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Evaluation of Various Properties of Amaranthus (*Genus Amaranthus L.*) **Based Composite Flour Blends for Preparation of Gluten-Free Biscuits**

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| ARTICLE INFO | ABSTRACT |
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| Article history: Received: February 24, 2020 Accepted: September 8, 2020 | This research was conducted to investigate the pasting, rheological and functional properties, and gluten-free biscuit making potential of a composite flour prepared from grains of amaranthus, sorghum and finger millet. The |
| <i>Keywords</i> : composite flours gluten-free amaranthus sorghum finger millet | formulation for the composite flour was obtained from D-optimal mixture design ratio using Design-Expert. The rheological and pasting properties of the composite flours were determined, while the proximate composition, physical dimensions, mineral concentration and sensory quality attributes of the biscuits were assessed. The results showed that there were significant (p<0.05) differences in the pasting profile of the control and amaranthus based composites flour except for pasting temperature. Water absorption capacity and water soliblity index increased as the blending ratio of amaranthus flour increased, while oil absorption capacity decreased. The proximate composition evaluation 13.75, 2.04, 1.77 and 31.75% were found to be the highest values of the biscuit samples in terms of protein, crude fiber, ash and crude fat, respectively. Mineral analaysis was carried out and there was a significant (p<0.05) difference in Fe, Ca, Zn and P content among the biscuit samples made from the composite flour blends. Similarly, the sensory evaluation indicated that there was a significant (p<0.05) differences in apperance, colour, texture, flavour and overall acceptability among the composite biscuit samples. In a nut shell this research revealed that a nutritionally dense gluten-free biscuits can be formulated without affecting the quality attributes of the biscuit. Thus, the composite flours can be used for the preparation of gluten free food products in africa, where the crops have not |
| | been effectively utilized in food processing industries. |

Introduction

Baking industry is considered to be one of the major segments of food processing in the present days and, because of their availability and reasonably good shelf life, baked products are gaining popularity. Wheat is a major cereal grain used for preparation of many baked goods. However, most of these foods are poor in terms of nutritional quality (Omobolanle et al., 2017). Baked products could be produced from cassava, sorghum, finger millet and other composite flours (Manley,

Amaranth is a pseudo-cereal with high nutritional value, particularly for its balanced amino acid content, dietary fiber content and antioxidant activity. Its nutritional quality have attracted the attention of researches about the use of amaranth as functional ingredient (Cornejo et al., 2019). Grain amaranth can be used as seeds or flour to make products such as cookies, biscuits, and other bakery products (Muyonga

^{2000).} A considerable attention has been given to the development of baked goods with better nutritional quality.

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et al., 2008). Beside its availability, amaranthus can also contribute to fighting malnutrition, especially in developing countries like Ethiopia. Sorghum has been utilized differently in different parts of Ethiopia and the grains are used in the preparation of porridge, infant food, and in the preparation of local beverages (Belton and Taylor, 2002). They are promising source of useful compounds because of their nutritional properties (Anonymous, 2016). In Ethiopia, finger millet is grown mainly as a sole crop in rotation with other annual crops, preferably legumes. Unleavened bread, thin-or thick porridge, fermented porridge, makingnjera', "a flat levened Ethiopian most widly used traditional foodare some of the different food types prepared from finger millet (Belton and Taylor, 2002). The grain's protein content (7.4%) is comparable to that of rice (7.5%). Finger millet is also a rich source of minerals (Desai et al., 2010; Tsehaye et al., 2006).

Amaranth, sorghum, and finger millet are all glutenfree cereal grains and are considered to be better alternatives for those who have been suffering from celiac disease or gluten - sensitive enteropathy, which is the case for some grains like wheat, barly and rye. But, the processing of these grain flours into baked products has several limitations as the grains lack these storage protein (gliadins and glutenin), which is responsible for gelatinization of flour and is considered to be one of the qualities of baked products as it influences elasticity, chewiness etc. These characteristics of baked products are mainly expressed by the rheological and pasting properties of the flour (Adevemi and Ogazi, 1985; Belton and Taylor, 2002). Therefore, the aim of this research was to assess the rheological and pasting properties of composite flours prepared from grains of amaranthus, sorghum and finger millet, and the nutritional quality and sensory acceptability of biscuits developed thereafter.

Materials and methods

Raw material

Amaranthus grain (Amaranthus caudatus) samples were purchased directly from a local market in Konso, Ethiopia. Finger millet (Whitey/Necho variety) and sorghum (Melkam variety) samples were provided by Adet Agricultural Research Center and Melkassa Agricultural Research Centers of Ethiopia, respectively. The collected samples were transported to the laboratory in surface sterilized polythene bags. As described in Figure 1, the grains were sorted, cleaned and washed to remove immature seeds, sand and soil and sun dried for 24 hours. Wheat flour was collected from KOJJ Food Processing PLC., Addis Ababa, Ethiopia and used as a control.



