

Effect of cowpea flour inclusion on the storage characteristics of composite wheat-cowpea bread

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Abstract. Substitution of refined wheat flour (WF), the principal ingredient for bread making with cowpea flour (CF) could encourage diverse utilization of cowpea and reduce overdependence on wheat importation. However, the effect of cowpea flour inclusion on the storage characteristics of composite wheat-cowpea (CWC) bread which is essential for the commercial success of the product needs to be studied. Hence, this study was conducted to determine the effect of cowpea flour inclusion on the storage characteristics of CWC bread. CF was blended with wheat flour (WF) at 5, 10, 15 and 20% substitution levels. Flour samples were analyzed for proximate composition, rheological and functional properties. The CWC dough after proofing was cut into uniform sizes (300 g) and baked at 220°C for 30 min. The physical, chemical, microbiological and sensory qualities of the freshly baked bread samples were also determined. Changes in these qualities were also determined in the bread samples stored at ambient conditions (27 ± 2°C, 79 ± 3% RH) over a period of 5 days. Mould count ranged 1.00 to 3.00, 6.50 to 24.00 and 33.50 to 50.50 cfu/g for day 1, 3 and 5, respectively during storage. The order of overall acceptability of the CWC bread over the storage period was 5% > 10% > 15% > 20%. The softness index ranged between 16.40-34.80, 12.00-31.75, 10.25-29.65, 7.90-26.40 and 7.25-23.20 mm for day 1, 2, 3, 4 and 5, respectively. The CWC bread was acceptable to the sensory panelist for the first three days of its production. Bread made from the 100% WF was more shelf stable than the one made from the CWC flour.

Keywords: Composite wheat-cowpea, pasting properties, proximate composition, rheological, sensory qualities, softness index.

INTRODUCTION

Bread is one of the most widely consumed food products in the world. It has relatively low cost and gives some of the nutrients missing in majority of carbohydrate foods (Foster, 2008). Bread is mainly produced from wheat flour since it was introduced as food in the nation. Yeast-raised bread is highly favoured worldwide because of its desirable sensory attributes. Quality attributes such as large loaf volume and fine, even crumb texture require formation of a well developed, elastic dough structure. This is made possible by the quantity and quality of the wheat flour proteins, gliadins and glutenins, collectively known as gluten (Shukla, 2001). The concept of blending

wheat flour with non-wheat, high-protein flours has been employed in situations where wheat flour may be in short supply or where nutritional quality requires enhancement as reported by Sadowska et al. (1999).

In 2011, Nigeria produced 165,000 tonnes of wheat and imported 4,054,000 tonnes. Wheat flour has been the principal ingredient in bread baking. Nigeria's consumption per capita for wheat was 15.0 kg/year and the country produced only 80,000 tons (FAO, 2013). Recently, Federal Government of Nigeria encouraged the use of composite flour for baking purpose so as to reduce high importation rate of wheat flour.

Table 1. Dough recipe and formulation (composition) used for the baking experiment.

Ingredients (g)	5%	10%	15%	20%	Control
Wheat flour	285	270	255	240	300
Cowpea flour	15	30	45	60	-
Salt (2%)	6.0	6.0	6.0	6.0	6.0
Sugar (6.0%)	18.0	18.0	18.0	18.0	18.0
Yeast (5.0%)	15.0	15.0	15.0	15.0	15.0
Fat (3.0%)	9.0	9.0	9.0	9.0	9.0
EDC (0.3%)	0.9	0.9	0.9	0.9	0.9
Water	165.0	165.0	165.0	165.0	165

Adapted from Shittu et al. (2007). % values are based on the total flour weight (300 g). Values in parenthesis denotes percentage ingredient.

Cowpea (*Vigna unguiculata*) is a high-protein, starchy legume that can easily be processed into flour and blended with wheat flour. In addition to being a good source of B-vitamins, cowpea contains substantial quantities of lysine and when blended with cereals, produces mixtures with complementary amino acid profiles and improves nutritional quality (Mensa-Wilmot et al., 2001). Nigeria produced 2,500,000 metric tons of cowpea (FAO, 2013).

Substitution of wheat flour (WF), the principal ingredient for bread making with cowpea flour (CF) could encourage diverse utilization of cowpea and reduce overdependence on wheat importation. However, the impact of such substitution on the composite dough and loaf qualities must be properly understood.

Moreover, limited studies have reported on the effect of cowpea flour on the storage characteristics of cowpea-wheat bread which is also essential for the commercial success of the product. Therefore, this study was conducted to determine the effect of cowpea flour inclusion on the storage characteristics of composite cowpea-wheat bread (CWC) bread.

MATERIALS AND METHODS

Materials

The baking ingredients used in this study include wheat flour (Honeywell Flour Mills, Lagos), beans flour from Nigerian Stored Products Research Institute, Ibadan. Other materials used include granulated sugar (Dangote Nigeria Plc., Lagos, Nigeria), Fermipan baking yeast (DSM bakery ingredient, Dordrecht-Holland), salt, Simas margarine (PT Intiboga Sejahtera, Jakarta, Indonesia) and Edlen Dough Conditioner (EDC 2000; Edlen International Inc., GA, USA).

Physical, chemical and functional properties of composite wheat-cowpea flour

The flour samples (wheat and composite wheat-cowpea

flour) were analyzed for proximate composition, rheological, pasting and functional properties.

Bread baking

The straight dough method described in Shittu et al. (2008) for composite cassava wheat bread was used in dough preparation. The two flours (wheat and cowpea flour) were blended together in percentages as shown in Table 1. The bread dough was prepared using the main ingredients (cowpea and wheat flour) in varying percentages while other ingredients are based on composite flour weight shown in Table 1. The mixing was done manually for 15 min prior to kneading, which was also done manually 3 to 5 min until smooth dough was obtained. The dough was then divided into uniform sizes (300 g). Proofing of the dough was done in the pan at ambient conditions ($29 \pm 2^\circ\text{C}$, 79% RH) for 2 h. Baking was done with an electric oven (Gallenkamp, UK) at 180°C for 25 min. The weights of the loaves were determined with the aid of a weighing balance with accuracy of 3 d.p. (Mettler Toledo, Switzerland).

