

QUALITY EVALUATION OF SORGHUM - MALTED COWPEA FLOUR BLENDS AND BISCUITS USING ALBINO RATS

ABSTRACT

The study investigated the quality evaluation of sorghum and malted cowpea flour blends for biscuit making. Sorghum -malted cowpea flour blends were used to produce biscuits and the proximate composition, functional, anti-nutrient, sensory and haematological qualities were carried out using standard laboratory procedures. The results of the proximate composition showed an increase in the protein, fat and moisture contents from 18.25 to 30.52%, 0.63 to 2.66% and 18.31 to 20.21%, respectively with decrease in the ash, fiber and carbohydrate contents from 2.21 to 1.82%, 16.10 to 14.85% and 45.23 to 29.78%, respectively. The phytate and tannin levels decreased from 76.42 to 45.12 mg/100 and 0.15 to 0.02 mg/100. The sensory mean scores for taste, aroma, color, texture and general acceptability ranged from 6.53 to 6.60, 3.93 to 4.06, 7.40 to 7.53, 7.33 to 7.40, 6.40 to 6.60. Biscuit samples with 10% malted cowpea inclusion were most preferred. The foaming capacity, water absorption capacity, oil absorption capacity, swelling capacity and bulk density ranged from 2.00 to 2.50, 0.15 to 0.43, 25.5 to 16.90, 3.954.04 to and 2.75 to 3.90mg/100g. The Hemoglobin (HB), Packed Cell Volume (PCV),Red Blood Cell (RBC),White Blood Cell (WBC) are as follows; 8.10 to 9.70 g/dl, 25.50 to 31.50%, 29500 to 37500 L⁻¹, 4425.0 to 12425.0 L⁻¹ respectively over the duration of four (4) weeks of feeding the albino rats. Generally, the inclusion of malted cowpea flour has improved the nutrition assessed parameters as well as that of the consumers.

Running Title: Effect of Sorghum- malted cowpea flour blends and biscuits on the nutritional and haematological parameters of fed albino rats

INTRODUCTION

Biscuits are confectionary products usually dried to low moisture content. They are ready-to-eat and convenient food product that is consumed in between meal or as a breakfast item among all age groups in many countries (Chinma *et al.*, 2012; Usman *et al.*, 2015). They represent the most popular and largest category of snack item among bakery product because of their affordable price, shelf-stable, convenience and nutritive value (Akubor, 2003). Biscuits are nutritious, contributing valuable quantities of iron, calcium, calories, fibre and some of the B-vitamins to our diet and daily food requirement.

In most developing countries like Nigeria, the importation of wheat had an increasingly adverse effect on the balance of trade and the consumption of ready-to-eat baked product is continually growing and there has been an increase in reliance on imported wheat (Akpapunam and Darbe, 1999). Moreover, Nigeria grows less wheat but grows other staple crops such as pigeon pea, sweet potato, cassava, yam and cereals that could be used or substituted for bakery foods. For this reason and more, FAO and developing countries initiated replacing wheat needed for making baked goods, wholly or partly with flour obtained from local staples (Gernah *et al.*, 2010) like tubers, legumes and cereals, hence reducing the reliance on its importation and thus enhance the industrial utilization of local crops.

Sorghum (*Sorghum bicolor*) is an annual or perennial grass in the family Poaceae grown primarily for its grain. Sorghum is a staple food source in Africa and Asia where the grains can be boiled and eaten in a manner similar to rice, roasted or popped. Sorghum grain can also be used to produce flour which can be used to make bread. Sorghum grain is also used extensively as animal feed and as fodder. Sweet sorghum varieties can be processed into syrups and molasses sorghum is rich in a variety of nutrients. Including B-vitamins, this plays an essential role in metabolism, nerve cell development and healthy hair and skin.

Cowpea (*Vigna unguiculata L.Walp*) is a member of the *Phaseoleae* tribe of the *Leguminosae* family. Members of the *Phaseoleae* include many of the economically important warm season grain and oilseed legumes, such as soybean (*Glycine max*), common name (*Phaseolus vulgaris*) and mungbean (*Vigna radiate*). Cowpea plays a critical role in the lives of millions of people in Africa and other parts of developing world, where it is a major source of dietary protein that nutritionally complements staple low – protein cereals and tuber crops , and is a valuable and dependable commodity that produces income for farmers and trader (Singh,2002;Langyintuo *et al.* 2003).

Composite flour is a mixture of varying proportion of two or more flour which may contain or may not contain wheat flour and used for production of bread, pastries, cake and other confectionery products that are conventionally produced from wheat flour with the intention of increasing the essential nutrients in human diet and increase the economic relevance of indigenous crops (Okoye and Obi, 2017). The use of composite flours have among other advantages for developing countries such as Nigeria in terms of enhancement of nutritional

quality of food, utilization of under-exploited crops, and reduction in the importation of wheat flour, thereby saving of foreign exchange (Hasmadi *et al.*, 2014).

The consumption of biscuit is considerably high in Nigeria and other African countries and mostly by children. Wheat, which is the main raw material for biscuit production is expensive as its importation calls for search for alternative crop materials. Sorghum a better alternative, which is largely grown, relatively poor in nutrient, is underutilized. Cowpea grow abundantly in Nigeria and very rich in nutrient but contain some anti- nutrients.

Cowpea contains good source of B- vitamin, with substantial quantity of lysine (Okaka and Potter, 2009) while sorghum is an excellent source of protein with 48% of fiber without any implication of celiac disease. The acceptability of sorghum and malted cowpea in the food production of biscuit will lead to reduction in the importation of wheat and in the enrichment of the product. The objective of the research work is to evaluate the quality of Sorghum-malted cowpea flour blends and biscuits.

MATERIALS AND METHODS

3.1 Material

Cowpea and sorghum were purchased from Wukari new market, Taraba state Nigeria. Albino rats (15) were purchased in Biochemistry Department, Federal University Wukari, Taraba State, Nigeria. The cage for keeping the rats were made locally and kept in Federal University Wukari, Taraba State safe site for study.

Methodology

Preparation of cowpea and sorghum flour

The cowpea was cleaned by removing pods, sticks, soaked in water for about 5 min. at room temperature (28°C) so as to loosen the seed coats, and then dehulled manually. The dehulled beans were malted by soaking in 200 ppm of 5.25% sodium hypochlorite (to control microbial growth) for 6 h. The seeds were allowed to germinate at 30°C for 48 h, (to reduce flatulence-producing oligosaccharides and minimize rootlet development). Sprouted seeds were dried in hot air oven at 50°C for 12-20 h to reduce the moisture content up to 10% (wb). Seed coats were separated from sprouted cowpea seeds manually by gently rubbing the seeds between palms and fingers and then removed away by fan. Dried malted seeds were milled to flour, packed and stored at 4°C prior to further analysis. Sorghum grains were cleaned and

sorted (bad ones were sorted and thrown away). The sorted sorghum grains were milled using an attrition mill and then sieved by passing through 0.3 μm mesh size and then packaged in Zip-lock polyethylene bags prior to analysis.

Formulation of malted cowpea- sorghum flour blends

The malted cowpea flour was substituted into sorghum flour at different percentages (0, 5, 10, 15, 20, 25 and 100%) while 100% malted cowpea and sorghum flours were used as control samples. The flours were thoroughly mixed using a Kenwood blender to a uniform blend (Ayo and Gidado 2018).

Preparation of Sorghum-malted cowpea Flour Blend and Biscuits

Sorghum-malted cowpea flour blends were mixed with 30, 20, 1.5 and 1.0% of fats, sugar, baking powder and salts, respectively and used to produce biscuits. The procedural method described by Chinma *et al.* (2011) was used for biscuit production. The fat and sugar were first blended for 1 min., followed by the addition of sodium bicarbonate and salt which were dissolved in part of the water to be used, then with the addition of liquid milk. The flour blend was transferred into the blender and mixed well with the addition of the remaining water. Mixing time was between 5 and 7 min. The dough was rolled on a flat rolling board sprinkled with the same flour to a uniform thickness of 0.5 cm using a wooden rolling pin, cut into rectangular shapes using a moulding shell and then placed on a greased baking tray. Baking was done in an oven at 160°C for 10 min.. The biscuits were allowed to cool, packaged in low density polyethylene bags of thickness 0.5 mm and stored in air tight containers for further analysis.

