04$	$0.28^{f} \pm 0.01$	$8.21^{b} \pm 0.02$	$0.29^{d} \pm 0.00$
II(70:30:0)	$200.81^{e} \pm 0.05$	$0.23^{e} \pm 0.01$	$0.85^{e} \pm 0.03$	$8.61^{a} \pm 0.31$	$0.56^{c} \pm 0.00$
III(65:30:5)	$2861.9^{d} \pm 0.03$	$0.30^{d} \pm 0.05$	$2.92^{d} \pm 0.05$	$7.48^{d} \pm 0.08$	$0.75^{b} \pm 0.03$
IV(60:30:10)	$2940.6^{c} \pm 0.18$	$0.34^{c} \pm 0.02$	$3.81^{c} \pm 0.02$	$7.91^{c} \pm 0.04$	$0.83^{a} \pm 0.01$
V(55:30:15)	$3078.7^{b} \pm 0.26$	$0.39^{b} \pm 0.00$	$4.19^{b} \pm 0.11$	$8.05^{bc} \pm 0.01$	$0.83^{a} \pm 0.01$
VI(50:30:20)	$3157.6^{a} \pm 0.36$	$0.44^{a} \pm 0.03$	$4.46^{a} \pm 0.04$	$8.02^{bc} \pm 0.11$	$0.86^{a} \pm 0.04$

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The vitamin C content of the samples ranged from 7.48 to 8.61 mg/100g, with the highest value observed in sample III produced from yellow maize and soybean, followed by the control (sample I). The amount of vitamin C decreased in extruded snacks with incorporation of amaranth leaf flour. The vitamin C content detected in this study was higher than the value of vitamin C (1.40 mg/100g) reported by [24] for African breadfruit-soybean-corn snack. This high value of vitamin C could be attributed to the materials used to produce these snacks. The vitamin E content of the extruded snacks ranged from 0.29 to 0.86 mg/100g. The lowest value was observed in control (sample I), while the highest value was observed in the snack sample which had high level of incorporation of amaranth leaf flour. The content of vitamin E increased as the levels of amaranth leaf flour was introduced. The high amount of vitamin E (2.60 mg/100g) was reported by [24] for African breadfruit-soybean-corn snack.

The result of evaluation of the anti-nutrients in the extruded snack samples is presented in Table 4. It revealed significant ($p < 0.05$) differences amongst the extruded snack samples due to formulations, extrusion cooking and baking treatments. Extrusion cooking has been shown to destroy anti-nutritional factors, especially trypsin inhibitors, haemagglutinins, tannins and phytates, all of which inhibit protein digestibility [37]. The values obtained for the anti-nutritional factors of the extruded snack samples were generally low. These low values may be attributed to extrusion cooking and baking which are known to reduce anti-nutrition factors. This indicates that the snacks could be utilized effectively since the anti-nutritional composition has been reduced and there would be no interference with the nutrient like protein and minerals in the food samples. Fermentation and other processing methods are known to improve the nutritional quality of legumes and cereals by causing significant changes in their chemical composition and elimination of anti-nutritional factors [38].

Table 4: Anti-nutritional factors of the extruded snacks

Extruded Snack Samples	Phytate (mg/100g)	Oxalate (%)	Saponin (mg/100g)	Lectin (HUI/mg)	Trypsin inhibitor (TIU/mg)
I(100:0:0)	$0.55^{b} \pm 0.31$	$0.04^{b} \pm 0.05$	$1.41^{e} \pm 0.04$	$0.03^{b} \pm 0.00$	ND
II(70:30:0)	$0.79^{b} \pm 0.04$	$0.05^{ab} \pm 0.04$	$1.16^{e} \pm 0.03$	$0.20^{ab} \pm 0.17$	$0.06^{a} \pm 0.06$
III(65:30:5)	$1.17^{a} \pm 0.06$	$0.07^{ab} \pm 0.04$	$3.73^{d} \pm 0.14$	$0.15^{ab} \pm 0.12$	$0.11^{a} \pm 0.11$
IV(60:30:10)	$1.24^{a} \pm 0.27$	$0.08^{ab} \pm 0.03$	$4.57^{c} \pm 0.09$	$0.22^{ab} \pm 0.06$	$0.21^{a} \pm 0.27$
V(55:30:15)	$1.33^{a} \pm 0.08$	$0.09^{ab} \pm 0.01$	$5.49^{b} \pm 0.37$	$0.20^{ab} \pm 0.01$	$0.24^{a} \pm 0.31$
VI(50:30:20)	$1.48^{a} \pm 0.08$	$0.10^{a} \pm 0.01$	$6.57^{a} \pm 0.02$	$0.24^{a} \pm 0.23$	$0.29^{a} \pm 0.38$

