Dough rheology and loaf quality of wheat-cassava bread using different cassava varieties and wheat substitution levels

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Abstract

Cassava flours obtained from 6 cassava varieties grown in Zambia were incorporated into wheat flour for bread making. The effect of cassava variety (CV) and cassava flour substitution level (CFSL) (wheat:cassava, 90:10, 80:20, 70:30) on dough rheology and bread quality were investigated. Dough rheology and bread baking were determined using the Brabender Farinograph and straight-dough method, respectively, while chemical composition was done using AOAC and AACC standard methods**.** There was a positive correlation (r = 0.60, $p<0.05$) between gluten content (7-13%) and water absorption capacity (WAC) (60-62%). Dough development time and stability time of the composite doughs ranged from 1.5-11 min and 6.3-12 min, respectively. Dough consistency (476-512 FU) positively correlated (r = 0.54) with gluten content. Bread specific volume (1.5-2.5 g/cm^3) varied significantly ($p<0.05$) with CV and CFSL, and correlated positively (r = 0.76, $p<0.05$) with gluten content. Flour particle size negatively correlated with WAC (r = -0.26, 0.001), bread specific volume (r = -0.72, $p<0.05$) and bread volume (r = -0.68, $p<0.05$). The flour particle size, WAC, and gluten content are significant flour properties influencing dough rheology and bread quality. The results show that wheat can be substituted with cassava flour from cassava varieties *Mweru*, *Kariba* and *Katobamputa* in bread making up to a level of 10%, without affecting bread quality negatively. Cassava inclusion generally led to reduced bread weight loss. Further work, however, needs to be done to explore use of higher levels of cassava in composite bread.

Keywords: bread quality, cassava, composite flours, farinograph, gluten, wheat substitutes

1. Introduction

Wheat is widely consumed in many African countries and ranks third after maize and cassava for daily caloric supply (Chapoto, 2010). However, continuous increases in the price of wheat in international markets, due to inflation and changes in exchange rate, is raising serious concern about the economic sustainability of large importation of wheat grain by some African countries, including Zambia. Thus, there is a growing interest to promote the use of locally produced staples for partial substitution of wheat flour in baking (Abass et al., 2016). Cassava flour has been identified for partial replacement of wheat flour (Eriksson et al., 2014).

The gluten proteins (glutenins and gliadins) in wheat are responsible for the unique viscoelastic dough that is suitable for leavened baked products (Ribeiro et al., 2018). During dough making, the hydration of glutenins and gliadins results in the development of the gluten structure, a viscoelastic network held together by covalent bonds, and to some extent non-covalent bonds (Jekle & Becker, 2015; Chen et al., 2018).

Gluten is not present in cassava flours. However, cassava flour has some attractive properties such as low tendency for starch retrogradation, good stability, high water binding capacity and good adhesive strength (Sriroth et al., 1999; Jyothi et al., 2005; Shittu et al., 2016), which could complement dough mixing properties and subsequent bread quality. Thus, it might be beneficial to determine the influence of partial substitution of wheat flour with cassava flour on dough rheology and consequently bread quality. Currently, there has been limited research on the effect of partially substituting wheat flour with cassava flour on dough rheology and bread quality, particularly on cassava varieties grown in Zambia.

Previous studies have observed that genotypes of both cassava and wheat significantly influenced the physical, chemical and functional characteristics of cassava-wheat composite flours and that bread quality varied with cassava genotype and substitution levels (Eriksson et al., 2014). Product physical characteristics were not only due to processing conditions but also varied with genotype (Ngobese & Workneh, 2018). The amylose content was reported to vary with genotype (Mejía-Agüero et al., 2012), and variations in starch types, proteins, lipids and fibre were found with different genotypes along with subtle influences of growing conditions (Halford et al., 2014; Zhu, 2015). However, the previous studies did not determine the chemical components of cassava flour that influenced dough rheology and bread quality. Differences in particle size distribution can affect water absorption capacity of flours, which subsequently influences dough rheological properties, and bread quality (Liu et al., 2015a). Sakhare et al. (2014) reported that when wheat flour was fractionated using sieving, the finer (<75 and 75–118 μm) fractions produced high-quality bread than the coarser (118–150 and >150 μm).

The cassava root is perishable due to physiological deterioration of the root immediately after harvest (Zainuddin et al., 2018). Thus, processing of cassava into flour for bread making is one strategy to reduce postharvest loss and greater diversification for cassava roots. In Zambia, cassava is a guarantee to food security and the most important staple crop after maize (Haggblade et al., 2012). The Zambian Government has prioritized improvement of cassava through breeding. One of the national agricultural strategies is to develop a viable cassava industry. As a result, a number of cassava varieties have been developed and released into the Zambian market. Nonetheless, there is no cassava variety that was developed for a specific culinary and/or food processing purpose, e.g., bread making. Therefore, there is a need to evaluate the developed cassava varieties for their potential and/or suitability for partial replacement of wheat flour in bread making. Positive evaluation of some of the cassava varieties for bread making would enhance their market value thereby encouraging their cultivation. Thus, in this work, the effects of cassava variety and substitution level on dough rheology and bread quality were measured.

2. Materials and methods

2.1. Source of materials

White bread wheat flour (Golden Cloud *since*1940™, Tiger Consumer Brands Ltd, JHB, South Africa) was procured from the local market of Pietermaritzburg, South Africa. Six cassava varieties (*Bangweulu, Katobamputa, Mweru, Kariba, Kampolombo* and *Chila)* were planted at the Mansa Root and Tuber Research Station, a branch of the Zambian Agriculture Research Station (ZARI), Mansa District, Luapula Province, Zambia, in a completely randomized block design in triplicates on a plots of 5 m with plant spacing of 1 m in January 2016, and were harvested at 18 months after planting (June 2017). The roots were collected from 5 cassava plants randomly selected from each block.

2.2. Processing of cassava flour and blending

2.2.1. Cassava flour

Cassava roots were processed into flour as described by Eriksson et al. (2014). The fresh cassava roots were cleaned to remove soil and debris. The cleaned roots were peeled by hand with a knife followed by washing in potable water. The peeled cassava were then chopped into small pieces and grated using motorized grater machine (locally fabricated) to obtain fine grated pulp. The pulp was then put into the clean polypropylene woven sacks and pressed using manually operated dewatering machine (vertical screw press). The dewatered cassava pulp was then granulated into small particles (grits) and spread on the mats placed on raised platforms. The grits were then sun-dried before drying using hot air circulation oven drier at 35 ^{o}C for 12 h. The dried grits were milled using a centrifugal mill (Retsch ZM200, Haan, Germany) at peripheral tip speed of 93 m/s.

2.2.2. Flour particle size

The particle size distribution at 90% (D90) finer particles of cassava flours was determined as described by Patwa et al. (2014) by sieving 250 g of sample for 5 min using 7 sieves with opening dimensions of 425, 300, 180, 150, 106, 90 and 38 µm. The sieves were serially stacked in the descending order with the receiver pan at the base on single shaker using vibratory mechanical shaker (DuraTap, Model DT168, Advantech Mfg. Co., New Berlin).

2.2.3. Blending of wheat and cassava flour

Three levels of wheat:cassava (90:10, 80:20, 70:30) composite flours were prepared as described in Aboaba and Obakpolor (2010). The cassava flour sample (25 g) was mixed with 225 g of wheat flour to obtain the ratio of 90:10 (wheat: cassava). Wheat flour (100 %) was used as a control in the analysis.

2.3. Chemical analysis

2.3.1. Proximate composition

Crude protein content was determined as described in Nuwamanya et al. (2010) using the Dumas combustion method of nitrogen content analysis (Leco Truspec Model FP-528, St Joseph Mi, USA) by taking about 0.3 g of sample. Crude protein was calculated as % N x 6.25. The moisture, lipid and fibre contents were determined as described in AOAC (2012) Methods 925.10, 920.39 and 962.09, respectively.

2.3.2. Amylose contents

The amylose content in cassava and wheat flour samples was determined using a Megazyme amylose/amylopectin assay kit (K-AMYL 12/16) (Megazyme International Co., Bray, Wicklow, Ireland). A flour sample (20 mg) was dispersed using dimethyl sulfoxide (Sisco Research Laboratories, Maharashtra, India) and precipitated in ethanol solution (95% v/v) (Sisco Research Laboratories, Maharashtra, India) using a centrifuge (Avanti® J-26XPI, Beckman Coulter, Inc., IN, USA). The precipitated starch sample in an acetate/salt solution and lectin concanavalin A (Con A) was precipitated to remove amylopectin by centrifugation using a mini centrifuge (Microfuge® 16, Beckman Coulter, Inc., Krefeld, Germany). The amylose, in the supernatant, was enzymatically hydrolysed to glucose by using a amyloglucosidase/α-amylase enzyme system. The absorbance of colour developed was measured at 510 nm by using a UV-Vis spectrophotometer (UV-1800PC, Shimadzu Corpor., Kyoto, Japan). The concentration of amylose in the starch sample was then estimated as the ratio of absorbance of the supernatant at 510 nm of the Con A precipitated sample to that of the total starch sample.

2.3.3. Gluten content

The gluten content was determined by the hand washing method using 2% sodium chloride solution (Sisco Research Laboratories, Maharashtra, India) by taking about 10 g flour sample as described in AACC (2011) Method 38-10. 2.8.

2.4. Rheological properties

The rheological properties of doughs from wheat flour alone (control) and composite wheat-cassava flours were determined with a Brabender Farinograph (Model 820603, Brabender OHG, Duisberg, Germany) at 30±0.2 °C using a 300 g mixing bowl operated at 63 rpm according to AACC (2011) Method 54-21.

2.5. Preparation of bread

Bread was baked according to the straight-dough bread-making method (AACCI Method 10-09) (AACCI, 2000). The flour (250 g), 25 g sugar, 3 g salt, 5 g baking fat (Margarine, Spar Group Ltd., PMB, South Africa) and 2.5 g baker's yeast (Gold Star, Anchor Yeast Ltd., JHB, South Africa) were weighed into a mixing bowl. About 150 mL water was added and the mixture was kneaded by hand and proofed for 45 min in the SelfCookingCentre® (Rational AG, Landsberg am Lech, Germany) at 30 °C and 100% humidity. After proofing, the dough was re-kneaded and divided into 70 g portions (three portions for each blend), moulded and placed in separate oil greased (Margarine, Spar Group Ltd., PMB, South Africa) baking pans. The baking took place in the SelfCookingCentre® in the temperature range of 178-193 ^{o}C for 16 min. The quality characteristics of the bread were determined after cooling at room temperature (20-22 ^{o}C) overnight.