Flours Packaging and labeling (air tight polyethylene bags)

Fig. 1. Flow chart for production of flour

All grains were made into flour by dry milling process using a laboratory mill 120, version 2.2; model MF 3170, Hagersten, Sweden with a mesh size of 0.5 mm (Fig. 1). The prepared flours were separately stored in a polyethylene bags at 20 °C until further analysis. The composite flours were prepared by mixing amaranth flour with finger millet and sorghum flours at different level of ration. D-Optimal mixture ratio design was used to determine the optimum mixture formulation and a 10run constrained D-optimal mixture experiment was generated using Design-Expert®, version 6.0.8, based on the lower and upper limits provided. For three components, the range of constrains was provided based on different literatures (Abdelghafor et al., 2011; Schoenlechner et al., 2006; Vijayakumar and Mohankumar, 2009). Constrained region in the simplex coordinate system was defined by the limits of $50 \le X_1 \le$ 100, $0 \le X_2 \le 50$, $0 \le X_3 \le 20$, $X_1 = Amaranths$, $X_2 =$ sorghum and X_3 = finger millet flour. A wheat flour was used as a control, i.e. 100% wheat flour (w/w on flour basis) with total runs in this experiment were 11 runs (Table 1).

Biscuit preparation process

Biscuits were produced from the prepared composite flours using the standard AACC Method No.10.52 (AACC, 2000) and the baking process was as described by Karki et al. (2016). The dough was prepared in a laboratory manual dough mixer. Ingredients used for the preparation of biscuit samples, like margarine, sugar, sodium bicarbonate, salt and ammonium bicarbonate, were purchased from the local market in Addis Ababa, Ethiopia and prepared prior to the baking process. The recipe for the formulation of gluten-free biscuits was presented in Table 2.

| Run | Code Component 1 A: Amaranth % | | Component 2 B: Sorghum % | Component 3 C: Finger millet% | |
|-----|-----------------------------------|--------|-----------------------------|----------------------------------|--|
| 1 | C-01 | 100.00 | 0.00 | 0.00 | |
| 2 | C-02 | 50.00 | 50.00 | 0.00 | |
| 3 | C-03 | 80.00 | 10.00 | 10.00 | |
| 4 | C-04 | 50.00 | 30.00 | 20.00 | |
| 5 | C-05 | 60.00 | 35.00 | 5.00 | |
| 6 | C-06 | 65.00 | 15.00 | 20.00 | |
| 7 | C-07 | 80.00 | 0.00 | 20.00 | |
| 8 | C-08 | 75.00 | 25.00 | 0.00 | |
| 9 | C-09 | 50.00 | 40.00 | 10.00 | |
| 10 | C-10 | 90.00 | 0.00 | 10.00 | |

| Table 2. Recipe for preparation of biscuit samples | Table 2. | Recipe for | preparation | of biscuit samples |
|---|----------|------------|-------------|--------------------|
|---|----------|------------|-------------|--------------------|

| 1Total flour2002Grinded sugar753Baking Soda/ Sodium bicarbonate54Ammonium bicarbonate35Table salt36Margarine/Shortening1007Water75mL | N⁰ | Ingredients | Amount (g) | |
|--|----|---------------------------------|------------|--|
| 3Baking Soda/ Sodium bicarbonate54Ammonium bicarbonate35Table salt36Margarine/Shortening100 | 1 | Total flour | 200 | |
| 4Ammonium bicarbonate35Table salt36Margarine/Shortening100 | 2 | Grinded sugar | 75 | |
| 5Table salt36Margarine/Shortening100 | 3 | Baking Soda/ Sodium bicarbonate | 5 | |
| 6 Margarine/Shortening 100 | 4 | Ammonium bicarbonate | 3 | |
| | 5 | Table salt | 3 | |
| 7 Water 75mL | 6 | Margarine/Shortening | 100 | |
| , , , , , , , , , , , , , , , , , , , | 7 | Water | 75mL | |

As shown in Fig. 2, all ingredients except flour and sodium bicarbonate were added with continuous mixing during the preparation of dough from the composite flours and mixed more for about 10 min. The mixing process continued till the mixture turned to a homogenous creamy desired form. Then, the prepared composite flour and sodium bicarbonate were added to the dough and mixed continuously to form the final dough. Doughs were placed on a plate and a manual shaping machine was used to cut the dough to form desired shapes and sizes. The shaped dough pieces were baked in baking oven at about 210 °C for 15 min and then allowed to cool, packed in Ziploc bags and stored prior to analysis.

