Quality Evaluation of Malted Pigeon Pea –Orange Fleshed Sweet Potato Flour Blends and Biscuits Using Albino (Wistar) Rats

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Original Research Article

ABSTRACT

The use of underutilized food crops and their fortification could add value to food products. Malted pigeon pea was produced and substituted into orange fleshed sweet potato at 0, 5, 10, 15, 20, and 100% to produce flour blends which was used to feed albino rats on addition of salt, bone meal, blood meal. The blend flour was also used to produce biscuits on addition of baking fat, sugar and salt. The biscuit was analysed for physical and sensory properties. The albino rat feed was analysed for nutrition and haematological qualities. The ash, protein and energy value content of

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the flour increased from 2.34 to 3.11, 4.64 to 13.27%, 379.83 to 384.19 Kcal/100g, respectively, with increase in added malted pigeon flour. The flavonoid, saponin, phenol, phytate, tannins contents increased from 2.11 to 3.43, 6.61 to 9.70, 0.03 to 0.07, 33.74 to 50.28, 0.04 to 0.08 mg/kg, respectively. The water absorption capacity and bulk density capacity decreased from 0.38 to 0.15% and 0.73 to 0.46 g, while the swelling, oil absorption and foam capacity increased from 1.60 to 3.65, 0.91 to 1.82 and 7.15 to 12.20%, respectively, with addition of malted pigeon pea. The highest break strength was observed in biscuit produced with 85:15 (85 % orange fleshed sweet potato, 15 % malted pigeon pea flour blends) (3000g). The Feed Efficient Ratio (FER) and the Protein Efficient Ratio (PER) of the feed albino rats improved with addition of malted pigeon pea. The Haemoglobin (HB), Park Cell Volume (PCV), Total White Blood Cells (TWBC), White Blood Cells (WBC), Red Blood Cell (RBC) of the feed albino rats generally increased with increase in the quantity of malted pigeon pea over the three weeks feeding duration. All the blend flour biscuits were generally accepted; however, the 15% malted pigeon pea biscuit was the most preferred. Quality and acceptable biscuits can be produced from malted pigeon pea –orange fleshed sweet potato.

1. INTRODUCTION

"Biscuits areready-to-eat, convenient, confectionery food products that are consumed in-between meals or as breakfast items among all age groups in many countries" [1,2]. "Biscuits represent the most popular and largest category of snack items among bakery products because they are affordable, shelf-stable and convenient" [3]. Biscuits are relatively nutritious, contributing valuable quantities of iron, calcium, calories, fibre and some of the B-vitamins to diets.

"Biscuits are usually produced from wheat flour. In most developing countries like Nigeria, the reliance on imported wheat has an increasingly adverse effect on the balance of trade, and the consumption of ready-to-eat baked products" [4]. "Additionally, Nigeria grows lower quantities of wheat, but produces other staple crops such as legumes including pigeon pea, and roots and tubers such as sweet potato, cassava and yam. These could be used or substituted for wheat in bakery foods. For these reasons, the Food and Agriculture Organization (FAO), and some developing countries have promoted the replacement of wheat in baked goods, wholly or partly with flour obtained from local staples like tubers, legumes and other cereals" [5]. This reduces the reliance on wheat importation and enhances the industrial utilization of local crops.

"Pigeon pea (*Cajanus cajan*) belongs to the Leguminosae family of flowering plants and is a drought-tolerant pulse/legume mainly grown in the semi-arid tropics for its seeds. It is an important food security crop for several African countries, and is used for local consumption and export" [6]. Lysine, which is limiting in most cereals, is supplemented when cereals are combined with legumes such as pigeon pea, which are rich in lysine. Pigeon pea and sweet potato are abundantly grown in Africa, but currently underutilized. Thus, they could be substituted into other food products.

"Orange-fleshed sweet potato (*Ipomoea batatas*) is a root and tuber crop, that plays a significant role in agriculture, and facilitates food security in many developing countries" [7]. "It is a potential energy contributor, considered as the fifth essential crop (fresh weight basis) after rice, wheat, maize, and sorghum" [8]. "Additionally, the orange-fleshed sweet potato possesses special characteristics including adaptability to a broader topography, ability to Thrive in a wide range of environmental conditions, good productivity within short time frames, and is a good source of various macro- and micronutrients" [9,10]. "Its high sensory acceptability makes it suitable in malnutrition management and facilitation of food security in underdeveloped nations" [11].

"Pigeon pea has been characterized with anti-nutrients which can be reduced by germinating the grains. Composite flour is a mixture of varying proportions of two or more flours which may or may not contain wheat flour. It is used for production of bread, pastries, cake and other confectionery products that are conventionally produced from wheat flour. The aim of using composite flour is to increase the essential

nutrients in human diets and improve the economic relevance of indigenous crops" [12]. "The use of composite flours has some advantages for developing countries such as Nigeria. These include the enhancement of nutritional quality of foods, utilization of under-exploited crops, thus, preventing them from going into extinction, and reduction in the importation of wheat flour, thereby saving foreign exchange" [13]. "The bakery products produced using composite flour are usually of good quality, with some characteristics similar to wheat-flour products, though the texture and the properties of the composite flour bakery products may be different from those made from wheat flour, with an increased nutritional value and distinct appearance" [13].

The importation of wheat flour with the present economic problem in the nation had called for research into possible alternatives such as blends of pigeon and orange fleshed sweet potato flour. The acceptability of malted pigeon pea–orange fleshed sweet potato flour blends in food products could improve the nutritional content and as well the sensory properties of the flour products [14-17]. The acceptability of these flour blends in food products could help reduce use of wheat flour, hence save cost and as well encourage the use of the underutilized materials that are abundantly available. The broad objective of this study: is to evaluate the nutritional quality of malted pigeon pea–orange fleshed sweet potato flour blends and biscuits using Albino (Wistar) Rats.

2. MATERIALS AND METHODS

2.1 Materials

Raw Materials and Sources: Pigeon pea (*Cajanus cajan*) and orange-fleshed sweet potato were purchased from Wukari new market in Wukari Local Government Area of Taraba State. Other ingredients including granulated sugar (Dangote Sugar, Nasarawa), fortified milk (Wangara Properties Dev, Co. Ltd, Jalingo), baking powder, baking fat and salt used in the production of the biscuit formulation were also obtained from the same market. Albino (Wistar) rats were procured from the Biochemistry Department at the Federal University, Wukari. The cage for keeping the albino rats were made locally and kept in a safe site at the Federal University, Wukari, Taraba State.

2.1.1 Preparation of malted pigeon pea flour

Pigeon pea flour was produced following the method described by Onwuka [18]. Pigeon pea seeds (*Cajanus cajan*) were sorted and cleaned to remove foreign materials, soaked in tap water for 10 hours, washed and drained. The drained seeds were germinated (covered in wet jute bag) for 5 days, sun-dried milled (attrition grinding machine) and sieved (0.3μm aperture size) into flour.

2.1.2 Preparation of orange fleshed sweet potato flour

Fresh, matured orange-fleshed sweet potato tubers were obtained, washed, sorted, peeled, sliced and soaked in citric acid for 30 min. The sliced orange-fleshed sweet potato tubers were dried at 50°C in an oven, milled (attrition, sieved (0.3μm aperture size), packaged in a polyethylene bag and stored (at temperature 5°C) [19].