2.6. Bread quality characteristics

2.6.1. Crumb and crust colour

The breadcrumb and crust colour; L* (degree of lightness), a* (redness to greenness) and b* (yellowness to blueness) were measured using a HunterLab ColorFlex instrument (Hunter Associate Laboratories Inc., VA, USA). The crumb samples were prepared by slicing 4 mm thickness of the cross section of the bread and slicing-off the crust area. The crust samples were obtained by thin sectioning (1-2 mm thickness) of the bread crust. The samples were placed directly on sample hold and covered with black cup (HunterLab Cup). The whiteness value of crumb and brownness of crust were calculated as described by Zhu et al. (2016) using the equation:

$$\text{Whiteness index} = \sqrt{[(100 - \mathrm{L})^2 + \mathrm{a}^2 + \mathrm{b}^2]} \tag{1}$$

$$\text{Brownness Index (BI)} = \frac{100 \text{ x } (\chi - 0.31)}{0.17} \tag{2}$$

where

$$\chi = \frac{\mathrm{a} + 1.75\mathrm{L}}{5.645\mathrm{L} + \mathrm{a} - 3.01\mathrm{b}} \tag{3}$$

2.6.2. Bread specific volume and density

Bread mass (W) was measured using a digital balance (Kern & Sohn, GmbH, Balingen, Germany). Bread volume (BV) was determined using a modification of the AACC Method 10-05 rapeseed replacement method using maize grit (1.0 mm) instead of rapeseeds (Eriksson et al. 2014). The bread was placed in a container of known volume (VC) and the basin filled to the brim with grits, bread was removed and the volume of the grits (VG) was measured using a measuring cylinder.

$$\text{Loaf volume, BV (cm}^3) = \text{VC} - \text{VG} \tag{4}$$

$$\text{Density (g/cm}^3) = \frac{\text{W}}{\text{BV}} \tag{5}$$

$$\text{Specific volume (cm}^3/\text{g}) = \frac{\text{BV}}{\text{W}} \tag{6}$$

2.6.3. Weight loss

The weight loss of the bread in percent was determined as described in Bakare et al. (2016).

$$\text{Weight loss (\%)} = \frac{\text{A}-\text{B}}{\text{A}} \text{ x } 100 \tag{7}$$

where A = weight of dough; B = weight of baked bread.

2.6.4. Breadcrumb pore size characteristics

The bread morphology and pore size of the crumb were studied using a scanning electron microscope (SEM) as described in Hayta and Ertop (2018) with modifications. The bread crumb samples of ~ 5 x 5 x3 mm were prepared and mounted on the SEM sample stubs with double adhesive tape. Freeze-drying and gold sputtering were skipped to obtain actual crumb pores (gas cells). The crumb pores were evaluated using the variable pressure mode (VPM), which allows viewing of fresh samples (uncoated specimens) using an environmental SEM (ESEM) (EVO LS15, Carl Zeiss Microscopy, Jena, Germany). The SEM images were subjected to image analysis for pore size (cross section area in mm^2) estimation using the Soft Imaging System (GmbH, Munster, Germany). The porosity was expressed as total pore area to the total surface area of the image.

2.7. Experimental design and statistical analysis

A completely randomized design with two factors: Cassava variety and blend ratio (cassava concentration) was used. Triplicate data were measured using two-way ANOVA. Pearson's correlation and multivariate principal component analysis (PCA) were done using GenStat (18th Edition software, VSN International Ltd., Hemel Hempstead, UK) and mean differences were determined using Fisher's Least Significance Difference (LSD) test at 5% significant level ($p<0.05$). Although $p<0.05$ was generally used, $p<0.01$ and $p<0.001$ were used for some of the data to indicate greater significance differences.

3. Results and Discussion

3.1 Proximate composition of cassava and wheat flours

The moisture content of the cassava flours ranged from 10 to 12% compared to 13± 0.2% for wheat flour (Table 1). The protein content of the cassava flours ~~was in the ~~ranged from 1.2–1.9% (Table 1). The protein content of the cassava flours was very low compared to that of wheat flour (11±0.3%). Wheat flour protein contain about 85% gluten proteins (glutenins and gliadins) (Avramenko et al., 2018; Ribeiro et al., 2018), while cassava flour protein is gluten-free (Chakrabarti et al., 2017). The lipid content of cassava flour ranged between 0.15 and 0.63%. The lipid contents in all cassava flour varieties were significantly ($p<0.05$) lower than in wheat flour (1.7±0.2%). The lipids reinforce gluten structure through lipid-protein interactions (Avramenko et al., 2018). The fibre content (0.03–0.6%) of the cassava flour was significantly ($p<0.05$) lower than that of the wheat flour (2.9±0.1%). Leavened aerated bread cannot be made without wheat flour because of viscoelastic dough making properties of wheat gluten proteins (Ceresino et al., 2018). Blending of cassava flours in wheat flour influences the blended dough rheological properties and bread nature by diluting wheat protein content and gluten proteins functionality.

3.2 Amylose content

The amylose content in cassava varieties ranged from 16–27% and 21±0.5% for wheat flour. The amylose content of cassava were reported previously, 19-20% (Morante et al., 2016), 23% (dos Santos et al., 2018), and 17-26% (Liu et al., 2019). The amylose content is the basis of classifying starches into waxy, semi-waxy, normal/regular and high-amylose types when amylose content is 0-2~~%~~, 3-15~~%~~, 16-35, and >35% of the total starch, respectively (Tester et al., 2004; Morante et al., 2016; Botticella et al., 2018). The result showed that all the cassava flour varieties including wheat flour were generally classified as normal/regular starches. The amylose content in *Katobamputa* was significantly different ($p<0.05$) from the other cassava varieties. There was no significant difference ($p>0.05$) between the amylose content of the wheat flour and that of any of the cassava varieties. High amylose content can reduce starch granules swelling and significantly increase the level of resistant starches (Hallström et al., 2011).

3.3 Particle size of cassava varieties and wheat flours

Cassava varieties flour average particles size ranged from 250–333 μm (Table 1) and varied among varieties. The average particle size of the wheat flour was low compared to the particle size of cassava flours. The particle size of flour is an important factor that can affect the baking properties and end product quality (Vouris et al., 2018). Particle size is influenced by the milling technique applied and inherent hardness differences of wheat grain and cassava flour varieties (Liu et al., 2015b). Reduction of flour particle size during milling can result in a high proportion of damaged starch granules leading to high water absorption capacity of the flour and high susceptibility of starches to enzymatic hydrolysis, both of which can affect bread quality (Wang et al., 2017).

3.4. Water absorption capacity (WAC)

The moisture content of the composite flour blends ranged from 13 to 14% (Table 2) and increased with increase in CFSL. The WAC results for the flour blends were in the range 60–62, 61–62, and 60, at 10, 20, and 30%, respectively, and negatively correlated with CFSL ($r = -0.65$, $p<0.05$) suggesting that higher CFSL resulted in decreasing WAC, in part, due to large particle size of cassava flour with low water absorption capacity. There was a weak positive correlation between WAC and protein ($r = 0.34$, $p<0.01$) and fibre ($r = 0.36$, $p<0.01$) contents. The high protein and fibre levels in wheat flours are significant contributors toward water absorption. The crude protein contents were generally very low in cassava with no significant difference among the cassava varieties ($p>0.05$) (Table 1). The fibre contents of the cassava varieties (≤0.6%) were low compared to wheat flours (2.9%) and hence the contribution of cassava fibre to WAC was likely to be low. Nevertheless, the difference in fibre content can bring a difference in water absorption of wheat flours. A study by Struck et al. (2018) observed that addition of almond fibre significantly reduced WAC of wheat flour.

In a similar study on potato-wheat flour, higher protein contents increased WAC of wheat flour (Sarker et al., 2008). According to Liniņa et al. (2014), water absorption of weak flour is below 55%, of medium flour 54–60%, and strong above 58%. The WAC of the flour blends in the present study was characteristic of strong flours and showed significant correlations with gluten content ($r = 0.60$, $p>0.05$), an indication that high gluten content resulted in a high WAC. There was a weak negative correlation between WAC and flour particle size ($r = -0.26$, $p<0.01$), which indicates that smaller particle size flours had higher water hydration capacity.

3.4.3. Gluten content

The dry gluten content of wheat flour was 13±1% and decreased with increase in CFSL. The mixing of wheat flour with water transforms gluten proteins into viscoelastic gluten structures that ultimately determine the quality of the final bread product (Sissons & Smit, 2018). The negative correlation between protein content and CFSL (r = -0.77, p<0.05) suggests that inclusion of cassava flour resulted in decreasing gluten proteins of wheat flour. Cassava flour does not contain proteins found in wheat and hence has a diluent effect during wheat gluten development, an effect also observed by Collar andArmero (2018). Thus partial replacement of wheat flour with cassava flour could reduce bread volume because of dilution in wheat gluten functionalities (Šárka et al., 2017). Flour particle size had a significant negative correlation (r = -0.53, p<0.05) (Table 3) with gluten development implying that smaller particles hydrate faster and thereby promote migration of excess water to the gluten network.

3.5. Dough rheological properties

3.5.1 Dough development time (DDT)

The DDT of composite flours was in the range 1.5-11 min, and increased with increase in CFSL. The DDT of the control (wheat) flour (2.1±0.1 min) was not significantly different from that of composite flours of *Katobamputa, Mweru* and *Kampolombo* at 10%, Kariba at 20%, and *Mweru* at 30%. The DDT showed weak negative correlation with WAC (r = -0.44, p<0.01), gluten content (r = -0.28, p>0.001) and positive correlation with flour particle size (r = 0.45, p<0.05) (Table 4). Reduced WAC inhibits gluten development. Additionally, excess water beyond that required for gluten development can cause weakening of gluten matrix leading to delayed dough development (Jafari et al., 2018). Higher WAC increases hydration of gluten and hence contribute to quicker dough development. Jafari et al. (2018) reported that decreased gluten hydration is the main reason for high DDT. The high DDT observed in this study could be attributed to decreasing gluten content with increasing cassava flour content, which might have disrupted the formation of the gluten network (Zhang et al., 2018), thus increasing dough development time (Eduardo et al., 2013). DDT is influenced by protein content (Huang et al., 2016). The positive correlation between DDT and flour particle size suggest that large particle size is a significant contributor to the low hydration capacity that might have delayed gluten development resulting in extended period of dough development.