Ingredient Weighing

Creaming of Ingredients (sugar, vegetable fat, water, salt)

Ingredient Mixing and final dough (flour and Sodium bicarbonate)

Dough Shaping/Forming (Circle Shape)

Baking (210 °C, 15 min)

Cooling (30-35 °C)

Packaging and Flour Storage

Fig 2. Flow chart for biscuit preparation adapted from (Gallagher, 2008; Kabuo et al., 2018)

Chemical analysis

Determination of pasting and rheological properties of composite flours

Pasting property of composite flours was evaluated with Rapid Visco-Analyzer (RVA-4500) Perten instruments Pty. Ltd., Macquarie Park NSW,2012, Australia, according to method 76-21 of AACC (2000). Viscosity profile indices were recorded and obtained from a Thermocline for Windows (TCW) Version 3 software, provided with the RVA as described by Amoo et al. (2014) and Bourekoua et al. (2016).

The effect of different flour levels on dough rheology was determined according to Brabender ICC BIPEA 300 by using the Brabender® Farinograph®-E, model 810130 USB/230V/50-60Hz, Duisburg, Germany and results obtained as graphic output (farinograms) from a computer software provided with the Brabender Farinograph.

Determination of functional properties of composite flours

Water absorption capacity (WAC) of the samples was determined by using the method of Sulieman et al. (2019). The same procedure was repeated for oil absorption (Adebowale et al., 2005). WAC and OAC were expressed as the weight of sediment/initial weight of flour sample (g/g). The method of Awolu et al. (2015) was used to determine water solubility index of the flours. The swelling index (SI) was determined

by using the method of Ezeocha and Okafor (2016) and expressed as the ratio of the final over the initial volume (mL/mL). bulk The density (BD) measurement was determined by measuring the material volume, which was compacted in a cylinder of 25 mL and the results were calculated as g/mL. The gluten content was determined according to the method of ICC (2000), standard No. 137/1, using perten gluten index®, version 1.0 E.N, Hagersten, Sweden, 2012.

Determination of proximate composition of composite flour blends and biscuit

The proximate composition of the flours and prepared biscuit samples were determined using the standard methods of AOAC (2000). The samples were analyzed for moisture, ash, crude fiber, crude protein, crude fat, and carbohydrates using the procedure described by AOAC (2000) method 925.09, 923.03, 962.09, 979.09 4.5.01, respectively. Determination and of carbohydrate was carried out using estimation by difference. The heating value for the three groups of nutrients, which provide the body energy, was estimated in kJ by multiplying the percentage of crude protein, crude fat and carbohydrates by the energy values for gross nutrients conversion factors.

Determination of mineral composition of composite flour blends and biscuit

The analyses for essential minerals were carried out by using atomic absorption spectrophotometric method. A sample of digest was used to determine some elements (calcium (Ca), iron (Fe) and Zinc (Zn)) on the atomic absorption spectrophotometer and phosphorus (P) on flame photometry. The digestion of the sample and minerals were quantified according to AOAC (2000), method 985.35. Standard solutions and analytical curves were used for each element and the results were expressed in mg 100 g of the product on a dry – weight basis.

Determination of physical properties of biscuits samples

Physical properties of the biscuit samples were determined according to AOAC (2000). An electronic digital caliper was used to determine the diameter (D) and thickness (T) of biscuit samples. The diameter of the samples was measured as average value of placing four biscuits edge to edge by using an electrical digital caliper having 0.05 mm accuracy. Thickness was also measured by taking the average of stacking four biscuits on top of each other. Weight of biscuit samples was measured as average value of four individual biscuits with the help of an electronic analytical balance (PASS, FA-2004, Germany) of 0.01 g sensitivity.

The spread factor was determined by calculating the thickness and the diameter of the prepared samples according to AOAC (2000) method 10-50D. Spread ratio was calculated by dividing the average value of diameter by average value of thickness of biscuits (Sulieman et al., 2019).

Determination of colour dimension of biscuit samples

Color measurements, L* (lightness), a* (rednessgreenness) and b* (yellowish-bluish), of the biscuit samples were carried out using a colour measuring instrument spectrophotometer (Model CM-600d, Konica Minolta, INC, Japan, 2012). The instrument was initially standardized (L*=90.29, a*=1.37, b*=0.06) using a white reference standard (white duplicating paper sheet, 80 g/m²).