Physical properties of composite cowpea-wheat bread

Oven spring was determined by recording the height of the fermented dough and height of the baked bread samples. Oven spring was determined as the difference in the dough height and baked bread height. The loaf volume was determined after baking process using volume displacement method in which millet seed was used instead of rapeseed.

The specific volume was calculated as:

$$\text{Specific volume (cm}^3\text{/g)} = \frac{\text{loaf volume (cm}^3\text{)}}{\text{loaf weight (g)}}$$

Textural analysis was determined with the aid of a cone penetrometer to measure bread crumb softness. Six centimeters thick bread slice was placed on the base of

the penetrometer (Central Model, Central Ignition Co., London) and the tip of the cone was adjusted to touch the central core surface of the bread crumb. The cone was then released to penetrate into the crumb under gravitational force. The readings were done in duplicate. Penetration was measured at 3 s (range 0 to 400 units, equivalent to 0 to 40 mm penetration). Higher penetration units indicate increased crumb softness. The tristimulus color parameters L^* (lightness), a^* (redness to bluishness), b^* (yellowness to greenness) of the baked loaves crumbs were determined using a digital colorimeter (Color Tec PCM, Accuracy Micro Sensor Inc., USA). The instrument has spot diameter view of 15 mm. To determine the color of bread crust, the top crust was divided into three regions while the tristimulus color parameters L^* , a^* , b^* were determined at each point in duplicate. For crumb moisture determination, 1 g of bread was obtained from five different portion of a slice and weighed into a previously weighed Petri dish. The Petri dish and the samples were transferred into the oven set at 105°C to dry to constant weight for 2 h. At the end of the 2 h, the Petri dish and sample were removed from the oven and transferred to desiccators and cooled and weighed.

Bread crumb color and cell structure analysis

This was conducted as described by Shittu (2007). Images of the sliced breads were captured using flatbed Mercury Scanner 1200U (SCAMXX, Mercury, China; <http://www.kobian.com>). The images were scanned full scale at 300 dots per inch and analyzed in grey scale. A 200 × 200 pixel square field of view (FOV) was evaluated for each image. This FOV captured the majority of the crumb area of each slice. Seven digital images were processed and analyzed for each bread sample, giving a total of 70 images. Image analysis was performed using the Image 1.32j software (National Institute of Health, USA). The crust images were cropped processed and analyzed using Corel PHOTO-PAINT 12 software (Corel Corporation, USA) as described in Shittu et al. (2008).

Sensory analysis

Panelists consist of untrained fifty persons among whom 28 were males and 22 females. The panelists were students and staff of the Moshood Abiola Polytechnic, Ojere, Abeokuta, Ogun State, Nigeria participated in the sensory preference test. The age range of the participants was from 20 to 45 years with about 70% of them having post secondary school educational training. The composite bread samples were coded and packaged in low density polyethylene bags before presenting them to the panelist not later than 3 h post-baking period. The panelists rated the bread samples for appearance, crust

colour, texture, flavor, overall acceptability based on 9 point scale where 1 and 9 represent 'like extremely' and 'dislike extremely', respectively.

Textural analysis and storage study

The textural analysis was carried out to determine both the fresh crumb softness and reduction in crumb softness (hardness) of stored bread samples. Modified method of Shittu et al. (2007) was used to determine the crumb softness. This experiment was repeated daily for five days as a measure of loss of softness (hardness) during storage. Fresh loaves were packaged in polythene bag (nylon 6) and placed on a stainless steel shelf free from insects and rodents and at room temperature ($27 \pm 3^\circ\text{C}$). Shelf stability study was carried out in which the texture and product quality (microbiological safety) of the composite bread samples were assessed as described by Tessi et al. (2002).

Data analysis

The loaf characteristics and sensory attributes data were subjected to statistical analysis using the SPSS 16.0 version (SPSS Inc. USA). Means of the sensory data were separated using Duncan's multiple range test.

RESULTS AND DISCUSSION

Physical, chemical and functional properties of composite cowpea-wheat flour

Table 2 shows the physical, chemical and functional properties of composite wheat-cowpea flour. Increasing the levels of CF inclusion in the composite increased the protein (11.00 to 12.70%), ash (0.74 to 0.78% DMB), swelling power (10.43 to 11.57%), water absorption index (19.36 to 26.44%), water solubility index (5.80 to 9.80%), breakdown viscosity (78.88 to 99.63 RVU) and pasting temperature (70.10 to 79.95°C). In contrast, peak viscosity (203.84 to 221.63 RVU), trough (105.46 to 142.75 RVU), setback viscosity (116.13 to 139.79 RVU), final viscosity (221.58 to 282.55 RVU), and peak time (5.37 to 6.10 min) decreased with increasing CF inclusion. There was significant difference ($p < 0.05$) in pasting properties and water absorption which could be attributed to replacement of wheat with cowpea flour and lower flour moisture, respectively.

Physical properties of composite cowpea-wheat bread

Table 3 shows the physical properties of composite wheat-cowpea bread. The oven spring ranged from -0.20

Table 2. Physical, chemical and functional properties of composite wheat-cowpea flour.

