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Proximate Composition, Amino acid and Sensory profiles of bread made from wheat, monkey kola and differently-processed cowpea

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Abstract:
This work investigated the proximate composition, amino acid (AA) and sensory profiles of bread loaves made from blends of wheat, monkey kola and cowpea flours. Monkey kola pulps were extracted from fruit pods and processed into flour. Cowpea seeds were cleaned and separately processed (sprouted, soaked and blanched), dehulled, oven-dried, dry milled and finely sieved into flour. Composite flour samples were made by partially substituting wheat with inclusion levels of 10% monkey kola flour and 20% processed cowpea flour respectively. The composite flours were mixed with other ingredients and used to produce bread. The bread loaves were subjected to amino acid, proximate and sensory analyses. Proximate analysis revealed thus: Protein (8.66 to 14.74%), fat (19.48 to 20.83%), ash (1.15 to 2.03%), crude fibre (0.79 to 2.31%), moisture (21.39 to 27.05%), and carbohydrate (33.94 to 44.36%). Eighteen (18) amino acids were detected amongst the composite flour bread samples. The nine essential amino acids were present at a range of 30.52g/100g protein to 33.07g/100g protein; the total AAs ranged from 72.34g/100g protein to 77.04g/100g protein. Sprouting increased the values of most of the amino acids. The 70:10:20 wheat-monkey kola-sprouted cowpea bread was most accepted by the sensory panel, whilst also having the highest amounts of protein, ash, crude fibre, moisture and a remarkable AA profile amongst the bread samples. The presence of cowpea in the blend affected consumers' responses due to the beany flavour. Potentials exist for enhancing bread quality and variety via the inclusion of monkey kola and cowpea flours.

Keywords: Bread, Amino acid analysis, Proximate analysis, Cowpea, Monkey kola.

I. INTRODUCTION

Bread is one of the oldest staple foodstuffs, which is made and eaten in most countries around the world. It may be described as a fermented confectionary product produced mainly from wheat flour, water, yeast, and salt by a series of processes involving mixing, kneading, proofing, shaping and baking. Bread-making is fundamentally a temperature-dependent procedure involving a two-step progression, consisting of fermentation, in which Carbon IV oxide (CO2) production linked with yeast activity is manifested in dough structure with the development of dough volume during baking where yeast activity is ended and the bread structure is finalized (Dewettink et al., 2008).

Wheat flour has been the major ingredient used in the production of pastry products. In Nigeria, reliance on wheat flour in the pastry and bakery industries has over the years restricted the use of other cereals and tuber crops available to domestic use. In recent years, government has through intensive collaboration with research institutes encouraged the use of composite flours in the production of bread and related food products such as biscuit. This initiative has enhanced the use of flours from cassava, sweet potato bread fruit, plantains and other underutilized crops that are good sources of flour. The adoption of these locally produced flours in the bakery industry will increase the utilization of indigenous crops cultivated in Nigeria and also lower the cost of bakery products (Oyeyinka et al., 2014).

Cowpea, which is also addressed as black-eyed pea or southern pea, is an important grain legume in the tropics and is a highly beneficial constituent of the local cropping systems in the semi-dry and tropical regions (Lum et al., 2018). Cowpea is a critical source of income in the developing countries of the tropics, which is germane to the survival of millions of indigent segments of the population (FAO, 2002). Agbogidi and Egho (2012) reported that all sections of the plant utilized as food possess nutrient density, and supply proteins and vitamins; as juvenile leaves, green-coloured pods and greenish seeds are ingested as vegetables, while dry seeds are employed in the preparation of multiple snacks and primary meals/cuisines.

Cowpea is a major essential diet in African and Asian Continents (Awe, 2008). It plays a significant role in the livelihoods of countless millions of persons in the African continent and many other regions of the developing world, where it serves as the primary source of dietary protein that equilibrates the nutritional capacities of the staple low-protein tubers and cereal crops (Singh, 2002). In addition, Cowpea is a major source of minerals, vitamins and dietary fibre in the daily diets in Africa, and thus, it positively affects the health of men, women and children (Tariku, 2018). Calcium and iron contents of cowpea are greater than those of fish, meat and eggs; and their iron contents are equal to that of dairy milk. The magnitudes of the water-soluble vitamins (especially thiamin, riboflavin and niacin) present in cowpea compare favourably with those inherent in fish and lean meat (Agbogidi, 2010).

With the increased advocacy on the consumption of functional foods by world nutrition bodies due to different health

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problems related to food consumption such as celiac disease, diabetes and coronary heart disease, the recent WHO recommendations to reduce adverse trends in nutrition is the consumption of low carbohydrate diets, including slowly-digested food products as well as increased intake of functional foods (Apotiola and Fashakin, 2013).