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The phytate content of the extruded snack samples ranged from 0.50 to 1.48 mg/100g. There was no significant ($p > 0.05$) difference between the extruded snack sample I, and the extruded snack sample II. The values were low in these snacks when compared with the levels detected in the extruded snack samples with amaranth leaf flour incorporation. The extruded snack samples III, IV, V, and VI showed no significant ($p > 0.05$) differences between them. The amount of phytate in these snacks increased with increasing amaranth leaf flour incorporation. However, these values were lower than the values (19.25, 18.01 and 15.21 mg/100g) detected by [33] for fermented popcorn-African locust bean blend; fermented popcorn-bambara groundnut blend; and fermented popcorn-African locust-bambara groundnut blend, respectively. The phytate content of other extruded snack samples were much lower than the value (0.28 %) reported by [34] for extruded product from pigeon pea and unripe plantain blends. In general, the values obtained in this study were below 1–3 % phytic acid reported for soybean and soybean meals [39]. The oxalate content of the extruded snack samples were generally low and ranged from 0.04 to 0.10 %. The The extruded snack sample I which served as control, had the lowest value. The other extruded snack samples revealed no significant ($p > 0.05$) differences between them. The oxalate content of the extruded snack samples incorporated amaranth leaf flour increased with increasing levels of incorporation. The values were lower than the value (1.07 %) reported by [23] for extruded acha-soybean blends but also compared well with the value (0.07 %) found in recent study by [40] for complementary food gruels from ginger modified starch and soybean flour blends. Oxalates like phytates limit the availability of calcium in the body (being calcium binders) by forming insoluble calcium oxalates, hence decreasing the utilization of the mineral (calcium) by the bones and tissues [41]. The saponin content ranged from 1.41 and 6.57 mg/100g. There were no significant ($p > 0.05$) differences between the extruded snack sample I and snack II in the saponin content of the extruded snack samples, which were lower than those observed in the extruded snack samples with amaranth leaf flour incorporation. Therefore, incorporation of amaranth leaf flour into these extruded snack samples increased their level of saponin content. The values were very low compared to the value (0.09 %) observed by [34] for extruded product from pigeon pea and unripe plantain blends. The lectin content of the extruded snack samples which ranged from 0.03 to 0.24 HUI/mg were significantly ($p < 0.05$) different from each other. The extruded snack sample I which had wheat flour only, had the lowest value, followed by the extruded snack sample III. The highest values were observed in the extruded snack samples with high level of amaranth leaf flour incorporation. Extrusion has been shown to be very effective in reducing or eliminating lectin activity in legume flour [42]. Thus, extrusion cooking is more effective in reducing or eliminating lectin activity compared with the traditional aqueous heat treatment. The trypsin inhibitor content which reduced with formulations, ranged from 00.00 to 0.29 TIU/mg. Trypsin inhibitor was not detected in the extruded snack sample I containing wheat flour. Other extruded snack samples showed an increase in trypsin inhibitor as the level of incorporation of amaranth leaf flour increased.