Feeding of Albino Rats

The feeding of albino rats was done for 21 days in a cage which was divided into 5 segments with each segment housing three albino rats. The rats were fed with sorghum- malted cowpea flour blends to which salt (1%), bone meal (2%) and blood meal (2%) were added and thoroughly mixed. The formulated feed (100g) was weighed into plate in each of the segmented houses; leftover feeds were measured to know the amount of feed consumed by the albino rats. Plastic bottles were constructed for water intake on each segment of the cage with opening that permits sucking from the bottle. The daily weight of the albino rats was measured. After every five days, blood was collected and taken for analysis on the haematological parameter.

METHODS

Determination of Proximate composition

Proximate composition

The moisture content, crude protein, ash, crude fat, carbohydrate and crude fibre contents of the flour blends were determined as described in AOAC (2012).

Determination of Anti-nutrients Factors

Determination of Phytate Content

The phytate content of the flour blends was determined according to the method described by Hassanein *et al.*, (2011). The sample (0.5 g) was extracted with 10 ml 2.4% HCL for 1h at ambient temperature and centrifuged (3000 rpm) for 30 min. The clear supernatant was used for phytate estimation. The wade reagent (0.03% solution of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ (1ml) containing 0.3% sulfosalicylic acid in water) was added to 3 ml of the sample solution mixture were centrifuged. The absorbance at 500nm was measured using UV- VIS spectrophotometer. The phytate concentration was calculated from the difference between the absorbance of the control (3ml of water + 1ml wade reagent) and that of assayed sample. The concentration of phytate was calculated using phytic acid standard curve and result was expressed as phytic acid in mg per 100g dry weight. To prepare the phytic acid standard curve, a series of standard solution were prepared containing 5-40mg/ml phytic acid in water. 3ml of standard was pipetted into 15ml centrifuged tubes with 3ml of water used at a zero level. To each tube was added 1ml of wade reagent, and the solution was mixed using a vortex mixer for 5 secs. The mixture was centrifuged for 10 min. and the supernatant was read at 500nm by using water as a blank. By plotting the calibration curve (absorbance vs concentration), the slope and intercept was located. The phytate content was calculated using equation 1

$$\text{Phytate} \left(\frac{\text{mg}}{100\text{g}} \right) = (\text{absorbance} - \text{intercept}) \times 10\% (\text{slope} \times \text{density} \times \text{wt sample}) \times 3 \quad (1)$$

Determination of Tannin content

The tannin content of the flour blends was determined according to the method described in AOAC (2012). Aliquots of about 400 μL of the phenolic extract were treated with polyvinyl pyrrolidone (40g, 100 mg/mL) at 4 $^{\circ}\text{C}$ for 20 min. After centrifugation at 10,000 rpm (4 $^{\circ}\text{C}$,

10 min), non-tannin phenolic (supernatant) was determined as above by the Folin-Ciocalteu method (2011). Total tannins were calculated by subtracting non-tannin phenolic from total phenolic.

Determination of functional properties of the flour blends

Determination of Water and Oil Absorption Capacity of flour blends: The absorption was determined using the method described by Beuchat (2007). Distilled water (10ml) was added to 1g of Sorghum and malted cowpea flour blend samples in a weighed centrifuged tube. The tube was agitated on a vortex mixer for min. The clear supernatant was decanted and discarded. The adhering drops of water were removed and weighed. The water absorption capacity was expressed as the weight of water bound by 100g of dried flour.

The oil absorption was determined by using the method described by Beuchat (2007). Sorghum and malted cowpea flour blend (1 g) was mixed with 10ml of refined vegetable oil and was allowed to stand at ambient temperature for 30 min. The oil and adhering drops of oils decanted and discarded. Oil absorption capacity was expressed as per cent oil bound per gram flour.

Determination of Bulk density, Foaming capacity and Swelling capacity of the flour blends: The bulk density was determined by using the method described by Okaka and Potter (2009). Sorghum and malted cowpea flour blend (5g) was poured into a 10 ml dry measuring and the volume was recorded as the loose bulk density. The bottom of the cylinder was tapped 50 times on the laboratory table and the volume was recorded for packed bulk density.

The volume of the sample was calculated using equation 4.

$$\text{Bulk density (g/cm}^3\text{)} = \frac{\text{Weight of sample}}{\text{volume of sample after tapping}} \times 100 \quad (4)$$

The foaming capacity was determined according to the method described by Narayana and Narasinga Rao (2007). Two grams (2 g) of sorghum and malted cowpea flour blends was added to 50ml of distilled water at $30 \pm 2^\circ\text{C}$ in a 100 ml graduated cylinder. The suspension was mixed and shaken manually for 5 min to foam. The volume of foam at 0second after whipping was expressed as foaming capacity using equation 5.

$$\text{Foam capacity} = \frac{\text{volume of foam after whipping}}{\text{volume of mixture}} \times 100 \quad (5)$$

The volume of foam was recorded at different time intervals (5, 10, 15 and 20 secs) after whipping to determine the foam stability as per cent of the initial foam volume.

The swelling capacity was determined using the method described by Leach *et al.* (2009). Sorghum and malted cowpea flour blend (1g) was mixed with 10ml of water in a weighed centrifuge tube. The tube was heated in water bath at 85°C for 15 min and then centrifuged at 2000 rpm for 30 min. The clear supernatant was decanted and discarded. The adhering drops of water was removed and then weighed. Swelling capacity was expressed as per cent swelled per gram flour.

Determination of Physical Properties of Biscuit Samples

Determination of Weight, Diameter, Thickness and height of the blend biscuits: Weight of biscuits was measured as average values of six individual biscuits with the aid of an analytical weighing balance (Ayo *et al.*, 2007). Average value for weight was reported in grams. Diameter of biscuits was determined by placing four biscuit samples edge to edge and measuring with a digital Vernier caliper (Bala *et al.*, 2015). An average of six values was taken for each set of samples. Average value for diameter was reported in millimeter.

Thickness of biscuits was determined by measuring the diameter of four biscuit samples placed edge to edge with a digital Vernier caliper (Bala *et al.*, 2015). An average of six values was taken for each set of samples. Average value for thickness was reported in millimetre.

Determination of Spread ratio and break strength of the blend biscuits: The spread ratio was determined using the method of Gomez *et al.*, (1997). Three rows of five well-formed biscuit were made and the height measured. Also the same was arranged horizontally edge to edge and sum diameter measured. The spread ratio is calculated as diameter/ height.

The break strength of the biscuit was determined using the method of Okaka and Isieh (1990). Biscuit of known thickness (0.4cm) was placed between two parallel wooden bars (3.0cm apart). Weights were added on the biscuit until the biscuit snapped. The least weight that caused the breaking of the biscuit was regarded as the break strength of the biscuit.

Evaluation of Bioassay Parameter

21 albino rats were weighed and randomly distributed into five groups. Allocation to group was based on the initial weight of the rats and group mean weight. Rats were acclimatized for four days, after which group A (control) was given a diet containing 75.0g of vitamin and minerals premix, groups B, C and D were given diet containing (2% blood meal, 2% bone meal and 1% salt) respectively, fed for 21 days (Table 1).

Housing and feeding

The rats were housed in metabolic cages and provided diets. Feeds were weighed each morning and recorded. At the end of the day, the troughs were weighed and subtracted from the total weight of feed provided for the day to get the daily feed intake. Water was provided and the weights of the animals were taken before and during the experiment, and records kept weekly.

Feed efficiency ratio (FER) and Protein efficiency ratio (PER)

The Feed efficiency ratio (FER) and Protein efficiency ratio (PER) were determined (AOAC 2010). Feed consumption is the difference between the quantity of the leftover feed and the quantity of the feed provided. The body weight was determined weekly. It represents the difference between the weight in the current week and that of the previous week.