Parameter	5%	10%	15%	20%	Control
M.c. (%)	13.05 ± 0.07 ^b	12.60 ± 0.14 ^a	12.40 ± 0.00 ^a	12.55 ± 0.07 ^a	13.35 ± 0.07 ^c
A.c. (DMB)	0.75 ± 0.01 ^{ab}	0.76 ± 0.01 ^{abc}	0.77 ± 0.00 ^{bc}	0.78 ± 0.01 ^c	0.74 ± 0.01 ^a
Prot. (%)	11.35 ± 0.07 ^a	12.20 ± 0.14 ^b	12.40 ± 0.00 ^{bc}	12.70 ± 0.00 ^c	11.00 ± 0.28 ^a
L.g.c. (%)	6.43 ± 0.21 ^{ab}	6.20 ± 0.03 ^{ab}	6.57 ± 0.41 ^{ab}	6.70 ± 0.28 ^b	5.45 ± 0.84 ^a
S. p. (%)	11.07 ± 0.57 ^a	11.57 ± 0.42 ^a	11.12 ± 0.66 ^a	10.97 ± 0.55 ^a	10.43 ± 0.64 ^a
W. a. (%)	194.80 ± 1.13 ^a	264.40 ± 16.40 ^c	229.60 ± 18.10 ^b	210.00 ± 0.00 ^{ab}	193.60 ± 0.57 ^a
W. s.i.	5.80 ± 1.98 ^a	8.60 ± 1.41 ^{ab}	9.80 ± 1.41 ^b	8.40 ± 0.00 ^{ab}	7.20 ± 0.00 ^{ab}
M.c. (%)	13.05 ± 0.07 ^b	12.60 ± 0.14 ^a	12.40 ± 0.00 ^a	12.55 ± 0.07 ^a	13.35 ± 0.07 ^c
A.c. (DMB)	0.75 ± 0.01 ^{ab}	0.76 ± 0.01 ^{abc}	0.77 ± 0.00 ^{bc}	0.78 ± 0.01 ^c	0.74 ± 0.01 ^a
P. v (RVU)	215.08 ± 0.71 ^{bc}	206.29 ± 2.65 ^{ab}	203.83 ± 7.31 ^a	206.58 ± 3.30 ^{ab}	221.62 ± 2.65 ^c
Trg (RVU)	129.83 ± 2.00 ^c	117.46 ± 0.83 ^b	105.46 ± 5.95 ^a	106.96 ± 0.29 ^a	142.75 ± 2.00 ^d
B. v. (RVU)	85.25 ± 2.71 ^{ab}	88.83 ± 0.83 ^b	98.38 ± 1.36 ^c	99.63 ± 3.01 ^c	78.88 ± 4.66 ^a
F.v (RVU)	260.00 ± 0.41 ^c	239.42 ± 0.83 ^b	221.58 ± 12.37 ^a	224.00 ± 0.35 ^a	282.54 ± 1.59 ^d
S. v. (RVU)	130.21 ± 1.59 ^b	121.96 ± 1.00 ^a	116.12 ± 6.42 ^a	117.04 ± 0.06 ^a	139.79 ± 0.41 ^c
P. t. (min)	5.87 ± 0.09 ^c	5.63 ± 0.05 ^b	5.47 ± 0.09 ^{ab}	5.37 ± 0.05 ^a	6.1 ± 0.05 ^d
P. tem (°C)	70.10 ± 1.13 ^a	71.35 ± 0.64 ^a	79.00 ± 1.13 ^b	79.95 ± 0.00 ^b	70.13 ± 0.04 ^a

Results are expressed as mean + standard deviation. Mean values followed by different superscript letter across a row are significantly different ($p \leq 0.05$). P.v.: Peak viscosity; Trg: Trough; B.v.: Breakdown viscosity; F.v.: Final viscosity; S.v.: Setback viscosity; P.t.: Peak time; P.tem: Pasting temperature; M.c.: Moisture content; A.c.: ash content; Prot.: Protein; L.g.c.: Least gelation concentration; S.p: Swelling power; W.a.: Water absorption; W.s.i.: Water solubility index

Table 3. Physical properties of composite wheat-cowpea bread.

Parameter	5%	10%	15%	20%	Control
Ov. s. (cm)	0.62 ± 0.74 ^c	-0.25 ± 0.27 ^a	-0.20 ± 0.48 ^{ab}	0.22 ± 0.98 ^{bc}	2.12 ± 0.41 ^d
L.w. (g)	254.00 ± 4.23 ^b	260.50 ± 7.2 ^c	251.17 ± 3.97 ^{ab}	247.50 ± 3.56 ^a	254.83 ± 3.97 ^{bc}
S.l.v. (cm ³ /g)	3.95 ± 0.07 ^d	3.53 ± 0.10 ^c	3.18 ± 0.05 ^b	3.03 ± 0.04 ^a	4.12 ± 0.07 ^e
L.v. (cm ³)	1000.00 ± 0.00 ^a	920.00 ± 0.00 ^a	800.00 ± 0.00 ^a	750.00 ± 0.00 ^a	1050.00 ± 0.00 ^a
S.i. (mm)	26.07 ± 9.85 ^a	28.97 ± 6.05 ^a	26.53 ± 1.46 ^a	26.03 ± 6.23 ^a	20.83 ± 7.72 ^a
M.c. (%)	26.70 ± 0.99 ^a	30.90 ± 3.54 ^a	26.90 ± 4.38 ^a	30.20 ± 1.70 ^a	29.10 ± 0.71 ^a
C.d. (g/cm ³)	0.28 ± 0.01 ^a	0.30 ± 0.01 ^a	0.31 ± 0.04 ^a	0.31 ± 0.01 ^a	0.33 ± 0.02 ^a
C.p.	0.83 ± 0.08 ^a	1.07 ± 0.24 ^{ab}	1.12 ± 0.17 ^{ab}	0.92 ± 0.14 ^a	1.47 ± 0.19 ^b
S.d. (g/cm ³)	0.83 ± 0.14 ^a	1.07 ± 0.02 ^{ab}	1.12 ± 0.00 ^{ab}	0.92 ± 0.01 ^a	1.47 ± 0.02 ^b
Crust l*	46.86 ± 7.10 ^a	56.84 ± 5.15 ^a	56.33 ± 6.81 ^a	56.35 ± 4.13 ^a	44.35 ± 13.57 ^a
Crust a*	13.68 ± 7.05 ^a	14.67 ± 8.77 ^a	16.22 ± 9.77 ^a	16.65 ± 9.95 ^a	13.76 ± 9.05 ^a
Crust b*	26.73 ± 4.11 ^a	30.84 ± 2.72 ^a	31.19 ± 3.72 ^a	32.76 ± 3.96 ^a	24.65 ± 8.79 ^a
Crumb l*	71.57 ± 0.00 ^{bc}	70.25 ± 0.00 ^b	68.05 ± 0.01 ^a	73.19 ± 0.02 ^c	75.23 ± 0.13 ^d
Crumb a*	0.06 ± 0.01 ^a	0.29 ± 0.00 ^b	0.67 ± 0.02 ^c	0.25 ± 0.00 ^{ab}	0.1 ± 0.01 ^{ab}
Crumb b*	10.18 ± 0.02 ^a	11.47 ± 0.00 ^b	12.99 ± 0.01 ^c	10.05 ± 0.03 ^a	10.04 ± 0.00 ^a