2.1.3 Formulation of flour blends

The malted pigeon pea was substituted into the orange-fleshed sweet potato at 0, 5, 10, 15, 20% to produce flour blends. A Kenwood mixer was used for mixing samples at speed 6 for 3 minutes to achieve uniform mixing [20].

The method as described by Chinma et al. [21] was used for biscuit production. The flour blends were mixed with 30, 20, 1.5 and 1.0 % of the baking fats, sugar, baking powder and salt, respectively. The fat and sugar were first blended for 1 min. This was followed by the addition of a solution of sodium bicarbonate and salt in part of the water to be used. Thereafter, liquid milk was added. The flour blend was transferred into the blender and blended well, adding the remaining water. Mixing time was between 5 and 7 minutes. 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 and cut into rectangular shapes using a moulding shell. Cut biscuit dough was placed on a greased baking tray and baked in an oven at 160°C for 10 minutes. The biscuits were allowed to cool and packaged in low density polyethylene bags of thickness 0.5 mm and stored in air tight containers for further analysis.

2.1.4 Rat feeding experiment

Male albino rats of an average age of 5-6weeks and weight of 90 -110g were used for the

experiment. The feeding of albino rats was done for 21 days in a cage which was divided into 5 segments, of three albino rats each. The rats were fed with malted pigeon pea-orange fleshed sweet potato flour blends. 100g of the formulated feed were weighed into plates in each segment. Leftovers of the feed 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 openings that permit sucking from the bottle. The daily weight of the albino rats was measured for 21 days, after each rat feeding episode.

2.2 Methods

2.2.1 Determination of chemical composition of the malted pigeon pea-orange fleshed sweet potato flour blends

2.2.1.1 Determination of Proximate composition

Determination of Proximate Composition: The proximate composition of samples (moisture content, crude protein, ash, crude fat, carbohydrate and crude fiber) was determined via methods documented by AOAC [22]. The calorific content was calculated from the values obtained for protein, fat and carbohydrate, using Atwater factor method as expressed below (AOAC 2012).:

Calorific value (Kcal/100g) = (P X 4.0) + (F X 9.0) + (C X 3.75).

Where;
Protein content (%) =P, Fat content (%) = F and Carbohydrate content (%) = C

2.2.1.2 Determination of Phytochemical Composition of the Malted pigeon pea-Orange fleshed sweet potato flour blends

Determination of flavonoids: This was determined as described by AOAC [22]. A 5 g sample was boiled in 50 ml of 2 M HCl solution for 30 minutes under reflux. It was allowed to cool and then filtered through a filter paper. A weighed filter paper was used for recovery by filtration of a measured volume of the extract. The resulting difference was the weight of the flavonoid in the sample.

Determination of saponins: The method of AOAC [22] was used for the determination of saponins.

Determination of carotenoids: The carotenoid content was determined as described by Onyeka and Nwambekwe [23]. A measured weight of the sample was homogenized in methanol using a laboratory blender. (1:10, sample: methanol). The homogenate was filtered to obtain the initial crude extract using about 20 ml of distilled water in separating funnel. The other layer was recovered and evaporated to dryness at low temperature ($35-50^{o}C$) in a vacuum desiccator. The dry extract was then saponified with 20 ml of ethanoic potassium hydroxide and left overnight in a dark cupboard. After one day, the carotenoid was taken up in 20 ml distilled water. The carotenoid extract (ether layer) was dried in a desiccator and treated with a light petroleum (Petroleum spart) and allowed to stand overnight in a freezer. The next day, the precipitated steroid was removed by centrifugation and the carotenoid extract evaporated to dryness in a desiccator and weighed.

The weight of carotenoid was determined and expressed as percentage of the sample weight.

2.2.1.3 Determination of Anti-nutritional Composition

Determination of phytates: The phytate content was determined by modified method of AOAC [22]. 0.5g of sample was extracted with 10 ml 2, 4% HCl for 1hr at ambient temperature and centrifuged (3000 rpm) for 30 min. The clear supernatant was used for phytate estimation. 1ml of wade reagent (0.03% solution of $FeCl_3. 6H_2O$ containing 0.3% sulfosalicylic acid in water) was added to 3 ml of the sample solution and the mixture was centrifuged. The absorbance at 500 nm 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.

$$Phytate\ (mg/100g) = \frac{(Absorbance-intercept)\times10}{(Slope\ \times density\ \times wt.sample)\times3}$$

Determination of tannins: The determination of tannins was conducted using modified method of AOAC [22]. The phenolic compound was first extracted with chloroform. 1 mL of the acetone phase was pipetted into a test tube marked No. 1 (n°1). 5 mL of distilled water was successively added together with 1 mL of ammonia, then shaken for a few seconds with the vortex mixer.

1 mL of the acetone phase was pipetted into a test tube marked No. 2 (n°2). 5 mL of distilled water was pipetted and 1 mL of the iron (III) ammonium citrate solution was also added. The solution was then shaken for a few seconds using the vortex mixer. Both solutions n° 1 and n° 2 were transferred into the measuring cuvettes (1 cm) and the absorbances were measured with a spectrophotometer at 525 nm against water, 10 min after the end of operations 1 and 2. The result is the difference in absorbances.

2.2.2 Determination of functional properties of the malted pigeon pea-orange fleshed sweet potato flour blends

The following functional properties was studied on the flours: Bulk density, Water absorption capacity, oil absorption capacity, foam capacity and swelling power.

2.2.2.1 Determination of bulk density, water absorption capacity and oil absorption capacity

Bulk density, water absorption and oil absorption capacity were determined as described by Onwuka [24].

The bulk density: Fifty grams (50g) of flour was poured into a 100 ml measuring cylinder and tapped to a constant volume.

$$\text{Bulk density} \left(\frac{\text{g}}{\text{cm3}}\right) = \frac{\text{weight of sample}}{\text{Volume of sample after tapping}}$$

Water absorption capacity: One gram of samples (flours) was mixed with 10 ml distilled water in a pre-weighed 20 ml centrifuge tube. The slurry was agitated for 2 min and allowed to stand at 28°C for 30 min and then centrifuged at 500 rpm for 20 min. The clear supernatant was decanted and discarded. The adhering drops of water in the centrifuge tube were wiped with cotton wool and the tube weighed. Percentage water absorption capacity was calculated.

Oil absorption capacity: One gram of samples (flours) was mixed with 10 ml refined palm oil in a pre-weighed 20 ml centrifuge tube. The slurry was agitated for 2 min, allowed to stand at 28°C for 30 min and then centrifuged at 500 rpm for 20 min. The clear supernatant was decanted and discarded. The adhering drops of oil in the centrifuge tube was removed with cotton wool and the tube weighed, the weight of oil absorbed

by 1 g of flour or protein was calculated and expressed as oil absorption capacity stock.