Formula of PER = Total weight gain (g) / Total amount of protein in total feed intake

Formula = Total weight gain (g) / Total feed intake (g)

Determination of the Haematological Parameter

Red blood cell (RBC) and white blood cells (WBC) of the flour blend of the rat fed blood: The method described by researcher was used for red blood count determination. This is an improved Neubauer method. The erythrocyte diluting pipette was used to draw blood from the vein to exactly 0.5 marks. The tip of the pipette was wiped free of blood before inserting into the erythrocyte diluting fluid and the fluid drawn into the pipette up to the 101 mark above the bulb. The pipette was gently rotated and allowed to stand for 2 min. The first few drops from the pipette were discarded before being used to change the counting chamber. The ruled area of the hemacytometer was thoroughly and carefully cleaned to remove grease.

The cover slip was thereafter changed with the fluid from the pipette. The chamber was left for 2min and cells in 5 of 25 small squares were counted less than 40x objective of light microscope. The number of the red cells counted was multiplied by 10,000 to give the number of the red blood cells in millions per cubic millimetre.

The method of Coles (1986) was used for the white blood cells count. The white blood cell pipette was used to draw blood to 0.5 marks. The tip of the pipette was wiped free of blood and used to draw WBC diluting fluid to 11 marks above the bulb. The pipette was shaken thoroughly to mix the contents and then be allowed to stand for 3min. The counting chamber was changed with the diluting fluid after discarding the first few drops. 1min after changing the chamber the cells was counted with the aid of a light microscope at 40x objective. The cells in the four corners squares will be counted and multiplied by 1000 to give the total number of the cells counted in thousand per cubic millimetre.

Packed cell volume (PVC) and Leucocytic count in rat blood fed with the flour blend

The PVC was determined using the method described by Coles (1986). Blood from the tail of the vein of the rat was allowed to run into a microchematocrit tube by capillary action until it is about three-quarter full. The end of the tube in contact with the blood was sealed with plasticine and placed in a micro-heamatocit centrifuged operated at the rate of 3,000 revolution per min for 5 min, thereafter, the capillary tube was placed in a micro- heamatocit reader and the PVC read and expressed as percentage.

A dry micropipette was used to suck blood from the snipped parts of the rats tail, a small drop of blood was applied to one end of a slide and quickly placed on the bench holding it in position, the end of the second slide was then placed in the drop and held there until the blood has spread across it. The blood is dried and stained with giemsa, washed with distilled water, dried for 2min and then examined under a microscope at a low and high magnification power for cellular appearance (Osim *et al.*, 2004).

Determination of sensory properties of the biscuits

The sensory properties of sorghum-malted cowpea biscuits was evaluated using twenty untrained panels, randomly selected from the staff and students of the Federal University Wukari, Nigeria. The flour blends, appropriately coded (ACH, BSA, CAU, DON, EFU and FDA) at the same size and temperature ($29 \pm 3^{\circ}\text{C}$) were placed in white plastic plates. The panels rinsed their mouth with bottled water after tasting each sample and were not allowed to make comment during evaluation to prevent influencing others. Structured questionnaires were presented to individual panel, using a nine-point Hedonic scale with one (1) representing “extremely dislike” and nine (9) “extremely like”. The attributes assessed were colour, texture, taste, flavour and general acceptability.

Statistical Analysis

All analyses were conducted in duplicates in completely randomized design. The data was subjected to analysis of variance using Statistical Package for Social Science (SPSS) software version 23, 2007. The average that was significantly different were separated by the Least Significant Difference (LSD) test and significances were accepted at $p < 0.05$.

RESULTS AND DISCUSSION

Proximate composition of Sorghum-malted cowpea flour blends

The results of the proximate composition are presented in Table 1. The moisture content of the flour blends ranged from 18.31 to 22.40%. Increase in moisture could be attributed to the processing technique adopted, which is by germination of the cowpea (Kumkum *et al.*, 2010).

The ash content of the flour blends varied between 1.82 and 2.21%, indicating that the samples are poor sources of minerals. Food with high ash content is expected to have high concentration of various mineral elements (McWatters *et al.*, 2004).

The protein content of the samples increased from 18.25 to 30.52% as the substitution level of the malted cowpea flour increases in the blend. However, this shows the practical significance of adding malted cowpea to the blend as the protein content were higher than that reported by (Ezeocha and Onwuka, 2010). Fasasi (2009) reported that germinated and fermentation improves the protein content and quality of products.

The carbohydrate content of the flour blends ranged from 29.78% to 45.23%. The decrease in the carbohydrate content of the samples could be due to the presence of high protein content in the malted cowpea.

The fat content of the samples varied between 0.63% and 2.66%. This value agrees with 4.8 to 9.42 reported by (Ikujenkola and Fashakin, 2005). Although, high fat content is notionally advantageous because it increases the energy level of diet, although, it can reduce the shelf life and stability of food products during storage since unsaturated oils are vulnerable to oxidative rancidity (Adebayo *et al.*, 2012).

The fiber content of the flour blends ranged from 16.10 to 20.50% with increase in the substitution level of malted cowpea. Emphasis has been placed on the importance of low fiber content in diets owing to the fact that the gastrointestinal system of the infants is not well developed to digest high fiber diets, because they impair protein and mineral digestion and absorption in human (Anwa *et al.*, 2006).

The energy value of the flour blends ranged from 258.9 to 265.14 Kcal/100g, with increasing substitution level of malted cowpea. The energy values were significantly different ($p < 0.05$) at all levels of substitution and increased as the inclusion level of malted cowpea increases in the flour blends. The energy value of food helps to provide physiological functions to human body.

Table 1: Proximate composition of Sorghum and Malted cowpea flour blends

SG/MC	Fat (%)	Fiber (%)	Protein (%)	Moisture (%)	Ash (%)	CHO (%)	Energy Value Kcal/100g
100:0	0.63 ^c ±0.1	16.10 ^c ±0.1	18.25 ^c ±0.3	18.31 ^d ±0.4	2.21 ^a ±0.2	45.23 ^a ±0.1	258.9
95:5	1.00 ^d ±0.0	17.21 ^d ±0.0	22.91 ^d ±0.2	20.16 ^d ±0.1	1.15 ^c ±0.1	36.71 ^b ±0.1	247.48
90:10	1.63 ^c ±0.1	17.70 ^c ±0.0	26.24 ^c ±0.1	21.01 ^c ±0.1	2.02 ^{ab} ±0.2	34.03 ^c ±0.1	255.76
85:15	2.05 ^b ±0.0	18.05 ^b ±0.1	28.25 ^b ±0.3	21.80 ^b ±0.2	1.86 ^b ±0.1	30.97 ^d ±0.1	255.33
0:100	2.66 ^a ±0.2	20.50 ^a ±0.1	30.52 ^a ±0.3	22.40 ^a ±0.0	1.82 ^b ±0.3	29.78 ^e ±0.0	265.14

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test. SG=Sorghum MC=Malted Cowpea.

Anti-nutrients composition of Sorghum-malted cowpea flour blends

The effects of processing on phytic acid and tannin contents of the flour blends are shown in Table 2. The phytate and tannin contents decreased from 76.42 to 45.12 and 0.15 to 0.02mg/100g with increase in the inclusion levels of malted cowpea. However, previous research works had shown that malting and fermentation could reduce phytate significantly up to 40 and 77%, respectively. Mohammed *et al.* (2011) reported the utilization of phytate as source of inorganic phosphate for germination, phytase activity, and leaching loss during soaking may result in reduction in phytate. The inherent phytase activity of cowpea during germination results in reduction in phytic acid (Reddy *et al.*, 2012). Lactic acid bacteria exhibit phytase activity during fermentation and may degrade phytate (Sreeramulu *et al.*, 2016). High reduction in phytate content of fermented cowpea flour compared to germinated cowpea flour may be due to low pH of fermented slurry and high activity of phytase in pH 4.5–5.0 (Valencia *et al.*, 2009).