Results are expressed as mean + standard deviation. Mean values followed by different superscript letter across a row are significantly different ($p \leq 0.05$). Ov.s.: Oven spring; L.w.: Loaf weight; S.l.v.: Specific loaf volume; L.v.: Loaf volume; S.i.: Softness index; M.c.: Moisture content; C.d.: Crumb density; C.p.: Crumb porosity; S.d.: Solid density; Crust l*: Crust lightness; Crust a*: Crust redness; Crust b*: Crust yellowness; Crumb l*: Crumb lightness; Crumb a*: Crumb redness; Crumb b*: Crumb yellowness

cm in sample with 20% level of substitution to 2.11 cm in the control sample, the oven spring was highest in the control sample and was decreasing as the substitution level is increasing. Loaf weight ranged from 247.5 g in sample with 20% level of substitution to 260.50 g in sample with 10% level of substitution. Loaf weight increase in substituted samples during baking is a

desirable economic quality at the bakers end as consumers often get attracted to bread loaf with higher weight believing that it has more substance for the same price (Hallen et al., 2004; McWatter et al., 2004).

The bread samples varied significantly ($P < 0.05$) in volume of sample with the 20% level of substitution having the least specific volume of 3.03 cm³/g and the

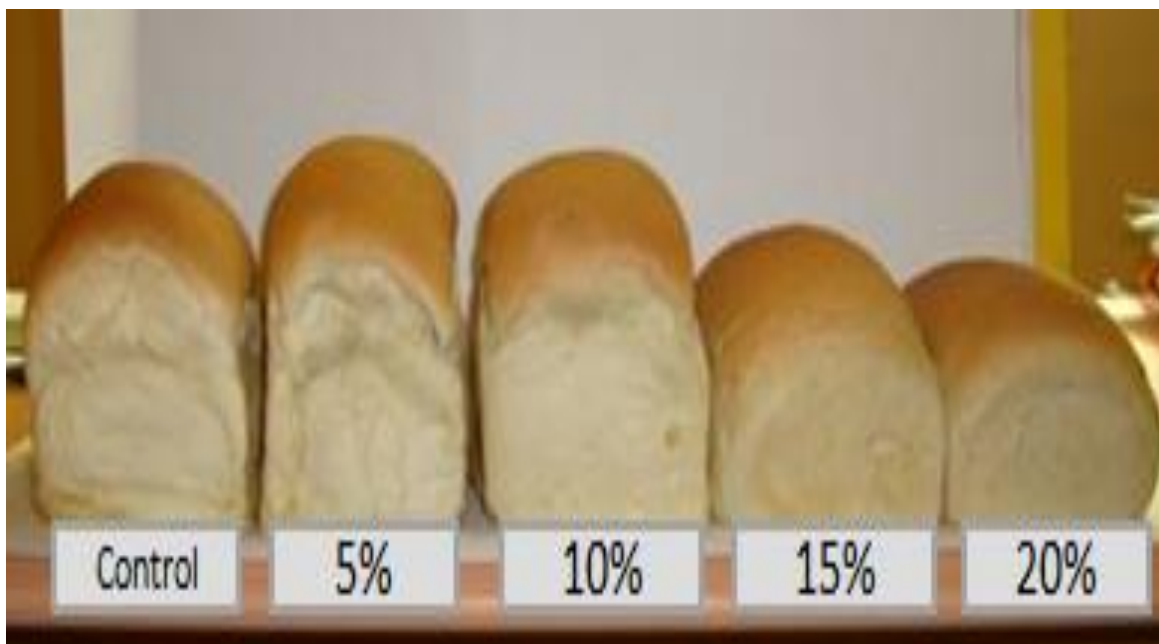


Figure 1. Effect of wheat flour substitution with cowpea flour on loaf size.

control sample with 0% level of substitution having the highest specific volume of $4.12 \text{ cm}^3/\text{g}$. It is apparent that the specific volume of the composite bread reduces at the increasing level of substitution of wheat with cowpea. As the level of cowpea content increases, the loaf volume and the specific volume decreased (Figure 1). A significant reduction in loaf volume was recorded in blend at a substitution level of 15 and 20%. This observed volume reduction in wheat structure forming proteins and low ability of dough to entrap air (poor gas retention due to the dilution effect on gluten with the addition of wheat-less flour to wheat flour (McWatter et al., 2004). Legume starch usually exhibit a restricted-swelling pattern (Reddy et al., 1984), depending on starch content as well as the presence of impurities (e.g. proteins and lipids) and pre-treatment or processing history. Physical appearances of fresh samples are shown in Figure 1. Crumb moisture ranged from 26.7% in sample with 5% level of substitution to 30.9% in sample with 10% level of substitution. Crumb moisture varied insignificantly ($P > 0.05$) in wheat-cowpea bread. Crumb moisture was comparatively higher in wheat substituted bread samples than that of wheat (control) bread. This may be attributed to increase in water absorption capacity of the cowpea-wheat flour sample McWatter et al. (2004).

The moisture content of the flour correlated significantly ($p < 0.05$) with specific loaf volume; notably, the protein and ash content of the flours had a significant negative correlation ($p < 0.05$) with specific loaf volume of the breads. The present findings are similar with the results reported by Hafiz et al. (2011) and Sharma et al. (1999) who reported that incorporation of cowpea flour in the dough had a certain negative effect on loaf volume of

bread. Loaf volume is affected by the quantity and quality of protein in the flour (Ragaei and Abdel-Aal, 2006) as well as proofing time (Zghal et al., 2002) as reported by Shittu et al. (2007). The specific volume, which is the ratio of two properties; loaf volume and weight, has been generally adopted in literature as a more reliable measure of loaf size (Shittu et al., 2007).