3.5.3. Consistency

Cassava variety and main interaction (CV x CFSL) had significant (p<0.05) influence on the consistency of dough. The peak consistency value of the wheat dough (512 FU) was higher than those of the composite doughs, which varied among flour blends, showing a decreasing trend with increasing CFSL (r = -0.65, p<0.05). This indicates that inclusion of cassava flour resulted in decrease in dough consistency. Dough consistency positively correlated with gluten content (r=0.54, p<0.05), which is expected because gluten is largely responsible for dough structure and strength. Dough consistency positively correlated with water absorption capacity (r = 0.45, p<0.01) and protein content (r = 0.60, p<0.05) indicating that strong, high gluten content dough would be of high dough consistency. The dough consistency correlated negatively with flour particle size (r = -0.48, p<0.01) implying that doughs with smaller particles had higher consistency than doughs made with flour of large particle size.

3.5.4. Dough stability time (DST)

The DST at 10% cassava flour substitution level was in the range 7.1–11 min and decreased when cassava flour substitution level was increased to 20% except for varieties *Bangweulu* and *Kariba*. DST showed weak negative correlation with CFSL (r = -0.27, p<0.001) suggesting that high DST were associated with low CFSL. The DST of the composite doughs increased when CFSL was increased to 30%, and DST values were similar (p>0.05) to the DST of the control (12.2±0.2). Dough stability time indicates the tolerance of the dough to mixing stress. Flour with a DST >10 min is resistant to mechanical stress (Edun et al., 2018), and is classified as flour of excellent quality, and flour of poor quality has stability time of about less than 3 min (Liniņa et al., 2014). DST had a negative correlation (r = -0.51, p<0.05) with particle size implying that DST increased with reduced particle size, presumably because smaller particle size favour uniform and high water absorption that in turn enhances gluten development. Wang et al. (2017) reported that reducing the particle size strengthened the gluten network, and resulted in shorter development time and longer mixing stability of the dough. DST showed significant correlation with protein (r = 0.64, p<0.05) and fibre content (r = 0.61, p<0.05). The DST of the composite flours were generally poor at 10% and 20% CFSL, but improved at 30% substitution level. Increased fibre along with starch content might have contributed to an increase in the water absorption required for the development of gluten structure (Hrušková & Švec, 2018). The increased starch-protein interaction may have contributed to increased hydrogen bonding in the starch-gluten interaction and thus contributed to the stability of the gluten network. A similar, observation was made by Zhang et al. (2018) on tapioca starch-wheat composite flours.

3.5.5. Mixing tolerance index (MTI)

The MTI ranged from 11–40 FU across varieties for all blend ratios and did not vary with CFSL (p>0.05). The MTI for the control was 24±1 FU and was not significantly different from *Katobamputa* (25±1 FU) at 10%, *Kariba* (22±1) at 20%, *Kariba* and *Kampolombo* at

30%. Mixing tolerance index (MTI) indicates the degree of dough softening over a period of mixing (Srikanlaya et al., 2018). The lower the MTI value the better quality. According to Liniņa et al. (2014), dough mixing quality is considered satisfactory if mixing tolerance is below 70 FU. Doughs with mixing tolerance values higher than 110 FU are considered weak and are characterised by difficulties in mechanical handling during dough making. Depending on its quantity and composition (Gómez et al., 2003), dietary fibre can have a dilution effect on gluten proteins (Ho & Aziah, 2013). All the composite flour samples in this study, irrespective of variety and CFSL, had satisfactory mixing tolerance, which indicates good dough mixing quality. Values obtained in this study were in the range considered satisfactory for good dough mixing quality. However, the mixing tolerance showed poor correlation with other mixing properties. Similar results were reported by Isah (2017) in the study of African locust bean pulp flour incorporated into wheat flour at different ratios. It seems likely that high levels of cassava flour contributed a large amount of starches which may have weakened gluten structure.

3.6. Bread quality

3.6.1. Bread volume (BV) and bread specific volume (SV)

Table 4 shows the bread volume, specific bread volume and density of bread made from six different cassava varieties-wheat flour blends. The BV of the control (0% cassava flour) was 148±10 cm^3. The BV decreased with increase in CFSL. The volume of bread made from flour blends ranged from 103–140, 103–120, and 92–105 cm^3 at 10, 20, and 30% CFSL, respectively. Bread volume (BV) correlated strongly and positively with gluten content ($r = 0.78$, $p<0.05$), protein ($r = 0.77$, $p<0.05$), WAC ($r = 0.66$, $p<0.05$), dough consistency ($r = 0.60$, $p<0.05$) and negatively with DDT ($r = -0.51$, $p<0.05$). The correlations indicate that flour samples with relatively high gluten content had good mixing properties, including good consistency, short mixing time and resistance to stress and yielded quality bread with respect to BV. Further, the BV showed negative correlation ($r = -0.68$, $p<0.05$) with particle size (Table 4), indicating flours of small particle size produced bread of large volume. Similarly, Jacobs et al. (2018) reported that flours of smaller particle size produced large volume bread. This observation can be attributed mainly to the observation described earlier, that flours of small particle size have high water absorption capacity (WAC), which promotes good gluten development.

The SV for bread from 100% wheat was the highest (2.5±0.2 g/ cm) and the value decreased as cassava flour level increased. The SV correlated positively with WAC ($r = 0.63$, $p<0.05$), gluten content ($r = 0.76$, $p<0.05$), DST ($r = 0.39$, $p<0.01$), and negatively to DDT ($r = -0.52$, $p<0.05$). The highest SV was obtained at 10% cassava flour substitution of *Katobamputa*, *Mweru* and *Kariba*, and was not significantly different ($p>0.05$) from that of control wheat bread. These results are similar to those of Eriksson et al. (2014), who observed insignificant variation in cassava variety effect on SV of bread. The decrease in bread SV with an increasing amount of cassava flours has been reported by several authors (Eggleston et al., 1993; Aboaba & Obakpolor, 2010; Eriksson et al., 2014). The SV values obtained in the previous studies were higher than the SV values of the present study, which was probably influenced by additional ingredients. Ingredients such as concentrated milk have an enhanced emulsifying effect (Julianti et al., 2017), which promotes emulsification of the shortening, which improves bread quality. The negative correlation between flour particle size and SV ($r = -0.72$, $p<0.05$) again indicating that smaller flour particle size is associated with large bread volume likely due to high WAC of the flour.

3.6.2. Bread density (BD)

The density of wheat bread (control) was the lowest (0.4±0.03 g/cm^3) and BD values increased with increase in CFSL. Increased CFSL favoured starch-starch/starch-protein system more than protein-protein interactions, which can weaken gluten structure. During baking, starch granules lose birefringence properties through swelling and leaching of amylose and this results in an increase in viscosity and migration of plasticising water from gluten to starch (Verbauwhede et al., 2018). Higher absorption capacities due to damaged starch of cassava flour (Nindjin et al., 2011) can deplete water to a level lower than is required for gluten structure, which can lead to inhibition of expansion of gas cells and hence a dense (less foam) crumb structure at the end of the ovenspring. The bread density positively correlated with flour particle size ($r = 0.67$, $p<0.05$) indicating that flours of larger particle size had low expansion capacity when processed into dough and then bread. Bread density was significantly negatively correlated with gluten content ($r= -0.72$, $p<0.05$) and WAC ($r = -0.65$, $p<0.05$) but positively correlated with DDT ($r = 0.58$, $p<0.05$), indicating that high BD values were associated with flours of low gluten content, low WAC and high dough development time.

3.6.3. Weight loss (WL)

The WL of wheat bread (control) was 14±1%, and was similar to the WL of the bread containing the cassava varieties *Bangweulu* at 20%, *Chila* at 20% and *Bangweulu* at 30% CFSL. Weight loss ranged from 10-13% across all CFSL, and there was no clear pattern of change in WL. Vouris et al. (2018) reported weight loss of wheat bread in the range of 16-18%, values somewhat higher than those obtained in this study. Weight loss occurring during the baking stage of bread processing may be due to both fermentation processes, evaporation of water as well as volatilization of low molecular weight compounds produced during fermentation, including ethanol (Bakare et al., 2016; Verbauwhede et al., 2018) during baking (Shittu et al., 2007b).

3.7. Bread crumb colour and crust colour

The bread crumb lightness values (L* values) for flour blends ranged from 73–75, 72–73, and 71–76 at 10, 20 and 30% CFSL, respectively (Table 5), and did not vary with CFSL. The L value (74±0.02) of the control was similar ($p>0.05$) to the L values of bread samples containing cassava flour at 20% CFSL for *Mweru*, 30% CFSL for *Kampolombo* and 30% for *Chila*. These results indicate that cassava flour of different cassava varieties can be incorporated at different substitution levels to obtain bread with lightness similar to that of the control. The bread crumb redness-greenness (a^*) ranged between -0.4 (green) and 0.7 (red), and varied significantly ($p<0.05$) across blend ratios. However, their values were too low to significantly reduce lightness. These traces of weak red and green could be attributed to carotenoid pigments in wheat flour (Zhai et al., 2018) and residual pigment due to reddish peels of cassava. The crumb yellowness (b^*) ranged between 20 and 21 across the blend ratios, and varied significantly ($p<0.05$). Crumb yellowness could be attributed, in part, to the non-enzymatic reaction between reducing sugar and proteins to develop a yellow-brown colour. Flour yellowness was most probably as a result of the accumulation of carotenoids in the wheat grain flour used (Zhai et al., 2018). A combination of lower a* and higher b* values increased the whiteness index.

The whiteness index ranged between 65 and 68 and did not vary with CFSL ($p>0.05$). The whiteness of the control was 67±0.01 and was similar ($p>0.05$) to the whiteness of *Bangweulu, Kariba, Katobamputa,* and *Chila* at 30% CFSL. The whiteness of the crumb significantly correlated positively ($r = 0.85$, $p<0.05$) with lightness, implying that increased lightness produced a high level of whiteness. Nevertheless, whiteness is affected by the yellowness of the crumb. The increased crumb yellowness reduced lightness resulting in decreased whiteness. Crumb colour affects consumer preference of bread because it is perceived as a measure of quality. Crumb colour is influenced by the colour of flours and other ingredients (Shittu et al., 2007a), as well as non-enzymatic reactions, which can contribute to yellowness or brownness. The desired colour of wheat flours for industrial applications is a high value for lightness (L^*) and low value for Chroma (Sankhon et al., 2014; Vasconcelos et al., 2017). The Chroma was in the range 19-22 and varied ($p<0.05$) with CFSL. However, there was no clear trend in the variations across the flour blend ratios. Chroma is influenced by redness-greenness and yellowness. Chroma positively correlated with a* ($r = 0.74$, $p<0.05$) and b* ($r = 1$, $p<0.05$), suggesting that higher levels of crumb a* and b* increased the chroma.

