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# Production of Tuwo from Sorghum-Cowpea and Millet-Cowpea Composite Flours: Assessment of Flour Quality Parameters and the Textural Properties and Acceptability of Tuwo

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## ABSTRACT

*Sorghum-Cowpea and Millet-Cowpea composite flours were produced at 70:30 cereals to legume ratios. Functional properties, proximate composition, amino acids profile and protein quality of the flours were determined. The flours were then used to produce tuwo (a stiff porridge made from flours of cereal grains, consumed in different forms across Africa and Asia), and the textural properties and acceptability of the tuwo were evaluated. Cowpea addition at 30% has significantly increased Water Absorption Capacity, Least Gelling Concentration and Gelatinisation Temperature but has no significant effect on Swelling Capacity and Bulk Density. The treatment has significantly increased the ash and protein contents but has no significant effect on fat content. Curiously, Carbohydrate content and Energy Value were significantly reduced. Methionine, tryptophan and valine contents of the samples were significantly increased by 30% cowpea addition.*

*Protein Efficiency Ratio (PER), Essential Amino Acids Index (EAAI), Biological Value (BV) and Nutritional Index (NI) all showed significant increases at 30% cowpea addition. The textural properties of the tuwo samples showed remarkable trends. The cooling time of tuwo and its index of hardness correlated positively ( $r < 0.9$ ) and fit well into a linear polynomial curve.*

*The hardness of tuwo at any temperature can be estimated from the linear equation with good accuracy. The overall acceptability of sorghum and millet tuwo with 30% cowpea addition did not show any significant variation from control samples indicating that tuwo made from*

*complemented cereal flours is acceptable to the consumer.*

**Keywords:** tuwo, composite flours, functional properties, tuwo texture.

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## I. INTRODUCTION

The term complementation is used with respect to proteins when the relative deficiency of an amino acid in one is compensated by a surplus from another protein consumed at the same time. A combination of cereals and legumes where one complements the other with the deficient amino acid creates mutual balance resulting in nutritional complementation. The protein quality is greater than either protein source used alone.

This principle of nutritional complementarities of cereal and legume proteins seems to have been discovered empirically on all continents; rice with soybean in South-East Asia, sorghum or millet plus chickpea or pigeon pea in South Asia, and rice with cowpea in tropical Africa (Hulse, 1991).

Chemical analysis has shown that the amino acids deficient in the legume are generally adequately compensated by the protein of cereals and vice versa. The mutual compensation is closest to ideal when the ratio by mass of cereal to legume is roughly 70:30, in which proportion each provides about equal parts by mass of protein. Eating cereal and legumes together in this proportion has

been demonstrated to have greater complementary benefit than if the two are eaten in the similar quantitative proportions, but at different meal times (Hulse, 1991). The nutritional response of combining two proteins has been classified into four groups using rat bioassay in which *Type III* is reported to have the true complementary effect because one protein source has considerably higher concentration of the most limiting amino acid than in the other, producing a synergistic effect such as maize and soy or sorghum and soy (Bressani *et al.*, 1972).

Sorghum and millet are rated as poor cereal grains due to their high levels of anti-nutritional factors like phytic acid, polyphenols, and tannins that readily forms complexes with monovalent and multivalent cations of potassium, calcium, iron, zinc, magnesium and other cations, reducing their bioavailability and creating a deficit in their absorption (Boncompagni, *et al.*, 2018). This is in addition to the fact that they are rich in the essential sulphur-containing amino acids, methionine and cystine but deficient in lysine.

Therefore the combination of cereal grains and legumes in traditional food preparation complement each other since the latter are found to be rich in lysine but deficient in both methionine and cystine (Nkama and Sopade, 1990). Thus, the cereal-legume blends serve as major sources of calorie, proteins, minerals and vitamins. Consequently, the complementation of the grain legumes has been suggested as one way of improving the protein quality of cereal based diets (Nkama, *et al.*, 1995). Production of shelf stable maize-cowpea composite flour can increase the availability of the product for wider application in many cereal-based traditional foods like *tuwo*, *danwake*, *masa* etc. The evaluation of the functional properties, proximate composition, amino acid profile and protein quality of sorghum-cowpea and millet-cowpea composite flours can contribute to general efforts for traditional food products development and industrialisation drive for the traditional food sub-sector.

The regional ICRISAT West African trials of the International Sorghum Food Quality Trials found

four important quality criteria for *tuwo* to include taste, colour, texture and overnight keeping quality (Scheuring *et al.*, 1982). However, consumption temperature and aroma are equally regarded as quality factors. *Tuwo* is normally served immediately after preparation as hot as it were, though it may be allowed to stay in the open dish or plate for some minutes under the prevailing ambient condition for the temperature to drop enough to be comfortably cut with the bare hand. This cooling process, as insignificant as it may seem, is an important aspect of consideration in *tuwo* processing as it determines the final eating texture of the product. Upon cooling the gelatinized starch of the *tuwo* undergo retrogradation and also losses moisture which reduce the stickiness of the product and increase the *Tuwo* hardness. The desired final *tuwo* hardness determines the water: flour ratio to be mixed during production, taking into consideration the moisture loss during cooling and the subsequent texture modification.

However, research on the cooling behaviour of *tuwo* was not given enough attention probably due to fact that *tuwo* preparation is still a household process that is yet to be industrialized.

*Tuwo* texture preference depends to a large extend on individual choice and community practice. However, people generally tend to prefer non-sticky *tuwo* with moderate hardness. The gel should be stiff, but not dense. A person should be able to dip in his fingers, scoop out a piece and readily manipulate the piece with the forefingers and thump without the gel adhering to the fingers (Scheuring, 1982).

The colour of *tuwo* is normally the colour of the grain and the method adopted for preparing the flour. However white colour *Tuwo* is generally considered attractive and qualitative. In some practices, spent steep water is used to improve the white colour appearance by soaking the grains overnight. The taste of *tuwo*, like its colour, depends on the grain from which it is prepared and the method of flour preparation. Fermented grains tend to produce sour taste while *tuwo* prepared with added ash leachate gives characteristic tardy taste. Generally bland taste is considered most acceptable. *Tuwo* aroma is an

important consideration in the processing. In some cases the formed gel is allowed to stay on fire until it starts caking from the bottom of the pot. The resulting caramel flavor of the caramelized starch adds to the aroma of the *tuwo*.

## II. MATERIALS AND METHODS

### 2.1 Preparation of Composite Flours

Three kilograms each of sorghum and millet grains were tempered with water using a quantity of 3% (v/w) followed by decortication of the grains in commercial dehulling machine (previously cleaned), where the germs and hulls of the grains were removed. The decorticated grains were aspirated manually to remove adhering hulls and then ground into flour using a Laboratory Hammer mill. Cowpea flour was produced by first cleaning and sorting the cowpea sample. The cleaned sample was then soaked in water at room temperature for 1hr and subjected to thermal treatment at 80°C for 20 minutes in a steam bath. The seeds were manually dehulled and dried in a solar dryer. The dried seeds were then dry-milled using a Laboratory Hammer mill.