Determination of texture profile of biscuit samples

The texture profile of baked biscuit samples was determined using a texture analyzer (TA1 Series Analysis Machine (TA1SH-203V, texture AMETEK® Test and Calibration Instruments, LLOYD Materials Testing, 2016, England) according to AOAC (2000). Biscuit hardness was measured by means of a cutting-shear test using a stainless steel probe, which runs perpendicular to the major dimension of the sample, placed on a slot surface (4 mm wide), at a constant head speed of 2 mm/s. The maximum force (N) required to shear the sample was taken as a measure of hardness. The distance at the maximum force was also recorded. All measurements were carried out in triplicate.

Sensory evaluation of biscuit samples

The evaluation and testing preferences of the baked biscuit samples were analyzed by a panel of twenty (20) semi-trained panelists randomly selected from Addis Ababa Science and Technology University. The panelists were from both sexes, and from different ages. They were requested to taste each sample separately without comparing it with another sample. Sensory evaluation was performed 24 hours after baking, using a 9-hedonic scale of points, where 1 corresponds to the statements "I dislike it extremely" and 9 corresponds to "I like it extremely." The samples were evaluated for desirability for appearance, colour, taste, crispness, flavour, texture, and overall acceptability of the baked samples. Coded product samples were arranged in a random order on white plates and served to the panelists. The panelists were given a 20 min orientation about the procedure of sensory evaluation. Potable water was provided to rinse the mouth between evaluations and covered cups were also provided if panelists did not wish to swallow the samples.

Experimental design and statistical analysis

The analyses were designed to use replication techniques for each treatment and determined by triplicate. Each treatment was repeated a number of times (three) to obtain a valid and more reliable estimate. The composite flours were formulated based on a constrained mixture D-optimal design using Design-Expert®, version 6.0.8 (Stat-Ease, Inc. 2021 East Hennepin Ave., Suite 480 Minneapolis, USA, 2002). The tatistical analysis, means and standard deviation (SD) were calculated using SPSS statistical software. One way Analysis of Variance (ANOVA) was used for data analysis with a significance (p<0.05) difference for comparison of means.

Results and discussion

Pasting properties of a composite flours

Viscosity profile or pasting properties of the composite flour sample indices are presented in Table 3. There were no significant ($p \ge 0.05$) differences in pasting temperatures between various treatments of composite flours and the control wheat flour, but in general, the pasting temperature in composite flours was lower than that of the control (100% wheat flour),

which shows that the gluten in wheat flour held more water, so that some of the water is not available for starch, which resulted in the reduction of gelatinization. The reduced gelatinization temperature indicates the better availability of starch to amylolysis enzymes during baking process, which is desirable in baked products like bread, but not that significant for biscuits (Dautant et al., 2007).

The results of the pasting characteristics indicated that the higher level of finger millet flour increased the peak viscosity (PV), break down viscosity (BDV), and setback ratio (SBR) of composite flours. This is due to the reduced presence and interaction of components like fat and protein from finger millet starch that increase the viscosity (Dautant et al., 2007). There was a change in the pasting profile of the composite flours compared with its 100% flour. It was found that the PV decreased as finger millet flour decreased, but the ratio of sorghum flour and amaranth flour only had a little effect on peak viscosity of composite flours and there was a significant (p<0.05) difference among the composite flours. According to Morris et al. (1997), the differences in the starch and protein composition of composite flours could affect pasting viscosity and properties.

The lower BDV was found in composite flours, as compared with the control wheat flour with the lowest (874cP) and the highest (2157cP) BDV exhibited for C-01 and 100% SF flour, respectively, showing a significant (p<0.05) difference among the composite flour blends. The decrease in BDV were shown as the amount of Amaranthus flour increased in the composite flour, which can be related with the protein damage in amaranthus flour in the involvement of heat.

| Table 3. Pasting properties (Rapid visco-analyser parameters |) of composite flour blends |
|--|-----------------------------|
|--|-----------------------------|

| Samples | | | Pas | ting Property (1 | neters | | |
|----------------------|---------|---------|----------|------------------|--------|----------|------------------------|
| - | PV (cP) | TV (cP) | BDV (cP) | FV (cP) | SBR | PT (min) | P _{Temp} (°C) |
| 100% SF ^a | 3040 | 1962 | 1078 | 7184 | 5222 | 5.33 | 75.98 |
| 100% FMF | 3939 | 1782 | 2157 | 4468 | 2686 | 5.60 | 76.42 |
| 100% WF | 2648 | 1253 | 1395 | 3626 | 2373 | 5.40 | 78.56 |
| C-01 | 1967 | 1093 | 874 | 1639 | 546 | 5.49 | 76.20 |
| C-02 | 2199 | 1224 | 975 | 2344 | 1120 | 4.67 | 79.20 |
| C-03 | 2249 | 1146 | 1103 | 1885 | 739 | 4.33 | 76.75 |
| C-04 | 2305 | 1094 | 1211 | 2329 | 1235 | 5.01 | 78.80 |
| C-05 | 2265 | 1212 | 1053 | 2174 | 962 | 4.60 | 78.30 |
| C-06 | 2398 | 1067 | 1331 | 2022 | 955 | 4.47 | 76.75 |
| C-07 | 2432 | 1116 | 1316 | 1941 | 825 | 4.33 | 75.90 |
| C-08 | 2090 | 1129 | 961 | 1946 | 817 | 4.47 | 78.30 |
| C-09 | 2391 | 1177 | 1214 | 2422 | 1245 | 4.96 | 77.45 |
| C-10 | 2107 | 1091 | 1016 | 1718 | 627 | 5.04 | 76.65 |