Sample with 5% level of substitution had the least crumb density of 0.28 g/cm^3 while the control sample (0% substitution) had highest crumb density of 0.33 g/cm^3 . The results showed that the crumb density increased as the level of substitution of cowpea increases. Crumb porosity varied significantly between 0.66 and 0.78. Both crumb density and crumb porosity increased at increasing level of substitution of wheat with cowpea. The fact that crumb density significantly ($P < 0.05$) increased from 0.28 in sample with 5% level of substitution to control sample 0.33 showed that addition of cowpea slightly reduce the crumb density of the composite bread. It shows that cowpea flour is denser than wheat flour and that was quite apparent in the structural and textural development of the crumbs. Gas (CO_2) retention and moisture diffusivity greatly determine porosity of bread samples (Wiggins, 1998). The crumb density correlated significantly ($p < 0.05$) with the protein content of the flours (Table 6).

The perceived variation in crumb moisture content, density and porosity of samples could be attributed mainly to poor gas retention and moisture diffusion abilities of the dough with progressive substitution of wheat flour with cowpea flour (McWatter et al., 2004). Oven spring, which takes place in the early period of baking, is a measure of dough strength or stability that is

basically dependent on certain factors such as thermal regime (heating rate and duration), type of flour and ingredients used in dough formulation (Gandikota and MacRitchie, 2005; Sumnu et al., 2006).

The oven spring was highest in the wheat bread sample (control), while it decreases as the level of substitution increases. Surprisingly, sample with 10% and 15% had negative values of -0.25 and -2.0 respectively. The moisture content of the flour correlated significantly ($p < 0.05$) with oven spring of the bread samples. From the works of Luyten and van Vliet (1995), it was indicated that variation in the size distribution and morphology of starch granules in the flours could have significant effect on the formation and physical properties of the crumb cell wall. In addition, polymeric changes such as starch gelatinization and protein denaturation which take place in the oven affect dough viscosity and further determine the amount of stress exerted by gas on the cell wall (Blanshard, 1987). Furthermore, excessive stress on the gas cell could lead to tensile failure and opening up of the cell wall faces (Bloksma, 1990; Fan et al., 1999), thereby leading to gas cell coalescence (dough collapse) as observed in sample with 10 and 15% wheat substitution.

Bread crust color is an important index for the initial acceptability of bread by the consumer. Unlike bread crumb color that may be similar to the color of the ingredients at dough formation, crust color is formed during baking as a result of Maillard and Caramelization reactions. Previous works have shown that instrumental measurement of baked products' color is an inevitable quality check that could be used in determining the effects of ingredient or product formulation, process variable as well as storage conditions on baked products (Erkan et al., 2006; Gallagher et al., 2003; Sanchez et al., 1995). Most of these works reported the tristimulus CIE color parameters (L^* , a^* , b^*) for the respective products' crust and crumb. The color of bread is related to physico-chemical characteristics of the raw dough and chemical reactions that take place during baking which are dependent on operating conditions, such as Maillard reactions and caramelization which causes browning of baked products during baking (Qunyi et al., 2010). However, substituted bread samples were darker when compared with the control. This darker color could also be attributed to higher Maillard reaction between reducing sugars and high protein content in cowpea (McWatter et al., 2004; Hafiz et al., 2011; Phillips et al., 2003).

In this study, L^* , a^* and b^* values, for bread crust color ranged from 46.06, 11.62 and 25.57 to 56.90, 19.71 and 32.47, respectively. One way ANOVA showed non-significant ($p > 0.05$) in L^* , a^* and b^* of the bread crusts' color. At increasing level of substitution from 5 to 20%, the lightness increases. However L^* , a^* and b^* , for bread crumb color ranged from 68.05, 0.06 and 10.04 to 75.23, 0.67 and 11.47, respectively. One way ANOVA showed that there was significant difference ($p < 0.05$) in terms of

L^* , a^* and b^* for the crumb.

Microstructure of composite cowpea-wheat bread

The actual crumb cell distribution in these samples is depicted in Figure 2, while the crumb porosity of the bread samples are shown in Figure 3. The total number of cells (TNC) and the number of small cells (NSC) per cm^2 ranged from 36.24 to 52.54 and 23.26 to 33.67, respectively. The control (non-substituted) sample had the least total number of crumb cells and small crumb cells. Increasing trend was observed from 10 to 20% wheat substitution for both total and small crumb cells of cowpea-wheat bread samples. There was no significant difference in the crumb structure of most bread samples ($p > 0.05$) except at 10% substitution level which had slightly higher total cell count (TNC) than the control sample. It was observed that at increasing substitution level, the total number of crumb cells (small and large) also increased.

Knowledge of the structure and properties of bread crumb is necessary to optimize its quality and consequently its acceptability. The averages of frequency of gray color intensity for each crumb area (200×200 squared pixels) were obtained while the coefficient of variation (CV) of GL intensity was taken as a measure of uniformity of crumb structure. This approximation was assumed since there is a relationship between cellular structures of bread crumb and the intensity of light reflected during image acquisition.

The region with finer structure reflect more light (lower gray level intensity) while regions with coarser texture reflect less light. The higher the CV value the less uniform the crumb structure (Shittu et al., 2007). ANOVA showed that the bread crumb structure were not significantly different ($P > 0.05$) from each other, but as observed, sample A with 5% level of substitution had similar cell crumb structure in comparison with the control sample in which the small cells and the larger crumb cells are loosely packed while in the samples with 10, 15, and 20% the crumb cells are closely packed; also the control sample and sample with 5% were finer in structure (lower gray level intensity) than samples 10, 15 and 20% and therefore reflect more light. The finer crumb cell structure tends to reduce as the level of substitution increases.

The differences in the crumb structure of the control (wheat) and the wheat-cowpea (composite) bread could be attributed to the dough composition such as gluten quality, which is reduced due to the diluting effect of the wheat substitution with cowpea flour (Ragae and Abdel-Aal, 2006; Tohver et al., 2005), starch pasting characteristics (Ragae and Abdel-Aal, 2006), reduced alpha amylase activity of flour (Tohver et al., 2005). Higher water content in bread consequently increased the CO_2 bubbles, hence resulted in coarser crumb of the composite bread crumb structure.