Complementation was carried out at cereal to cowpea ratio 70:30 as recommended by Nkama, 1993. The right quantities each of the sorghum and millet flours were thoroughly mixed with the right quantities of cowpea flours using a laboratory mixer and sieved using a standard sieve with 300µm aperture and then kept in airtight polythene bags until needed.

### 2.2 Determination of Functional Properties

**Determination of Water Absorption Capacity (WAC):** Water absorption capacity was determined using the method of Sathe and Salunkhe (1981) with slight modifications. 10 ml of distilled water was added to 1.0 g of the sample in a beaker. The suspension was stirred using a glass stirrer for 5 min. The suspension obtained was thereafter centrifuged at 3555 rpm for 30 minutes and the supernatant measured in a 10 ml graduated cylinder. The density of water was taken as 1.0 g/cm<sup>3</sup>. Water absorbed was calculated as the difference between the initial

volume of water added to the sample and the volume of the supernatant.

**Determination of Least Gelling Concentration (LGC):** The least gelation concentration was determined by the method of Sathe *et al.* (1981). Test tubes containing suspensions of 2, 4, 6, 8 up to 20% (w/v) flour in 5 ml distilled were heated for 1 h in boiling water, followed by cooling in ice and further cooling for 2 h at 40C. The least gelation concentration was the one at which the sample did not fall down or slip when the test tube was inverted.

**Determination of Gelatinization Temperature (GT):** GT was determined according to the method described by Shinde (2001). 1 g of flour sample was weighed accurately in triplicate and transferred to 20 ml screw capped tubes. 10 ml of water was added to each sample. The samples were heated slowly in a water bath until they formed a solid gel. At complete gel formation, the respective temperature was measured and taken as gelatinization temperature.

**Determination of Swelling Capacity (SC %):** This was determined with the method described by Leach *et al.* (1959) with modification for small samples. One gram of the flour sample was mixed with 10 ml distilled water in a centrifuge tube and heated at 80°C for 30 min. This was continually shaken during the heating period. After heating, the suspension was centrifuged at 1000×g for 15 min. The supernatant was decanted and the weight of the paste taken. The swelling power was calculated as:

Swelling power = weight of the paste / weight of dry flour.

**Determination of Bulk Density:** This was carried out using the procedure of Narayana and Narasinga (1984). A specified quantity of the flour sample was transferred into an already weighed measuring cylinder ( $w_1$ ) and gently tapped to eliminate spaces between the flour. The level was noted to be the volume of the sample and then weighed ( $W_2$ ). The study was conducted in triplicate and Bulk Density obtained as follows:

Bulk density (g/cm<sup>3</sup>) =  $(W_2 - W_1) / \text{Vol. of Sample}$

### 2.3 Proximate Analyses

**Moisture Content:** moisture content of samples was determined by hot air oven drying method as recommended by AOAC (1990).

**Crude Protein:** the Kjeldahl Digestion method was used to estimate the nitrogen in the sample which was then multiplied by the nitrogen conversion factor 6.25 to obtain the percentage protein (AOAC, 1990).

**Fat Content:** This was carried out according to (AOAC, 1990). Diethyl ether at 50°C was used for the extraction under reflux for 5h using a Soxhlet apparatus.

**Total Ash:** The ash content in the sample was determined by incineration with the furnace at 550°C (method 923-03, AOAC, 1990) after a period of 31 - 32 h a white ash was removed and placed in a desiccator for 1 h and then weighed.

Ash content (%) = (Weight of Ash/ Weight of sample) x 100%

**Carbohydrate:** Percentage Carbohydrate was obtained by difference (AOAC, 1990). The sum of Percentage Moisture, Fat, Protein and Ash was subtracted from 100 and the balance was recorded as percentage Carbohydrate.

**Energy Value (kcal.):** The sample calorific value was estimated in kcal/g by multiplying the percentages of crude protein, crude fat and carbohydrate with the recommended factors (% Protein x 2.44, % Fat x 8.37 and % CHO x 3.57) as proposed by Martin and Coolidge [1978].

### 2.4 Amino Acids and Protein Quality

**Determination of Amino Acids:** Amino Acids were profiled by the Isocratic HPLC-2 methods (AOAC, 2000). Samples were solubilized, centrifuged and filtered through a 0.22 µm membrane. The filtrate was then used for the experiment. Standard solutions of the amino acids (both essential and non-essential) were prepared and serially diluted to give 25µmol of each amino acid derivative. Chromatographic separation of samples was carried on a Buck scientific BLC10/11-model HPLC equipped with UV 338nm detector.

**Amino Acid Score:** The amino acid score was calculated using the ratio of a gram of the limiting amino acid in the food to the same amount of the corresponding amino acid in the reference diet multiplied by 100. The scoring patterns suggested by the FAO/WHO/UNU6 for children of 1-2 years of age were used for this purpose.

$$\text{Amino Acid Score (\%)} = \frac{\text{Value of EAA in Food Sample } \left(\frac{g}{100g \text{ Protein}}\right)}{\text{FAO Ref. Value for Essential Amino Acids}} \times 100$$

**The Essential Amino Acid Index [EAAI]:** was calculated using the method of Labuda et al. (1982) as reported by Steve, I.O., 2012, according to the following equation:

$$EAAI = \sqrt[9]{\frac{[Lys \times Threo \times Val \times Meth \times Isoleu \times Leu \times Phenylal \times Histi \times Trypt]a}{[Lys \times Threo \times Val \times Meth \times Isoleu \times Leu \times Phenylal \times Histi \times Trypt]b}}$$

Where: [lysine, tryptophan, isoleucine, valine, threonine, leucine, phenylalanine, histidine and methionine]a in test sample and [lysine, tryptophan, isoleucine, valine, threonine, leucine, phenylalanine, histidine and the sum of methionine and cystine]b content of the same amino acids in standard protein [%] [egg or casein] respectively.

**Protein Efficiency Ratio (PER):** Protein Efficiency Ratio of the samples were calculated according to the equations developed by Alsmeyer et al., (1974) as used by Ogunmodimu et al., (2015)

$$PER = 0.06320 [X_{10}] - 0.1539$$

Where: X<sub>10</sub> = Thr + Val + Met + Ile + Leu + Phe + Lys + His + Arg + Tyr

**Biological Value (BV):** Biological Values were computed according to the methods of Mune-Mune et al., 2011 as a function of EAAI.

$$BV = 1.09 (\text{EAA Index}) - 11.7.$$

**Nutritional Index (NI):** The nutritional index of the food samples was calculated using the formula below as described by Crisan and Sands (1978).

$$\text{Nutritional Index}(\%) = \frac{\text{EAAI} \times \% \text{ Protein}}{100}$$

## 2.5 Tuwo Preparation

Tuwo preparation method as described by Bolade *et al.* (2002) was used throughout the experiment. The overall ratio of flour to water used was 1:3.5 (w/v). Cold slurry of the flour was first prepared by mixing 20% of the desired quantity of flour (1 kg) with 25% of the desired quantity of water (3.5 l). This was followed by bringing 60% of the water into boiling and the cold slurry initially prepared was added to this boiling water coupled with vigorous stirring, using a wooden stirrer to form a consistent gruel. The remaining quantity of the flour (80% of the desired total) was then added incrementally to the boiling gruel with continuous stirring so as to avoid lumps formation and to ensure a homogenous gel is obtained. The remaining quantity of water (15% of the desired total) was finally added to the formed gel, covered properly without stirring, and allowed to cook for about 5 - 7 min, after which it was stirred vigorously to ensure smoothness of the gel. The final product so obtained is the desired *Tuwo*.