The functional properties of the snack samples were comparable to that of the control snack sample which was prepared from wheat flour only. The result of the functional properties of the extruded snack samples indicated significant ($p < 0.05$) differences in some tested parameters. Functional properties of food materials are very important for the appropriateness of diet, particularly, for the growing children [43]. The bulk density of the extruded snack which is of importance in packaging [44], ranged from 0.72 to 0.86 g/ml. The highest value was observed in the extruded snack sample VI, while snack sample I had the lowest value. Increasing of bulk density may be due to increase of both sugars and dietary fibers as the starch content is reduced by replacement [45]. Also, [46] reported that the density of extrudates increased as a result of liquification of sugar via melting during extrusion cooking. The results compared well with the range (0.74 - 0.94 g/ml) reported by [47] for extrudates from white yam and bambara nut blends.

Table 5: Functional properties and pH of the extruded snacks

Extruded Snack Samples	**Bulk density (g/ml)**	**Water absorption capacity (ml/g)**	**Oil absorption capacity (ml/g)**	**pH**	**Expansion ratio**
I(100:0:0)	$0.72^{c} \pm 0.01$	$2.35^{a} \pm 0.15$	$1.25^{ab} \pm 0.12$	$6.3^{cd} \pm 0.11$	$9.0^{a} \pm 0.48$
II(70:30:0)	$0.76^{bc} \pm 0.00$	$1.97^{b} \pm 0.12$	$1.10^{b} \pm 0.04$	$6.1^{d} \pm 0.00$	$7.3^{b} \pm 0.15$
III(65:30:5)	$0.83^{ab} \pm 0.06$	$2.20^{a} \pm 0.10$	$1.08^{b} \pm 0.02$	$6.5^{c} \pm 0.23$	$7.5^{b} \pm 0.12$
IV(60:30:10)	$0.84^{a} \pm 0.05$	$1.87^{b} \pm 0.06$	$1.50^{a} \pm 0.27$	$6.8^{b} \pm 0.10$	$7.3^{b} \pm 0.57$
V(55:30:15)	$0.84^{a} \pm 0.04$	$1.88^{b} \pm 0.13$	$1.20^{b} \pm 0.20$	$6.9^{b} \pm 0.06$	$6.0^{c} \pm 0.00$
VI(50:30:20)	$0.86^{a} \pm 0.03$	$1.91^{b} \pm 0.09$	$1.23^{ab} \pm 0.06$	$7.5^{a} \pm 0.00$	$7.3^{b} \pm 0.35$

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The water absorption capacity which is an index of the maximum amount of water that a food product would absorb and retain [48] ranged from 1.88 to 2.35 ml/g. With respect to water absorption capacity, [49] reported that the microbial activities of food products with low water absorption capacity are reduced significantly. Hence, the shelf-life of such product would be extended. There were no significant ($p > 0.05$) differences between the extruded snack samples I and III. The highest values were detected in the extruded snack samples I and III, while the lowest value was observed in the extruded snack sample IV. There were no significant ($p > 0.05$) differences between the extruded snack sample II, IV, V, and VI, implying that the extruded snack samples with low values, will have reduced microbial load and hence increased shelf-life, if properly packaged. These values were lower than the range (3.8 -4.7 g/g) from the study conducted by [50] for bean-corn extrudates. The oil absorption capacity ranged from 1.08 to 1.50 ml/g. The highest value was observed in the extruded snack sample IV, followed by the extruded snack sample I, while the lowest value was observed in the extruded snack sample III. The oil absorption capacity denotes how much oil is bound to matrices in a food system, which could be used as the index of hydro-phobicity of the food [51]. The pH of the extruded snack samples ranged from 6.1 to 7.5. This result compared well with the healthy body's pH range (6.1 to 8.0), qualifying the snacks as alkaline foods which are good for health [52]. The lowest values were observed in the extruded snack samples I and II, produced from wheat flour only and yellow maize-soybean flour blend, respectively. The highest value was that of the extruded snack sample VI containing high level of amaranth leaf flour incorporation. The increase of pH in the extrudates could be attributed to amaranth leaf flour incorporation which is alkaline food in their nature. The recent study by [53] mentioned that pulses and all cereals (wheat, maize, rice etc.,) are acidic foods while all green, leafy and root vegetables except peas and beans are alkaline foods. Expansion index is an important characteristic of the extruded snacks; which describes the degree of puffing undergone by the sample as it exits the die of the extruder [54]. The expansion ratio of the extruded snacks which ranged from 6.0 to 9.0 were not significantly ($p > 0.05$) different from each other except the extruded snack samples I and V. The lowest value was observed in the extruded snack sample V, while sample I had the highest value, which may be due to the fact that, extruded wheat dough has high starch content. According to [55], the maximum expansion index is closely related to the starch content of the dough, because maximum expansion values were obtained with pure starches. However, expansion ratio of the extruded snack samples, were superior to those (3.5 to 5.7) reported by [56] for extruded biscuits of sour cassava starch powder with fibers and also compared well with the result of previous study by [57] who reported a range of 2.75 to 6.80 for extruded acha/soybean blends.