^a100% SF: 100% Sorghum Flour, 100% FMF: 100% Finger Millet Flour, 100% WF: 100% Wheat Flour, samples coded with C-01 up to C-10: are composite flours obtained from D-optimal result for composite flour formulation and preparation of biscuit samples accordingly as presented in table 1; PV = Peak Viscosity, BDV = Breakdown Viscosity, TV = Trough Viscosity, SBR = setback ratio, FV = Final Viscosity, PT = Peak Time and P_{Temp} = Pasting temperature.

| Sample Codes | es Farinographic characteristics | | | | | | | | |
|----------------------|----------------------------------|--------|-----------|---------|---------|--------------|-----|--|--|
| | C (FU) | WA (%) | DDT (min) | S (min) | DS (FU) | DS (FU), ICC | FQN | | |
| 100% SF ^a | 119 | 50.5 | 1.0 | 0.8 | 71 | 42 | 17 | | |
| 100% FMF | 327 | 55.7 | 2.5 | 1.8 | 68 | 174 | 25 | | |
| 100% WF | 482 | 55.6 | 2.3 | 0.9 | 88 | 109 | 27 | | |
| C-01 | 946 | 67.2 | 1.2 | 0.2 | 389 | 33 | 13 | | |
| C-02 | 612 | 58.8 | 1.8 | 1.7 | 183 | 192 | 20 | | |
| C-03 | 674 | 60.4 | 2.2 | 1.3 | 155 | 177 | 22 | | |
| C-04 | 303 | 55.1 | 1.9 | 1.3 | 177 | 187 | 20 | | |
| C-05 | 381 | 57 | 1.5 | 1.2 | 153 | 151 | 16 | | |
| C-06 | 579 | 62 | 3.4 | 2.1 | 181 | 188 | 26 | | |
| C-07 | 617 | 62.8 | 2.2 | 0.6 | 266 | 277 | 22 | | |
| C-08 | 786 | 67.2 | 2.5 | 0.6 | 324 | 306 | 27 | | |
| C-09 | 491 | 59.8 | 2.4 | 0.9 | 200 | 193 | 23 | | |
| C-10 | 952 | 71.3 | 1.7 | 1.4 | 379 | 383 | 21 | | |

Table 4. Rheological property (Farinographic characteristic) of composite flour blends

 $^{a}100\%$ SF: 100% Sorghum Flour, 100% FMF: 100% Finger Millet Flour, 100% WF: 100% Wheat Flour and samples coded with C-01 up to C-10 are composite flours obtained from D-optimal result for composite flour formulation and preparation of biscuit samples accordingly as presented in table 1. C = Consistency, WA = Water Absorption, DDT = Development Time, S = Dough Stability, DS = Degree of Softening, FQN = Farinographic Quality Number.

Generally, the decrease in BDV and FV values shown as the amount of amaranth flour increased, which indicates the ability of the flour to form a viscous paste or gel after cooking and cooling as well as the resistance of the paste to shear stress during stirring (Lee et al., 2012). Composite flours showed significantly lower setback ratio than that of wheat flour, except for 100% SF, which obtained the the highest (5222). The blending gave the flours a longer paste peak times, and hence the starch granules swelled gradually and had better resistance to mechanical damage.

Rheological properties of composite dough blends

The results obtained from farinograph characteristics for dough made from a composite flours are presented in Table 4. Doughs consistence for composite flours (303-786 FU) showed a significant (p<0.05) difference and it decreased comparing to the 100% flours, but it was close (\pm 100 FU) to the desired consistence, mostly 500 FU for wheat, which exhibited 482 FU. The decrease in consistency of the flours may be due to the availability of bound water that occurs as a result of the absence of gluten proteins in the flours and the action of enzymes (protease and α -amylase) on the dough components (Vizitiu et al., 2012).