Monkey kola is a common name given to a number of minor relatives of the Cola spp. that produce edible tasty fruits. The pod of the yellow variety is roundish, while the white variety has more cylindrical shape. Monkey kola is identified by various local names in Southern Nigeria ("achicha" or "Ohirichia" in Igbo and "ndiyah" in Efik). Record shows that the yellow variety pulp is a good source of crude protein, crude fiber, crude fat, Ca, Mg, Zn, Cu, β-carotene and niacin while, C. lepidota (the white variety) pulp is a good source of ash, starch, carbohydrate, K, P and Se contents (Okudu et al., 2016).

Fibre (roughage) is an indigestible part of plants mostly made up of cell walls of plants. During the process of food digestion, it absorbs water and food residues, speeding up the passage of food through the gut and producing bulk in the stools. A fibre-rich diet is ideal for people seeking weight reduction and patients with diverticulitis and constipation (Okaka, 2010).

Composite flour has been defined as a mixture of several flours obtained from roots and tubers, cereals, legumes – with or without the addition of wheat flour; that is created to satisfy specific functional characteristics and nutrient composition. Flour confectionery preparations are exorbitant baking operations in the food industry, premised on the costly nature and insufficiency of their constituents which are predominantly imported in Nigeria (Ohaegbulam et al., 2021).

Processing of plant foods via sprouting, fermentation, soaking, blanching, amongst other methods, is essential for the improvement of digestibility, increasing the bioavailability of nutrients, modifying the nutritional value, enhancing sensory properties, eliminating antinutrients, amongst others (Okaka et al., 2011)

Conventional bread loaves are low in micronutrients, fibre and protein but high in calories. The physiological value of plain bread, as a source of essential nutrients is small. They contain large amounts of fat (from 5 to 35%), carbohydrates (47 to 100%), the main part of which is sucrose (39.6 to 100%), starch (34.7 to 66%) and small amounts of protein (from 3.2 to 10.4%). Excessive consumption of these products interferes with the balance of the diet in terms of nutrients as well as energy value. Children accustomed to such diets may no longer seek the meals that are beneficial to them. A significant drawback of the consumption of plain bread is the virtual absence of important biologically-active substances, like proteins, vitamins, carotenoids, macro- and microelements. In Nigeria, monkey kola is mostly consumed fresh and like most fruits and vegetables, it has a very short life span probably due to its high moisture content. In addition, its hard texture limits its consumption particularly among the vulnerable group (young children, the aged) due to poor dentition. There is therefore a need to process monkey kola fruit into a more stable, widely-accepted and easy-to-use form (Bread). Cowpea's beany flavour and flatulence-inducing capabilities, as well as its less-regarded social status amongst consumers, makes it less-exploited, in spite of its immense potentials – especially its reported nutritional benefits. This study would be of importance, in that it will strongly encourage the reduction in the underutilization of these local food items (cowpea and monkey kola), thereby enhancing their consumption, diversification and optimization. The use of composite blends of wheat flour, cowpea and monkey kola may result in the production of bread loaves that are less expensive, acceptable and highly nutritive than those produced from wheat only. This research work would also serve economic benefits at the industrial level by adding value to an item (bread) which has commercial viability, to the advantage of the local cultivators and processors of the indigenous food materials – cowpea and monkey kola. This study is aimed at evaluating the proximate composition, amino acid profile and sensory characteristics of wheat-monkey kola-cowpea seed composite flour bread loaves. This will be achieved by:

i. processing the cowpea seeds and monkey kola pulp into flour.
ii. producing bread loaves from wheat, cowpea seeds and monkey kola pulp flour mix in varying proportions.
iii. evaluating the proximate composition of the bread samples.
iv. analysing the amino acid profiles of the bread loaves.
v. evaluating the sensory properties of the bread samples.

Bread loaves are sweetened, baked products, they are broadly characterized by dough properties and then further classified by the process used to form and place them on the oven band. Although, bread loaves vary in their shape, sizes and composition, the three main ingredients are always flour, sugar and fat (butter or vegetable shortenings). Commercial bread normally constitutes 50% calories from fat and carbohydrates, with over 400 calories per 100g in plain bread. The four main processes employed to make bread are mixing, cutting, baking and packaging. Bread needs precise preparation to be a successful product. The ingredients are mixed to form a dough using mixers that are either operated manually or using a pre-set mixing programme. As the dough is mixed, the protein molecules form long strands of gluten resulting in an elastic web which, essentially, controls the quality of wheat flour-based products. Once the dough is mixed, it is then made into different shape and sizes. This process leads to an increase in the stress on the gluten structure (Klunklin and Savage, 2018).