## 2.6 Textural Properties of Tuwo

**Determination of Cooling Rate of Tuwo:** 10 equal quantities (30g) of each *Tuwo* sample was taken immediately after preparation with the aid of a stainless steel die to obtain uniform sample size and surface area. The die was lined with vegetable oil to avoid sticking of the *Tuwo* sample on the inner surface of the die. The samples were kept in covered sample holders at the prevailing ambient temperature ( $34 \pm 1^\circ\text{C}$ ). Successive temperatures of the *tuwo* samples were recorded with a thermometer at intervals of 10 minutes for the first six readings and then twenty minutes subsequently until constant temperature is attained.

**Determination of Index of Hardness:** Index of hardness was estimated from the depth of penetration data (recorded as mm of penetration) obtained by Hand Penetrometer while maintaining constant force of penetration. Ten equal quantities of each *Tuwo* sample was taken immediately after preparation with the aid of a stainless steel die to obtain uniform sample size

and surface area. The die was lined with vegetable oil to avoid sticking of the *Tuwo* sample on the inner surface of the die. The samples were kept in covered sample holders at room temperature ( $34 \pm 1^\circ\text{C}$ ). Successive temperatures and hardness were measured using a thermometer and a Hand Penetrometer respectively at 10 minutes intervals for the first one hour and 30 minutes intervals subsequently until constant temperature and hardness were recorded. The drop in temperature and increase in hardness with time were recorded as cooling behaviour.

## 2.7 Sensory Evaluation of Tuwo

A group of twelve people generally familiar with *tuwo* were trained for the sensory evaluation as described by Santacruz *et al.*, 2009. A seven point hedonic scale was used to rank the degree of like and dislike of the *tuwo* samples by the twelve semi-trained panelists. Point seven represented like extremely and point one represented dislike extremely. Samples were coded with random numbers and presented to the assessors.

## 2.8 Statistical Analysis

The results were analyzed by 3-way ANOVA and mean separation carried out by the Tukey-Kramer hsd using MATLAB statistical software [MATLAB7.12.0 (R2011a)]. The correlation of the *tuwo* temperature and index of hardness of *tuwo* samples were carried out by multiple regression analysis, curve fitted by linear polynomial curve fitting, and the constants evaluated from the curves were used to predict the index of hardness from  $30^\circ\text{C}$  to  $80^\circ\text{C}$ . The predicted values were used to plot the regression lines to show the impact of the treatments on the cooling behaviour of the samples.

# III. RESULTS AND DISCUSSION

## 3.1 Quality Parameters of Composite Flours

**Functional Properties:** Table 1 shows the measured values of the Functional Properties of the flour samples. The influence of 30% cowpea addition on the functional properties of the samples is evident from the values recorded. The values of control samples compare favourably

with the data recorded by Chandra and Samsher, 2013, and also demonstrate the influence of cowpea addition on the functionality of cereal flours.

**Table 1:** Functional Properties of Flours

Property	Sorghum Flour		Millet Flour	
	Single	Composite	Single	Composite
WAC (%)	189.7 ± 2.1 <sup>g</sup>	192.7 ± 1.53 <sup>h</sup>	192.3 ± 1.8 <sup>g</sup>	194.2 ± 2.02 <sup>h</sup>
LGC (%)	8.3 ± 0.8 <sup>g</sup>	9.5 ± 1.0 <sup>h</sup>	9.7 ± 0.8 <sup>g</sup>	10.5 ± 0.5 <sup>h</sup>
GT (°C)	62 ± 1.7 <sup>g</sup>	63.7 ± 1.15 <sup>h</sup>	64.3 ± 1.2 <sup>g</sup>	65.3 ± 0.58 <sup>h</sup>
SC (%)	15.7 ± 0.6 <sup>g</sup>	16.1 ± 0.23 <sup>g</sup>	15.7 ± 0.3 <sup>g</sup>	16.0 ± 1.0 <sup>g</sup>
BD (Kg/m <sup>3</sup> )	0.53 ± 0.02 <sup>g</sup>	0.54 ± 0.01 <sup>g</sup>	0.52 ± 0.02 <sup>g</sup>	0.49 ± 0.02 <sup>g</sup>

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

**Water Absorption Capacity (WAC %):** In cereal grain flours, WAC indicates the relative availability of starch and fibre which is important in dietary formulation (Singh, 2001). From table 1, it is observed that 30% cowpea addition caused significant increase in WAC ( $p < 0.05$ ) in both sorghum and millet. This might have been caused by the combined WAC of the cereal and that of the legume. Each of the flour (i.e. Cereal and the Legume) has its own absorption capacity, the combination of which resulted in the observed increase in WAC of the four samples (Chandra & Shamsar, 2013). The WAC of the control samples is the reflection of true nature of the grain without any treatment. **Least Gelling Concentration LGC (%)** is the lowest flour concentration at which starch gelatinization takes place which is an indication of the nature of starch and its density in a given sample which may vary from one grain to another. Both sorghum and millet exhibited similar LGC ( $p > 0.05$ ). However, 30% cowpea addition led to significant increase ( $p < 0.05$ ) in the value probably due to increase in non-starch components brought in by cowpea addition. millet exhibited the higher **Gelatinization Temperature (GT °C)** compared to sorghum, probably because

corneous endosperms tend to exhibit higher GT than floury endosperm (Cagampang and Kirleis, 1985). The GT values recorded in this work compared favourably with that of Chandra and Samsher, 2013, but with slight disparities. Such differences may be brought about by the agronomic history of the different samples and even by the measurement precision of the various methods adopted different researchers. The extent to which a particular starch sample swells under given set of conditions is the **Swelling Capacity (SC)** of the starch which is important in process specification and material balance. 30% cowpea addition did not significantly ( $p > 0.05$ ) affect the SC of the flours. **Bulk Density (kg/m<sup>3</sup>)** of food materials gives a measure of the amount of matter contained in the sample. Like the case of SC, cowpea addition did not significantly ( $p > 0.05$ ) affect the BD of the flours. Values of starch density recorded in literature ranges from 1.4g/cm<sup>2</sup> to 1.6g/cm<sup>2</sup> (Rooney and Pflugfelder, 1986).

**Proximate Composition and Energy Value:** Table 2 shows the proximate composition of the composite flours as determined in this work.