The effect of processing on tannin content of sorghum-malted cowpea flour is shown in Table 2. Malting and fermentation reduced the tannin content of sorghum-malted cowpea flour to 0.15 mg/g and 0.02 mg/g, respectively. Ogbonna *et al.* (2012) revealed a decrease in tannin content due to leaching loss during steeping. Alkali treatment may also reduce tannin content (Babiker and Tinay, 2013). Romo-parada *et al.* (2015) reported a 92% reduction in tannin content. Microbial activity may reduce the tannin content of fermented sorghum-malted cowpea flour and tannin acyl hydrolases is responsible for tannin reduction in fermented sorghum-malted cowpea flour.

Table 2: Anti nutrients composition of Sorghum-Malted cowpea Flour Blends

SG-MC	Phytates(mg/100)	Tannin(mg/100)
100:0	76.42 ^a ±1.4	0.15 ^a ±0.0
95:5	71.77 ^b ±2.2	0.03 ^a ±0.1
90:10	66.12 ^c ±1.4	0.02 ^a ±0.2
85:15	59.13 ^d ±1.4	0.03 ^a ±0.1
0:100	45.12 ^e ±1.4	0.02 ^a ±0.1

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test. SG=Sorghum MC=Malted Cowpea.

Functional Properties of sorghum-malted cowpea flour blend

The results of the functional properties of the flour blends are displayed in Table 3. The bulk density of the flour sample ranged from 2.75 to 3.90 with increase in the cowpea flour. Bulk density is a measure of heaviness of solid samples, which is important for determining packaging requirements, material handling and application in the food industry. Flours with high bulk densities (>0.7 g/mL) are used as thickeners in food products (Akubor and Badifu, 2004). Bulk density depends on the combined effects of interrelated factors such as the intensity of attractive inter-particle forces, particle size, and number of contact points the higher the bulk density the greater the quantity of material that can be packaged within a specified packaging space (Fagbemi, 1999).

Water absorption capacity refers to the water retained by food products following filtration and application of mild centrifugal force. The water adsorption capacity of the flour blends shows significant differences ($p<0.05$) which increases from 0.15 to 0.43%. The increase in the water absorption capacity of the samples could be due to changes in the quality of protein upon germination and also breakdown of the polysaccharide molecules.

The foaming capacity of the flour blends varied from 2.50 to 4.00%. The increase could be due to the presence of soluble proteins with the addition of malted cowpea in the flour blends.

The swelling capacity of the flour blends ranged between 3.95 and 4.04%. Swelling implies the ability to increase in volume when protein flours are mixed in water. Both protein and starch have been reported to be responsible for the swelling of legume flour with protein playing dominant role at low temperature and starch at high temperature (Henshaw and Adebawale, 2004). The reduced swelling index of cowpea flour could be attributed to the degradation of some carbohydrate components such as cellulose, hemi-cellulose, starch granules and denaturation of some protein molecules, resulting from the heat dissipated when using the dry milling method. However, several native proteins which are insoluble in water may occur in greater amount in the cowpea flour thereby reducing its ability to swell (Okorie, 2002).

The oil absorption capacity is the attribute of the physical entrapment of oil which is considered important as flour retainer and improves the mouth feel of food products. The oil absorption capacity of the flour blends ranged from 25.5 to 16.9% and there was significant differences $p<0.05$ among the samples.

Table 3: Functional properties of Sorghum-malted cowpea flour blend

SG-MC	Foaming (%)	WAC (%)	OAC (%)	Swelling (%)	Bulk Density (g/ml)
100:0	2.50 ^a ±2.12	0.15 ^a ±0.00	25.5 ^b ±0.42	4.04 ^a ±0.21	2.75 ^{ab} ±0.21
95:5	2.65 ^a ±1.41	0.24 ^b ±0.14	23.05 ^c ±0.21	4.16 ^c ±0.00	2.50 ^{ab} ±0.49
90:10	3.01 ^a ±0.70	0.27 ^b ±0.20	22.01 ^b ±0.21	3.90 ^a ±0.07	2.40 ^{ab} ±0.28
85:15	3.50 ^a ±1.41	0.33 ^c ±0.07	21.05 ^c ±1.27	4.05 ^a ±0.07	2.41 ^a ±0.00
0:100	4.00 ^a ±1.41	0.43 ^d ±0.00	17.90 ^a ±0.14	3.95 ^a ±0.07	1.50 ^a ±0.70

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test. SG=Sorghum MC=Malted Cowpea, WAC=Water absorption capacity, OAC=Oil absorption capacity

Cumulative Feed Intake and Weight Gain of Rats Fed with Sorghum-malted cowpea Flour Blends

The feed intake/weight gain of rats fed with sorghum- malted cowpea flour blends are shown in Tables 4 and 5. Result revealed that rats fed with the 95:5% malted cowpea had the highest feed intake/weight gain over the duration of 21days, followed by those fed with 90:10% malted cowpea. There was a steady increase in feed intake/weight gain in rats fed with the 15% malted cowpea.

The feed intake is the single most important piece of information that a nutritionist can use to minimize feed costs, while ensuring performance is maintained. The factors affecting feed intake by dairy cows may include nutrition, milk production, rumen health, heat stress, balanced diet, age, pregnancy, and level of exercise (Avonda *et al.* 2007). Feed intake is important in attaining target growth rates in animals and has a significant impact on efficiency of production (Whittington *et al.* 2022). Weight gain is an increase in body weight. This can involve an increase in muscle mass, fat deposits, excess fluids such as water or other factors. Weight gain is important in lowering the risk of heart disease, stroke, diabetes and high blood pressure (Eliassen, 2006).

Table 4: Cumulative Feed Intake of albino rats fed with sorghum-malted cowpea flour blends

DAYS	5	10	15	21
100	133.07 ^d ±0.14	323.03 ^c ±0.14	496.09 ^{bc} ±0.28	705.4 ^a ±0.21
95:5	217.03 ^c ±1.41	458.4 ^{bc} ±2.12	671.80 ^b ±0.14	940.10 ^a ±0.70
90:10	194.08 ^b ±0.70	415.50 ^c ±0.42	631.30 ^b ±0.28	872.03 ^a ±0.70
85:15	194.90 ^d ±0.70	343.90 ^c ±0.70	481.50 ^{ab} ±0.70	697.30 ^a ±0.28
100	89.80 ^d ±0.14	236.70 ^c ±0.14	332.6 ^d ±0.21	441.08 ^a ±0.78

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test

Table 5: Cumulative weight gain of albino rats fed with sorghum-malted cowpea flour blends

DAYS	5	10	15	21
100	0.003 ^c ±0.14	0.030 ^d ±0.14	0.042 ^{ab} ±0.72	0.060 ^b ±0.78
95:5	0.3 ^d ±0.21	0.064 ^d ±0.28	0.093 ^a ±0.28	0.113 ^d ±0.28
90:10	0.026 ^c ±0.28	0.08 ^d ±0.42	0.112 ^b ±0.07	0.143 ^a ±0.64
85:15	0.018 ^d ±0.14	0.048 ^b ±0.35	0.057 ^{ab} ±0.57	0.092 ^a ±0.14
100	0.001 ^d ±0.21	0.028 ^c ±0.07	0.024 ^b ±0.07	0.030 ^a ±0.28

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test

Feed Efficiency Ratio (FER) and Protein Efficiency Ratio (PER) of Rats fed with Sorghum-Malted Cowpea

The feed and protein efficiency ratio of the albino rats are shown in Figure 1. The feed efficiency ratio (FER) value increased from 0.02 to 0.05 while the protein efficiency ratio increased from 0.15 to 0.35 respectively with increase in the percentage of the malted cowpea. The effect of adding malted cowpea is significant, $p < 0.05$. The 10% malted cowpea (90:10) had the highest value for the feed efficiency ratio (FER). The protein efficiency ratio increased at the substitution levels of the malted cowpea with sorghum increased.