Cowpea (Vigna unguiculata L. Walp or Vigna sinensis), predominantly called "beans" in Nigeria, is a major grain pulse in Nigeria, the western part of the African continent, other tropical countries as well as the United States of America (Enwere, 1998). It belongs to the family Papilionadeae and it is an ancient Africa domesticate. Cowpea is amongst the most commonly-grown cultivars of Vigna from

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the 25 - 26 cultivated pulses isolated from 600 - 700 clans (genera) and more than 18,000 species of the Leguminoseae family of the flowering plants. According to Timko et al. (2007), cowpea is affirmed to have originated from West Africa by some workers, because both undomesticated and cultivated species proliferated in the region. Cowpea (Vigna unguicalata (L) Walp) is a principal food legume in underdeveloped countries, majorly in sub-Saharan part of Africa, Central and South America, and in Asia (Ngompe-Deffo et al., 2017). It is extensively cultivated in 16 countries in Africa, with Africa generating over 66% of the total of the globe's supply (Agyeman et al., 2014). Cowpea is the most extensively grown, traded, distributed and consumed legume because the nutritional and health value to man and livestock is of importance (Agbogidi and Egho, 2012). Cowpea is a major essential diet in Africa and Asian continents (Awe, 2008). The seeds contribute immensely to the several urban and rural families' overall intake of protein and hence Agbogidi (2010) refers to cowpea as being the major source of protein for the common man.

According to Saka et al. (2018), cowpea is an indigenous Africa grain legume rated as one of the most economically important crop and a veritable source of plant protein. It is therefore considered crucial for reduction of malnutrition among children and resource poor rural households. Cowpea has been regarded as "poor man's meat" owing to its high protein content (20-25%) as posited by Ngompe-Deffo et al. (2017). According to Makoi (2019), the protein and carbohydrate content of cowpea is high and its amino acid pattern which compliments that of a cereal grains, makes it important nutritionally in the diet of humans. Cowpea is a dependable crop in semi-arid regions used by resource poor farmers and it is highly consumed virtually in so many parts of the world due to its protein content which is of high quality. Cowpea has in recent times got a whole lot of attention from different stakeholders globally because of its publicized health properties which is of benefit, and which includes its anti-cancer, anti-hyperlipidemic, anti-diabetic, anti-hypertensive and anti-inflammatory properties (Makoi, 2019).

Cowpea seeds, according to Davies and Zibokere (2011), contain an average of 23.4% protein, 11% moisture, 3.6% ash, 1.3% fat and 56.8% carbohydrate. Agbogidi and Egho (2012) reported that the magnitudes of the water-soluble vitamins (especially thiamin, riboflavin and niacin) present in cowpea compare favourably with those inherent in fish and lean meat. Many researchers have made known that when 100-135g of dry beans is consumed on a daily basis, there is a reduction in serum cholesterol level by 20%, and this lessens to 40%, the risk of having coronary heart diseases.

Research is discovering new ways to combat the factors that contribute to the limitation of cowpea seeds use. The long preparation time is a factor, it is highly inconvenient and causes much fuel to be exhausted or used during its cooking. During post–harvest storage of the cowpea seeds, many changes occur and this can lead to seed-hardening, moisture absorption, mould growth, discolouration of the seeds and development of off-flavour. Certain anti-nutrients including lectins and protease inhibitors can obstruct the digestion process. Another effect which is undesirable is the factor which encourages flatulence (CIAT, 2013). The nutritional and industrial potential of cowpeas is unknown in the communities where they are grown in Nigeria (Audu *et al.*, 2013).

Cola parchycarpa K. Schum (Family *Malvaceae)* is a perennial tree commonly described as monkey kola. Monkey kola is a popular nomenclature for the lesser known members of the *Cola* species that yield edible tasty fruits. They are a close relative to the familiar West Africa kola nuts (*C. nitida and C. acuminata*), cultivated for their masticatory and stimulating nuts. In southern Nigeria and Cameroon, the fruit pulp is eaten by humans as well as some wild primate animals especially monkeys, baboons and other species (Essien and Udousor, 2017). The mature pulp is crisp and sweet with moderate calorie of 65 calories/100grams fruit pulp. This makes its consumption ideal for all age groups though people's demand for this fruit tends to have decreased, compared to other fruits such as garden egg, cucumber, banana etc. (Bob, 2017).