**Table 2:** Proximate Composition

Property	Sorghum Flour		Millet Flour	
	Single	Composite	Single	Composite
Moisture (%)	11.0 ± 1.0 <sup>g</sup>	11.3 ± 0.6 <sup>h</sup>	10.6 ± 0.5 <sup>g</sup>	11.3 ± 0.6 <sup>h</sup>
Ash (%)	1.5 ± 0.5 <sup>g</sup>	1.7 ± 0.4 <sup>h</sup>	1.1 ± 0.2 <sup>g</sup>	1.6 ± 0.4 <sup>h</sup>
Fat (%)	1.5 ± 0.2 <sup>g</sup>	1.6 ± 0.4 <sup>g</sup>	2.0 ± 0.2 <sup>g</sup>	2.1 ± 0.2 <sup>g</sup>
Protein (%)	12.0 ± 1.0 <sup>g</sup>	15.0 ± 1.0 <sup>h</sup>	11.3 ± 1.2 <sup>g</sup>	15.3 ± 1.5 <sup>h</sup>
Carbohydrate (%)	74.0 ± 2.3 <sup>g</sup>	70.4 ± 1.8 <sup>h</sup>	75.0 ± 1.0 <sup>g</sup>	69.7 ± 2.5 <sup>h</sup>
Energy Value (Kcal)	305.7 ± 5.1 <sup>g</sup>	301.5 ± 2.1 <sup>h</sup>	312.0 ± 2.0 <sup>g</sup>	303.5 ± 4.5 <sup>h</sup>

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

*Moisture Contents* of the samples did not show significant variation between both single flours or the composite. The slight disparities recorded may be a function of the drying conditions. Moisture values of 10.82%, 10.53% for cereal flours were reported in literature (Adeoti *et al.*, 2013, Adenike *et al.*, 2014). *Percentage Ash* contents of the samples were observed to have increased due to cowpea addition which may be attributed to the increase in total dry matter of the samples. On the other hand, cowpea addition has no significant influence on the *fat content* of the samples ( $p > 0.05$ ). Cowpea addition has significantly increased the protein content ( $p < 0.05$ ) of all the flours.

*Percentage Carbohydrate* contents and *Energy Value* of the composite flours were significantly higher than single flours. As the calorific values are obtained by multiplying the proximate values with the recommended factors, variations in the energy values of the samples follow logically from the variations in the proximate values as they are affected by the treatment.

**AMINO ACIDS PROFILE:** The results of amino acids profiling were presented in table 3. The result of statistical analyses showed that the amino acids were differently affected by the treatment as discussed below.

*Table 3:* Essential Amino Acids of Samples (g/100g Protein)

Property	Sorghum		Millet	
	Single	Composite	Single	Composite
Histidine	0.72 ± 0.014 <sup>s</sup>	0.63 ± 0.014 <sup>s</sup>	1.05 ± 0.014 <sup>s</sup>	1.19 ± 0.014 <sup>s</sup>
Isoleucine	0.96 ± 0.014 <sup>s</sup>	1.785 ± 0.007 <sup>s</sup>	2.115 ± 0.007 <sup>s</sup>	2.575 ± 0.007 <sup>s</sup>
Leucine	1.865 ± 0.021 <sup>s</sup>	1.725 ± 0.007 <sup>s</sup>	2.345 ± 0.007 <sup>s</sup>	2.76 ± 0.014 <sup>s</sup>
Lysine	2.045 ± 0.007 <sup>s</sup>	2.32 ± 0.014 <sup>s</sup>	2.035 ± 0.021 <sup>s</sup>	1.715 ± 0.007 <sup>s</sup>
Methionine	1.245 ± 0.007 <sup>s</sup>	1.445 ± 0.007 <sup>h</sup>	3.415 ± 0.007 <sup>s</sup>	3.64 ± 0.014 <sup>h</sup>
Phenylalanine	1.045 ± 0.007 <sup>s</sup>	0.855 ± 0.007 <sup>s</sup>	1.525 ± 0.007 <sup>s</sup>	1.74 ± 0.014 <sup>s</sup>
Threonine	1.055 ± 0.007 <sup>s</sup>	1.355 ± 0.007 <sup>s</sup>	1.515 ± 0.007 <sup>s</sup>	1.63 ± 0.014 <sup>s</sup>
Tryptophan	0.875 ± 0.007 <sup>s</sup>	1.185 ± 0.021 <sup>h</sup>	2.01 ± 0.014 <sup>s</sup>	1.76 ± 0.014 <sup>h</sup>
Valine	0.35 ± 0.014 <sup>s</sup>	0.46 ± 0.014 <sup>h</sup>	1.05 ± 0.014 <sup>s</sup>	1.35 ± 0.014 <sup>h</sup>
TEAA	10.16	11.76	17.06	18.36

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

*Leucine:* it was observed that there is no significant difference at  $p < 0.05$  in the leucine content of the two grains, though millet (2.35) still exhibited higher leucine content compared to sorghum (1.87) but the differences are not statistically significant. *Histidine, Isoleucine, Lysine, Phenylalanine and Threonine* were observed to differ significantly between the two grains, but no significant difference was observed in their contents on the part of cowpea addition.

The histidine contents of millet (1.05g/100g protein) is higher than that of sorghum (0.72). This observation is consistent with reported literature where millet is generally held to be of higher nutritional value than sorghum. Significant difference was observed in the isoleucine content of the two grains with millet still having the higher value (2.12g/100g) compared to sorghum (0.96g/100g). Lysine content observed in this work for sorghum and millet do not vary

significantly. Similarly, cowpea addition has slightly increased the lysine content of the samples albeit statistically insignificant at  $p < 0.05$ . Phenylalanine content of sorghum (1.05g/100g) is significantly lower than that of millet (1.53g/100g). It was also observed that cowpea addition has significantly increased phenylalanine in the samples with millet still maintaining the lead. Threonine content of millet also showed a higher value of 1.52g/100g.

*Methionine, Tryptophan, Valine:* the contents of these three amino acids were significantly affected by the type of grain. The methionine content of millet was observed to be significantly the higher (3.12g/100g) than that of sorghum (1.25g/100g). Tryptophan contents of the samples vary significantly with grain type with millet having the higher (2.01g/100g), followed by sorghum (0.88g/100g). Cowpea addition has significantly increased tryptophan in the samples. Valine

contents of the samples vary significantly with grain type; and cowpea addition significantly increased valine.

**Amino Acid Score:** The chemical score of each EAA expressed as the percentage of that amino acid in the sample to the same amino acid in egg

which is used as the standard, is presented in table 4. The limiting amino acids observed from all the samples are tryptophan, methionine and valine. Each sample showed its unique LAA either one or any two of the three.