The bread crust lightness values (L*) ranged from 51–64, 51–62, and 54–64 at 10, 20, and 30% CFSL, respectively (Table 6). The bread crust colour of the control (59±0.2) was similar to the bread processed with the inclusion of cassava flour at 30%, CFSL for *Mweru* and at both 10% and 20% CFSL for *Kampolombo* ($p>0.05$). Crumb colour was lighter than crust colour. The acceptable range of lightness (L^* value) for bread crust is 54 to 62 (Fu et al., 2018) which was achieved by most bread samples except for *Mweru* at 10 and 20% CFSL. Crust redness values (a*) ranged from 12 to 17, and varied ($p<0.05$) with CFSL without a clear trend. Crust yellowness (b*) values were in the average range of 35-39, and varied ($p<0.05$) across CFSL. The colour shift from crumb to crust was characterised by increased a* and b* values. It is generally acceptable to have darker crust than crumb, therefore the relative colour of the crust and crumb of the bread samples of the current study are acceptable.

The brownness index of the wheat bread (control) was 109±2, and the value was similar ($p>0.05$) with bread samples containing flours of the cassava varieties *Bangweulu* and *Katobamputa* at 20% CFSL, and *Chila* at 30% CFSL($p>0.05$). The reduced lightness and increased chroma resulted in an increased brownness index. The BI correlated negatively with WL ($r = -0.59$, $p<0.05$) and BV ($r = -0.35$, $p>0.01$) and positively ($r = 0.59$, $p<0.05$) with the weight of the bread (Table 7). In addition, the weight of the bread was strongly correlated negatively ($r = -1.00$, $p<0.05$) with weight loss. The high BI of the bread was typical of reduced BV and WL with increased bread weight. Browning can be ascribed to the products of the Maillard and caramelisation reactions that occur during dry heating as in baking (Shen et al., 2018). The BI showed weak negative correlation with protein content ($r = -0.24$, $p<0.001$). The flour blends with higher CFSL had lower protein content and higher BI.

Increased starch contents in the flour blends may possibly lead to higher levels of reducing sugars available for the Maillard reaction that results in high BI of the bread (Buckman et al., 2018). Crust chroma of the control was 39±0.4, and for crust chroma of the bread samples containing cassava flour was in the average range of 37-42, and varied across CFSL, without a clear pattern. Compared to the crumb, the crust colour had higher a* and b* values.

3.8. Bread crumb pore area

The pore area of the control was 0.47 mm^2, whilst the average crumb pore area of experimental breads increased with cassava flour substitution level (CFSL) from 0.4-0.8, 0.4-0.7, and 0.3-0.9 mm^2 at 10, 20, and 30%, respectively, across the varieties (Fig 1). The porosity of the control 71±0.5% (Fig. 2) differed significantly ($p<0.05$) from the porosities of experimental bread, which decreased with increasing CFSL. Crumb porosity was negatively correlated ($r = -0.57$, $p<0.05$) with cassava flour particle size. This indicates that flours with smaller particles produced bread of higher porosity. Crumb porosity correlated positively with SV ($r = 0.66$, $p<0.05$) and gluten ($r = 0.64$, $p<0.05$). (Table 7). The bread of large volume was associated with higher gluten content, which promotes appreciable pore formation and better gas retention during proofing. Fig. 3 shows photos of typical crust and crumb structures. Espinosa-Ramírez et al. (2018) reported that large bread volume is related to better retention of carbon dioxide during proofing. Increasing CFSL led to increased bread density and a decrease in crumb porosity. Analysis of the crumb structure of the experimental bread, which had low porosity, showed that the microstructures of the bread crumbs were characterised by a continuous dense mass (Fig. 4). The increased levels of cassava flour seem to have weakened the gluten network by disrupting the intermolecular disulphide bonds in the glutenin and gliadins molecules, thereby limiting protein-protein interaction. The fat and protein contents at higher cassava flour substitution possibly increased hydrophobicity (Muoki et al., 2015; Uthumporn et al., 2017), thereby limiting available water for gluten development. Hydration is primarily responsible for the development of the gluten network (Chen et al., 2018), hence any other flour component with strong water absorption capacity is likely to limit gluten development by reducing the hydration of the gluten proteins and consequently affect bread quality negatively.

4. Multivariate analysis

Principal component analysis (PCA) was used to provide in-depth analysis of the differences among the cassava varieties. The scree plot (Fig. 5A) showed that the cassava flour properties had low percentage variations resulting in no significant differences among the cassava varieties ($p<0.05$). This implies that the cassava flour properties did not exhibit distinct separation among the cassava varieties. The scree plot (Fig. 5B) showed that partial replacement of wheat with cassava flour yielded high percentage variations resulting in distinct separation among cassava varieties. This suggests that wheat flour properties were significant contributors to differences among cassava varieties across the blend ratios. The dough mixing properties (Fig. 5C) showed wheat clustered separately from cassava varieties. It is worth noting that based on experimental design and the plot, all cassava varieties at zero (0%) CFSL overlapped with the actual coordinates of 100% wheat flour resulting in over clouding (hence black shade). Based on the coordinates of the plot *Mweru* formed distinct separation from other varieties and closely associated with *Katobamputa* and *Kariba*. These varieties (*Mweru*, *Katobamputa* and *Kariba*) were near the coordinates of wheat, and did not cluster together with other varieties. The variety *Kampolombo* was disparate but strongly overlapped with *Bangweuru* and *Chila*. For the bread quality characteristics (Fig. 5D), bread baked from composite flour of *Mweru* could be distinguished from other varieties and was close to the coordinates of wheat bread without interference from other varieties. Similarly, *Katobamputa* and *Kariba* clustered separately on the same coordinates as those for wheat bread, however with little overlaps with the other varieties. The PCA plot (Fig. 5E) described the effect of flour properties on dough mixing characteristics. According to Mtunguja et al. (2016), based on coordinates of the plot, values close to the origin have smaller impact on the plot pattern, while those further away are significant contributors. The plot showed that the impact of flour properties on dough characteristics varied with CFSL. The control (wheat) dough was distinguished from cassava varieties. The flour proximate properties (protein, fibre and lipids) closely associated (clustered together) on the PCA plot and strongly correlated with D90-axis but in the opposite direction. This indicates that proximate contents had similar effect on dough mixing properties (Equations 8 and 9), and are impacted negatively by D90. Gluten content, flour particle size and water absorption capacity were distinctly separated on the plot, suggesting that these properties impacted dough mixing differently. The DDT is closely associated with WAC but in the opposite direction, indicative of negative effect of water absorption capacity on dough development time. The Mixing dough properties of cassava varieties *Mweru*, *Katobamputa*, *Kampolombo* and *Kariba* (clustered in the bottom left of plot) were strongly influenced by WAC and gluten content. However, all cassava varieties clustered strongly along D90-axis, indicative of variations due to flour particle size. The PCA plot (Fig. 5F) explained the effect of flour properties on bread quality characteristics. Flour particle size, water absorption capacity and gluten contents showed different correlation coefficients (Equations 10 and 11) and were distinct on the plot suggesting that they were significant source of variations on the bread quality. The proximate contents (protein, fibre and lipid) were clustered together in the plot suggesting similar correlations and contributions to the bread quality characteristics. The coordinates of bread volume and bread specific volume were close together, indicative of similar response to flour properties. The bread made from *Mweru* and *Katobamputa* clustered separately along the axis of WAC and gluten, respectively, indicative of strong influence from water absorption and gluten content. The bread from all cassava composite varieties showed strong presence along and towards D90-axis. This suggests that flour particle size distribution were the source of variations among the cassava varieties.

$$\mathrm{PC1} = 0.09X_1 - 0.37X_2 + 0.40X_3 + 0.33X_4 + 0.40X_5 + 0.40X_6 + 0.22X_7 + 0.30X_8 - 0.20X_9 + 0.27X_{10} - 0.12X_{11} \qquad (8)$$

$$\mathrm{PC2} = 0.10X_1 - 0.07X_2 + 0.08X_3 - 0.21X_4 + 0.09X_5 + 0.09X_6 - 0.43X_7 - 0.30X_8 + 0.38X_9 + 0.47X_{10} - 0.52X_{11} \qquad (9)$$

where X_1=Amylose, X_2=D90, X_3=Fibre, X_4=Gluten, X_5=Lipids, X_6=Crude protein, X_7=Water absorption capacity, X_8=Dough consistency, X_9=Dough development time, X_{10}=Dough stability time, X_{11}=Mixing tolerance index

$$\mathrm{PC1} = 0.06X_1 - 0.32X_2 + 0.34X_3 + 0.31X_4 + 0.35X_5 + 0.35X_6 + 0.23X_7 + 0.35X_8 - 0.34X_9 + 0.35X_{10} + 0.11X_{11} \qquad (10)$$

$$\mathrm{PC2} = 0.19X_1 - 0.32X_2 + 0.13X_3 - 0.27X_4 + 0.17X_5 + 0.16X_6 - 0.53X_7 - 0.16X_8 + 0.12X_9 - 0.09X_{10} - 0.60X_{11} \qquad (11)$$

Where X_8=Bread volume, X_9=Bread density, X_{10}=Bread specific volume, X_{11}=Weight loss

5. Conclusion

Cassava variety and CFSL had varying effects on dough rheology and bread quality. The differences among cassava varieties were attributed to variation in cassava flour particle size, a parameter that affected the WAC~~)~~ and SV~~)~~. The bread SV was high (2.1, 2.1, and 2.3 cm^3/g), specifically at 10% CFSL of *Mweru*, *Kariba,* and *Katobamputa*, respectively. The SV for the bread containing 10% *Katobamputa* cassava flour was insignificantly different from wheat bread (2.5 cm^3/g). The bread with acceptable quality can be processed from these varieties at about 10% CFSL. Further studies could target the effect of particle size distribution (fractionated flour) on rheology and bread quality characteristics to ascertain effective flour size for substitution into wheat flour.

The bread SV is a response function of flour particle size and WAC. Particle size can be a differentiating genetic trait among cassava varieties. Given that cassava genotypes were cultivated simultaneously in a single plantation and harvested at the same time, and milling conditions were the same across varieties. Thus, the variation in flour particle size were attributed to genetic differences among the cassava varieties (Vasconcelos et al., 2017). Particle size distribution influences the hydration properties of flours, which in turn, is responsible for gluten development in dough system and ultimately bread quality. Optimising the flour particle size between the blends of wheat and cassava flours in response to WAC can be the basis for formulating composite flours of improved properties for dough performance and processing of bread quality.