Research has shown that juice and jam can be developed from the pulp of the monkey kola. Reports from other studies have suggested that the husk and white shell of slimy kola nut (*C. vesticillata*) could serve as a blend in animal feed. *In vivo* studies also revealed that about 50% of kola nut husk meal could replace maize diet for rabbits (Essien and Udousoro, 2017).

Monkey kola has nutritional and medicinal values. Record shows that the yellow variety pulp is a good source of crude protein, crude fiber, crude fat, Ca, Mg, Zn, Cu, β-carotene and niacin while *C. lepidota* (the white variety) pulp is a good source of ash, starch, carbohydrate, K, P, and Se contents. Nutrients present in this fruit are vitamins, mineral elements and others which are beneficial to human health in many ways. A concise assessment is as follows:

i) Like some other fruits, Cola parchycarpa contains soluble fiber which contributes to the health of gastrointestinal tract and weight management. The beta-carotene content in yellow monkey kola contributes to a healthy skin and mucous membrane. The body converts the better-carotene in the fruit into vitamin A which helps to keep the eye healthy for better sight.
ii) Riboflavin which is also known as vitamin B2 is present in the fruit and it helps to lower cardiovascular risks and improve cholesterol levels due to the presence of niacin (vitamin B3) known as nicotinic acid.
iii) Intake of Cola parchycarpa moderately can help to reduce the risk of heart disease because it is rich in flavonoids which are metabolites that provide essential benefits to the heart and health in general.
iv) Essential mineral elements have been proven to be present in the vegetable and such that are needed for healthy bones, protein synthesis, proper functioning of the cells, organs, tissues and overall body function and development.

The low caloric effect has made it a better item for a weight loss diet. Nutrition experts have discovered that fruits and vegetables that are high in fiber and low in calories serve best for weight loss (Okudu et al., 2016).

Composite flour is a welcomed development in developing countries to promote high yield of native plant species and better use of domestic agricultural product and thus prevent them from going into extinction (Adeboye et al., 2014). The use of composite flour to produce baked goods, where feasible, helps to lessen total dependence on imported wheat (Hasmadi et al., 2020). Composite flour is desirable in this regard because it improves the nutritional value of food products such as bakery products.

II. METHOD

Whole, mature and ripe monkey kola (Cola lepidota) fruits were sourced from monkey kola trees planted in some compounds in Ezuhu/Amadi Nguru, Nguru Nweke Autonomous Community domiciled in Aboh Mbaise Local Government Area of Imo State Nigeria. Cowpea (Vigna unguiculata) seeds were sourced from Relief Market in Owerri, Imo State. Selected quantities of the cowpea seeds and monkey kola fruits were subsequently taken to the Department of Agricultural Technology, Federal Polytechnic, Nekede, Owerri, Imo State, for identification. The other ingredients used in the production of bread were also sourced from Relief Market in Owerri, Imo State.

The methods of Baryeh (2001), Uneanya and Ohaegbulam (2017), Ohaegbulam et al. (2018) and Uzodinma et al. (2018) were adopted in the production of the Cowpea (Vigna unguiculata) seed flour. The cowpea seeds were cleaned manually to remove all foreign matters such as dust, dirt, stones and chaff, as well as immature and broken seeds. Thereafter, the seeds were separated into three portions of One (1) kilogramme each, subjected to blanching, soaking and sprouting respectively.

SPROUTING METHOD

This fraction of cowpea seeds was steeped for a day, with water discarded from the steeped seeds and changed every eight hours. After steeping, the seeds were spread on a tray and covered with wet muslin cloth and kept in a humid and airy place, with water sprinkled intermittently every two (2) hours. Germination was stopped on the 3rd day by oven drying the malts at a temperature of 60°C for 3hours. The testa, rootlets and minor caulms were removed and the malts were allowed to cool for thirty (30) minutes before milling and sieving to fine flour.

SOAKING METHOD

One kilogramme (1 kg) of cowpea seeds was soaked in 2 litres of potable water at room temperature (28 ± 2oC) for 10 minutes to soften the testa and manually dehulled by rasping between palms. The dehulled beans were oven-dried at 60oC for 24 h with frequent turning for uniform heat distribution, milled and finely sieved through a 0.2mm mesh screen to obtain cowpea flour. The flour samples were kept in high density polythene packaging films until analysis.

BLANCHING METHOD

One kilogramme (1 kg) of cleaned cowpea seeds was blanched with 2 litres of potable water at 100oC for 1 minute. The seeds were manually dehulled by rasping between palms. The dehulled beans were oven-dried at 60oC for 24 h with frequent turning for uniform heat distribution, milled and finely sieved through a 0.2mm mesh screen to obtain cowpea flour. The flour samples were kept in high density polythene packaging films until analysis.