**Table 4:** Percentage Amino Acid Score (AAS %) & Limiting Amino Acids (LAA)

Property	Sorghum Flour		Millet Flour	
	Single	Composite	Single	Composite
Histidine	32.73 <sup>g</sup>	28.64 <sup>g</sup>	47.73 <sup>g</sup>	54.1 <sup>g</sup>
Isoleucine	17.78 <sup>g</sup>	33.06 <sup>g</sup>	39.17 <sup>g</sup>	47.69 <sup>g</sup>
Leucine	21.69 <sup>g</sup>	20.06 <sup>g</sup>	27.27 <sup>g</sup>	32.1 <sup>g</sup>
Lysine	29.21 <sup>g</sup>	33.14 <sup>g</sup>	29.07 <sup>g</sup>	24.5 <sup>g</sup>
Methionine	13.39 <sup>g</sup>	15.54 <sup>h</sup>	36.72 <sup>g</sup>	39.14 <sup>h</sup>
Phenylalanine	22.23 <sup>g</sup>	18.19 <sup>g</sup>	32.45 <sup>g</sup>	37.02 <sup>g</sup>
Threonine	22.45 <sup>g</sup>	28.83 <sup>g</sup>	32.23 <sup>g</sup>	34.68 <sup>g</sup>
Tryptophan	18.62 <sup>g</sup>	25.21 <sup>h</sup>	42.77 <sup>g</sup>	37.45 <sup>h</sup>
Valine	5.30 <sup>bdg</sup>	6.97 <sup>bdh</sup>	15.90 <sup>adg</sup>	20.45 <sup>adh</sup>
Limiting Amino Acid	Valine, methionine		Valine	Valine

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

**Histidine, Isoleucine, Lysine, Phenylalanine and Threonine:** the chemical score of these amino acids varies significantly between the grains and did not show any significant variation with cowpea addition. The chemical score of lysine in sorghum and millet did not show any significant difference. The score of phenylalanine in sorghum is significantly lower than the chemical score of the amino acid in millet. Similar observations can be deduced for the chemical score of histidine, isoleucine and threonine from table 4.

**Leucine:** the chemical score of leucine in the samples did not show any significant variation with neither the rain nor the treatment. the chemical scores of *Methionine, Tryptophan, Valine* vary significantly with grain type and with cowpea addition.

**Calculated Protein Quality:** Calculated Protein quality of the samples are shown in table

**Table 5:** Calculated Protein Quality of Samples

Property	Sorghum Flour		Millet Flour	
	Single	Composite	Single	Composite
PER (g/100g)	0.53 <sup>g</sup>	0.64 <sup>h</sup>	0.91 <sup>g</sup>	1.05 <sup>h</sup>
EAAI (%)	18.41 <sup>g</sup>	21.3 <sup>h</sup>	32.34 <sup>g</sup>	34.96 <sup>h</sup>
BV (%)	8.37 <sup>g</sup>	11.52 <sup>h</sup>	23.55 <sup>g</sup>	26.41 <sup>h</sup>
Nutritional Index	2.21 <sup>g</sup>	3.2 <sup>h</sup>	3.65 <sup>g</sup>	5.4 <sup>h</sup>

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

**Protein Efficiency Ratio (PER):** varies significantly among grain types and with cowpea supplementation, but neither pregelatinization nor fermentation has affected PER significantly. Like most of the amino acids quality parameters, millet recorded highest PER value of 0.91g/100g, followed by sorghum (0.53) then maize (0.29). Cowpea supplementation has increased by 75.9%, sorghum by 20.8%, and millet by 15.4%.

**Essential Amino Acids Index (EAAI):** the EAA index was observed to vary significantly with grain type and with cowpea supplementation. Supplementation increased the EAAI of maize by 57.1%, sorghum by 15.7% and millet by 8.1%.

**Biological Value (BV):** BV of the samples varies significantly with grain type with millet still maintain the lead. Pregelatinization and

fermentation did not significantly affect the BV, but cowpea supplementation has significantly increased the BV of all the samples. The value for maize was increased by 2,530%, (from 0.27 to 7.1), sorghum by 37.7%, and millet by 12.1%.

**Nutritional Index (NI):** NI of the samples varies significantly with all the three factors but not with any of their interactions. Pregelatinization has increased the NI of maize *tuwo* by 15.6%, but decreased that of sorghum and millet by 9% and 14.8% respectively. Fermentation, on the other hand, has increased the NI of maize *tuwo* by 130%, sorghum by 21.7% and millet by 15.9%. Cowpea supplementation has equally increased

the NI of maize by 212%, sorghum by 44.8% and millet by 47.9% which are all significantly higher than those of non-supplemented samples.

### 3.2 Textural Properties and Acceptability of *Tuwo*

**Cooling Behaviour:** One of the most important changes that take place immediately after *tuwo* preparation is the hardening of the texture as the temperature drops with time.

**Temperature Drop:** Cooling rates of the different *tuwo* samples were estimated as the record of temperature fall with time under the prevailing ambient conditions. Table 6 shows the rate of temperature drop of the food samples with time.

**Table 6:** Temperature Readings (°C) with Time for Various *tuwo* Samples

Grain	Flour	Cooling Time in Minutes									
		0	10	20	30	40	50	70	90	110	130
Sorghum	Single	82.5	73	64.5	61.5	54.5	51.5	41.5	34	33.5	33
	Composite	82	73.5	65	59.5	55.5	50.5	45	36	33.5	34
Millet	Single	80	72.5	66.5	61	56.5	50.5	44.5	36	34.5	33.5
	Composite	80.5	71	65	59	54	49	44	36	33	34.5

*Values are means of two readings*

**Index of Hardness:** As *tuwo* is subjected to natural cooling process, moisture is being lost to the environment as starch retrogradation also takes place causing increase in the forces of attraction between the molecules leading to

increase in hardness with time. Tables 7 give the penetrometer readings of the *tuwo* samples with time. The penetrometer readings recorded was observed to vary in inverse proportion to the cooling time until a constant value was attained.

**Table 7:** Penetrometer Readings in mm as Index of Hardness of *tuwo* Samples

Grain	Flour	Cooling Time in Minutes									
		0	10	20	30	40	50	70	90	110	130
Sorghum	Single	15.1	14.8	12.1	10.1	9.8	8.5	7	5.5	5	4.9
	Composite	13.8	13.7	12	9.3	9.6	9	6.7	4.9	4.1	4
Millet	Single	18.5	17.1	15.3	13.5	11.5	10.1	8.1	6.6	6.2	6.2
	Composite	18.5	17.1	15.3	13.5	11.5	10.1	8.1	6.6	6.2	6.2

*Values are means of two readings*

Hardness of cereal flour meals is attributable to the inherent associative forces within the starch molecules such as amylose/amylopectin ratio, and the level of chemical transformation during *tuwo* preparation (Bolade, 2010)). Mua and Jackson 1997 observed that higher amylose content and longer amylopectin chains could contribute to the hardness of a food gel from maize. Mua and Jackson further observed that flour preparation methods could affect the inherent associative

forces within the starch molecules and by extension that of the food prepared from such flour. This is seen in the differences in penetrometer readings between supplemented and non-supplemented samples

**Correlation of Temperature and Hardness with Time:** The temperature and index of hardness of non-supplemented samples recorded with time all show positive correlation (Table 8). That means as

penetrometer readings between supplemented and non-supplemented samples

*Correlation of Temperature and Hardness with Time:* The temperature and index of hardness of non-supplemented samples recorded with time all show positive correlation (Table 8). That means as the temperature of the samples decreases with time so does the index of hardness. But the index of hardness is the extent of the penetration of the spindle into the *tuwo* sample meaning that the harder the sample the less the penetration.