The water absorption at a given 14% moisture content had shown a significant (p<0.05) difference between the composite flours as compared to the control wheat flour, since increasing amount of water absorption was experienced in the composite flour (55.1-71.3%), while the control wheat flour exhibited a water absorption of (55.6%). Sample C-10 had absorbed a higher amount of water (71.3%),

while 100% SF flour absorbed the lowest amount of water (50.5%).

Dough development time (DDT) reflects the time between the first addition of water and the time when the dough seems to have optimum elastic and viscous properties for the retention of gas (Vizitiu et al., 2012). In this case, sorghum flour exhibited the minimum (1.0 min) time and sample C-06 had the maximum (3.4 min). Compositing the flours had shown a significant (p<0.05) difference and an increase in DDT which has a positive effect on doughs quality. Doughs stability for the composite flours was ranged between the lowest (0.2 min) for raw amaranth flour and the highest (1.8 min) for 100% finger millet flour. Finger millet flour showed the better stability for mixing which resulted in an stability for the composite flours increase of containing a higher amount of it (Miralbes, 2004). Minimum stability was obtained for the most of the composite flours that differ significantly from the control wheat flour. But there was an improvement in dough stability as the amount of finger millet flour in the composite flours increased, which is similar with the findings of Vijayakumar and Mohankumar (2009).

Degree of softening (DS) parameter had ranged between 33 and 383 FU for raw amaranth flour and sample C-10, respectively. The values for DS showed a significant (p<0.05) difference among the composite flour blends. Raw amaranth flour and sample C-08 exhibited the lowest (13) and the highest (27) FQN, respectively, resulting in a significant (p<0.05) difference among the composite flour blends. The study showed that amaranth flour is a weak flour and not able to stay long without breaking, while sorghum and finger millet flour had a better quality in terms of FQN.

| Samples | | | | Parameters | | | |
|----------------------|------------------------|------------------------|---------------------------|-------------------------|----------------------|-------------------------|-----------------------|
| | BD (g/mL) | WAC (g/g) | OAC (g/g) | SI (mL/mL) | SP (g/g) | WSI (g/g) | WGC (%) |
| 100% SF ^a | 0.61 ± 0.8^{b} | 2.27 ± 0.8^{b} | 1.40 ±0.2 ^a | 5.63 ±0.1 ^{bc} | 5.89 ± 0.8^{b} | 0.06 ± 0.7^{b} | Nil |
| 100% FMF | $0.59 \pm 0.6^{\circ}$ | $2.32 \pm \! 0.2^{ab}$ | $1.32\pm\!\!0.2$ a | $5.75 \pm 0.4^{\circ}$ | 4.77 ± 0.5^{d} | $0.20 \pm 0.7^{\circ}$ | Nil |
| 100% WF | 0.57 ± 0.9^{d} | $1.78 \pm 0.4^{\circ}$ | 1.09 ± 0.2^{b} | 4.75 ± 0.67^{a} | 7.05 ± 0.6^{ab} | 0.14 ± 0.2^{ab} | $31.5\pm0.14^{\rm a}$ |
| C-01 | $0.87\pm0.7^{\rm a}$ | 2.38 ± 0.2^a | 1.39 ± 0.2^{a} | 4.40 ± 0.6^{a} | $8.03 \ {\pm} 0.5^a$ | 0.18 ± 0.8^{a} | Nil |
| C-02 | $0.57 \pm 0.9^{\circ}$ | 2.32 ± 0.7^{ab} | 1.21 ±0.2 ^a | 5.59 ±0.1° | 4.51 ± 0.6^d | 0.23 ±0.1° | - |
| C-03 | $0.58\pm\!0.7^{\circ}$ | 2.33 ± 0.9 ab | 1.28 ± 0.2 a | $4.80\pm0.7a$ | 6.48 ± 0.4^{ab} | 0.18 ± 0.6^{a} | - |
| C-04 | $0.58\pm 0.8^{\circ}$ | 2.45 ± 0.7 ab | 1.31 ± 0.2 a | 4.27 ± 0.2^{a} | 4.56 ± 0.7^{d} | $0.20\pm 0.8^{\circ}$ | - |
| C-05 | $0.57 \pm 0.2^{\circ}$ | 2.68 ± 0.4 ab | 1.21 ±0.2 ^a | $3.92 \pm 0.4^{\rm d}$ | 5.72 ± 0.9^{bc} | $0.22 \pm 0.2 ^{\circ}$ | - |
| C-06 | $0.57 \pm 0.5^{\circ}$ | 2.47 ± 0.3 ab | 1.30 ± 0.2 a | 3.60 ± 0.3^d | 5.27 ±0.7° | 0.23 ±0.1 ° | - |
| C-07 | $0.57 \pm 0.1^{\circ}$ | 2.56 ± 0.2 ab | 1.78 ± 0.2 ° | $3.96 \pm 0.4^{\rm d}$ | 7.37 ± 0.6^{ab} | 0.17 ±0.3 ^a | - |
| C-08 | 0.57 ±0.1° | 2.48 ± 0.7 ab | 0.98 ± 0.2 a | 3.79 ± 0.6^d | 6.44 ±0.9e | $0.29 \pm 0.9^{\circ}$ | - |
| C-09 | 0.64 ± 0.6^d | 2.45 ± 0.9 ab | 1.32 ± 0.2 a | 4.08 ± 0.5^d | 4.72 ± 0.9^{d} | 0.17 ± 0.5^{a} | - |
| C-10 | $0.58\pm\!0.9^{b}$ | 2.48 ± 0.1 ab | $1.02 \pm 0.2 \ ^{\rm b}$ | $3.91 \pm \! 0.4^{d}$ | 5.39 ± 0.3^{bc} | 0.11 ± 0.3^{ab} | - |