Conflicts of interest

No competing conflicts of interest were expressed by any authors.

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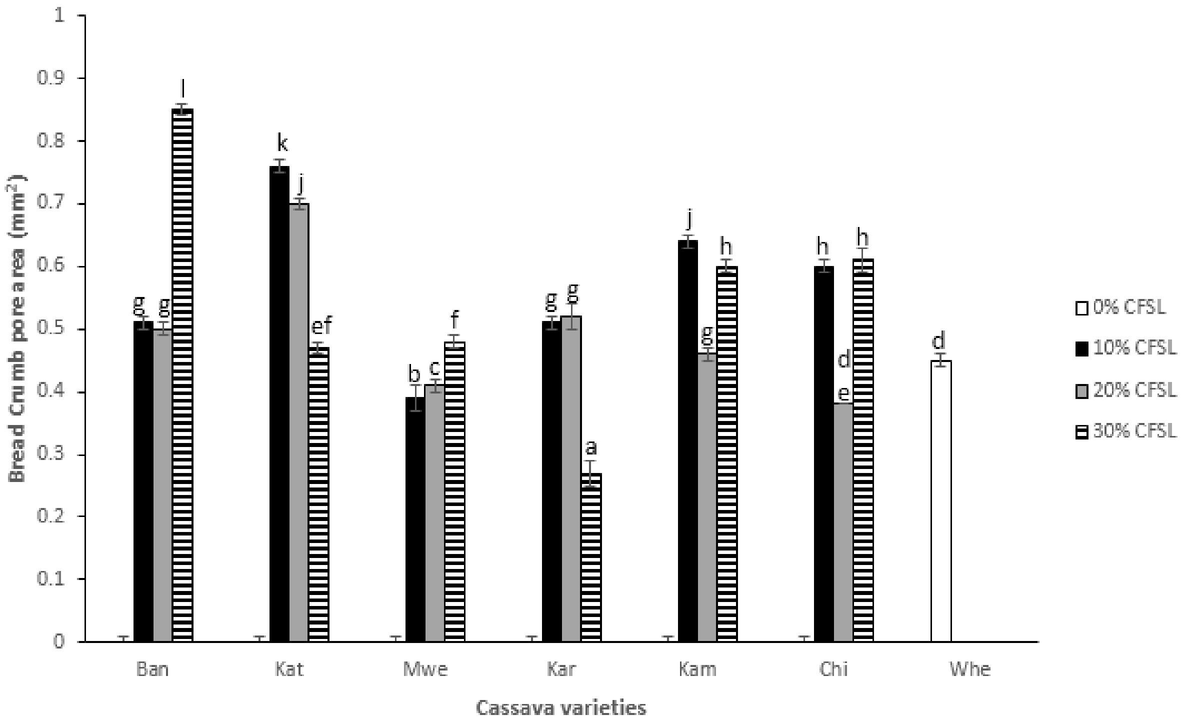

Fig. 1. Bread Crumb pore cross surface area (mm^2) per cassava flour substitution level (CFSL). Ban=Bangweulu, Kat=Katobamputa, Mwe=Mweru, Kar=Kariba, Kam=Kampolombo, Chi=Chila, Whe=Wheat (Control). Values are presented as mean ± standard deviation ($n = 3$). Error bars represent standard deviations based on 3 replicates. Bars marked with different letters within the same CFSL indicated significant difference ($p<0.05$) among cassava varieties.

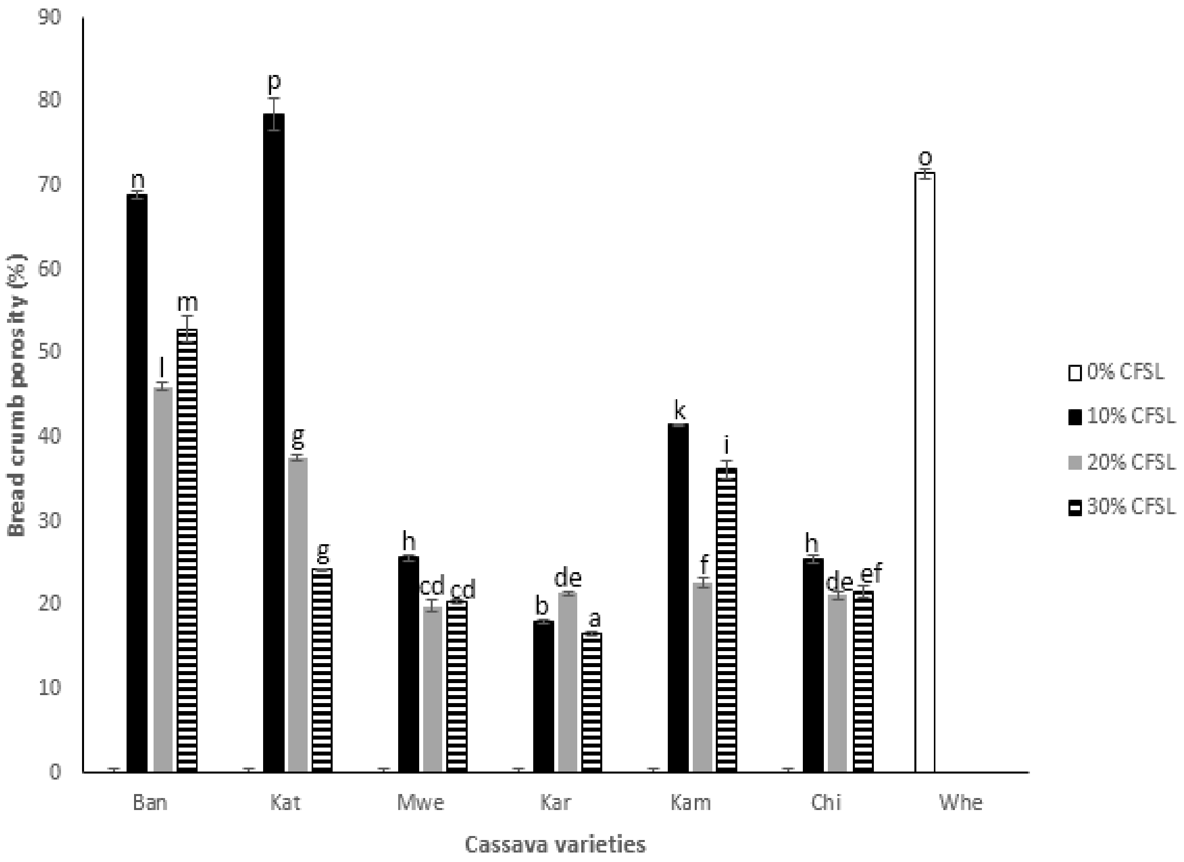

Fig. 2. Bread crumb porosity (%) per cassava flour substitution level (CFSL). Ban=Bangweulu, Kat=Katobamputa, Mwe=Mweru, Kar=Kariba, Kam=Kampolombo, Chi=Chila, Whe=Wheat (Control). Values are presented as mean ± standard deviation (n = 3). Error bars represent standard deviations based on 3 replicates. Bars marked with different letters within the same CFSL indicated significant difference ($p<0.05$) among cassava varieties.

Fig. 3. Bread crumb and crust from flour blend of cassava/wheat in Kampolombo at 10, 20 and 30% CFSL. Photos at scale 10 mm obtained using microscope (Leica MZ16) with Leica camera (Leica DFC450C), Leica Application Suite (LAS).

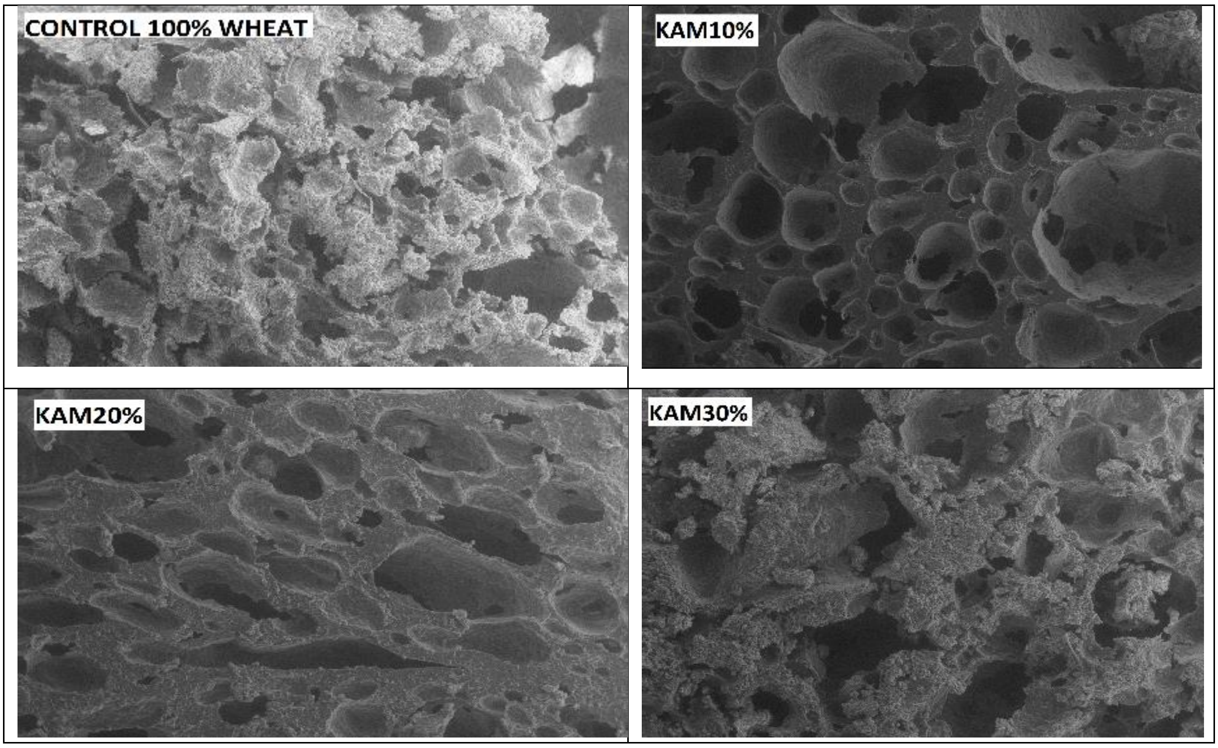

Fig. 4. Scanning microscopic images of bread made from cassava-wheat flour blends at 10%, 20% and 30% CFSL. Variety KAM: Kampolombo Control: 100% Wheat flour. Image properties: Signal A = VPSE G3, EHT = 20 kV, Chamber = 2.07e-001 Torr, I Probe = 253 pA, Spot Size = 442, Mag = 100X, Scale: 200µm