2.2.2.2 Determination of foam capacity and foam stability

The method described by Onwuka [18] was used for the determination of foam capacity and foam stability. Two (2) g of flour sample was added to 50 mL distilled water at 30 ± 2 °C in a 100 mL measuring cylinder. The mixture was stirred and the volume was noted. The suspension was blended using a warring blender (Model HGBTWT; Warring Commercial, Torrington, USA) at $160 \times g$ for 5 min to create foam, then returned to the measuring cylinder and the volume of the foam after 30 s was recorded. The foam capacity was expressed as a percentage increase in volume using the formula of Abbey and Ibeh [25]. Foam volume was recorded in 1 h after whipping to determine the foam stability as a percentage of the initial foam volume.

2.2.3 Physical properties of the malted pigeon pea-orange fleshed sweet potato flour blends biscuits

2.2 3.1 Weight and diameter of biscuits

Weight of biscuits was measured as average values of six individual biscuits with the help of an analytical weighing balance [26]. 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 [27]. An average of six values was taken for each set of samples. Average value for diameter was reported in millimeter.

2.2.3.2 Spread ratio and break strength

The spread ratio was determined using the method of Gomez et al. [28] Three rows of five well-formed biscuit was 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 [29]. Biscuit of known thickness (0.4cm) was placed between two parallel wooden bars (3.0cm apart). Weights was 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.

2.2.4 Sensory evaluation of the malted pigeon pea-orange fleshed sweet potato flour blends biscuits

The sensory characteristics of the prepared biscuits after baking and storage at room temperature were assessed in the Department of Food Science and Technology, Federal University, Wukari. The samples were evaluated for desirability in color, texture, taste, flavor and overall acceptability using a 9-hedonic scale test as described by Ayo et al [30], varying from 9 (which means like extremely) to 1 (which means dislike extremely). Water was used to neutralize the taste between samples testing.

2.2.5 Nutritional and hematological qualities albino rats fed with malted pigeon pea-orange fleshed sweet potato flour blends

2.2.5.1 Body Weight Measurement

The increase in body weight was measured each day for the 21 days study period with a standard weighing balance.

2.2.5.2 Feed Efficiency Ratio (FER) and Protein Efficiency Ratio (PER)

The feed efficiency ratio (FER) was mathematically calculated as follows using the recommendations of AOAC [22].

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

The protein efficiency ratio (PER) was also mathematically calculated as follows using the recommendations of AOAC [22]

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

2.2.5.3 Hematological properties of rats

The blood samples were collected from the rat tail veins by ocular method. The haematological properties (haemoglobin, packed cell volume, total white blood cell, red blood cell, white blood cell, lymphocytes, eosinophils, basophils and monocyte) of fed animal (rat) were determined using standard procedures.

Haemoglobin: The haemoglobin concentration was determined as described by Ayo, et al. [31].

5 ml of Cyanmethemoglobin reagent was pipetted into different labelled tubes. 20 μl of the appropriate sample was added into each tube. The tubes were allowed to stand for 10 minutes. The absorbance (A) was read in the spectrophotometer at 540 nm, zeroing the spectrophotometer with the Blank solution. The plot of Absorbance vs. Haemoglobin Concentration in grams % was done on linear graph paper.

Packed cell volume: Packed cell volume (PCV) was determined by the microhematocrit method [31]. Heparinised capillary tubes were ¾ filled with the blood sample and sealed at one end with plasticine. The capillary tubes were centrifuged using SH120 high speed microhematocrit centrifuge at 12,000 g for five minutes and then read using microhematocrit reader.

Red blood cell: Manual cell counts (RBC, nucleated cells or platelets) are performed using a haemocytometer. This is specifically done on body cavity fluids that are poorly cellular (<1000 cells/uL) because most automated analysers (impedance or laser-based) are insensitive to such low values. This is particularly the case with cerebrospinal fluid, where the red blood cell count should be low (unless there is haemorrhage into the fluid or concurrent blood contamination during collection).

White blood cell: Manual absolute WBC counts were determined in a hemacytometer chamber after lysis of red blood cells with 2% acetic acid. For differential counting, blood smears were fixed with 100% methanol for 5 minutes and then stained with Wright-Giemsa stain modified according to the manufacturer's recommendations [32].

Lymphocytes, Eosinophils, Basophils and Monocyte were identified by their staining properties under optical microscopy.

2.3 Statistical Analysis

All the analyses were conducted in duplicates in completely randomized design. The data were subjected to analysis of variance using Statistical Package for Social Science (SPSS) software version 23, 2017. Post-hoc analysis was conducted via the least significant difference (LSD) test. Significance was accepted at $p < 0.05$.

3. RESULTS AND DISCUSSION

3.1 Chemical Composition of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends

3.1.1 Proximate composition of malted pigeon pea – orange fleshed sweet potato flour blends

The proximate composition of malted pigeon pea– orange fleshed sweet potato flour blends is presented in Table 1. The flour replacement at 100:0, 95:5, 90:10, 85:15, 80:20 and 0:100 of malted pigeon pea and orange fleshed sweet potato flour blends significantly affected the moisture, ash, crude protein, crude fat, crude fibre, carbohydrate and energy values of the flour blends produced.

The moisture content of the flour blends decreased from 5.92 to 5.23 % with increase in malted pigeon pea in the composite flour. This is lower than the 12% recommended. The lowest moisture was observed in 0:100 (100% malted pigeon pea). "The values in the present study were lower than the moisture content of 5.70 to 8.57 % for cookies produced from composite flours of wheat, Bambara nut and orange fleshed sweet potato" [33]. "The low moisture content reported in this study indicates that the product could have high storage stability. Moisture content in food is very important because it enhances storage stability, and low moisture inhibits the survival and growth of microorganisms in food products" [34].

The ash content of the flour blends ranged from 2.34 to 3.11 % with increase in malted pigeon pea. The values were statistically the same ($p<0.05$). The highest ash content was observed in 0:100 (100% malted pigeon pea). The values for the ash content reported in this study were similar to those (1.93 -2.73%) reported in the work of Ayo and Gidado [20]. Ash content indicates the presence of mineral matter in food. The high ash content observed in 100% malted pigeon pea showed that the product could contain higher percentage of ash and will be a good source of minerals.

The crude protein content of malted pigeon pea – orange fleshed sweet potato flour blends increased from 4.64 to 13.27 % with increase in malted pigeon pea. The highest crude protein was observed in 0:100 (100% malted pigeon pea) and were significantly different ($p<0.05$).

The crude protein content of the flour blends increased with increasing proportions of malted pigeon pea, likely due to the high protein content of malted pigeon pea. Crude protein reduced upon addition of OFSP. Proteins play a part in the organoleptic properties of the sample and also act as a source of amino acids in the food. Ubbor et al. [33] reported "crude protein ranging from 9.62 to 11.93 % for cookies produced from composite flours of wheat, bambara nut and orange fleshed sweet potato".

The crude fat of the flour blends decreased from 4.95 to 4.21 % with increase in malted pigeon pea, and differed significantly ($p<0.05$). The lowest fat (4.21 %) was recorded in 0:100 (100 % malted pigeon pea). Bello et al. [35] reported higher fat (16.21 to 18.13 %) for cookies produced from sprouted sorghum, pigeon pea and orange fleshed sweet potato flour blends. However, the relatively low-fat content (4.21 - 4.95%) observed in this work could be an advantage in extending the shelf life, as possibility of development of rancid could be very low in comparisons to that observed by Bello et al [35]. Crude fat is an essential component of the tissues. It also acts as a flavour retainer and helps improve sensory properties of baked products [36].