Feed efficiency ratio (FER) is the mass of the input divided by the output (thus mass of feed per mass of milk or meat). FER is important because it helps to know how much amount of feed will be required in the growth cycle of animals. This serves as a powerful tool by letting one know what choices he would make in order to maximize the profitability of his business. The factors affecting FER could be parity order, body weight, body condition score, rumen acidosis, genetics and reproduction.

Protein efficiency ratio (PER) is the weight gain of test group/ protein consumed by the test group. PER measure the nutritive value of protein sources. The higher the PER value of a protein, the more beneficial it is to the animal. Per is an important factor because an abnormally increased intake of one of the essential amino acids can exert toxic effects in the body. Thus a protein will have a high biological value if it has the following characteristics: It should contain all the essential amino acids in sufficient amounts (Folio3, 2021). The factors affecting PER could be disproportionate amounts of amino acids in the diet (excess or shortage), protein source, feed processing and antagonism.

Table 6: Effect of adding Malted cowpea on the PER and FER of albino rats

SG-MC	FER	PER
100:0	0.02 ^a ±0.00	0.15 ^a ±0.0
95:5	0.03 ^{ab} ±0.01	0.17 ^a ±0.1
90:10	0.04 ^c ±0.01	0.32 ^{bc} ±0.2
85:15	0.04 ^{bc} ±0.01	0.27 ^{ab} ±0.1
0:100	0.05 ^e ±0.01	0.35 ^{ab} ±0.1

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different (p<0.05) from each other using Duncan multiple range test

Haematological Parameters of Albino Rats fed with Sorghum-malted cowpea Flour Blend Biscuits

The haematological properties of albino rats fed with biscuits produced from sorghum-malted cowpea flour blends are shown in Table 7 and 8. Hematological parameters such as Haemoglobin (HB), Packed Cell Volume (PCV), Total White Blood Cell (TWBC), Red Blood Cell (RBC), White Blood Cell (WBC), Lymphocytes (L), Eosinophil (E), Basophils (B) and Monocytes (M) were investigated in this study. The effect of malted cowpea on these haematological parameters of albino rats are significant (p<0.05). The results showed that at

day 5, HB, PCV, RBC, Neutrophils increased from 8.10 to 9.70 g/dl, 25.50 to 31.50%, 29500 to 37500 L⁻¹ 64.00 to 76.00% respectively. TWBC decreased from 3750.0 to 3625.0 L⁻¹ Lymphocytes, Eosinophil, and Monocytes also decreased from 29.00 to 19.00% , 3.50 to 1.75% , 1.25 to 0.65% Basophils values were not affected as the level of malted cow pea was increased.

On day 10, HB, PCV, RBC, WBC increased from 7.70 to 9.35 g/dl, 24.35 to 28.50% 289250.0 to 36650.0 L⁻¹, 4425.0 to 12425.0 L⁻¹ Neutrophils, Eosinophil and Monocytes decreased from 65.58 to 58.5%, 2.25 to 1.25%, 1.15 to 1.10% respectively Lymphocytes increased from 32.50 to 37.50% respectively Basophils values were not affected as the level of malted cowpea increased.

On day 15, HB, PCV, RBC, WBC, Lymphocytes decreased from 10.35 to 8.50 g/dl, 31.50 to 27.50%, 393250.0 to 336500.0 L⁻¹, 5750.0 to 4450.0 L⁻¹ 29.50 to 24.50% respectively. Neutrophils had no significant increase in it, Eosinophil increased from 2.25 to 6.25% Monocytes and Basophils there was change in the value from 0.00 to 1.10%, 0.00 to 1.15%.

On day 21, HB, TWBC, RBC, Neutrophils, Eosinophil increased from 9.15 to 9.70 g/dl, 3340.0 to 4450.0 L⁻¹, 327250.0 to 340500.0 L⁻¹ 68.50 to 70.50% 1.10 to 3.10% respectively PCV decreased from 28.50 to 27.50% Lymphocytes, Monocytes and Basophils decreased from 29.50 to 25.50%, 1.15 to 0.05%, 1.05 to 0.00%.

The effect of adding malted cowpea on the haematological parameters was significant, $p=0.05$, for WBC, PCV, RBC and HB. The high concentration of PCV, HB, RBC and TWBC of the experimental rats fed with malted cowpea further established the nutritional and sensory quality of these products while in the neutrophils, higher values was observed and recorded lymphocytes, eosinophil, monocytes, and basophils had a low significant increase in values. This finding agrees with the report of Sandu *et al* ., (2016) who established that diets containing quality protein and iron usually enhance the production of haemoglobin and immunity in animals.

Haemoglobin (HB) values reported in this work increased over the period of 21 days as the level of malted cowpea was increased except at day 15 were there was a decrease in the HB value. The HB values reported in this study were lower than the recommended HB range of 10-15g/dl documented in Merck Manual (2012). Haemoglobin is the iron-containing oxygen-transport metalloprotein in red blood cells of almost all vertebrates as well as the tissues of some invertebrates. Generally, a normally range is considered for male: 2.09-2.71 mmol/L

and Female: 1.86-2.48 mmol/L. The expected values for normal fasting blood glucose concentration age, administration of drugs, anti-aflatoxin treatment and continuous supplementation of vitamins affect the blood profile of healthy animals.

Packed Cell Volume (PCV) values increased and decreased over the period of 21 days as the level of malted cowpea increased. The PCV values reported in this study were lower than the recommended PCV range of 30-45% documented in Merck Manual (2012). Red blood cells (RBC) are the most common types of blood cell and the vertebrate's principal means of delivering oxygen to the body tissue via blood flow. PCV is a measurement of the proportion of blood that is made up of cells. Generally, a normal range is considered to be:- 38.3 to 48.6% for men and 35.5 to 44.9% for women.

Total White Blood Cell (TWBC) values increased at day 10 and day 21 of feeding the rats with increased malted cowpea level. However, at day 5 and 15, it decreased with increase in malted pigeon pea substitution. White blood cell (WBC) is a type of blood cell made in the bone marrow and found in the blood and lymph tissue. A white blood cell (WBC) count of less than $4 \times 10^9/L$ indicates leukopenia. A WBC count of more than $11 \times 10^9/L$ indicates leucocytosis. The major function of the white blood cell are to fight infections, defend the body by phagocytosis against invasion by foreign organisms and to produce or at least transport organisms and distribute antibodies in immune response. Animals with low white blood cell are exposed to high risk of disease infection, while those with high count are capable of generating antibodies in the process of phagocytes and have high degree of resistance to low diseases (Soetan *et al.* , 2013) and enhance adaptability to local environmental and disease prevalent condition (Iwuji *et al.*, 2012).

Red Blood Cells (RBC) values increased over the period of 21 days but decrease at day 15. The RBC values reported in this study were lower than the recommended RBC range of $5.0-10.0 \times 10^6/mm^3$ documented in Merck Manual (2012). Red blood cells serve as a carrier of haemoglobin. This haemoglobin reacts with oxygen carried in the blood to form oxy-haemoglobin during respiration. The normal range for RBC for male is: $4.3-5 \times 10^{12} /L$ Female: $3.5-5.5 \times 10^{12}$. A reduced red blood cell count implies a reduction in the level of oxygen that would be carried to the tissues as well as the level of carbon dioxide returned to the lungs (Ugwuene *et al.*, 2011).

Lymphocytes (L) values decreased at day 5, increased at day 10 then decreased at day 15 and 21. In general, an increase in Lymphocytes was observed with decrease in malted cowpea.

Lymphocyte is a type of white blood cell in the immune system of most vertebrates and is responsible for both humoral and cellular immunity. These cells help protect the body from infection. When lymphocytes levels are low, there is higher risk of infection.

Eosinophil (E) values decreased at day 5, decreased at day 10 then decreased at day 15 and increased at day 21. In general, an increase in Eosinophil was observed. Eosinophil are a variety of white blood cells and one of the immune system components responsible for combating multicellular parasites and certain infections in vertebrates. High number of eosinophil can be an emergency and if untreated, may cause damage to multiple organs.