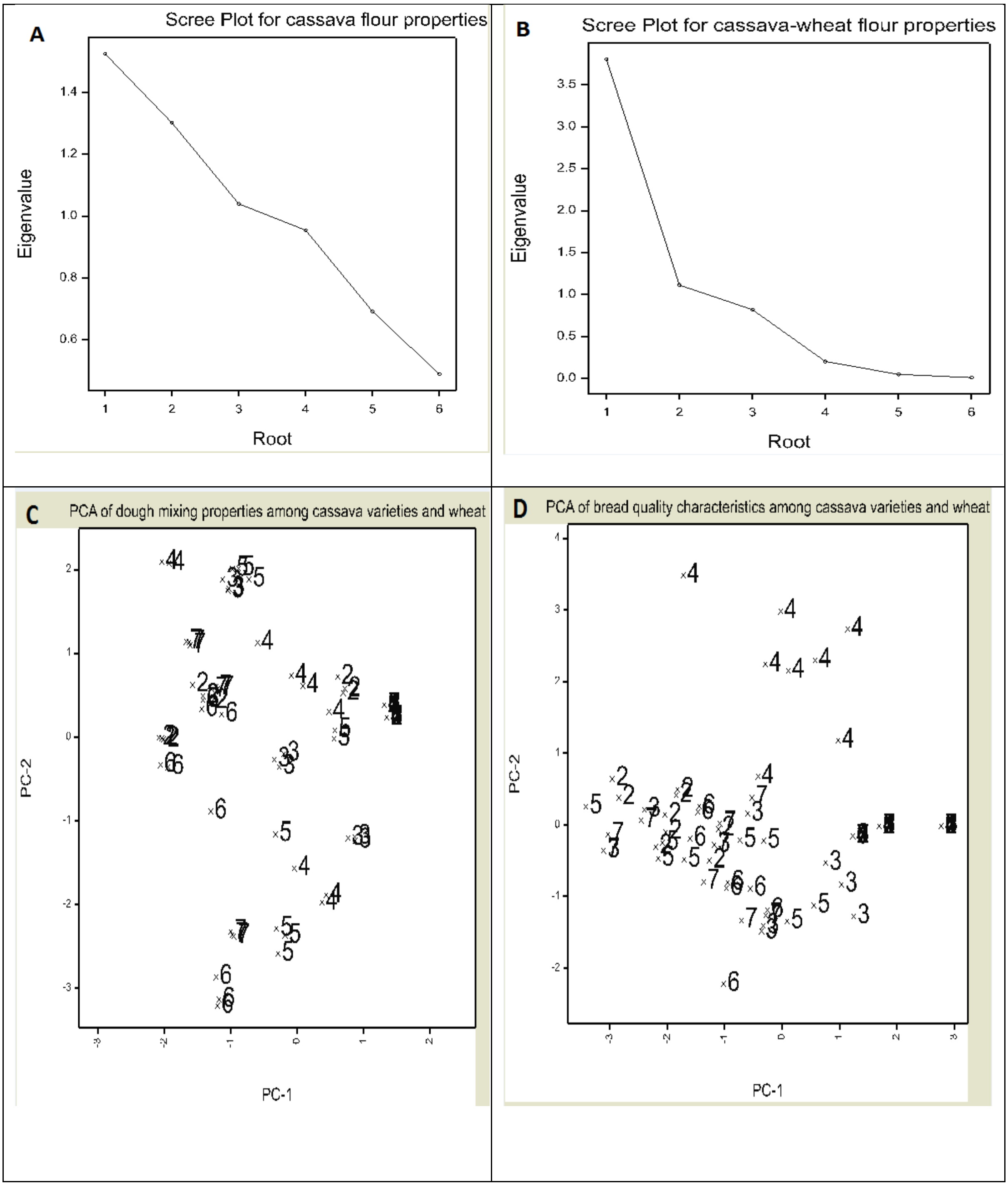

Fig. 5. Principal component analysis (PCA). (A) Scree plot for cassava flour properties, Latent roots PC 1 = 1.5, PC2 = 1.3. Percentage variation PC1 = 25, PC2 = 22. (B) Scree plot for cassava flour with wheat flour, Latent roots PC1 = 3.7, PC2 = 1.1. Percentage variation PC1 = 62, PC2 = 19. (C) PCA of dough mixing properties and (D) PCA of bread quality characteristic among cassava varieties and wheat. Varieties: 1=Control (Wheat shaded black), 2=Bangweulu, 3=Katobamputa, 4=Mweru, 5=Kariba, 6=Kampolombo and 7=Chila.

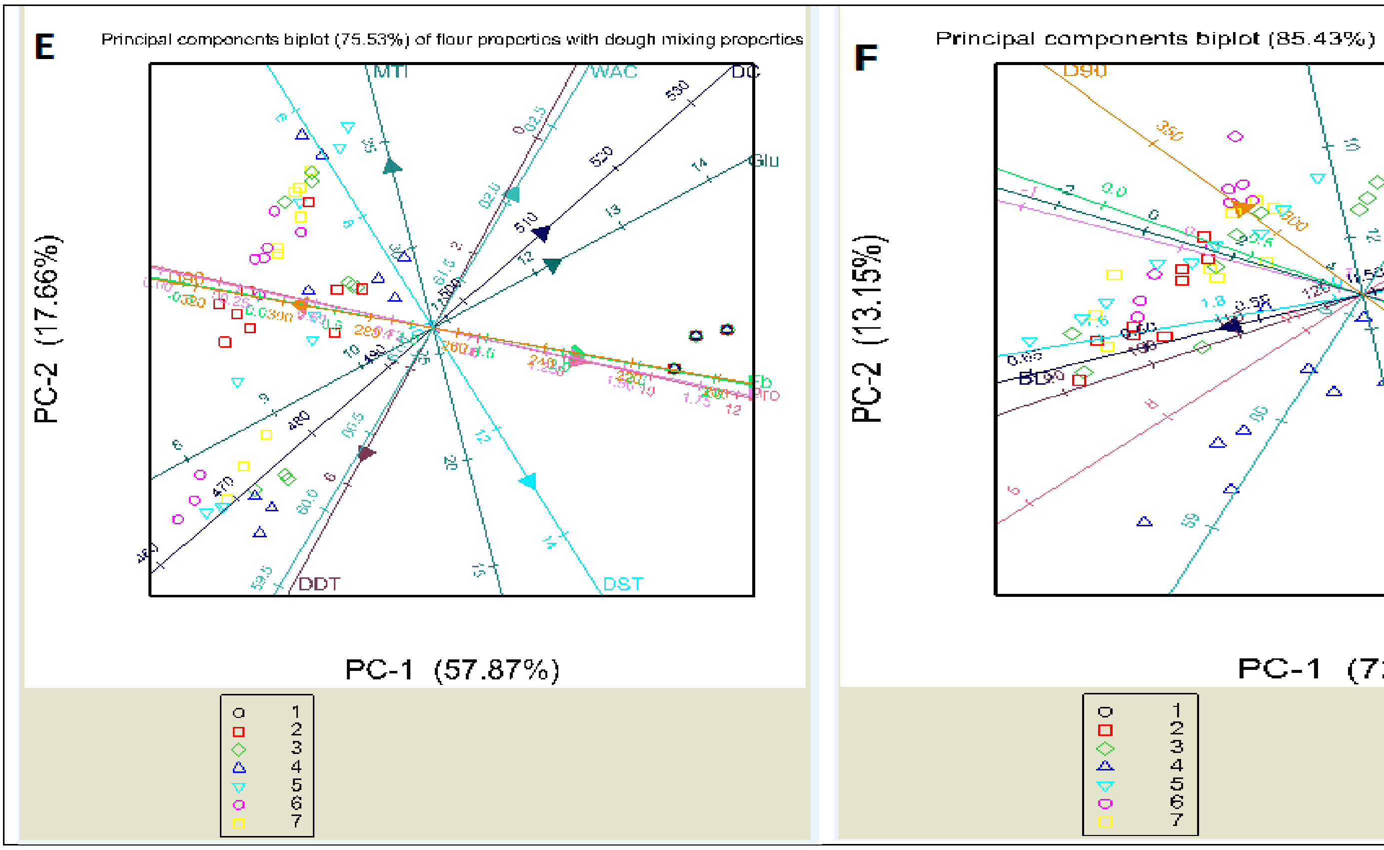

Fig. 6. (E) Principal components biplot of flour properties with dough mixing properties. Latent roots: PC1 = 5.8, PC2 = 1.8. Percentage variations: PC1 = 53, PC2 = 16 (F) Principal components of biplot of flour properties with bread quality characteristics. Latent roots: PC1 = 7.3, PC2 = 1.3. Percentage variations: PC1 = 66, PC2 = 12. Flour properties WAC=water absorption capacity, D90=flour particle size, Pro=protein, Li=Lipid, Fb=Fibre, Glu=Gluten. Dough properties: DDT=dough development time, DC=dough consistency, DST=dough stability time, MTI=mixing tolerance index. Bread characteristics: BV=bread volume, SV=bread specific volume, BD=bread density, and WL=weight loss. Varieties: 1=Wheat, 2=Bangweulu, 3=Katobamputa, 4=Mweru, 5=Kariba, 6=Kampolombo and 7=Chila.

Table 1. Moisture, protein, lipid, and amylose contents, and particle size of cassava flours from six cassava varieties grown in Zambia

Variety	Moisture (%)	Protein (%)	Lipid (%)	Fibre (%)	Amylose (%)	Size (µm)
Bangweulu	11 ± 1^{ab}	1.9 ± 1^{b}	0.4 ± 0.04^{bc}	0.6 ± 0.5^{b}	22 ± 3^{ab}	312^{a}
Katobamputa	11 ± 1^{ab}	1.5 ± 0.03^{ab}	0.4 ± 0.1^{bc}	0.2 ± 0.1^{a}	27 ± 2^{b}	283 ± 0.02^{c}
Mweru	12 ± 2^{b}	1.8 ± 0.3^{ab}	0.6 ± 0.2^{cd}	0.1 ± 0.1^{a}	18 ± 8^{a}	250 ± 0.03^{b}
Kariba	11 ± 1^{ab}	1.4 ± 0.4^{ab}	0.6 ± 0.1^{d}	0.04 ± 0.02^{a}	16 ± 1^{a}	333 ± 0.02^{e}
Kampolombo	11 ± 1^{a}	1.6 ± 0.2^{ab}	0.3 ± 0.2^{ab}	0.03 ± 0.02^{a}	18 ± 7^{a}	334 ± 0.01^{e}
Chila	10 ± 0.4^{a}	1.2 ± 0.1^{a}	0.2 ± 0.04^{a}	0.2 ± 0.1^{a}	16 ± 4^{a}	278^{c}
Wheat (control)	13 ± 0.2^{d}	11 ± 0.3^{c}	1.7 ± 0.2^{c}	2.9 ± 0.1^{c}	21 ± 0.5^{ab}	207 ± 10^{a}

Values are presented as mean ± standard deviation ($n = 3$). Within the same column, the values with different letters are significantly different at $p<0.05$ by LSD test.