The crude fibre content of malted pigeon pea – orange fleshed sweet potato flour blends decreased from 1.00 to 0.55 % with increase in malted pigeon pea ($p<0.05$). The highest crude fibre was observed in 100:0 (100% orange fleshed sweet potato). Higher crude fibre values (3.54 to 4.95 %) were reported in the work of Chinma et al. [1]. "The low crude fibre recorded in this research (0.6-1.0%) may be as result of the poor dietary fibre content in OFSP" as observed by Villanueva-c et al., (2003). "Crude fibre helps to lower the risk of constipation, it also serves as a useful tool in the control of oxidative processes in food products and as well as functional food ingredients which protect against cardiovascular disease and obesity" [37].

The carbohydrate content of the flour blends decreased from 80.53 to 70.96 % and was lowest in 0:100 (100% malted pigeon pea). The carbohydrate content reported in this study were higher than those reported in the work of Bello et al. [35] which ranged from 58.97 to 69.32 % for cookies produced from sprouted sorghum, pigeon pea and orange fleshed sweet potato flour blends.

Table 1. Proximate composition of malted pigeon pea – orange fleshed sweet potato flour blends

OFSP:MPP	Moisture (%)	Ash (%)	Crude Protein (%)	Crude Fat (%)	Crude Fibre (%)	Carbohydrate (%)	Energy Values (KJ)
100:0	5.92[a]±0.03	2.93[b]±0.05	4.64[d]±0.03	4.95[a]±0.00	1.00[a]±0.01	80.53[a]±0.05	379.83[c]±1.50
95:5	5.90[a]±0.01	2.89[b]±0.01	5.54[d]±0.03	4.85[a]±0.01	0.85[b]±0.01	80.05[a]±0.03	380.82[b]±0.15
90:10	5.83[b]±0.06	2.84[c]±0.01	6.44[c]±0.06	4.79[b]±0.01	0.70[b]±0.00	79.54[b]±0.65	381.81[b]±1.25
85:15	5.80[b]±0.01	2.83[c]±0.00	7.16[b]±0.01	4.35[c]±0.02	0.65[c]±0.00	78.11[c]±0.00	384.19[a]±0.65
80:20	5.86[ab]±0.03	2.34[d]±0.01	6.49[c]±0.04	4.28[d]±0.00	0.60[c]±0.01	79.08[b]±0.23	380.62[b]±1.45
0:100	5.23[c]±0.01	3.11[a]±0.04	13.27[a]±0.04	4.21[e]±0.01	0.55[d]±0.01	70.96[d]±0.03	380.57[b]±0.96

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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

Table 2. Phytochemical Composition of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends

OFSP:MPP	Flavonoid (mg/100g)	Saponin (mg/100g)	Carotenoid (mg/100g)	Phenol (mg/100g)
100:0	2.11[d]±0.04	6.61[d]±0.04	4.40[a]±0.01	0.07[a]±0.00
95:5	2.31[c]±0.02	7.62[c]±0.01	4.05[a]±0.02	0.06[ab]±0.02
90:10	2.33[c]±0.00	7.89[c]±0.02	3.40[b]±0.01	0.06[ab]±0.00
85:15	2.55[b]±0.01	8.63[b]±0.05	1.31[c]±0.02	0.06[cb]±0.01
80:20	2.37[c]±0.04	8.70[b]±0.01	1.09[d]±0.04	0.05[c]±0.01
0:100	3.43[a]±0.02	9.70[a]±0.01	0.98[e]±0.01	0.03[d]±0.00

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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

Table 3. Anti-nutritional Composition of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends

OFSP:MPP	Phytate (mg/100g)	Tannins (mg/100g)
100:0	33.74[e]±0.06	0.04[d]±0.01
95:5	34.95[e]±0.17	0.05[c]±0.00
90:10	41.22[d]±0.03	0.05[c]±0.00
85:15	42.94[c]±0.13	0.06[cb]±0.01
80:20	46.16[b]±0.28	0.07[b]±0.01
0:100	50.28[a]±0.03	0.08[a]±0.00

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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

The energy value of the flour blends ranged from 379.83 to 384.19 Kcal/100g with increasing level of malted pigeon pea. Energy value was lowest in 0:100 (100% OFSP). The energy values were significantly different ($p<0.05$) at all levels of substitution and increased as the level of malted pigeon pea was increased in the flour blends. The energy value of food helps to provide physiological functions of a human body.

3.1.2 Phytochemical Composition of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends

The phytochemical composition of malted pigeon pea – orange fleshed sweet potato flour blends is shown in Table 2. The flavonoid content of the malted pigeon pea – orange fleshed sweet potato flour blends increased from 2.11 to 3.43 mg/100g. Highest flavonoid was observed in 0:100 (100% malted pigeon pea). The result showed a significant ($p<0.05$) difference between the samples. This indicates that malted pigeon pea has low flavonoid content and thus decreased the flavonoid content of the flour blends. Flavonoids represent the most common and widely distributed group of plant phenolics and are among the most potent antioxidants [38].

The saponin content of the malted pigeon pea – orange fleshed sweet potato flour blends increased from 6.61 to 9.70 mg/100g and was highest in 0:100 (100% malted pigeon pea). The result showed a significant ($p<0.05$) difference between the samples. This indicates that malted pigeon pea has high saponin content and thus increased the saponin contents of the flour blends. Saponins are phytochemicals that have been shown to exhibit a wide spectrum of activity as antifungal and antibacterial agents. They help to lower blood cholesterol and inhibit cancer growth [39].

The carotenoids content of the malted pigeon pea – orange fleshed sweet potato flour blends decreased from 4.40 to 0.98 mg/100g and was highest in 0:100 (100% OFSP). A signsificant difference ($p<0.05$) was observed between the samples. This indicates that OFSP has high carotenoid content and thus addition of malted pigeon pea decreased the carotenoid contents of the flour blends. Carotenoids help to protect the body from disease and enhance the immune system [40].

The phenol content of the malted pigeon pea – orange fleshed sweet potato flour blends decreased from 0.07 to 0.03 mg/100g and was highest in 0:100 (100% OFSP). Samples were significantly different ($p<0.05$).

3.1.3 Anti-nutritional Composition of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends

Table 3 shows the anti-nutritional composition of malted pigeon pea – orange fleshed sweet potato flour blends. The phytate content of the flour blends increased from 33.74 to 50.28 mg/100g and was significantly different ($p<0.05$) between the samples. Highest phytate was recorded in 0:100 (100% malted pigeon pea). The result shows that the increase in phytate may result from the addition of malted pigeon pea. Bello et al. [35] reported lower phytate contents of 3.11-6.11 mg/g. Phytate is the primary storage form of both phosphate and inositol in plant seeds. Phytate also negatively impacts protein and lipid utilisation.

The tannin content of the flour blends increased from 0.04 to 0.08 mg/kg and were significantly different among the samples. Tannins were highest in 0:100 (100% malted pigeon pea). The increase was observed with increase in malted pigeon pea. The low tannin content reported in this study showed that the flour blends are good for consumption. Bello et al. [35] reported higher tannins in cookies produced from sprouted sorghum, pigeon pea and orange fleshed sweet potato flour blends (0.31 to 14.21 mg/kg). Tannins are water-soluble complexes and may be destroyed by heat and some may have leached into the boiling water. Tannins reduce bioavailability of minerals, decrease the digestibility and palatability of proteins and carbohydrates by forming insoluble complexes with them.