Therefore the positive correlation observed between decreasing temperature and decreasing index of hardness means a negative correlation between the temperature drop and actual hardness of the sample: meaning as temperature of the sample decreases, the actual hardness increases. This is attributable to several factors (Mua & Jackson, 1997); at lower temperatures, the forces of attraction between the molecules are higher than at elevated temperatures leading to harder texture. Starch retrogradation as an inherent property of plant starches also lead to harder textures at lower temperatures.

**Table 8:** Correlation Coefficients (r) of Temperature & Index of Hardness for Non-supplemented Samples

	1	2	3	4	5	6
SCN (P)	0.97910	0.97682	1			
SCN (T)	0.99572	0.99079	0.99221	1		
MiCN (P)	0.99767	0.98801	0.97381	0.99443	1	
MiCN (T)	0.99007	0.99732	0.97719	0.99340	0.99377	1

**Table 9:** Correlation Coefficients (r) of Temperature and Index of Hardness for Supplemented Samples

FLOUR	1	2	3	4	5	6
SCS (P)	0.97285	0.97559	1			
SCS (T)	0.99381	0.98821	0.95716	1		
MiCS (P)	0.98704	0.98586	0.98998	0.98386	1	
MiCS (T)	0.99430	0.99321	0.97939	0.99445	0.99624	1

*Curve Fitting [Index of Hardness (mm) Against Temperature (°C)]:* As strong correlation was established between index of hardness and temperature, a linear polynomial curve was fitted to enable the prediction of hardness at any given temperature. The slope of the curve  $\beta$  and the intercept  $\epsilon$  were evaluated, while r-square indicated the goodness of fit of the data to the

curve. These constants were presented in table 4.10. The values of rsquare for all the samples approached zero indicating that curves were all well fitted to the data. This means that the values of hardness predicted from these constants will approximate closely to experimental data.

**Table 10:** Table of Constants for the Linear Regression Curves

Grain	Flour	$\beta$ Slope of the Curve	E Intercept of the curve	$r^2$ Goodness of Fit	Adjusted $r^2$
Sorghum	Single	0.2197	-2.5650	0.9845	0.9806
	Composite	0.1798	-1.2970	0.9161	0.8952
Millet	Single	0.1933	-1.1510	0.9575	0.9469
	Composite	0.2101	-2.7480	0.9925	0.9906

The constants evaluated from the curves were used to predict the index of hardness between 30 to 80°C to observe the effects of grain type,

processing methods and cowpea supplementation on the hardening pattern of *tuwo*.

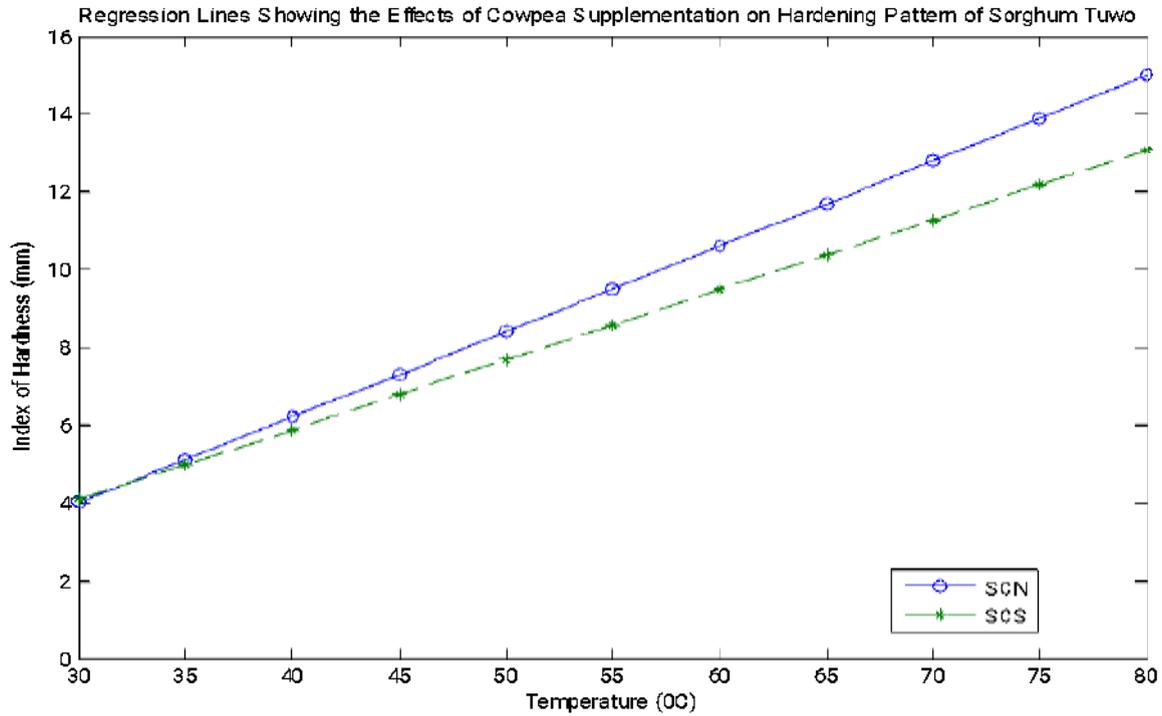


Figure 1: Regression Lines Showing the Effects of Supplementation on the Hardening pattern of Sorghum *Tuwo*