Table 5. Functional properties of composite flour blends

^a100%SF: 100% Sorghum Flour, 100% FMF: 100% Finger Millet Flour, 100% WF: 100% Wheat Flour, samples coded with C-01 up to C-10: are composite flours obtained from D-optimal result for composite flour formulation and preparation of biscuit samples accordingly as presented in table 1.^{a-d} All data are means of three replicates \pm SD. Means with the same superscripts in a column do not differ significantly (p < 0.05) among the composite flours.

Functional properties of composite flours

The functional property evaluations of the composite flours were presented in Table 5. There was a significant (p<0.05) difference among them with a range of 0.57-0.87 g/mL, 1.78-2.68 g/g, 0.98-1.78 g/g, 3.60-5.75 mL/mL, 4.52 up to 8.03 g/g and 0.02-0.29 g/g for bulk density (BD), water absorption capacity (WAC), oil absorption capacity (OAC), swelling index (SI), swelling power (g/g) and water solubility index (WSI), respectively.

The BD ranged between 0.57 and 0.87 g/mL and the control wheat flour exhibited the lowest along with some of the composite flours with C-01 having the highest BD value. There was no significant (p<0.05) difference among the composite flour blends and the higher BD of the composite flours demonstrated grater compactness and possible mixed effect caused by the interaction of the molecules of the composite flours. The higher BD observed for C-01 (0.87 g/mL) implies that a solid, thick and compact packaging material may be required for this product as bulk density can influence the selection of packaging materials which relates to the sample particle size (Adeleke and Odedeji, 2010).

The WAC varied between 1.78 and 2.68 g water/g flour. The control wheat flour (100% WF) had the lowest (1.78 g water/g flour) and sample C-05 had the higher (2.68 g/g) WAC than the rest of the composite flours blends showing no significant (p<0.05) difference. The variations of the composite flours in particle size distribution may have influenced the WAC for the composite flours. According to Abu et al. (2006) the physical entrapment of oil within the flour starch structures significantly influences the oil absorption in flours starch as it is not able to possess nonpolar sites as compared to those flour components found in proteins.

The composite flour had better with sample C-08 exhibited the lowest (0.98 g oil/g flour) and C-07 the highest (1.78 g oil/g flour). According to Taiwo et al. (2017), the effect of OAC can be seen at the storage ability of a flour sample which is very important to take into consideration when developing a new food product as it can have an influence on the shelf life stability of the products. Thus, the biscuit samples prepared from the composite flours could have a relatively lower shelf life as compared to the control.

There were significant (p<0.05) differences in SP among the composite flours and the control wheat flour. The swelling power of composite flour blends ranged from 4.52 up to 8.03 g/g. C-01 had the highest SP, while C-02 had the lowest value. The increased amount of amaranthus flour had increased the swelling power in composite flours. As Carcea and Acquistucci (1997) indicated, the water absorption of the starch granules in the flour could be influenced by SP/SI capacity of the flours. The wet gluten content (WGC) determination resulted nil for 100% grain flours and composite flour blends, while the control exhibited (31.48%) and this result is comparably the same with the result of Thorat and Ramachandran (2016). This proves that the grains used in this study are gluten free.