The microbial load of the extruded snacks is shown in Table 6. The microbial load of any food material is a useful index of quality, revealing the potential safety status especially for read-to-eat extruded food products. The result of microbialogical evaluation of the extruded snack samples revealed low levels of bacteria, mould and coliform growth. The bacteria count ranged from 1.08 to 9.4×10 cfu/g. The mould count ranged from 0.00 to 3×10 cfu/g, while coliform was not detected in all the extruded snack samples. The microbial analysis of extruded snacks was conducted after seven days of production, which may have influenced the presence of microorganisms, due to contamination of the samples after production. The maximum permissible level of total aerobic colony of ready-to-eat foods recommended by Fylde Borough Council [58] was 10^4 to less than 10^6 cfu/g for ready-to-eat food products. However, the obtained levels were within acceptable limits. The level of bacteria count detected in this study were lower than the level (4.5×10^5 cfu/g) detected from the study by [22] for biscuit from a blend of maize and pigeon pea flour. In addition, the level of mould counts in the extruded snack samples were very low compared to the levels (2×10^2 cfu/ml and 1.5×10^2 cfu/ml of mould) reported by [31] for weaning food product with or without fishmeal A and B, respectively prepared from fermented maize, rice, soybean and fishmeal blends. Total viable counts have been reported to be a useful monitor in processing of food products and may therefore reflect poor handling or storage at retail level [59]. Bacillus cereus and coliforms have however been identified as the major organisms causing spoilage in extruded food products and the total plate count must also be very low in order to rule out the possibility of formation of preformed toxins in the extruded food products [58].

Table 6: Microbial profile of the extruded snacks

Extruded Snack Samples	Bacteria count (cfu/g)	Mould count (cfu/g)	Coliform count (cfu/g)
I(100:0:0)	9.4 × 10	3.0 × 10	ND
II(70:30:0)	1.3 × 10	ND	ND
III(65:30:5)	1.18 × 10	ND	ND
IV(60:30:10)	1.08 × 10	ND	ND
V(55:30:15)	8.2 × 10	1.0 × 10	ND
VI(50:30:20)	5.8 × 10	1.0 × 10	ND

Values are mean ± standard deviation of triplicate determination. Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

ND= No Detected

The sensory evaluation of the extrudates is shown in Table 7. The result shows that there was significant ($P < 0.05$) difference in all the sensory attributes, which may be due to formulation and consequently, composition of each extrudate. Addition of 15 % amaranth leaf flour lowered the scores for all the samples in respect to taste, aftertaste, mouthfeel, aroma, texture, crust colour, crumb colour, and overall acceptability in comparison to formulations I, II, III, and IV. Incorporation of amaranth leaf flour at 20 % levels further lowered the scores of all the attributes of the product. The taste, aftertaste, mouthfeel, and crust colour had the lowest scores (< 5) due to incorporation of amaranth leaf flour at 20 % level (formulation VI).