Basophils (B) values decreased over the period of 21 days. The variation was significant. Basophils release enzymes to improve blood flow and prevent blood clots. A high basophil count signifies infection or a more serious medical condition like leukaemia or autoimmune disease.

Monocytes (M) values were not affected over the period of 21 days. A monocyte is a type of white blood cell and a type of phagocyte. A normal monocyte count is between 2% and 8% of the white blood cell count. Low levels of monocytes indicate that the body is more susceptible to infection.

Neutrophil is found in the white blood cell and helps immune system to fight infection and heal injuries. They are formed in the bone marrow and travel throughout the blood, tissues and lymph nodes. If the neutrophil is too high it may lead to the development of leucocytosis.

Generally, haematological factors are affected by several factors which include physiological, environmental conditional, dietary content, fasting. Chineke *et al.*, (2006) also reported that apart from genotype, age and sex difference in haematological indices may be caused by nutritional, environmental and hormonal factors. According to Radostits *et al.*, (1994), low nutritional, grassland, pasture, stress, parturition, and climatic factors greatly alter the blood values of goats, sheep and other farm animals.

Table 7: Haematological Parameters of Albino Rats fed with Sorghum -Malted Cowpea Flour Blend Biscuits

Day	Parameter	100:0	95:5	90:10	85:15	0:100
5	HB(g/dl)	8.10 ^d ±0.1	8.50 ^c ±0.01	8.70 ^c ±0.01	9.10 ^b ±0.1	9.70 ^a ±0.1
	PCV (%)	25.50 ^b ±0.7	26.00 ^b ±1.4	28.50 ^c ±0.7	29.50 ^a ±0.7	31.50 ^a ±35.3
	TWBC(L ⁻¹)	3750.0 ^{ab} ±353.5	2350.0 ^c ±70.7	4025.0 ^a ±35.3	3350.0 ^b ±70.7	3625.0 ^{ab} ±35.3
	RBC(L ⁻¹)	29500.0 ^{cd} ±0.14	31500.0 ^c ±0.70	32100.0 ^b ±0.14	36400.0 ^{ab} ±141.4	37500.0 ^a ±707.1
	N(%)	64.00 ^a ±1.4	79.50 ^a ±13.4	64.50 ^a ±0.7	71.50 ^b ±0.7	76.00 ^b ±0.1
10	HB (g/dl)	7.70 ^b ±0.01	9.25 ^a ±0.3	9.55 ^a ±0.07	9.65 ^a ±0.07	9.35 ^a ±0.07
	PCV (%)	28.35 ^b ±0.07	15.35 ^b ±17.8	29.50 ^a ±0.07	30.50 ^a ±0.7	28.50 ^a ±0.7
	TWBC(L ⁻¹)	4425.0 ^d ±35.3	3550.0 ^c ±70.7	6750.0 ^b ±70.7	5550.0 ^c ±70.7	12425.0 ^a ±35.5
	RBC(L ⁻¹)	289250.0 ^d ±353.5	349250.0 ^b ±353.3	374500.0 ^c ±707.1	315500.0 ^c ±21920.2	36650.0 ^a ±70.7
	N(%)	65.35 ^c ±0.7	63.45 ^c ±0.7	57.70 ^c ±0.1	70.50 ^a ±0.7	58.50 ^b ±0.7
15	HB(g/dl)	10.35 ^a ±0.7	9.75 ^b ±0.7	9.55 ^{bc} ±0.07	9.35 ^b ±0.70	8.50 ^d ±0.7
	PCV (%)	31.50 ^a ±0.7	30.50 ^a ±0.7	31.25 ^a ±0.7	27.50 ^b ±0.7	27.50 ^b ±0.7
	TWBC(L ⁻¹)	5750.0 ^c ±70.0	12825.0 ^b ±35.3	14825.0 ^a ±35.3	4575.0 ^c ±1378.8	4450.0 ^c ±70.7
	RBC(L ⁻¹)	393250.0 ^a ±353.3	37250.0 ^a ±3535.5	26400.0 ^a ±236.6	362500.0 ^a ± 3535.3	336500.0 ^c ± 707.1
	N(%)	68.50 ^a ±0.7	49.50 ^b ±0.7	48.50 ^b ±13.4	70.50 ^a ±39.5	68.50 ^a ±0.71
21	HB(g/dl)	9.15 ^b ±0.2	9.15 ^b ±0.70	8.35 ^c ±0.02	9.85 ^a ±0.7	9.70 ^a ±0.1
	PCV (%)	28.50 ^a ±0.71	29.50 ^a ±0.71	20.50 ^a ±2.12	29.00 ^a ±1.41	27.50 ^a ±0.71
	TWBC(L ⁻¹)	3340.0 ^a ±14.1	4025.0 ^a ±35.3	1720.0 ^a ±237.8	3775.0 ^a ±883.8	4450.0 ^a ± 70.7
	RBC(L ⁻¹)	327250.0 ^c ± 14.1	342750.0 ^b ± 35.3	30550.0 ^d ± 237.8	372250.0 ^a ± 883.8	340500.0 ^b ± 70.7
	N(%)	68.50 ^a ±0.7	64.50 ^a ±0.7	63.50 ^a ±0.7	34.05 ^b ±39.5	70.50 ^a ±0.7

Values are mean ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($P \leq 0.05$) from each other using Duncan multiple range test. HB = Hemoglobin, PCV = Packed Cell Volume, TWBC = Total White Blood Cell, RBC = Red Blood Cell, WBC = White Blood Cell

UNDER PEER REVIEW

Table 8: Haematological Parameters of Albino Rats fed with Sorghum-malted Cowpea Flour Blend Biscuits

Day	Parameter	100:0	95:5	90:10	85:15	0:100
5	Lymphocytes (%)	29.00 ^{ab} ±1.4	27.50 ^b ±0.7	30.50 ^a ±0.7	20.50 ^c ±0.7	19.00 ^d ±0.1
	Eosinophils (%)	3.50±0.07	3.05±0.00	1.25±0.00	1.30±0.04	1.75±1.00
	Basophils (%)	0.00 ^b ±0.00	0.00 ^b ±0.00	0.00 ^b ±0.00	1.15 ^a ±0.02	0.50±0.7 ^{ab}
	Monocytes (%)	125 ^a ±0.03	0.00 ^a ±0.00	1.00 ^a ±1.4	0.00 ^a ±0.00	0.65±0.09 ^a
10	Lymphocytes (%)	32.50 ^c ±0.7	34.50 ^c ±0.7	41.50 ^a ±0.7	26.50 ^b ±0.07	37.50 ^d ±0.71
	Eosinophils (%)	2.25 ^{bc} ±0.03	2.05 ^{bc} ±0.7	5.65±0.1	5.60 ^a ±6.2	2.25 ^c ±2.5
	Basophils (%)	0.00 ^a ±0.00	0.00 ^a ±0.00	0.00 ^a ±0.00	5.55 ^b ±6.2	0.00 ^a ±0.00
	Monocytes (%)	1.15 ^a ±0.2	0.00 ^a ±0.00	0.00 ^a ±0.00	5.65 ^a ±6.1	1.10 ^a ±0.1
15	Lymphocytes (%)	29.50 ^c ±0.07	41.50 ^a ±0.71	36.50 ^b ±0.7	25.50 ^b ±0.7	24.50 ^b ±0.71
	Eosinophils (%)	2.25 ^c ±0.3	4.80 ^b ±0.4	2.25 ^c ±0.3	1.10 ^d ±0.1	6.25 ^a ±0.3
	Basophils (%)	0.00 ^a ±0.00	0.00 ^a ±0.00	1.10 ^b ±0.1	1.15 ^b ±0.2	1.10 ^b ±0.1
	Monocytes (%)	0.00 ^c ±0.00	1.05 ^b ±0.07	2.25 ^b ±0.2	1.20 ^b ±0.02	1.15 ^a ±0.2