3.2 Functional Properties of Malted Pigeon Pea Orange Fleshed Sweet Potato Flour Blends Biscuits

Table 4 shows the functional properties of malted pigeon pea – orange fleshed sweet potato flour blends. The values of the functional composition of the flour blends were statistically different at $p<0.05$ and increased with increasing level of OFSP.

The water absorption capacity (WAC) of the flour blends decreased from 0.38 to 0.15% with increase in malted pigeon pea addition and the values were significantly different ($p<0.05$).

Highest WAC was observed in 100:0 (100% OFSP) and was lowest in 0:100 (100% malted pigeon pea). The values reported are higher than the water absorption capacity as reported by Agume et al. [41]. High WAC of flours suggests that the flours can be used in formulation of some foods such as sausage, dough, processed cheese and bakery products (Abu et al., 2006).

The bulk density capacity decreased from 0.73 to 0.46 g with increase in malted pigeon pea addition. The values for bulk density were significantly different (p<0.05). The bulk density of flour blend produced with 100% OFSP was the highest. Increase in malted pigeon pea thus reduced the bulk density of the flour blend. Bulk density provides information about packaging, transportation and digestion of the product. Low bulk density suggests high weight and require large packaging material. Low bulk density in food also helps to ease digestion and improve the release of nutrients in the body.

The swelling capacity of the flour blends increased from 1.60 to 3.65 % with increase in malted pigeon pea addition and the values differed significantly (p<0.05). 100% malted pigeon pea had the highest swelling capacity. Swelling capacity is regarded as a quality criterion in some good formulations such as bakery products and as thickeners for soups and puree [42].

The oil absorption capacity increased from 0.91 to 1.82% with addition of malted pigeon pea, with significant differences in the samples (p<0.05). 100% malted pigeon pea had higher oil absorption capacity than other flour blends. The oil absorption capacity denotes how much oil is bound to matrices in a particular food system, which could be used as the index of hydrophobicity of the food [43].

The foam capacity of the flour blends increased from 7.15 to 12.20% with addition of malted pigeon pea. 100% malted pigeon pea had higher foam capacity (12.20%) than other flour blends and there was no significant difference. Foam formation and stability depend on the type of protein, pH, processing method, viscosity and surface tension. This is important since the usefulness of whipping agents depend on their ability to maintain the whip as long as possible as applied in confectioneries and dairy products [44].

3.3 Physical Properties of Biscuits Produced from Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends Biscuits

The physical properties of biscuits produced from malted pigeon pea – orange fleshed sweet potato flour blends is shown in Table 5.

The weight of the flour blends biscuits ranged from 5.68 to 7.68 g with increase in the added malted pigeon pea. The weight increased with the level of flour substitution. The biscuit produced with 0:100 (100% malted pigeon pea) was the heaviest and bulkiest among the samples.

The diameter ranged from 11.30 to 12.04 cm with increase in the level of malted pigeon pea substitution. The biscuit produced with 100:0 (100% OFSP) had the highest diameter. The thickness increased from 4.03 to 5.24 cm with increase in the level of malted pigeon pea substitution. The result revealed that the highest thickness was observed in biscuit produced with 0:100 (100% malted pigeon pea).

The break strength ranged from 2000.00 to 3000.00 g with increase in the level of malted pigeon pea substitution in the biscuit. The highest break strength was observed in biscuit produced with 85:15 (85 % orange fleshed sweet potato, 15 % malted pigeon pea flour blends). 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 2.99 to 2.27 cm with increase in the level of malted pigeon pea substitution. Biscuit produced with 100:0 (100% OFSP) showed the highest spread ratio (2.99). The decrease could be as a result of relatively high fat content and low starch content of malted pigeon pea.

3.4 Nutritional Quality of Rats Fed with Malted Millet-Orange Fleshed Sweet Potato Flour Blends

3.4.1 Cumulative feed intake and weight gain of rats fed with malted millet-orange fleshed sweet potato flour blends

The cumulative feed intake and weight gain of rats fed with malted millet-orange fleshed sweet potato flour blends are shown in Table 6. The feed intake increased from 125.30 to 190.90, 170.00 to 398.70, 221.70 to 457.50 and 259.75

to 500.30 with increase in malted pigeon pea at 0, 5, 10 and 15% respectively and thereafter decreased at above 15%. This is an indication that increase in malted pigeon pea affected the cumulative feed intake of the albino rats and this increase could be due to taste and flavour of the malted pigeon pea added. The weight gain increased from 125.30 to 110.80, 170.00 to 398.70, 221.70 to 457.50 and 259.75 to 500.30g with increase in malted pigeon pea at 0, 5, 10 and 15% respectively and thereafter decreased at above 15%. The increase in weight gain may be attributed to the high amount of feed consumed by the albino rats over the duration of 21 days. Albino rats fed 85:15 (85% orange fleshed sweet potato, 15% malted pigeon pea flour blends) had higher cumulative feed intake (500.30g) and had higher cumulative weight gain (1240.10g).

3.4.2 Hematological parameters of albino rats fed with malted pigeon pea – orange fleshed sweet potato flour blend biscuits

The haematological properties of albino rats fed with malted pigeon pea – orange fleshed sweet potato flour blend biscuits are shown in Tables 7 and 8. Haematological parameters such as Haemoglobin (HB), Packed Cell Volume (PCV), Total White Blood Cell (TWBC), Red Blood Cell (RBC), White Blood Cell (WBC), Lymphocytes (L), Eosinophils (E), Basophils (B) and Monocytes (M) were investigated in this study. The effect of malted pigeon pea on these haematological parameters of albino rats are significant ($p < 0.05$).

The result showed that at day 5, HB, PCV, TWBC, RBC, Lymphocytes, Eosinophils increased from 7.6 to 9.6 g/dl, 24 to 30%, 68000 to 188000 L-1, 2800000 to 3480000 L-1, 34 to 37 (x 109L-1), 1 to 6 (x 109L-1), respectively as a result of malted pigeon pea substitution. WBC decreased from 63 to 51 L-1. Basophils and Monocytes values were not affected as the level of malted pigeon pea was increased.

At day 10, HB, PCV, RBC, WBC increased from 7.4 to 9.8 g/dl, 23 to 31%, 2780000 to 3680000 L-1, 47 to 56 L-1. TWBC, Lymphocytes, Eosinophils and Basophils values decreased from 144000 to 122000 L-1, 49 to 40 (x 109L-1), 3 to 2 (x 109L-1) and 1 to 2 (x 109L-1) respectively as a result of OFSP substitution. Monocyte values changed as the level of malted pigeon pea was increased (1 to 2).

At day 15, HB, PCV, RBC, Lymphocytes, Eosinophils increased from 9.8 to 11.4 g/dl, 31 to 37%, 3800000 to 4180000 L-1, 30 to 38 (x 109L-1), 1 to 3 (x 109L-1), respectively with increase in malted pigeon pea. TWBC, WBC decreased from 98000 to 85000 L-1, 68 to 57 L-1. Basophils value was observed in 5% OFSP and 100% OFSP and was not recorded in other feed treatment. Monocytes values increased from 1 to 2 (x 109L-1) and later reduced with increase in malted pigeon pea.