Mature monkey kola fruits were washed with water, peeled and sliced into thin slices of about 5mm thickness using a knife. The pulp was drained and dried in an oven at 55oC for 10 hours with frequent turning. The dried monkey kola slices were milled using an attrition mill. The flour samples were sieved and packaged in low-density polyethylene bags.

An overall flour weight of 250g was adopted for the production of bread loaves (made of composite flour) using the following ingredients: Wheat flour, monkey kola flour, cowpea flour, margarine, sugar, salt, yeast and water.

The modified Straight dough method was used in baking of the bread loaves following the method described by Ohaegbulam et al. (2021). Flour and all other ingredients were weighed accordingly and placed into an electric mixer (Kenwood® brand). All the ingredients were properly mixed within twenty-five (25) minutes to form dough. The dough was kneaded with a kneading machine repeatedly, cut into pieces, rolled into a cylindrical shape, subsequently moulded into shapes and placed into individual previously-greased baking pans. The dough was then allowed to ferment (proof) for 90 min. Proofing was done at 38oC for 45 min in a proofing cabinet. Thereafter, the proofed dough was baked in an oven at 220oC for 45 min. The loaves were de-panned, allowed to cool, packaged and stored in a cool and dry place prior to analyses.

Moisture, protein, fat, ash and crude fibre contents of the samples were determined according to the methods described by AOAC (2005). The carbohydrate was obtained by difference as follows: Carbohydrate= 100 –(%Moisture + %Protein + %Fat + %Ash + % Crude fibre) (1)

The amino acid analysis was carried out using a HPLC amino acid analyzer (Sykam-S7130) according to the method of Nwanekezi et al. (2021) in which hydrolysates of the samples were obtained. The sample (a known weight mentioned in the calculation sheet) was transferred into a hydrolysis tube containing 5ml of 6N HCl and afterwards tightly closed and incubated for 24h at 110°C. After incubation and filtration, the filtrate was evaporated to dryness at 140°C for one hour and diluted with 1ml of 0.12N citrate buffers (pH 2.2) similar to the amino acid standards. Then, the sample hydrolysate (150 µl) was injected in a cation separation column at 130°C. Ninhydrin solution and an eluent buffer (containing solvent A, pH 3.45 and solvent B, pH 10.85) were delivered simultaneously into the high temperature 16m length reactor coil at a flow rate of 0.7ml/min. The buffer/ninhydrin mixture were heated in the reactor for 2 minutes at 130°C and the mixture was detected at wavelengths of 570nm and 440nm on a dual-channel photometer. The amino acid composition was calculated from the areas of standards obtained from the integrator and expressed as percentages of the total protein.

The tryptophan content was determined according to the method described by Maria et al. (2004). The tryptophan in the known sample was hydrolyzed with 4.2M Sodium hydroxide (NaOH). In general terms, the known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer.

DEFATTING SAMPLE

A known weight of the dried sample was weighed into the extraction thimble and the fat was extracted with chloroform/methanol (2:1 mixture) using soxhlet extraction apparatus as described by AOAC (2005). The extraction lasted for 15h.

NITROGEN DETERMINATION

The Nitrogen content was determined using the method adopted during proximate analysis.

HYDROLYSIS OF THE SAMPLE

A known weight of the defatted sample was weighed into a glass ampoule. Ten (10) millilitres of 4.2M NaOH was added and oxygen was expelled by passing nitrogen into the ampoule. The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at 105oC ± 5oC for 4 hours. The ampoule was allowed to cool before breaking it open at the tip and the content was filtered to remove the humins. The filtrate was neutralized to pH 7.0 and evaporated to dryness at 40oC under vacuum in a rotary evaporator. The residue was dissolved with 5ml of borate buffer (pH 9.0) and stored in plastic specimen bottles, which were kept in the freezer.

LOADING OF THE HYDROLYSATE INTO TSM ANALYZER

The amount loaded was 60 microlitres. This was dispensed into the cartridge of the analyzer. The period of analysis lasted for 45 minutes.

METHOD OF CALCULATING AMINO ACID VALUES

An integrator attached to the analyzer calculates the peak area proportional to the concentration of each of the amino acids.

The sensory evaluation of the bread samples was carried out using a twenty-member panel of judges drawn from the Polytechnic community. The procedure was explained to the panelists before commencement. The quality attributes of the bread samples to be assessed were appearance (colour).