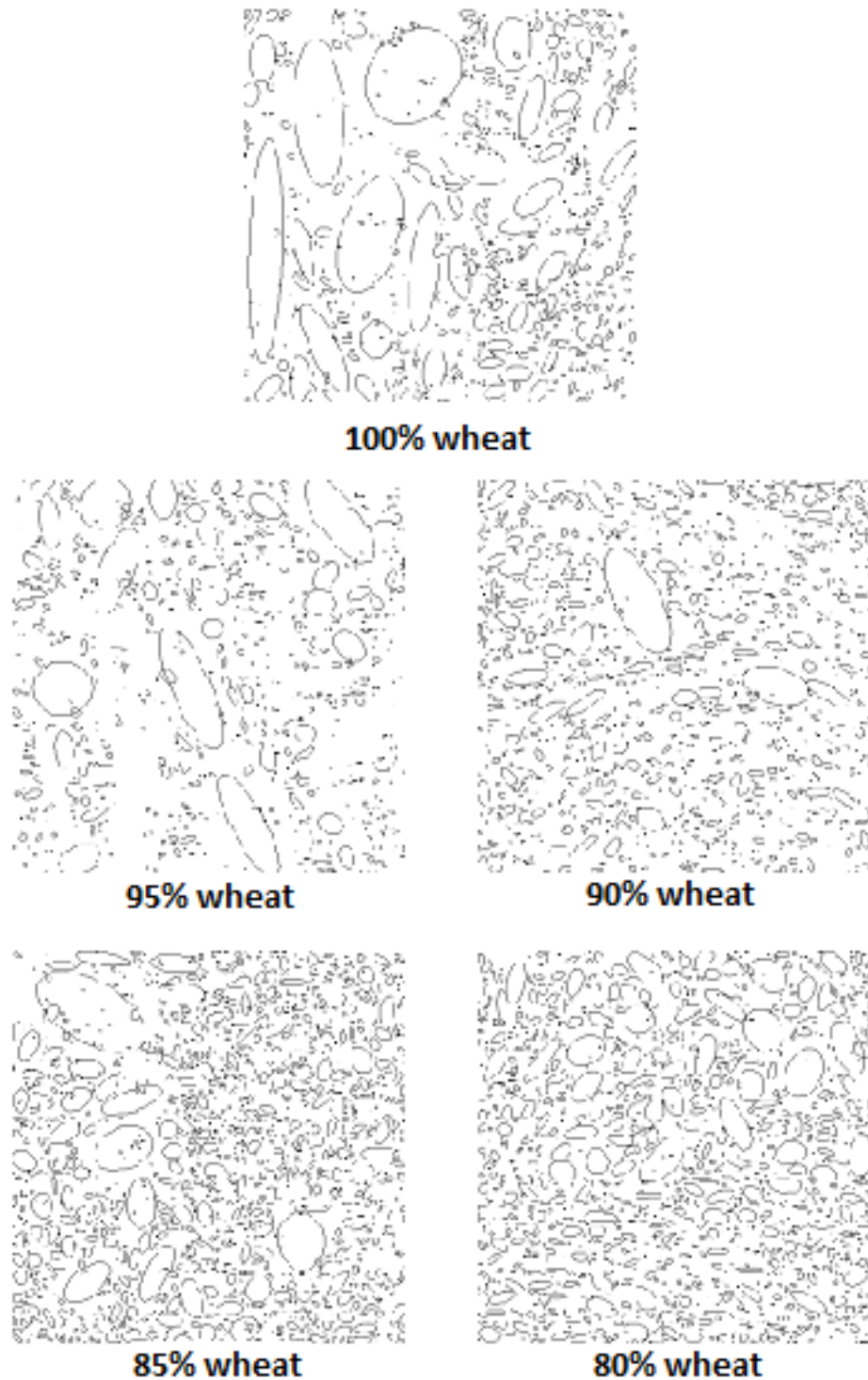


Figure 2. Crumb cell distribution at various wheat substitution levels.

Sensory acceptability scores

Table 4 shows the mean score of multiple comparisons of the composite wheat-cowpea bread samples with the control sample. There was significant ($P < 0.05$) difference in all the bread sample in terms of appearance

(5.87 to 8.40) and crust color (5.83 to 7.67), although, sample with 15% level of substitution was not significantly ($P > 0.05$) different from sample with 20% level of substitution in appearance while there was no significant ($P > 0.05$) difference in control samples and sample with 5% substitution. There was no significant difference ($P >$