At day 21, HB, PCV, TWBC, RBC, Lymphocytes increased from 7.4 to 7.8 g/dl, 24 to 26%, 81000 to 102000 L-1, 2820000 to 2800000 L-1, 34 to 39 (x 109L-1), respectively. WBC, Eosinophils and Basophils values decreased from 63 to 59 L-1, 2 to 1 (x 109L-1), respectively as a result of malted pigeon pea substitution. Monocytes values were the same with increase in malted pigeon pea.

Haemoglobin (HB) values reported in this work increased over the period of 21 days as the level of malted pigeon pea was increased. The HB values reported in this study were lower than the recommended HB range of 10-15g/dl documented in Merck Manual [45]. Hemoglobin is the protein inside red blood cells [46,47]. Haemoglobin is the iron-containing oxygen-transport metalloprotein in red blood cells of almost all vertebrates as well as the tissues of some invertebrates (Reference?). Low hemoglobin levels may be a symptom of several conditions, including different kinds of anemia and cancer [48,47]. Iron deficiency anemia in dogs and cats has been reported to be caused by chronic blood loss [49,46].

Packed Cell Volume (PCV) values increased over the period of 21 days as the level of malted pigeon pea was increased. The PCV values reported in this study were lower than the recommended PCV range of 30-45% documented in Merck Manual [45]. Packed cell volume (PCV) is a measurement of the proportion of blood that is made up of red blood cells (Anon, 2023a). Generally, in females, the normal range is 35.5 to 44.9%. and in males, 38.3% to 48.6% is the normal PCV range (Aon, 2023b). Anemia as indicated by the PCV of infected animals is very important in the symptomatology of blood scavenging parasites [50]. Dehydration is one of the most common causes of elevated PCV levels [51,46,48]. With adequate fluid intake, the levels return to normal, but it can also cause polycythemia, a condition in which there are more red blood cells than normal [47].

Table 4. Functional properties of malted pigeon pea – orange fleshed sweet potato flour blends

OFSP:MPP	Water Absorption (%)	Bulk Density (g/cm³)	Swelling Capacity (%)	Oil Absorption (%)	Foaming Capacity (%)
100:0	0.38ᵃ±0.02	0.73ᵃ±0.00	1.60ᶜᵇ±0.28	0.91ᵈ±0.21	7.15ᶠ±0.21
95:5	0.34ᵇ±0.00	0.65ᵇ±0.00	2.00ᶜᵇ±0.28	0.91ᵈ±0.21	7.20ᶠ±0.28
90:10	0.32ᵇ±0.00	0.63ᶜ±0.00	2.10ᶜᵇ±0.28	1.34ᵇ±0.21	8.15ᵉ±0.21
85:15	0.30ᶜ±0.00	0.61ᵈ±0.00	2.40ᶜᵇ±0.28	1.80ᵃ±0.28	9.20ᵈ±0.28
80:20	0.28ᵈ±0.00	0.52ᵉ±0.00	2.50ᶜᵇ±0.28	1.80ᵃ±0.28	10.20ᶜ±0.28
75:25	0.19ᵉ±0.00	0.51ᶠ±0.00	2.75ᵃᵇ±0 .21	1.81ᵃ±0.28	11.20ᵇ±0.28
50:50	0.15ᶠ±0.00	0.51ᶠ±0.00	3.60ᵃ±0.85	1.81ᵃ±0.28	11.20ᵇ±0.28
0:100	0.15ᶠ±0.00	0.46ᵍ±0.00	3.65ᵃ±0.35	1.82ᵃ±0.28	12.20ᵃ±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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

Table 5. Physical properties of malted pigeon pea – orange fleshed sweet potato flour blends biscuits

OFSP:MPP	Weight (g)	Diameter (cm)	Thickness (cm)	Break Strength (g)	Spread Ratio
100:0	5.83ᶜ±0.42	12.04ᵃ±0.06	4.03ᵇ±0.04	2500.00ᵃᵇ±0.00	2.99ᵃ±0.04
95:5	6.10ᵇ±0.05	11.90ᵃ±0.05	4.03ᵇ±0.10	2500.00ᵃᵇ±0.00	2.95ᵃ±0.09
90:10	6.37ᵇ±0.12	11.75ᵃᵇ±0.04	4.04ᵇ±0.06	2250.00ᵇ±353.55	2.91ᵃ±0.04
85:15	5.68ᶜ±0.25	11.30ᶜ±0.35	4.14ᵇ±0.20	3000.00ᵃ±0.00	2.74ᵇ±0.22
80:20	6.13ᵇ±0.04	11.38ᶜ±0.11	4.25ᵇ±0.42	2500.00ᵃᵇ±707.11	2.69ᵇ±0.24
0:100	7.68ᵃ±0.63	11.86ᵃᵇ±0.22	5.24ᵃ±0.13	2000.00ᶜ±0.00	2.27ᶜ±0.09

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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

Table 6. Cumulative feed Intake and Weight Gain of albino rat fed with malted pigeon pea-orange fleshed sweet potato

	OFSP:MPP			
DAYS	5	10	15	21
100	125.30ᵈ±0.14 (309.20ᵉ±0.14)	170.00ᵉ±0.14 (546.80ᵉ±0.14)	221.70ᵉ±0.28 (453.45ᵃ±0.72)	259.75ᵉ±0.21 (1159.85ᶜ±0.78)
95:5	133.20ᶜ±1.41 (306.05ᵈ±0.21)	233.40ᶜ±2.12 (590.70ᵈ±0.28)	260.80ᵈ±0.14 (790.70ᵃ±0.28)	279.10ᵈ±0.70 (1003.70ᵈ±0.28)
90:10	159.00ᵇ±0.70 (324.70ᵃ±0.28)	266.50ᵇ±0.42 (618.80ᵃ±0.42)	349.30ᵇ±0.28 (896.65ᵃ±0.07)	445.10ᵇ±0.70 (1204.35ᵉ±0.64)
85:15	190.90ᵃ±0.70 (322.70ᵇ±0.14)	398.70ᵃ±0.70 (598.15ᵇ±0.35)	457.50ᵃ±0.70 (892.60ᵃ±0.57)	500.30ᵃ±0.28 (1240.10ᵃ±0.14)
100	110.80ᵉ±0.14 (302.05ᶜ±0.21)	202.70ᵈ±0.14 (591.95ᶜ±0.07)	270.35ᶜ±0.21 (871.95ᵃ±0.07)	328.95ᶜ±0.78 (1208.60ᵇ±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. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea
*Weight gain are in parenthesis (brackets)

Table 7. Hematological parameters of albino rats fed with malted pigeon pea – orange fleshed sweet potato flour blend biscuits