III. RESULTS AND DISCUSSION

The results from the proximate composition of flour bread loaves made with wheat, cowpea and monkey kola composite flour samples are shown in Table 1. There were significant differences ($p>0.05$) amongst the samples across all parameters tested. The outcomes revealed that the protein content ranged from 8.66 to 14.74%. The 100% wheat flour bread (control) had the least protein content (8.66%), whilst the sample that had sprouted cowpea flour in its make-up had the highest protein content (14.74%). Several studies have shown that sprouting helps to increase protein content, as sprouts also tend to contain higher levels of essential amino acids, with certain individual amino acids increasing by as much as 30% in value (Petre, 2018). This agrees with the assertion of Devi et al. (2015) that sprouting of legumes enhances the bioavailability and digestibility of nutrients, and therefore plays an important role in human nutrition.

The fat content of the bread samples ranged from 19.48 to 20.83% with the 100% wheat flour (control) having the highest fat content (20.83%) while the sample having blanched cowpea in its blend had the least fat content (19.48%). Blanching may have resulted in the leaching out of fat from the interstitial spaces of the cowpea. The values were generally higher than the values (14.21 – 15.12%) obtained by Ohaegbulam et al. (2021) for wheat-African pear seed flour bread. The values of fat content suggest the samples' predisposition to spoilage via rancidity if not well managed.

The Ash content ranged from 1.15 to 2.03%. The 100% wheat flour (control) had the least ash content (1.15%) while the sample having sprouted cowpea in its constitution had the highest ash content (2.03%). The ash content of a food material could be used as an index of mineral constituents of the food (Ponka et al., 2015). This implies that the presence of the cowpea and monkey kola flours steadily increased the level of mineral matter in the bread loaves (Table 4.1). This indicates that the presence of cowpea and the method of processing it increased the Ash content in the bread samples.

The crude fibre content ranged from 0.79 to 2.31%. The 100% wheat flour (control) had the least crude fibre content (0.79%), while the bread sample with 70% wheat - 20% sprouted cowpea - 10% monkey kola had the highest crude fibre content (2.31%). The fibre content of bread loaves obtained in this study is attributable to the incremental replacement of wheat flour with cowpea seed and monkey kola pulp flours – both of which have been reported to possess high fibre contents. Fiber content of food contributes to bulk and encourages bowel movement, discourages constipation and piles, reduces blood cholesterol levels and helps prevent cancer of the colon (Duru et al., 2019). The fibre content in the bread loaves was influenced by the method in which the cowpea flour was processed. Sprouting uses up the starchy portion of the endosperm and forms shoots and roots – both of which are fibrous in nature (Nwanekezi et al., 2007). Thus, the sprouted cowpea flour had the greatest effect on fibre content of the bread loaves.

The moisture content of the bread samples ranged from 21.39 to 27.05%. The sample with soaked cowpea had the least moisture content (21.39%), whilst the sample with the highest moisture content was that which had sprouted cowpea in its composition (27.05%). This shows that the bread loaves supplemented with sprouted cowpea and monkey kola flours, due to the increase in fibre content, as fibre has a tendency to entrap water (Okaka, 2009), would have the lowest shelf life amongst the samples unless they are protected from absorbing moisture from damp surroundings or the atmosphere (Duru et al., 2019). The different moisture contents may be due to the different processing methods applied. The outcomes indicate

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that the bread loaves cannot be stored for long because of their high moisture content values.

The nitrogen-free extract (NFE) or carbohydrate content ranged from 33.94 to 44.36%. The bread containing sprouted cowpea had the least NFE content (33.94%) and the loaf with blanched cowpea in it had the highest carbohydrate content (44.36%). Blanching pre-gelatinised/pre-hydrolysed the starch in the cowpea, making it more readily available (Eke, 2003), whilst sprouting used up a good portion of the carbohydrates. The different processing methods used for producing the cowpea flour, certainly accounted for the trend.

TABLE 1: PROXIMATE COMPOSITION OF WHEAT-MONKEY KOLA-COWPEA BREAD LOAVES.

	Parameters (%)					
Sample	Protein	Fat	Ash	Crude fiber	Moisture	CHO
AOA	8.66^{d}±0.15	20.83^{a}±0.06	1.15^{d}±0.02	0.79^{d}±0.02	25.54^{b}±0.04	43.03^{b}±0.56
WOW	10.80^{c}±0.03	19.48^{d}±0.04	1.81^{c}±0.05	1.91^{b}±0.07	21.64^{c}±0.05	44.36^{a}±0.34
XOX	12.14^{b}±0.33	20.34^{b}±0.07	1.92^{b}±0.06	1.57^{c}±0.03	21.39^{c}±0.05	42.64^{c}±0.68
YOY	14.74^{a}±0.11	19.93^{c}±0.04	2.03^{a}±0.06	2.31^{a}±0.05	27.05^{a}±0.02	33.94^{d}±0.49
LSD	0.4313	0.2326	0.1066	0.2142	0.5713	0.8476