20	Lymphocytes (%)	29.50 ^c ±0.7	30.50 ^c ±0.7	34.50 ^b ±0.7	36.50 ^a ±0.7	25.50 ^d ±0.7
	Eosinophils (%)	1.10 ^b ±0.1	3.05 ^a ±1.14	1.10 ^b ±0.1	1.05 ^b ±0.7	3.10 ^a ±0.1
	Basophils (%)	1.05 ^a ±0.7	0.00 ^a ±0.00	0.50 ^a ±0.7	0.05 ^a ±0.7	0.00 ^a ±0.00
	Monocytes (%)	1.15 ^a ±0.3	0.05 ^a ±0.07	0.00 ^a ±0.00	0.00 ^a ±0.00	0.05 ^a ±0.7

Values are mean ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($P \leq 0.05$) from each other using Duncan multiple range test

UNDER PEER REVIEW

Sensory Evaluation of Sorghum-Malted Cowpea Flour Blends Biscuits

The sensory properties of biscuits produced with sorghum-malted cow pea flour blends are shown in Table 9. The average mean score for colour, texture and taste increased from 7.40 to 7.53, 7.00 to 4.75, 7.20 to 4.60, 7.33 to 7.40 and 6.53 to 6.60 with increase in malted cowpea while the aroma decreased from 4.06 to 3.93. Color and taste are important attribute that enables the acceptability of food products. The decrease in aroma could be due to the smell of the malted cowpea. The general acceptability was the 100:0 and 0:100 of the sorghum and malted cowpea but among the flour blend was the 90: 10.

Table 9: Sensory Evaluation of Sorghum- Malted Cow pea Flour Blends Biscuits

SG-MC	Taste	Aroma	Color	Texture	Gen accept.
100:0	6.53 ^a ±2.2	4.06 ^a ±2.2	7.40 ^a ±1.63	7.33 ^a ±1.4	6.60 ^a ±1.3
95:5	6.46 ^a ±1.72	4.06 ^a ±2.4	6.80 ^a ±2.3	6.67 ^a ±1.4	5.93 ^a ±1.4
90:10	6.66 ^a ±1.83	9.40 ^a ±19.0	7.40 ^a ±1.5	7.33 ^a ±0.8	6.53 ^a ±1.4
85:15	5.93 ^a ±2.0	4.00 ^a ±2.2	7.46 ^a ±1.4	6.67 ^a ±1.6	6.20 ^a ±1.9
0:100	6.60 ^a ±1.9	3.93 ^a ±1.8	7.53 ^a ±1.5	7.40 ^a ±0.8	6.40 ^a ±1.5

Values are mean ± standard deviation of twenty panellist . Means within each column not followed by the same superscript are significantly different ($P \leq 0.05$) from each other using Duncan multiple range test. SG=Sorghum, MC=Malted cowpea.

Physical Properties of Sorghum - Malted cowpea Flour Blends Biscuits

The physical properties sorghum and malted cowpea flour blends biscuits is shown in Table 10. The diameter ranged from 44.50 to 42.87 cm with increase in the level of malted cow pea substitution. Biscuit produced with 100:0 (100% Sorghum) had the highest diameter. The length ranged from 17.99 to 20.50cm with increase of the malted cowpea substitution in the sorghum. The thickness increased from 18.50 to 20.55 cm with increase in the level of malted cow pea substitution. The result revealed that the highest thickness was observed in biscuit produced with 0:100 (100% malted cow pea). The break strength ranged from 2.50 to 1.50 g with increase in the level of malted pigeon pea substitution in the biscuit. The highest break strength was observed in biscuit produced with 100:0 and 90:10. Addition of malted pigeon pea reduced the break strength of the biscuits and indicates that they have low carbohydrate/starch. The spread ratio of the biscuit decreased from 2.40 to 2.00 cm with increase in the level of malted cow pea substitution.

Table 10: Physical Properties of Sorghum - Malted cowpea Flour Blends Biscuits

SG-MC	Diameter	Length	Weight	Break	Spread	Thickness
100:0	44.50 ^a ±0.7	17.99 ^{bc} ±0.7	0.75 ^c ±0.7	2.50 ^a ±0.7	2.40a±0.2	18.50 ^a ±2.1
95:5	45.05 ^a ±2.7	18.75 ^{abc} ±1.0	6.75 ^{bc} ±0.2	2.25 ^a ±0.3	9.72a±10.3	18.55 ^a ±2.0
90:10	44.01 ^a ±1.4	19.52 ^{bc} ±0.7	8.20 ^b ±0.0	2.50 ^a ±0.7	2.31 ^a ±0.9	19.01 ^a ±1.4
85:15	44.80 ^a ±1.1	16.75 ^c ±1.0	7.90 ^b ±0.4	2.00 ^a ±0.0	2.54a±056	17.51 ^a ±0.7
0:100	42.87 ^a ±3.0	20.50 ^a ±0.7	10.60 ^a ±0.1	1.50 ^a ±0.7	2.00 ^a ±1.4	20.55±0.6

Values are Means ± standard deviation of physical analysis. Means within each column not followed by the same superscript are significantly different ($p < 0.05$) from each other using Duncan multiple range test. SG=Sorghum, MC=Malted Cowpea.

CONCLUSION

Suitable biscuits can be made from the addition of malted cowpea flour blends, however the most preferred among the blends made was the 10% added malted cowpea. The malted cowpea has shown to increase the physical properties, FER and PER eating quality and weight gain. Generally the results show a positive relationship between addition of malted cowpea and weight gain. The relative improvement in all the haematological properties accessed showed that malted cowpea content is high in nutrients. The addition of malted cowpea can be used to improve other baking product for healthy consumption.

The use of sorghum and malted cowpea in diet is highly recommended and could reduced the cost for the importation of wheat and the use of 90:10 (90% sorghum, 10% malted cowpea flour blends) in diet is highly recommended since it add value to food products.

REFERENCES

- Adebayo, E.A., Oloke, J.K., Ayandele, A.A., Adegunlola, C.O. (2012a). Phytochemical, antioxidant and antimicrobial assay of mushroom metabolite from *Pleurotus pulmonarius* –LAU 09 (JF736658). *J Microbiol Biotech Res.*, 2 (2), 366-374
- Akpapunam, M.A., and Darbe J.W. (1999). Chemical Composition and Functional Properties of Blend of Maize and Bambara Groundnut Flours For Cookies Production. *Plant Foods for Human Nutrition*, 46, 147-155
- Akubor, P. (2003). Functional properties and performance of cowpea/plantain/wheat four blend in biscuits. *Journal of Plant Food and Human Nutrition*; 58:1-8

Akubor IP and Badifu I G(2004). Chemical composition, functional properties and baking potential of African breadfruit kernel and wheat flour blends [International Journal of Food Science & Technology](#) 39(2):223 – 229

Anwar F, Ashraf M, Bhangar MI. 2005. Inter provenance variation in the composition of *Moringa oleifera* oilseeds from Pakistan. *J Am Oil Chem Soc* 82: 45–51

AOAC, (2010). *Official Methods of Analysis. 18th Edn.*, Association of Official Analytical Chemists (AOAC), Washington, DC., USA

Ayo, J.A., Ayo, V.A., Nkama, I. and Adeworie, R. (2007). Physiochemical, In vitro Digestibility and Organoleptic Evaluation of Acha-wheat Biscuit Supplemented with Soybean flour. *Niger. Food J.* 25(1):77-89

Avonda T., Seresinhe*, S. A. C. Madushika, Y. Seresinhe, P. K. Lal, E. R. Ørskov¹ (2007). Effects of Tropical High Tannin Non Legume and Low Tannin Legume Browse Mixtures on Fermentation Parameters and Methanogenesis Using Gas Production Technique *Asian-Australasian Journal of Animal Sciences (AJAS)* 2012; 25(10): 1404-1410.