Table 2. Moisture and gluten contents, and Brabender Farinograph dough mixing properties of wheat flour and cassava-wheat flour blends.

Variety	CFSL (%)	Moisture (%)	Water absorption capacity (%)	Gluten (%)	Development time (min)	Consistency (FU)	Stability (min)	Tolerance index (FU)
Bangweulu	10	13 ± 0.3^{ab}	60 ± 2^{abcd}	10 ± 0.04^{de}	1.9 ± 0.3^{bcde}	478 ± 2^{cd}	7.2 ± 0.1^{b}	30 ± 1^{hi}
Katobamputa	10	13 ± 0.1^{bcd}	62 ± 0.1^{e}	11 ± 0.01^{ef}	2.1 ± 0.1^{de}	478 ± 2^{cd}	9.9 ± 0.05^{ef}	25 ± 1^{efg}
Mweru	10	13 ± 0.2^{ab}	62 ± 0.2^{e}	10 ± 0.02^{de}	2 ± 0.1^{cde}	501^{h}	11 ± 0.1^{gh}	29 ± 4^{hi}
Kariba	10	13 ± 0.1^{abc}	61 ± 1^{cde}	11 ± 0.01^{ef}	1.7 ± 0.1^{ab}	516 ± 1^{i}	7.2 ± 0.3^{b}	35 ± 1^{jk}
Kampolombo	10	13 ± 0.1^{ab}	62 ± 0.5^{e}	11 ± 0.01^{ef}	2.1 ± 0.1^{de}	476 ± 0.5^{c}	9.4 ± 0.5^{de}	35 ± 1^{jk}
Chila	10	13 ± 0.1^{ab}	62 ± 0.3^{e}	11 ± 0.01^{ef}	2.2 ± 0.1^{e}	490^{fg}	7.1 ± 0.1^{b}	35 ± 1^{jk}
Bangweulu	20	13 ± 0.2^{a}	61 ± 1^{bcde}	9.6 ± 0.02^{cd}	1.8 ± 0.2^{abcd}	510 ± 1^{i}	8.2 ± 0.1^{c}	18 ± 1^{bc}
Katobamputa	20	14 ± 0.1^{ef}	61 ± 0.2^{cde}	8.6 ± 1^{bc}	1.8 ± 0.1^{abcd}	511 ± 1^{i}	6.3 ± 0.1^{a}	32 ± 1^{ij}
Mweru	20	13 ± 0.2^{ab}	61 ± 1^{bcde}	8.7 ± 1^{bc}	1.5 ± 0.1^{a}	501 ± 1^{h}	6.7 ± 1.1^{ab}	37 ± 10^{k}
Kariba	20	14 ± 0.1^{cde}	62 ± 0.5^{e}	7.8 ± 1^{ab}	2.1 ± 0.1^{de}	484 ± 20^{def}	10 ± 0.3^{fg}	22 ± 1.2^{k}
Kampolombo	20	13 ± 0.2^{av}	62 ± 0.2^{de}	12 ± 0.03^{fg}	7.8 ± 0.2^{h}	492 ± 2^{g}	6.6 ± 0.2^{ab}	28 ± 5^{a}
Chila	20	13 ± 0.2^{abc}	61 ± 1^{cde}	10 ± 0.02^{de}	1.7 ± 0.1^{abc}	482 ± 1^{cde}	6.9 ± 0.1^{ab}	28 ± 1^{gh}
Bangweulu	30	13 ± 0.2^{abcd}	60 ± 0.5^{a}	8.7 ± 0.02^{bc}	7.2 ± 0.1^{g}	493^{g}	9.1 ± 0.1^{d}	40 ± 1^{l}
Katobamputa	30	14 ± 1^{cde}	60 ± 0.1^{a}	8.1 ± 1.01^{bc}	4.0^{f}	488 ± 1^{i}	9.5 ± 0.1^{de}	11 ± 1^{a}
Mweru	30	13 ± 0.2^{ab}	60 ± 0.4^{a}	6.9 ± 1^{a}	2.1 ± 0.1^{de}	453 ± 0.5^{h}	11 ± 1.3^{hi}	15 ± 4^{b}
Kariba	30	14 ± 0.1^{f}	60 ± 0.2^{ab}	6.9 ± 1^{a}	9.4 ± 1^{j}	481 ± 1^{def}	12 ± 0.1^{k}	24 ± 1^{de}
Kampolombo	30	13 ± 0.2^{abcd}	60 ± 1^{abc}	9.2 ± 0.5^{c}	11 ± 0.5^{k}	462^{g}	11 ± 0.3^{ij}	24 ± 1^{def}
Chila	30	14 ± 0.1^{def}	60 ± 1^{ab}	10 ± 0.01^{de}	8.7 ± 0.1^{i}	467 ± 1^{cde}	12 ± 0.1^{jk}	27 ± 1^{fgh}
Wheat	0	13 ± 0.2^{abc}	62 ± 1^{e}	13 ± 1^{g}	2.1 ± 0.1^{de}	512.33 ± 0.5^{i}	12 ± 0.2^{k}	24 ± 1^{fgh}

Values are presented as mean ± standard deviation (n = 3). Within the same column, the values with different letters are significantly different at $p<0.05$ by LSD test.

Table 3. Correlation coefficients of dough mixing properties, bread quality, amylose and proximate contents

	CFSL	M	WAC	DDT	DST	MTI	DC	Amy	Prt	Lip	Fib	BV	WB	BD	SV	WL	PA	P	D90	G
CFSL	1																			
M	0.37*	1																		
WAC	-0.65***	-0.30*	1																	
DDT	0.62***	0.32*	-0.44**	1																
DST	-0.27*	0.19	0.00	0.19	1															
MTI	-0.06	-0.19	0.16	0.00	-0.48	1														
DC	-0.65***	-0.19	0.45**	-0.45	0.03	0.12	1													
Amy	-0.10	0.12	-0.12	-0.16	0.00	-0.21	0.18	1												
Prot	-0.77***	-0.14	0.34**	-0.28	0.64***	-0.25	0.60***	0.15	1											
Lip	-0.74***	-0.08	0.36**	-0.31	0.65***	-0.27	0.61***	0.14	0.96	1										
Fib	-0.76***	-0.15	0.36**	-0.29	0.61***	-0.23	0.60***	0.16	0.98	0.93	1									
BV	-0.87***	-0.21	0.66***	-0.51	0.36	-0.06	0.60***	0.16	0.77***	0.78	0.74	1								
WB	0.04	0.13	0.08	0.21	-0.27	0.25	-0.02	0.06	-0.23	-0.32	-0.20	-0.13	1							
BD	0.85***	0.24	-0.65***	0.58	-0.30	0.05	-0.57***	-0.13	-0.71***	-0.73	-0.66	-0.98	0.24	1						
SV	-0.86***	-0.22	0.63***	-0.52*	0.39	-0.09	0.59***	0.14	0.79***	0.81	0.75	0.99	-0.25	-0.98	1					
WL	-0.04	-0.13	-0.08	-0.21	0.27	-0.25	0.02	-0.06	0.23	0.32	0.20	0.13	-1.0***	-0.24	0.25	1				
PA	0.19	0.00	-0.05	0.12	-0.23	0.40	-0.20	0.26	-0.29	-0.36	-0.24	-0.16	0.38	0.20	-0.20	-0.38	1			
P	-0.71***	-0.23	0.35	-0.30	0.43	-0.09	0.41	0.40	0.75***	0.68	0.78	0.66***	-0.0	-0.58	0.64	0.01	0.18	1		
D90	0.64***	0.13	-0.26	0.45*	-0.51***	0.24	-0.48**	-0.16	-0.83***	-0.79	-0.81	-0.68***	0.50	0.67	-0.72	-0.50	0.29	-0.57	1	
G	-0.84***	-0.44	0.60	-0.28*	0.25	0.10	0.54***	0.08	0.70***	0.62	0.70	0.78***	0.07	-0.72	0.76	-0.07	-0.08	0.64	-0.53	1

WAC=Water Absorption Capacity, DDT=Dough Development Time, DST=Dough Stability Time, MTI =Mixing Tolerance Index, DC=Dough Consistency, VB= Volume of Bread, WB= Weight of Bread, DB= Density of Bread, SV= Specific Volume, WL= Weight loss, P= Porosity, D90=Particle Size Distribution at 90% finer particles pass, G= Gluten Significance differences at p<0.001* p<0.01**, p<0.05***

Table 4. Volume, specific volume and density of bread baked from cassava-wheat flours blends

Variety	CFSL (%)	Volume (cm^3)	Specific volume (cm^3/g)	Density (g/ cm^3)	Weight loss (%)
Bangweulu	10	103±10^{abcd}	1.7±0.1^{abc}	0.6±0.03^{fghi}	12± 0.3^{bcde}
Katobamputa	10	140±5^{gh}	2.3±0.1^{ij}	0.4±0.01^{ab}	12±1^{bcde}
Mweru	10	123±10^{ef}	2.1±0.1^{hi}	0.5±0.03^{bc}	17±1^{gh}
Kariba	10	128±10^{fg}	2.1±0.1^{ghi}	0.5±0.02^{bc}	12±1^{abcd}
Kampolombo	10	122±3^{ef}	1.9±0.1^{efgh}	0.5±0.01^{cd}	10±1^{a}
Chila	10	118±10^{ef}	1.9±0.1^{defg}	0.5±0.03^{cde}	11±0.4^{ab}
Bangweulu	20	103±10^{abcd}	1.7±0.1^{abcd}	0.6±0.03^{fgh}	13±0.4^{def}
Katobamputa	20	120±10^{ef}	1.9±0.1^{efgh}	0.5^{def}	11±1^{abc}
Mweru	20	115±5de	2±0.1^{fgh}	0.5±0.03^{def}	16±2^{g}
Kariba	20	112±10^{cde}	1.8±0.1^{cdef}	0.6±0.03^{def}	12±0.5^{cde}
Kampolombo	20	112±10^{cde}	1.8±0.1^{cdef}	0.6±0.02^{def}	12±1^{bcde}
Chila	20	112±3^{cde}	1.8±0.1^{cdef}	0.6±0.01^{def}	13±1^{def}
Bangweulu	30	93±10^{ab}	1.5±0.1^{ab}	0.7±0.04^{ij}	13±0.3^{ef}
Katobamputa	30	100±10^{abc}	1.6±0.2^{abc}	0.6±0.1^{ghij}	12±1de
Mweru	30	103±10^{abcd}	1.8±0.2^{cdef}	0.6±0.1^{def}	18±1^{h}
Kariba	30	95±10^{ab}	1.5±0.1^{ab}	0.7±0.1^{hij}	12±0.4^{bcde}
Kampolombo	30	105^{bcd}	1.7±0.1^{bcde}	0.5^{efg}	13±0.5^{def}
Chila	30	92±3^{a}	1.5± 0.01^{a}	0.7±0.02^{j}	12±0.5^{bcde}
Wheat flour	0	148± 11^{h}	2.5± 0.2^{j}	0.4± 0.03^{a}	14± 1^{f}

Values are presented as mean ± standard deviation (n = 3). Within the same column, the values with different letters are significantly different at $p<0.05$ by LSD test.