Values are means of duplicate determinations. Means with identical superscripts in the same column are not significantly different (p>0.05). a>b>c>d. LSD = Least significant difference; CHO = Carbohydrate
Key: AOA = 100% wheat flour bread (Control), WOW = 70:10:20 wheat-monkey kola-blanched cowpea composite flour bread, XOX = 70:10:20 wheat-monkey kola-soaked cowpea composite flour bread, YOY = 70:10:20 wheat-monkey kola-sprouted cowpea composite flour bread.

TABLE 2: AMINO ACID PROFILE OF WHEAT-MONKEY KOLA-COWPEA FLOUR BREAD LOAVES.

	SAMPLES/CONCENTRATION (g/100g)			
AMINO ACID	AOA	WOW	XOX	YOY
Leucine†	7.14	7.62	7.32	7.64
Lysine†	3.34	3.58	3.45	3.71
Isoleucine†	4.45	4.62	4.55	4.78
Phenylalanine†	3.55	3.55	3.64	4.34
Tryptophan†	0.84	0.92	0.89	0.97
Valine†	4.00	4.56	4.21	4.44
Methionine†	1.23	1.31	1.26	1.39
Proline	3.45	3.66	3.45	3.55
Arginine	4.99	5.33	4.99	5.16
Tyrosine	3.44	3.44	3.44	3.61
Histidine†	2.17	2.33	2.27	2.39
Cystine	1.33	1.39	1.20	1.45
Alanine	4.28	4.40	4.02	4.51
Glutamic acid	9.84	9.92	9.96	9.99
Glycine	3.80	4.02	3.61	4.42
Threonine†	3.80	3.22	3.16	3.41
Serine	3.48	3.62	3.51	3.56
Aspartic acid	7.32	7.51	7.41	7.72
TOTAL	**72.45**	**75.00**	**72.34**	**77.04**
∑EAA	*30.52*	*31.71*	*30.75*	*33.07*

Keys: AOA = 100% wheat flour bread (Control), WOW = 70:10:20 wheat-monkey kola-blanched cowpea composite flour bread, XOX = 70:10:20 wheat-monkey kola-soaked cowpea composite flour bread , YOY = 70:10:20 wheat-monkey kola-sprouted cowpea composite flour bread. † = Essential Amino Acid; ∑EAA = Sum total of the essential amino acids

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This Table 2 shows the amino acid profile of Wheat-Cowpea-Monkey kola composite flour bread. A total of eighteen (18) amino acids were detected in the samples. Amino acids are the building blocks of protein (Mudambi and Rajagopal, 2009). All the nine essential amino acids were present in the samples at a range of 30.52g/100g protein to 33.07g/100g protein, whilst the total amino acids present in the samples ranged from 72.34g/100g protein to 77.04g/100g protein – from the Control sample to the bread sample with sprouted cowpea in its make-up, and the bread sample with soaked cowpea flour to the bread sample with sprouted cowpea flour respectively (Table 2). Eke (2003) explained that protein amino acids are the twenty α–L-amino acids and some of their derivatives which serve as the building blocks of protein polypeptides. Eight of these are essential (because the body needs them, cannot synthesise them and must obtain them from the diet. They include isoleucine, leucine, lysine, methionine, valine, phenylalanine, threonine and tryptophan; histidine, the ninth one is needed by infants), while the rest are non-essential (because the body can synthesise them from a utilizable source of nitrogen, usually protein). Manay and Shadaksharaswamy (2008) affirmed that the "quality" or "balance" of a protein depends upon the kinds and amounts of amino acids it contains. A "balanced" or "high-quality" protein contains all the essential amino acids needed for the human body. When the dietary pattern of amino acids differs from the ideal pattern, it results in "amino acid imbalance", leading to depressed growth and impairment of mental capabilities in children. Except for mental impairment, all other defects can be overcome by supplementing the diet with the limiting amino acids (those not contained or least below the minimum level, in a protein). Thus, the value of protein as food depends upon its amino acid composition, especially that of essential amino acids. From this study, there seemed to be a gradual but marked increase in the level of essential amino acids based on the treatment applied (Table 2). Sprouting generally increased the values of most of the amino acids, concurring with the position of Petre (2018) that sprouting helps to increase protein content, as sprouts also tend to contain higher levels of essential amino acids, with certain individual amino acids increasing by as much as 30% in value.