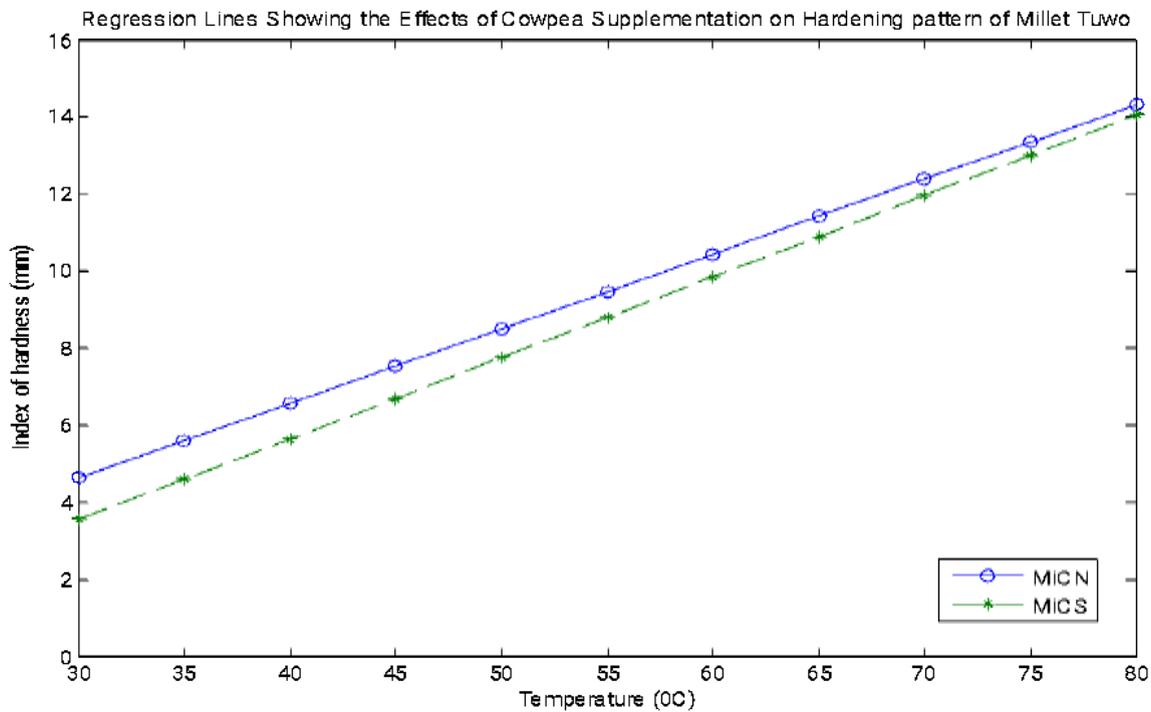


Figure 2: Regression Lines Showing the Effects of Supplementation on the Hardening pattern of Millet *Tuwo*

Table 10: Sensory Evaluation scores of Tuwo Samples

Attribute	Sorghum		Millet	
	Single	Composite	Single	Composite
Colour	4.83 ± 1.64 <sup>bdg</sup>	3.58 ± 1.08 <sup>bdg</sup>	4.17 ± 2.12 <sup>bdg</sup>	3.58 ± 1.17 <sup>bdg</sup>
Aroma	4.75 ± 1.22 <sup>adg</sup>	3.33 ± 1.50 <sup>adg</sup>	4.92 ± 1.88 <sup>adg</sup>	3.17 ± 0.84 <sup>adg</sup>
Taste	4.75 ± 0.75 <sup>bdg</sup>	3.67 ± 1.16 <sup>bdg</sup>	4.08 ± 2.23 <sup>bdg</sup>	3.25 ± 0.87 <sup>bdg</sup>
Texture	4.67 ± 1.44 <sup>bdg</sup>	3.92 ± 1.44 <sup>bdg</sup>	4.58 ± 2.02 <sup>cdg</sup>	4.17 ± 1.47 <sup>cdg</sup>
Overall Acceptability	5.0 ± 0.74 <sup>bdg</sup>	4.00 ± 1.13 <sup>bdg</sup>	4.67 ± 1.72 <sup>bdg</sup>	3.50 ± 1.17 <sup>bdg</sup>

Mean values in the same row with different superscript differ significantly ( $p < 0.05$ )

**Colour:** the colour ratings of the samples vary significantly only in grain type. Pregelatinization, fermentation and cowpea supplementation did not significantly affect the colour ratings. Among the grain types, the ratings of sorghum (4.83) and millet (4.17) do not differ significantly from each other but the ratings of the two are significantly lower than that of from maize (5.75). This may be a reflection of the survey data which indicated that maize is the most preferred grain for tuwo.

**Aroma:** the aroma ratings of the samples did not show any significant variation with any of the three factors studied in this work. This may be due the fact that tuwo aroma does not matter much to consumers since tuwo is normally eaten with soup.

**Taste:** the taste ratings of the samples vary significantly with grain type and processing method, and also in the interaction of processing method and supplementation. The taste ratings of sorghum (4.75) and millet (4.08) are not significantly different from each other but both ratings are significantly lower than that of maize (5.67). This may also be a reflection of the most proffered grain for tuwo. Pregelatinization has significantly reduced the taste ratings of all the grains probably due to the loss of soluble substances without generating any taste inducing substances. Fermentation, on the other hand, has significantly reduced the taste ratings of maize and sorghum but significantly increased that of millet, probably due to flavor modification induced by the fermentation process. The increased taste rating due to fermentation observed in millet but not in sorghum and maize may be due to the higher amino acid quality recorded in millet; the interaction of which

resulted in improved taste for the fermented millet tuwo.

**Texture:** the texture ratings of the samples differ significantly with grain type and processing methods, and with their interactions. Both pregelatinization and fermentation have significantly reduced the texture ratings of the samples, while cowpea supplementation did not have any significant effect on the texture ratings.

**Overall Acceptability:** on the overall, the acceptability of the tuwo samples varies significantly only with grain type and with the interaction of grain and processing method.

Pregelatinization, fermentation and cowpea supplementation all did not significantly affect the acceptability of the tuwo samples. This is an indication that improved cereal flours can be well accepted to the consumer.

#### IV. CONCLUSION

Cowpea supplementation significantly increased WAC, LGC, GT. Has no significant effect on SC and BD. Cowpea addition has significantly increased the ash and protein contents but has not significantly affected the fat content. CHO content and E.V were significantly reduced while Inv.D was significantly increased. Methionine, tryptophan and valine contents of the samples were significantly increased by cowpea addition.

Protein Efficiency Ratio (PER), Essential Amino Acids Index (EAAI) and Biological Value (BV) all showed significant difference among the grains and in cowpea supplementation. Nutritional Index (NI) showed significant variation with cowpea addition.

The cooling rate of *tuwo* and Index of hardness are positively correlated and fit well into a linear polynomial curve. The hardness of *tuwo* at any temperature can be estimated from the linear equation with good accuracy.

Cowpea addition did not significantly affect the colour ratings. Aroma and texture ratings of the samples did not show any significant variation with 30% cowpea addition. The overall acceptability of sorghum and millet *tuwo* with 30% cowpea supplementation did not show any significant variation from control samples indicating that *tuwo* made from improved cereal flours can be well accepted to the consumer.