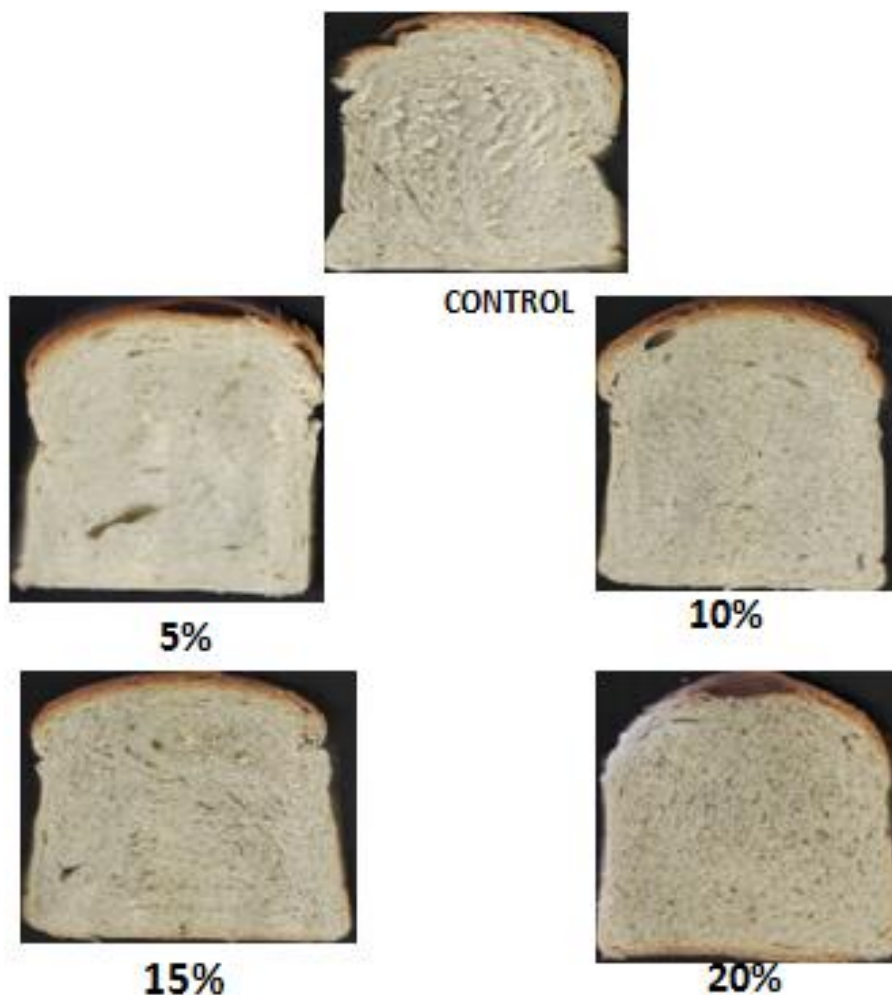


Figure 3. Crumb porosity of cowpea-wheat composite bread.

Table 4. Means (and standard deviations; n= 50) of sensory properties of composite wheat-cowpea bread.

Sub.L. (%)	Appearance	Texture	Crust color	Taste	Flavour	Overall acceptability
0	8.40 ± 0.97 ^d	8.09 ± 0.74 ^c	7.67 ± 0.99 ^c	7.80 ± 1.16 ^a	7.67 ± 1.06 ^c	8.30 ± 1.12 ^d
5	7.73 ± 0.58 ^c	7.53 ± 1.04 ^c	7.43 ± 0.85 ^c	7.23 ± 1.01 ^a	7.33 ± 0.96 ^c	7.63 ± 0.77 ^c
10	6.50 ± 0.73 ^b	6.50 ± 1.04 ^b	6.60 ± 0.89 ^b	8.67 ± 1.29 ^a	6.60 ± 1.10 ^b	6.83 ± 0.65 ^b
15	5.93 ± 1.14 ^a	6.13 ± 1.46 ^{ab}	6.13 ± 0.90 ^{ab}	6.20 ± 1.17 ^a	6.07 ± 0.97 ^b	6.40 ± 0.81 ^b
20	5.87 ± 1.41 ^a	5.67 ± 1.52 ^a	5.83 ± 1.44 ^a	5.77 ± 1.28 ^a	5.63 ± 1.38 ^a	5.93 ± 1.14 ^a

Results are expressed as mean + standard deviation. Mean values followed by different superscript letter within a column are significantly different ($p \leq 0.05$). Sub.L.: Substitution level.

0.05) in the flavour and fluffiness (texture) of samples from all the substitution levels, which precludes that wheat substitution with cowpea in this experiment, did not affect fluffiness of wheat-cowpea bread. The taste of the bread samples both wheat and composite are not significantly different, meaning all had acceptable taste as adjudged by the assessors.

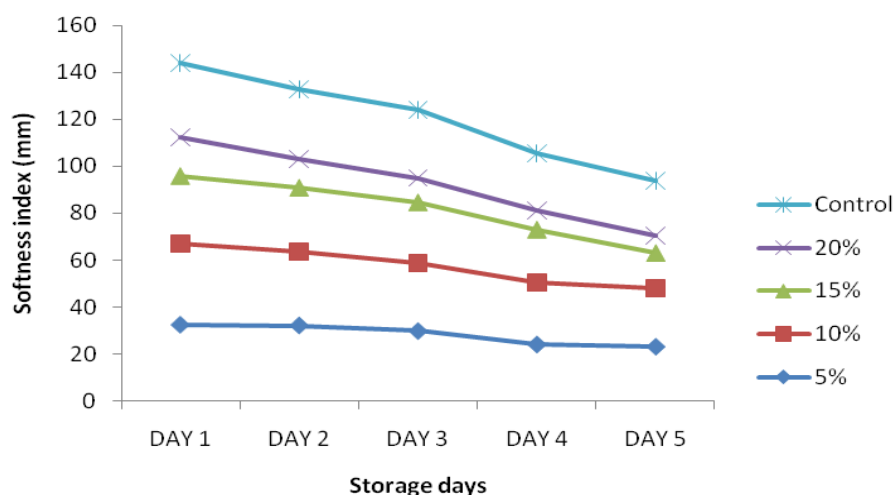
Generally, as adjudged by the assessors, the scores for the sensory quality parameters decreased

proportionally with the increasing level of substitution. Among the bread samples substituted with cowpea flour, sample with 5% substitution level was more acceptable to consumers than others, that is, control sample being the most preferred and the substituted samples followed the order 5% > 10% > 15% > 20%. The overall acceptability correlated significantly with the flour moisture, texture, crust color and flavor (fresh) but had significant negative correlation ($p < 0.05$) with protein and ash content of the

Table 5. Means (and standard deviations) mould count of composite wheat-cowpea bread.

Sub level (%)	Day 1 ($\times 10^3$) (cfu/ml)	Day 3 ($\times 10^3$) (cfu/ml)	Day 5 ($\times 10^3$) (cfu/ml)	Day 7 ($\times 10^3$) (cfu/ml)
0	1.5 \pm 0.14 ^b	7.00 \pm 0.00 ^a	34.00 \pm 0.00 ^{ab}	42.00 \pm 0.00 ^a
5	1.00 \pm 0.00 ^a	24.50 \pm 0.71 ^d	35.00 \pm 0.00 ^b	50.00 \pm 0.00 ^b
10	1.00 \pm 0.00 ^a	15.50 \pm 0.71 ^c	50.50 \pm 0.71 ^d	52.50 \pm 0.71 ^c
15	3.00 \pm 0.14 ^c	6.50 \pm 0.71 ^a	46.50 \pm 0.71 ^c	54.00 \pm 0.00 ^d
20	1.00 \pm 0.00 ^a	11.00 \pm 0.00 ^b	33.50 \pm 0.71 ^a	57.00 \pm 0.00 ^e

Results are expressed as mean + standard deviation. Mean values followed by different superscript letter within a column are significantly different ($p \leq 0.05$). Sub Level: Substitution level.