TABLE 3: SENSORY PROFILE OF WHEAT-MONKEY KOLA-COWPEA FLOUR BREAD LOAVES

Sample	Taste	Aroma	Colour	Mouthfeel	General Acceptability
AOA	8.40^{a}±0.40	7.30^{a}±1.16	8.10^{a}±0.60	7.50^{a}±0.90	8.50^{a}±0.23
WOW	5.70^{b}±1.70	4.60^{c}±1.20	5.20^{b}±2.20	5.40^{bc}±1.07	5.30^{c}±1.83
XOX	4.30^{c}±1.34	5.50^{bc}±0.97	5.10^{b}±1.60	4.60^{c}±2.01	5.30^{c}±0.67
YOY	6.50^{b}±1.18	6.10^{b}±1.45	5.40^{b}±2.22	6.20^{ab}±1.40	6.60^{b}±0.97
LSD	1.3909	1.0340	0.8900	1.3087	1.1556

Values are means ± SD of twenty replications. Means with different superscripts along a column are significantly different ($p < 0.05$). a>b>c>d. LSD = Least significant difference.
Keys: AOA = 100% wheat flour bread (Control), WOW = 70:10:20 wheat-monkey kola-blanched cowpea composite flour bread, XOX = 70:10:20 wheat-monkey kola-soaked cowpea composite flour bread, YOY = 70:10:20 wheat-monkey kola-sprouted cowpea composite flour bread.

Table 3 shows the outcome of sensory evaluation of bread produced from wheat-monkey kola-cowpea composite flour. The sensory score for taste varied from 4.3 to 8.5. The sample made with soaked cowpea flour had the least taste score (4.3), whilst the sample having sprouted cowpea flour in its make-up had the highest score amongst the blends (6.5). Sprouting appeared to have imparted desirable taste to the bread loaves as is depicted by the panelists' responses. The score for aroma ranged from 4.6 to 7.3, with the bread sample bearing sprouted cowpea having the highest score among the blends (6.1), indicating favourable preference by the panelists. Sprouting seemed to have reduced the beany aroma which usually deterred consumers. The score for mouthfeel (texture) ranged from 4.6 to 7.5, with the bread sample with soaked cowpea seeds having the least score (4.6). The bread sample containing sprouted cowpea had the greatest value amongst the blends (6.2), indicating favourable reception by the panelists.

The highest score for colour rating was recorded for the bread sample made with 100% wheat flour (8.1) while the lowest rating or value was observed in the samples made from blends of cowpea processed by soaking. There existed significant differences amongst the samples. The score for general acceptability ranged from 5.30 to 8.50. The least values were held by the loaves containing soaked and blanched cowpea flour (5.30). Amongst the blends, the loaf made with sprouted cowpea flour had the highest value. The members of the sensory panel seemed to prefer the bread loaves with sprouted cowpea flour, as this seemed to increase their interest in the bread sample.

IV CONCLUSION AND RECOMMENDATIONS

The production of bread loaves from wheat, monkey kola and differently-processed cowpea composite flours were successfully achieved in this study. The proximate, amino acid composition and sensory properties of the wheat-monkey kola-differently-processed cowpea bread loaves were successfully investigated. The quality assessment of the composite flour bread loaves indicated that the use of wheat, monkey kola and differently-processed cowpea flour blends in the production of bread improved nutritional values (both on proximate and amino acid compositional bases) and sensory qualities of the samples without adversely affecting most baking characteristics. The 70:10:20 blend of wheat-monkey kola-sprouted cowpea flours used to make bread was most accepted by the sensory panel, whilst also having the highest amounts of protein, ash, crude fibre, moisture and a remarkable amino acid profile amongst the bread samples. However, the presence of cowpea in the blend seemed to affect consumers' responses due to the unfamiliar taste and strong beany flavour when compared to the Control sample.

Based on the results of this study, it is recommended that bakeries should exploit and promote the use of sprouted cowpea and monkey kola flours in bread and confectioneries production due to their higher nutritive values.

Further studies should consider varying the levels of monkey kola and differently-processed cowpea flours, beyond the levels in this work, to assess the impact on the quality of the bread loaves produced. Good consumer education and public enlightenment campaigns on the nutritional benefits of incorporating cowpea and monkey kola flours in wheat flour supplementation efforts should be encouraged.

Also, storage trial and tests on packaging considerations should be conducted to determine the optimal shelf life and best packaging material for the composite flour bread loaves.

Further research on the mineral contents and phytochemical properties of wheat-cowpea-monkey kola composite flour bread loaves should be advanced. This will provide deeper understanding of the attributes of the bread loaves.

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