		OFSP-MMP					
Day	Parameter	100:0	95:5	90:10	85:15	80:20	0:100
5	HB(g/dl)	9.75[a]±0.21	9.50[a]±0.02	8.85[c]±0.07	9.25[b]±3.54	9.25[b]±0.35	7.65[a]±0.07
	PCV (%)	30.50[a]±0.71	29.45[a]±0.64	28.25[b]±0.35	28.45[b]±0.64	29.25[ab]±0.35	24.25[a]±0.35
	TWBC(L^{-1})	188250.00[e]±353.55	108550.00[e]±33.55	82025.00[b]± 35.36	168025.00[d]± 35.36	92025.00[c]± 35.36	68250.00[a]±353.55
	RBC(L^{-1})	3482500.00[a]±3535.53	3245500.00[a]±3435.04	3025000.00[c]±7071.07	3130000.00[c]±14142.14	3030000.00[b]±0.00	2825000.00[c]±35355.34
	WBC(L^{-1})	50.00[c]±1.41	56.47[c]±0.45	66.50[a]±0.71	64.00[a]±1.41	60.00[b]±1.41	64.00[b]±1.41
10	HB (g/dl)	9.85[d]±0.07	8.85[d]±0.14	7.35[a]±0.07	8.30[c]±0.14	7.65[b]±0.07	7.30[a]±0.14
	PCV (%)	31.00[c]±0.00	28.50[c]±0.35	23.50[a]±0.71	26.25[b]±0.35	24.25[a]±0.35	23.25[a]±0.35
	TWBC(L^{-1})	122250.00[d]±353.55	105250.00[d]±353.55	61250.00[a]±353.55	86250.00[b]±353.55	87400.00[c]±141.42	144250.00[e]±353.55
	RBC(L^{-1})	3682500.00[e]±3535.53	3422500.00[e]±3535.53	2842500.00[c]±3535.53	3162500.00[d]±3535.53	2740000.00[a]±14142.14	2782500.00[b]±3535.53
	WBC(L^{-1})	57.00[b]±1.41	60.50[b]±0.70	69.50[d]±0.71	65.50[c]±2.12	59.50[b]±0.71	48.00[a]±1.41
15	HB(g/dl)	11.60[c]±0.28	11.20[c]±0.14	9.55[a]±0.07	10.20[b]±0.28	10.90[b]±0.14	9.85[ab]±0.07
	PCV (%)	37.50[c]±0.71	35.25[c]±0.71	31.25[a]±0.35	33.25[b]±0.35	34.25[b]±0.35	30.75[a]±0.35
	TWBC(L^{-1})	85250.00[b]±353.55	85250.00[b]±353.55	208500.00[a]± 707.11	72250.00[e]±353.55	127750.00[d]±353.55	98250.00[c]±353.55
	RBC(L^{-1})	4177500.00[e]±3535.54	4177500.00[e]±3535.54	3965000.00[b]±7071.07	4015000.00[c]±7071.07	4060500.00[d]±707.17	3825000.00[a]±35355.34
	WBC(L^{-1})	57.50[b]±0.71	57.50[b]±0.71	46.00[a]±1.41	62.00[c]±1.41	58.50[b]±0.71	68.50[d]±0.71
21	HB(g/dl)	7.90[b]±0.14	7.90[b]±0.14	7.15[a]±0.07	9.55[c]±0.21	8.10[b]±0.14	7.45[a]±0.07
	PCV (%)	22.50[ab]±2.12	22.50[ab]±2.12	21.50[a]±2.12	31.00[a]±0.41	26.50[b]±0.71	24.50[ab]±0.71
	TWBC(L^{-1})	102250.00[d]±53.55	102250.00[d]±353.55	124250.00[e]± 353.55	67500.00[a]±707.11	94500.00[c]±707.11	81250.00[b]±353.55
	RBC(L^{-1})	2850000.00[b]±70710.68	2850000.00[b]±70710.68	2682500.00[a]±3535.53	3842500.00[d]±3535.53	3292500.00[a]±3535.53	2825000.00[b]±7071.07
	WBC(L^{-1})	60.00[ab]±1.41	60.00[ab]±1.41	59.50[ab]±0.71	54.00[a]±1.41	65.00[b]±7.07	64.00[b]±1.41

Values are mean ± 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. HB = Hemoglobin, PCV = Packed Cell Volume, TWBC = Total White Blood Cell, RBC = Red Blood Cell, WBC = White Blood Cell. OFSP-Orange fleshed Sweet Potato, MPP- Malted Pigeon Pea

Table 8. Hematological Parameters of Albino Rats fed with Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blend Biscuits

Day	Parameter	OFSP:MMP 100:0	95:5	90:10	85:15	80:20	0:100
5	Lymphocytes (%)	38.00[a]±1.41	35.50[b]±0.71	31.50[c]±0.71	29.50[d]±0.71	35.00[b]±1.41	34.50[c]±0.71
	Eosinophils (%)	6.00[a]±0.00	6.00[a]±0.00	1.00[d]±0.00	4.00[b]±0.00	3.00[c]±0.00	1.00[d]±0.00
	Basophils (%)	1.00[a]±0.00	1.00[a]±0.00	0.00±0.00	0.00±0.00	1.00[a]±0.00	1.00[a]±0.00
	Monocytes (%)	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00
10	Lymphocytes (%)	40.50[b]±0.71	38.50[c]±0.71	30.50[e]±0.71	36.00[d]±2.83	36.50[d]±0.71	49.50[a]±0.71
	Eosinophils (%)	2.35[b]±0.50	2.05[c]±0.50	1.25[d]±0.35	1.00[d]±0.00	2.25[b]±0.35	3.25[a]±0.35
	Basophils (%)	2.25[a]±0.35	2.15[b]±0.35	0.00[d]±0.00	1.25[c]±0.35	2.15[b]±0.21	0.00[d]±0.00
	Monocytes (%)	0.00[b]±0.00	0.00[b]±0.00	0.00[b]±0.00	0.00[b]±0.00	1.25[a]±0.35	1.25[a]±0.35
15	Lymphocytes (%)	39.50[c]±2.21	39.50[c]±2.21	45.00[a]±1.41	37.00[d]±1.41	41.00[b]±1.41	30.50[e]±0.71
	Eosinophils (%)	2.85[c]±0.21	2.85[c]±0.21	4.10[a]±0.14	3.00[b]±0.00	1.10[d]±0.14	1.25[d]±0.35
	Basophils (%)	1.25[a]±0.21	1.25[a]±0.21	1.00[b]±0.00	0.00[c]±0.00	1.00[b]±0.00	0.00[c]±0.00
	Monocytes (%)	1.00[c]±0.00	1.00[c]±0.00	2.30[a]±0.42	0.00[d]±0.00	1.00[c]±0.00	1.25[b]±0.35
20	Lymphocytes (%)	39.50[b]±0.71	39.50[b]±0.71	38.50[b]±0.71	46.00[a]±1.41	27.00[d]±1.41	34.50[c]±0.71
	Eosinophils (%)	1.10[d]±1.14	1.10[d]±1.14	2.25[b]±0.35	1.25[c]±0.35	3.25[a]±0.35	2.10[b]±0.14
	Basophils (%)	0.00[b]±0.00	0.00[b]±0.00	0.00[b]±0.00	0.00[b]±0.00	1.25[a]±0.35	0.00[b]±0.00
	Monocytes (%)	1.00[a]±0.00	1.00[a]±0.00	1.00[a]±0.00	1.00[a]±0.00	1.00[a]±0.00	1.00[a]±0.00