Babiker, E. E. And E.I, Tinay, A.H. (2013). Effect of Reconstitution and Na₂CO₃ on Tannin Content and In Vitro Protein Digestibility of Faba Bean Cultivars. *Plant Foods for Human Nutrition*, 44, 119–130.

Bala, A., Gul, K. and Riar, C. S. (2015). Functional and sensory properties of cookies prepared from wheat flour supplemented with cassava and water chestnut flours. *Cogent Food & Agriculture*, 1(1), 1019815.

Braide W, Odiong. I and Oranusi S(2012). Phytochemical and Antibacterial properties of the seed of watermelon. *Prime Journal of Microbiology Research* 292):99-104.

Chineke, C. A., Ologun, A. G., & Ikeobi, C. O. N. (2006). Haematological parameters in rabbit breeds and crosses in humid tropics. *Pakistan Journal of Biological Sciences*, 9(11), 2102-2106

Chinma, C.E., Igbabul, B.D. and Omotayo, O.O. (2012). Quality Characteristics Of Cookies Prepared From Unripe Plantain And Defatted Sesame Flour Blends. *American Journal Of Food Technology*; 7(7):398-408.

Chinma, C., James, S., Imam, H., Ocheme, O., Julian, A. and Yakubu, C. (2011). Physicochemical and Sensory Properties, and In-Vitro Digestibility of Biscuits Made from Blends of Tigernut (*Cyperus esculentus*) and Pigeon Pea (*Cajanus cajan*). *Nigerian Journal of Nutritional Sciences*. 32: 10.4314/njns.v32i1.67816.

Coles, E.H. (1986). *Veterinary Clinical Pathology, fourth edition*, WB. Saunders Co. Philadelphia. Pp 486.

Fagbemi T.N.F(1999) Effect of blanching and ripening on functional properties of plantain (Musa

aab) flour *Plant Foods Hum Nutrition*54(3):261-9

Fasasi O.S. (2009). Proximate anti-nutritional factors and functional properties of processed pearl millet (*Pennisetum glaucum*). *J. Food Technol.* 7(3): 92-97

- Folin Ciocalteu Method(2011). Application and Analysis of the Folin Ciocalteu Method for the Determination of the Total Phenolic Content from *Limonium Brasiliense* [Molecules](#) 18(6):6852-65
- Gernah, D. I., Senger, I. A. and Audu, J. O. (2010). Physicochemical And Sensory Evaluation Of Cookies Produced From Wheat And Brewers Spent Grain Composite Flour. *Nigerian Food Journal*, 28, 440– 447.
- Gutiérrez-Uribe J.A, Romo-Lopez I and Serna-Saldívar S.O (2011). Phenolic Composition and Mammary Cancer Cell Inhibition of Extracts of Whole Cowpeas (*Vigna Unguiculata*) and its Anatomical Parts. *Journal Functional Foods*. 3(4):290-297.
- Gomez, M.I., Obilance, A.B., Martin, D.F., Madzvanuse, M. and Many, E.S. (1997). Manual of Laboratory Procedures for quality evaluation of sorghum and Millet. *International crop Research Institute of the Semi Arid and Tropics (ICRSAT)*, India P.64.
- Henshaw, F.O and Adenekan, M.K . (2004),Functional Properties and Pasting Characteristics of Flour from Five Varieties of Nigerian Pigeon Pea (*Cajanus cajan*) J. of Biological Science and Bioconservation, Vol.6, No. 1, Pp. 73 – 84.
- Iwuji, T.C. and Herbert, U. (2012). Haematological and serum biochemical characteristics of rabbit bucks fed diets containing garcimiola kola seed meal (p.87-89). Proceedings of 37th Annual Conference of Nigerian Society for Animal Production
- Kumkum Agarwal RanjanaVarma(2010),1 **Antioxidant activity and Phytochemical analysis of *Hyptis suaveolens* (L.) Poit** Journal of Advanced Pharmacy Education & Research Oct-Dec 2013 Vol 3 Issue 4
- Langyintuo A.S, Lowenberg-Deboer J, Faye M, Lambert D, Ibro G, Moussa B, Kergna A, Kushwaha S, Musa S, Ntougam G (2003) Cowpea Supply And Demand In West Africa. *Field Crops Research* 82:215–231.
- Leach Opa, Alice Taylor and Flavia Sciota(2009). Vascular dysfunction in the diabetic placenta: causes and consequences L J. Anat. (2009) 215, pp69–76
- McWatter KH, Philips RD, Walker SL, McCullough SE, Mensah-Wilmot Y, Saalia FK (2004).** Baking performance and acceptability of raw and extruded cowpea flour breads. *J. Food Qual.* 27:337-351
- Merck Manual, (2012). Haematologic reference ranges. Mareck Veterinary Manual. Retrieved from <http://www.merckmanuals.com/>.
- Mohammed, N. A., Mohammed, I. E., & Barbiker, E. F. (2011). Nutritional Evaluation of Sorghum Flour (Sorghum Bicolour L. Moench) During Processing Of Injera. *International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering*, 5, 99–103.
- Narasinga-Rao, M.S. (2007). Functional properties of raw and heat processed winged bean flour. *J Food Sci.*; 47:1534–1538.
- Ogbonna, A. C., Abuajah, C. I., Ide, E. O., & Udofia, U. S. (2012). Effect of Malting Conditions on the Nutritional and Anti-Nutritional Factors of Sorghum Grist. *Food Technology*. 36(2), 64–72.

- Okaka, J.C. and Isieh, M.I. (1990). Development and Quality evaluation of Cowpea-wheat biscuit. *Nigerian Food Journal* 8:56-62.
- Okaka, J.C and Potter (2009). Physiochemical and Functional properties of cowpea powders processed to reduce benny flavour. *Journal of Food Science* 44: 1235-1240.
- Okorie, E.O. (2002) Influence of pH on the Adsorption of Trace Metals on Ecological and Agricultural Adsorbents. *Journal of Chemical Society of Nigeria*, 27, 95-98.
- Okoye, J.I and Obi, C.D(2017). Nutrient composition and sensory properties of wheat-African bread fruit composite flour cookies. *Sky Journal of Food Science* 6(3): 27-32.
- Onwuka G.I (2006). Soaking, Boiling and Anti nutritional Factors in Pigeon Peas (*Cajanus Cajan*) and Cowpeas (*Vigna Unguiculata*). *Journal Food Processing Preservation*. 30(5): 616-630.
- Osim, E.E., Mesembe, O.E. and Ibanga, I. (2004) The Effects of Fresh and Thermooxidized Palm Oil Diets on Some Haematological Indices in the Rat. *Nigerian Journal of Physiological Sciences*, 19, 86-91.
- [Sandhu K I](#), [Eoin Sherwin](#) -, [Harriët Schellekens](#) -, [Catherine Stanton](#) -, [Timothy G Dinan](#) -, [John F Cryan](#) (2016).³ Feeding the microbiota-gut-brain axis: diet, microbiome, and neuropsychiatry. *Review Transl Res* 179:223-244
- Soetan, K.O., Akinrinde, A.S., and Ajibade, T.O. (2013). Preliminary studies on the haematological parameters of cockerels fed raw and processed guinea corn (*Sorghum bicolor*) (p. 49-52). *Proceedings of 38th Annual Conference of Nigerian Society for Animal Production*.
- Reddy, N. R., Sathe, S. K., & Salunkhe, D. K. (2012). Phytate in Legumes and Cereals. *Advances in Food and Nutrition Research*, 28, 1032–1039.
- Singh B.B (2002). Recent Genetic Studies in Cowpea. In: Fatokun CA, Tarawali SA, Singh BB, Kormawa PM, Tamo M (Eds) *Challenges And Opportunities For Enhancing Sustainable Cowpea Production*. *International Institute of Tropical Agriculture*, Ibadan, Nigeria, Pp 3–13.
- Valencia, S., Svanberg, U., Sandberg, A. S., and Ruales, J. (2009). Processing of quinoa, Effects on In Vitro Iron Availability and Phytate Hydrolysis. *International Journal of Food Science and Nutrition*, 50, 203–211.