**Figure 4.** Change in softness index over storage period.

flour.

Storage characteristics of composite wheat-cowpea bread

At day 0, there was no significant difference ($P > 0.05$) in the total viable count of all the wheat-cowpea bread samples. The count however varied significantly ($P < 0.05$) from 1.00 to 3.00 $\times 10^3$, 6.50 to 24.50 $\times 10^3$, 33.50 to 50.50 $\times 10^3$, and 42.00 to 57.00 $\times 10^3$ cfu/ml at day 1, 3, 5 and 7, respectively (Table 5). Total count increased progressively with storage days but the rate of increase in the wheat substituted samples was higher than the control bread sample. Wheat-cowpea bread at 20% level of wheat substitution had the highest count at the end of the storage period (day 7) while the control sample had the least count at the end of the storage period when compared with wheat-cowpea bread samples. Most microbiological spoilage of baked products especially bread are attributed to mould growth (Cauvain and Young, 2007).

The fact that that there was no significant difference ($P > 0.05$) in the microbial count of all the bread (control and cowpea-wheat bread) samples at day 0 (freshly

produced) was due to the lethal effect of baking temperature on microorganisms. The observed low total viable count in the control sample when compared with CWC bread samples at day 1, 3, 5 and 7 could be attributed to the denser and compact crumb structure of the CWC bread which reduced the moisture migration from within the crumb to the outer surface (crust), whereas the flurffy/loose structure of the control bread samples gives air spaces which allowed moisture migration from within the bread crumb to outer surface (crust) of the bread (Figure 2). Bread ageing (staling) tendency is higher in the control sample than the CWC bread over the storage period.

In all the bread samples (control and CWC), softness index fell consistently during storage (Figure 4). Moisture migration from the crumb to the crust surface could result into stiffness within the crumb of the bread. The dense (closed) crumb cell structure that was formed at wheat replacement with cowpea and pasting characteristics of the wheat-cowpea flour could slow down the rate of stiffening in the composite bread. This is reflected in significant negative linear correlation ($p < 0.05$) between dough stability and softness index. Crumb hardness is occasioned by staling (Miyazaki et al., 2005) which is a reaction that takes place in gelatinized starch when the

Table 6. Correlation between rheological properties of wheat-cowpea flour and product (wheat-cowpea bread) quality.

	WA	DDT	DST	MTI	DOS	PMAX	EXTE	GFAC	WKDO	DSTR	ELAS	MCF	PRO	ASH	CDEN	CPOR
WA	1.00															
DDT	0.46	1.00														
DST	0.26	.921(*)	1.00													
MTI	0.70	0.74	0.76	1.00												
DOS	0.59	.903(*)	0.73	0.74	1.00											
PMAX	0.12	-0.82	-0.85	-0.41	-0.68	1.00										
EXTE	-0.71	-.914(*)	-0.70	-0.70	-.95(*)	0.58	1.00									
GFAC	-0.71	-.913(*)	-0.70	-0.69	-.935(*)	0.57	.999(**)	1.00								
WKDO	-0.29	-.97(**)	-0.87	-0.58	-.88(*)	.897(*)	0.87	0.87	1.00							
DSTR	-0.29	-.97(**)	-0.87	-0.58	-.88(*)	.897(*)	0.87	0.87	1.000(**)	1.00						
ELAS	.936(*)	0.41	0.12	0.46	0.54	0.15	-0.72	-0.73	-0.30	-0.30	1.00					
MCF	-0.08	-0.76	-0.56	-0.14	-0.69	0.78	0.73	0.73	0.88	0.88	-0.25	1.00				
PRO	0.00	0.82	0.72	0.29	0.77	-.931(*)	-0.71	-0.70	-.935(*)	-.936(*)	0.08	-.942(*)	1.00			
ASH	0.17	.89(*)	0.77	0.49	.890(*)	-.914(*)	-0.80	-0.79	-.958(*)	-.958(*)	0.19	-0.87	.969(**)	1.00		
CDEN	-0.18	0.78	0.81	0.28	0.59	-.98(**)	-0.54	-0.54	-.880(*)	-.881(*)	-0.16	-0.84	.931(*)	0.87	1.00	
CPOR	-0.50	0.46	0.65	0.20	0.33	-0.87	-0.12	-0.11	-0.56	-0.56	-0.60	-0.44	0.68	0.65	0.82	1.00

*.Correlation is significant at the 0.05 level (2-tailed)

**Correlation is significant at the 0.01 level (2-tailed)

WA: Water absorption; DDT: Dough Development Time; DST: Dough Stability; DOS: Degree of Softening; PMAX : Maximum pressure; EXTE: Extensibility; GFAC: Gluten Factor; WKDO: Workdone; DSTR : Dough Strength; ELAS: Elasticity; MCF: Moisture Content of Flour; ASH: Ash; CDEN : Crumb Density; PRO: Protein; CPOR: Crumb Porosity

amylose and amylopectin chains realign themselves.

CONCLUSION

Increasing level of substitution of wheat with cowpea flour resulted into increased total number of cells (small and large). The finer crumb cell structure reduced as the level of substitution increases thereby affecting the fluffiness and shelf stability of the composite wheat-cowpea bread. Although acceptable composite bread in terms of taste, color, tenderness and looseness, could be made from wheat-cowpea composite flour up to

15% substitution of wheat flour, higher levels of cowpea substitution significantly affected baking performance and sensory acceptability of bread. Wheat substituted bread sample of 5% was the most preferred in terms of taste and overall acceptability. Bread made from the 100% wheat is more shelf stable than the one made from wheat-cowpea flour which is attributable to more moisture bound within the composite bread crumb and the compact nature of the cell structure of the composite bread that reduce moisture migration. The wheat-cowpea composite bread was acceptable sensory-wise to the consumer for the first three days of its production beyond which the quality was impaired. The microbiological status of

the composite bread in storage revealed that the bread made with wheat flour had lower microbial load when compared with the composited wheat-cowpea bread. Composite wheat-cowpea bread stored at ambient conditions ($27 \pm 2^\circ\text{C}$, $79 \pm 3\%$ RH) was wholesome for 72 h.

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