Values are mean ± 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 9. Sensory evaluation of malted pigeon pea – orange fleshed sweet potato flour blend biscuits

OFSP:MPP	Colour	Mouth feel	Aroma	Texture	Taste	General Acceptability
100:0	7.60[a]±1.19	7.00[a]±1.62	7.20[a]±0.89	7.55[a]±1.64	7.50[a]±1.36	7.70[a]±1.08
95:5	6.90[b]±1.22	6.97[a]±1.12	7.20[a]±1.62	7.15[b]±1.64	7.40[a]±1.64	7.30[b]±1.15
90:10	6.70[c]±1.64	6.95[a]±1.36	7.00[b]±1.12	6.55[c]±1.96	6.80[b]±1.58	7.20[b]±1.28
85:15	6.25[d]±1.33	6.45[c]±1.67	5.80[c]±1.64	6.45[c]±2.09	6.10[c]±1.71	6.60[c]±1.23
80:20	6.95[b]±1.23	6.25[c]±1.74	5.70[c]±1.66	5.90[d]±1.68	5.60[d]±2.52	6.35[d]±1.50
0:100	6.05[e]±2.52	4.75[b]±2.90	4.60[d]±2.21	5.20[d]±2.09	4.70[e]±2.68	5.70[a]±2.96

Values are Average Means scores ± standard deviation of sensory quality of twenty panellists. Means within each column not followed by the same superscript are significantly different (p<0.05) from each other using Duncan multiple range test. PPPP = 100:0, OFPP = 90:10, FSPP = 85:15, SPPP = 80:20 and OFSP = 0:100 malted pigeon pea – orange fleshed sweet potato flour blends

Red Blood Cells (RBC) values in this study increased over the period of 21 days as the level of malted pigeon pea was increased. The RBC values reported in this study were lower than the recommended RBC range of 5.0–10.0x106/mm3 documented in Merck Manual [45]. Generally, a normal range for RBC is considered to be 4.3-5.9x1012/L for male, 3.5-5.5x1012/L for female (Anon, 2023b) and 11.0-19.2g/dl. for rats (Anon, 2023c). RBC are the most common type of blood cells and the vertebrate's principal means of delivering oxygen to the body tissues—via blood flow. Red blood cells (erythrocytes) serve as a carrier of haemoglobin [46]. It is this haemoglobin that reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration [52].

Total White Blood Cell (TWBC) values increased at day 5 and day 21 of feeding the rats with increased malted pigeon pea level. However, at day 10 and 15, it decreased with increase in malted pigeon pea substitution. The decrease could be due to decrease in the feed efficient ratio. White Blood Cell (WBC) values decreased at day 5, increased at day 10 then decreased at day 15 and 21. WBC is type of blood cell that is made in the bone marrow and found in the blood and lymph tissue [47,42,48]. A white blood cell (WBC) count of less than 4x109/L indicates leukopenia [47,48]. A WBC count of more than 11x109/L indicates leucocytosis [48]. White blood cells function to fight infections, defend the body by phagocytosis against invasion by foreign organisms [48,47]. Animals with low white blood cells are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to diseases [55,56].

Lymphocytes (L) values increased at day 5, decreased at day 10 then decreased at day 15 and 21 which could be due to presence of antinutrient in the feed. Lymphocyte is a type of white blood cell in the immune system of most vertebrates and are responsible for both humoral and cellular immunity [53]. These cells help protect the body from infection [47]. When lymphocyte levels are low, there is higher risk of infection [53,54].

Eosinophils (E) values increased at day 5, decreased at day 10 then decreased at day 15 and 21 which could be due to presence of antinutrient in the feed. In general, an increase in Eosinophils was observed with increase in malted pigeon pea. Eosinophils are a variety of white blood cells and one of the immune system components responsible for combating multicellular parasites and certain infections in vertebrates [57]. High number of eosinophils can be an emergency and if untreated, may cause damage to multiple organs [57].

Basophils (B) values decreased over the period of 21 days as the level of malted pigeon pea was increased. The variation was significant. Basophils release enzymes to improve blood flow and prevent blood clots [54]. A basophil count that's too high may be a sign of an infection or a more serious medical condition like leukemia or autoimmune disease.

Monocytes (M) values were not affected over the period of 21 days as the level of malted pigeon pea was increased. A monocyte is a type of white blood cell and a type of phagocyte [46,51]. A normal monocyte count is between 2% and 8% of your white blood cell count [48]. Low levels of monocytes indicate that the body is more susceptible to infection [51].

The high concentration of HB, PCV, TWBC, WBC, and RBC of the experimental rats fed on 100% OFSP further established nutritional quality of these products. Low HB in rats may lead to poor production of hemoglobin and, hence, could cause anemia [58]. The low hemoglobin concentration could be due to poor utilization of the minerals like iron due to antinutrients Generally, haematological factors are affected by several factors which include physiological, environmental condition, dietary content, fasting, age, administration of drugs, anti-aflatoxin treatment and continuous supplementation of vitamin affect the blood profile of healthy animal [46]. Others include health of the animal, degree of physical activity, sex, breeds of animal, diseases and stress factors, climate, geographical location, season, day length, time of day, life habit of species, oestrus cycle, and pregnancy [54].

3.5 Sensory Evaluation of Malted Pigeon Pea – Orange Fleshed Sweet Potato Flour Blends Biscuits

Sensory properties of biscuits produced with malted pigeon pea – orange fleshed sweet potato flour blends is shown in Tables 4.9. Mean hedonic scores for all sensory attributes of the biscuits decreased with increasing addition of malted pigeon pea. The average mean score for colour, mouth feel, aroma, texture and taste

decreased from 7.60 to 6.05, 7.00 to 4.75, 7.20 to 4.60, 7.55 to 5.20 and 7.50 to 4.70 with increase in malted pigeon pea. Colour and taste are important attributes that enables the acceptability of food products. The decrease in colour and taste of the flour blends could be due to inherent attractive colour imparted by the carotenoid in the orange fleshed sweet potato [59].

The general acceptability of the flour decreased from 7.70 to 5.70 and were highest in flour produced with 100% OFSP. The overall acceptability score reported in this work were similar to those of 5.70 to 7.80 reported by Wadikar et al. [60]. This thus indicates that biscuits produced with 100% OFSP were therefore the most preferred biscuits. Biscuit produced with 90:10 (90% orange fleshed sweet potato, 10% malted pigeon pea) also had good acceptability of 7.20.

4. CONCLUSION

Suitable biscuits can be made from malted pigeon pea-orange fleshed sweet potato flour blends. However, 85:15 was the preferred ratio because of improved physical properties. The addition of malted pigeon pea improved FER, PER. The eating quality and weight gain have generalized the result to show a positive relationship between addition of malted pigeon pea and weight gain. The non-improvement in most of the properties assessed could be due to non-availability of the intended minerals such as iron and this could be due to the presence of antinutrient in the feed. The use of pigeon pea to improve baking products for healthy consumption by consumers may need to be subjected to further pre-treatments to destroy the possible ant nutrients.

The study recommends the utilization of malted pigeon pea – orange fleshed sweet potato blend flours for domestic and industrial use and the use of 85:15 (85% orange fleshed sweet potato, 15% malted pigeon pea flour blends) in diet.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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