Table 5. Colour parameters of the crumb of the bread baked from six cassava varieties-wheat flour blends

Variety	CFSL (%)	L*	a*	b*	Whiteness	Chroma
Bangweulu	10	74 ± 0.1^{fg}	0 ± 0.1^{c}	21 ± 0.1^{cd}	67 ± 0.2^{efg}	21 ± 0.1^{cd}
Katobamputa	10	73 ± 0.3^{cd}	0.1 ± 0.1^{ef}	21 ± 0.1^{fgh}	65 ± 0.1^{bc}	21 ± 0.1^{fgh}
Mweru	10	73 ± 0.1^{def}	0.6 ± 0.04^{l}	22 ± 0.1^{j}	65 ± 0.1^{ab}	22 ± 0.1^{j}
Kariba	10	73 ± 0.3^{fg}	0.04 ± 0.03^{cd}	21 ± 0.3^{cde}	66 ± 0.4^{ef}	21 ± 0.3^{cde}
Kampolombo	10	73 ± 0.3^{ef}	-0.4 ± 0.1^{a}	20 ± 0.4^{a}	67 ± 0.5^{ghi}	20 ± 0.4^{a}
Chila	10	75 ± 0.1^{j}	0.3 ± 0.1^{gh}	21 ± 0.2^{gh}	67 ± 0.4^{j}	21 ± 0.2^{gh}
Bangweulu	20	72 ± 1^{cd}	-0.3 ± 0.1^{ab}	20 ± 0.1^{c}	66 ± 0.4^{cd}	20 ± 0.1^{c}
Katobamputa	20	73 ± 0.3^{cd}	-0.3 ± 0.1^{b}	19 ± 0.5^{a}	66 ± 0.04^{bc}	19 ± 0.5^{a}
Mweru	20	75 ± 0.5^{hi}	0.7 ± 0.03^{jk}	22 ± 0.1^{j}	66 ± 0.3^{de}	23 ± 0.1^{j}
Kariba	20	71 ± 1^{a}	0.2 ± 0.03^{fg}	21 ± 0.2^{cd}	65 ± 1^{a}	21 ± 0.2^{cd}
Kampolombo	20	73 ± 0.2^{cde}	0.4 ± 0.01^{ijk}	21 ± 0.1^{ef}	66 ± 0.4^{cd}	21 ± 0.1^{ef}
Chila	20	73 ± 0.3^{cde}	0.1 ± 0.02^{de}	21 ± 0.1^{fg}	66 ± 0.3^{bcd}	21 ± 0.1^{fg}
Bangweulu	30	72 ± 1^{ab}	0.1 ± 0.1^{cde}	20 ± 0.6^{b}	65 ± 0.4^{fgh}	20 ± 1^{b}
Katobamputa	30	75 ± 0.01^{ij}	0.5 ± 0.02^{k}	21 ± 0.04^{def}	68 ± 0.03^{j}	21 ± 0.04^{def}
Mweru	30	75.6 ± 0.5^{j}	0.4 ± 0.1^{jk}	22 ± 0.1^{i}	67 ± 0.4^{ij}	22 ± 0.1^{i}
Kariba	30	74 ± 0.01^{fg}	0.3 ± 0.1^{hi}	21 ± 0.1^{c}	67 ± 0.03^{fg}	21 ± 0.1^{c}
Kampolombo	30	74 ± 0.5^{h}	0.6 ± 0.1^{lm}	21 ± 0.1^{cd}	67 ± 0.4^{hij}	21 ± 0.1^{cd}
Chila	30	74 ± 1^{gh}	0.7 ± 0.1^{m}	21 ± 0.3^{fg}	66 ± 0.4^{efg}	21 ± 0.3^{fgh}
Wheat	0	74 ± 0.02^{h}	0.4 ± 0.02^{ij}	22 ± 0.03^{hi}	67 ± 0.01^{fgh}	22 ± 0.03^{hi}

Values are presented as mean ± standard deviation (n = 3). Within the same column, the values with different letters are significantly different at $p<0.05$ by LSD test.

Table 6. Colour parameters of the crust of the bread baked with six cassava-wheat flour blends.

Variety	CFSL (%)	L*	a*	b*	BI	Chroma
Bangweulu	10	53 ± 0.4^{b}	17 ± 0.2^{k}	38 ± 0.1^{fgh}	137 ± 1^{j}	41 ± 0.01^{ij}
Katobamputa	10	58 ± 2^{d}	14 ± 1^{def}	37 ± 0.2^{def}	115 ± 10^{e}	40 ± 0.5^{cde}
Mweru	10	51 ± 1^{a}	15 ± 0.1^{gh}	34 ± 1^{a}	122 ± 0.4^{f}	37 ± 1^{a}
Kariba	10	58 ± 0.01^{d}	15 ± 0.04^{efg}	37 ± 0.1^{de}	115 ± 0.3^{e}	40 ± 0.1^{cd}
Kampolombo	10	60 ± 0.01^{efg}	15 ± 0.03^{fg}	39 ± 0.04^{i}	116 ± 0.1^{e}	42 ± 0.03^{j}
Chila	10	64 ± 0.01^{j}	12 ± 0.02^{a}	38 ± 0.1^{gh}	100 ± 0.4^{b}	40 ± 0.1^{cd}
Bangweulu	20	62 ± 0.5^{h}	13 ± 0.05^{b}	38 ± 0.04^{h}	106 ± 1^{cd}	40 ± 0.03^{def}
Katobamputa	20	61 ± 0.4^{g}	14 ± 0.4^{cd}	37 ± 1^{def}	108 ± 4^{cd}	40 ± 1^{cd}
Mweru	20	51 ± 0.1^{a}	15 ± 0.2^{gh}	35 ± 0.4^{b}	128 ± 2^{cd}	38 ± 0.40^{b}
Kariba	20	53 ± 0.04^{b}	16 ± 0.1^{j}	38 ± 0.1^{gh}	134 ± 0.4^{ij}	41 ± 0.1^{hij}
Kampolombo	20	60 ± 0.4^{fg}	14 ± 0.2^{c}	378 ± 0.1^{efgh}	109 ± 1^{d}	40 ± 0.1^{cd}
Chila	20	53 ± 1.1^{b}	16 ± 0.2^{jk}	37 ± 0.4^{de}	132 ± 3^{hi}	41 ± 0.3^{efg}
Bangweulu	30	54 ± 0.3^{c}	15 ± 1^{hi}	38 ± 0.5^{fgh}	128 ± 3^{gh}	41 ± 1^{fghi}
Katobamputa	30	64 ± 1^{j}	12 ± 0.5^{a}	36 ± 0.5^{c}	95 ± 3^{a}	38 ± 1^{b}
Mweru	30	59 ± 0.1^{e}	14 ± 0.1^{de}	38 ± 0.1^{gh}	113 ± 0.5^{e}	40 ± 0.2^{defg}
Kariba	30	63 ± 1^{ij}	13 ± 0.5^{b}	39 ± 0.4^{i}	105 ± 4^{c}	41 ± 0.5^{fghi}
Kampolombo	30	55 ± 1^{c}	16 ± 1^{i}	38 ± 0.3^{fgh}	127 ± 5^{g}	41 ± 0.5^{ghij}
Chila	30	62 ± 1^{hi}	13 ± 0.2^{b}	39 ± 0.4^{i}	105 ± 1^{cd}	41 ± 0.5^{efgh}
Wheat	0	59 ± 0.2^{ef}	14 ± 0.2^{def}	37 ± 0.3^{cd}	109 ± 2^{cd}	39 ± 0.4^{c}

Values are presented as mean ± standard deviation (n = 3). Within the same column, the values with different letters are significantly different at $p<0.05$ by LSD test.

Table 7. Correlation coefficients of colour parameters of crumb and crust of bread baked from flour blends of wheat and cassava flour

	Crumb						Crust		
Parameter	CFSL	L	a	b	Whiteness	Chroma	CFSL	L	a
CFSL	1						1		
L	-0.20	1					0.01	1	
a	0.08	0.51**	1				-0.12	-0.87	1
b	-0.32*	0.52**	0.74	1			0.32	0.46	-0.12
Whiteness	-0.05	0.85**	0.15	-0.01	1		0.32	0.46	-0.12
Chroma	-0.31	0.52	0.74**	1.00	0.00	1			
Bread volume	-0.87	0.23	0.04	0.35	0.06	0.35	-0.87	0.05	0.06
Weight of Bread	0.04	-0.31*	-0.48	-0.67	0.06	-0.67**	0.04	0.37	-0.15
Bread density	0.85	-0.24	-0.07	-0.42	-0.03	-0.42*	0.85	0.05	-0.12
Specific volume	-0.86	0.27	0.11	0.43	0.05	0.43*	-0.86	0.00	0.08
Weight loss	-0.04	0.31	0.48	0.67	-0.06	0.67**	-0.04	-0.37	0.15
Aver Pore Area	0.19	-0.33	-0.37	-0.50	-0.09	-0.50**	0.19	0.02	0.06

Significance differences at p<0.01*, p<0.05**