## REFERENCES

1. Adebowale, Y.A, Adeyemi, I.A. Oshodi, A.A., (2005). Functional and Physicochemical Properties of Flours of Six Mucuna Species. *Afr. J. Biotech.* 4(12), pp 1461 – 1468.
2. Adenekan, M.K, Oguntoyinbo, S.I, Odunmbaku, L.A and Nupo S.S. (2014). Effect of Sprouted Pigeon pea (*cajanus cajan*) on Proximate Composition and Sensory Value of *Tuwo*. *Journal of Sciences & Multidisciplinary Research.* Vol.6 (2).
3. Adeoti, O.A., Elutilo, O.O., Babalola, J.O., Jimoh, K.O, Azeez, L.A and Rafiu, K.A, (2013). Proximate, Mineral, Amino Acid and Fatty Acid Compositions of Maize *Tuwo*-Cirina Forda Flour Blends. *Greener Journal of Biological Sciences.* Vol. 3 (4), pp. 165-171. Available online at [www.gjournals.org](http://www.gjournals.org).
4. Alsmeyer RH, Cuningham AE, Hapich ML (1974). Equations predicted (PER) from amino acid analysis. *Food Technol.*, 28, 34 – 38. In: Ogunmodimu et al., 2015.
5. AOAC (2000). Association of Official Analytical Chemists. 18<sup>th</sup> ed.Official Methods of Analyses. Washington D.C.
6. AOAC (1990). Association of Official Analytical Chemists. 18<sup>th</sup> ed.Official Methods of Analyses. Washington D.C.
7. Bolade, M.K., Usman, M.A., Rasheed, A.A., Benson, E.L., Salifou, I., (2002). Influence of Hydro-thermal treatment of maize grains on the quality and acceptability of *tuwon masara* (traditional maize gel). *Food Chem.*, 79: 479-483.
8. Bolade, M.K., (2010). Evaluation of suitability of commercially available maize grains for *tuwo* production in Nigeria. *Afr. J. food. Sci.* vol. 4(6)pp 371 - 381.
9. Cagampang, G.B., and Kirleis A.W., 1985. Properties of Starches Isolated from Floury and Corneous endosperm. *Starch/Staerke* 37:253-257. In: Dendy, D.A.V., 1995. *Sorghum and Millets: Chemistry and Technology.* American Association of Cereal Chemists. Minnesota.
10. Chandra, S., and Samsher, 2013. Assessment of Functional Properties of Different Flours. *Afric. J. Agric. Res.* Vol. 8 (38), pp 4849-4852. Available online at [www.academicjournals.org](http://www.academicjournals.org).
11. Crisan E.V, Sands A (1978). Biology and Cultivation of Edible Mushrooms. Hangeri Academic Press, New York, 137-142. In: Ogunmodimu et al., 2015.
12. Cianciaruso B, Jones M.R, Kopple J.D., 1981. Histidine, an essential amino acid for adult dogs. *Journal of Nutrition*, 111:1074–1084.
13. Cox, R. E., and R. H. Higby. 1944. A better way to determine the jelling power of pectins. *Food Ind.* 16, 441.
14. Da, S, Akingbala, J.o, Rooney, L.W and Miller, F.R, (1982). Evaluation of to quality in sorghum breeding program. Pp 11-23 In: Proceedings of the International Symposium on sorghum grain quality. Rooney L.W and Murty, D.S eds. ICRISAT. India.
15. FAO/WHO/UNU, 2002: Joint FAO/WHO/UNU Expert Consultation on Protein and Amino Acid Requirements in Human Nutrition, Geneva, Switzerland. WHO Technical FAO Food and Nutrition Paper No.92, 2013: Dietary Protein Quality Evaluation in Human Nutrition. Report of an FAO Expert Consultation 31 March–2 April, 2011, Auckland, New Zealand.
16. Hamaker, B.R., Kirleise A.W., Butler, L.G., Axtell J.D., and Mertz E.T., 1987. Improving the in vitro protein digestibility of sorghum with reducing agents. *Proc. Nat. Acad. Sci. USA* 84:626-628. In: Dendy, D.A.V., 1995. *Sorghum and Millets: Chemistry and*

- Technology. American Association of Cereal Chemists. Minnesota.
17. Hulse, J.H, (1991). Nature, Composition and Utilization of Grain Legumes pp 11-27. In: ICRISAT, 1991.
  18. Kirleis, a.W., 1990. The prolamins of sorghum: their role in protein digestibility. In: Dendy, D.A.V., 1995. Sorghum and Millets: Chemistry and Technology. American Association of Cereal Chemists. Minnesota.
  19. Labuda J, Kacerovský O, Kovàè M, Štirba A (1982). Výživa a krmienie hospodárskych zvierat. Príroda, Bratislava. p. 164s. In: Steve, I.O., 2012.
  20. Leach, H.W., McCowen LD, Schoch TJ (1959). Structure of the starch granules. In: Swelling and Solubility patterns of various starches. Cereal Chem. 36:534-544.
  21. Martin, E.A., Coolidge, A.A., (1978). Nutrition in Action, 4th ed. Holt, R and Wilson Co. New York, USA.
  22. Mune-Mune, M.A., Minka, S.R., Mbome I.L., Etoa F.X. (2011). Nutritional Potential of Bambara Bean Protein Concentrate. In: Steve, I.O., 2012.
  23. Narayana, K., and Narsinga, R.S.M., (1982). Functional Properties of Raw and Heat Processed Winged Bean (*Psophocarpus tetragonolobus*) Flour. J. Food Sci. 42: 534-538.
  24. Nkama, I., (1993). Traditional Methods of Production of High Protein Energy Foods from Grain Legumes in the North-Eastern States of Nigeria. Annals of Borno 10: 138-148.
  25. Nkama, L, Ogbo, E.N., and Igene, J.O., (1994). Proximate Composition, Shelf Stability and Consumer Acceptance of Groundnut Butter. J Arid agric. 3:7, 66-75.
  26. Ogunmodimu, O. O, Ijarotimi, O. S and Fagbemi, T. N, 2015. Evaluation of nutritional properties of high protein-fiber based snacks formulated from wheat, soybean concentrate and cassava fiber. Sky Journal of Food Science Vol. 4(3), pp. 030–041. Available online <http://www.skyjournals.org/SJFS>.
  27. Rooney, L.W., and pflugfelder, R.L., 1986. Factors affecting starch digestibility with special emphasis on sorghum and corn. J. Anim. Sci. 63:1607-1623. In: Dendy, D.A.V., 1995. Sorghum and Millets: Chemistry and Technology. American Association of Cereal Chemists. Minnesota.
  28. Shinde, B.G., (2001). Isolation and characterization of starch horse grain. Unpublished M.Sc. Thesis. Mahatma Phule Krishi Vidyapeeth, Rahuri (India). pp. 16-17. In: Chandra and Shamsheer, (2013).
  29. Steve O.I and Olufunmilayo O.K, (2011). Determination of Amino Acid, Fatty Acid, Mineral, Functional and Choking Properties of Germinated and Fermented Popcorn (*Zea mays everta*) Flour. European Journal of Food Research & Review 1(2): 102-122. Available at [www.sciencedomain.org](http://www.sciencedomain.org).
  30. Steve, I.O., 2012. Influence of germination and fermentation on chemical composition, protein quality and physical properties of wheat flour (*Triticum aestivum*). Journal of Cereals and Oil seeds Vol. 3(3), pp. 35-47, November 2012. Available online at <http://www.academicjournals.org/JCO>.
  31. WHO/FAO/UNU (2007) Protein and Amino Acid Requirements in Human Nutrition; Report of a joint WHO/FAO/UNU Expert Consultation, WHO Tech. Rep. Ser. No.935. Geneva: WHO.