Table 7: Sensory profile of the extruded snacks from wheat, yellow maize, soybean, Carrot and amaranth leaf flour blends

Extrudate Samples	Attributes							
	Taste	Aftertaste	Mouth feel	Aroma	Texture	Crust colour	Crumb colour	Overall acceptability
I(100:0:0)	$8.04^{a} \pm 0.44$	$7.65^{a} \pm 0.85$	$7.35^{a} \pm 0.88$	$7.5^{a} \pm 0.62$	$7.87^{a} \pm 0.65$	$8.02^{a} \pm 0.50$	$8.09^{a} \pm 0.33$	$8.06^{a} \pm 0.46$
II(70:30:0)	$6.79^{b} \pm 0.74$	$6.3^{b} \pm 1.14$	$6.17^{b} \pm 0.95$	$6.59^{b} \pm 0.68$	$6.08^{cd} \pm 0.85$	$7.09^{b} \pm 1.04$	$6.92^{b} \pm 0.88$	$6.98^{b} \pm 0.59$
III(65:30:5)	$6.56^{bc} \pm 1.01$	$6.08^{b} \pm 1.24$	$6.12^{b} \pm 1.02$	$6.86^{ab} \pm 0.67$	$6.98^{b} \pm 1.10$	$6.95^{b} \pm 1.36$	$6.32^{bc} \pm 1.16$	$6.76^{bc} \pm 0.56$
IV(60:30:10)	$6.03^{cd} \pm 0.80$	$5.65^{b} \pm 0.75$	$5.57^{b} \pm 0.89$	$6.45^{b} \pm 0.80$	$6.71^{bc} \pm 0.83$	$5.73^{c} \pm 1.08$	$6.01^{c} \pm 1.03$	$6.39^{c} \pm 0.50$
V(55:30:15)	$5.49^{d} \pm 0.70$	$5.55^{b} \pm 0.75$	$5.42^{b} \pm 0.71$	$5.74^{c} \pm 0.64$	$5.80^{d} \pm 0.90$	$5.48^{c} \pm 1.00$	$5.06^{d} \pm 0.51$	$5.73^{d} \pm 0.63$
VI(50:30:20)	$4.79^{e} \pm 0.47$	$4.52^{c} \pm 0.47$	$4.42^{c} \pm 0.50$	$5.08^{c} \pm 0.97$	$5.50^{d} \pm 1.08$	$5.10^{c} \pm 0.74$	$4.91^{d} \pm 0.74$	$5.15^{e} \pm 0.82$

Values are mean ± standard deviation (n=10). Values followed by different superscripts across columns are significantly ($p < 0.05$) different. Samples=I (Wheat flour only), II (Yellow maize: Soybean flour), III (Yellow maize: Soybean: Amaranth leaf flour(5 %), IV (Yellow maize: Soybean: Amaranth leaf flour (10 %), V (Yellow maize: Soybean: Amaranth leaf flour (15 %), and VI (Yellow maize: Soybean: Amaranth leaf flour (20 %).

The taste, aftertaste, mouth feel, aroma, texture, crust colour, crumb colour, and overall acceptability were scored high for formulation I, which served as a control (wheat based) followed by formulation II. Sensory evaluation did not reveal significant ($p > 0.05$) differences between formulations II and III in terms of taste, aftertaste, mouth feel, aroma, and crust colour, crumb colour and overall acceptability. On the other hand formulations III and IV were not significantly ($p > 0.05$) different in terms of taste, aftertaste, mouth feel, aroma, texture, crumb colour, and overall acceptability. However, scores for formulations III, IV and V were higher due to lighter colour of the snacks, compared to formulation VI which had deep green colour, due to high level of incorporation of amaranth leaf flour.

IV. Conclusion

This study produced and evaluated extrudates that could provide protein-energy and micronutrient requirements for consumers. Extrusion did not deleteriously affect these nutrients. However, extrusion cooking and drying treatments depleted most of the anti-nutritional factors evaluated to acceptable levels. Amaranth leaf flour incorporation improved minerals and vitamins in the extrudates. However, higher level of incorporation above 10 % did not produce acceptable products. Extrusion of blends of yellow maize, soybean and amaranth leaf flour produced ready-to-eat snacks adequate in macro- and micronutrients and safe in its composition in terms of anti-nutritional factors and microbial load, for consumption by both children and adults.

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