Proximate and mineral composition of composite flour blends and biscuit samples

Proximate composition

As Table 6 shows, the proximate composition of biscuit samples produced from the composite flours varied significantly (p<0.05) with a range of 2.22-10.43%, 6.65-10.85%, 1.89-3.4%, 28.62-37.75%, 1.40-1.73% and 46.90-57.23% for moisture, protein, crude fibre, crude

fat, ash and carbohydrate, respectively. There was no significant (p<0.05) difference in the moisture content of the prepared biscuit samples. The moisture content of the biscuit samples were significantly lower comparing to the required moisture content for such products (<10%), which can result in a better shelf life and reduced effect on the quality attributes of the product. The protein content varies significantly (p<0.05) among the biscuit samples with sample C-01 having the highest (13.75%), while the control sample had the lowest (7.01%). All the formulated biscuit samples had higher protein content than the control, which is close and conformed to the minimum FAO/ WHO recommended value of 10%. The protein content of the samples had shown an increase as the proportional amount of amaranth (18.90%) and sorghum (11.73%) flour increased, which is comparably the same with the result of Belton and Taylor (2002) and Sousa et al. (2014). The fiber contents of the biscuit samples were significantly (p<0.05) different, but the values were well within the recommended range (5 g/100 g) of FAO/WHO. The carbohydrate content of biscuit samples increased with the addition of finger millet flour. This may be due to higher carbohydrate content in finger millet than in amaranthus and sorghum flour, which conforms with the results of Sousa et al. (2014) and Suma et al. (2014).

Mineral analysis

Ready to eat foods, like biscuits, have a potential to fulfill the recommended dietary allowance (RDA)

mg/100g) (600-1200 for minerals with а significantly lower consumption comparing to other kind of foods. This is found to be the most important for those who suffer from malnutrition and mineral deficiency. Most of biscuit samples prepared from the composite flour blends had significantly (p<0.05) higher iron content than the control (16.88) mg/100 g). The iron content ranged between 14.21 to 23.21 mg/100 g for the prepared biscuit samples as presented in Table 7. However, there was a significant (p<0.05) difference in the zinc content of biscuit samples and it ranged from 2.44 to 3.60 mg/100 g, where sample C-08 obtained the highest value. Phosphorus resulted in a significant (p < 0.05) difference between the composite and control wheat flour. It ranged from 252.76 to 277.39 mg/100 g for biscuit samples prepared from the composite flours and the increased amount of finger millet in the composite flours resulted better in phosphorous content. Biscuit samples prepared from the composite flour had a significant (p<0.05)difference as compared to the control in terms of calcium content. Sample C-03 exhibited the highest 27.54 mg/100 g and the sample C-09 (15.65 g/100 g) had the lowest mineral value for calcium. The results showed that biscuit samples prepared from composite flours have shown better results in terms of mineral content and conform with the findings of Belton and Taylor (2002).

| Sample | | | | Chemical Compos | sition | | |
|---------------------|--------------------------|---------------------------|--------------------------|--------------------------|----------------------------|--------------------------------|------------------------------|
| | Moisture Content (%) | Crude Protein (%) | Crude Fiber (%) | Crude Fat (%) | Ash (%) | CHO (%) | Energy Value (Kcal/100gm) |
| 100%AF ^a | 9.38 ± 0.21^{a} | 18.90 ± 0.02^{a} | 2.22 ± 0.03^{a} | $6.87\pm0.15^{\rm a}$ | $1.66\pm0.04^{\rm a}$ | $60.97\pm0.92^{\rm a}$ | 381.87 ± 0.48^a |
| 100%SF | 9.58 ± 0.75^{ab} | $11.73\pm0.04^{\rm c}$ | 2.48 ± 0.1^{b} | 3.00 ± 0.31^{bc} | $1.64\pm0.12^{\rm a}$ | 71.57 ± 1.03^{ab} | 360.20 ± 0.97^{ab} |
| 100%FMF | 10.01 ± 0.25^{b} | $8.23\pm0.01^{\text{de}}$ | 2.23 ± 0.02^{a} | $2.37\pm0.03^{\text{b}}$ | 1.72 ± 0.01^{ab} | 75.44 ± 0.67^{b} | 354.25 ± 1.04^{b} |
| 100%WF | 10.43 ± 0.31^{b} | 10.75 ± 0.01^{bc} | $1.80 \pm 0.02^{\rm d}$ | $3.54\pm0.21^{\circ}$ | $0.62\pm0.02^{\rm c}$ | 72.86 ± 0.59^{ab} | 366.30 ± 1.25^{ab} |
| C-00 | $2.79\pm0.13^{\circ}$ | $7.01\pm0.16^{\rm g}$ | 1.31 ±0.15° | 31.50 ± 0.30^{de} | $1.77\pm0.02^{\rm b}$ | $55.62\pm0.95^{\rm c}$ | $534.02\pm0.87^{\text{c}}$ |
| C-01 | $2.60\pm0.14^{\text{c}}$ | $13.75\pm0.12^{\text{e}}$ | 1.91 ±0.21 ^{de} | 28.75 ± 0.45^{de} | $1.59\pm0.10^{\text{e}}$ | $51.40 \pm 1.32^{\text{cd}}$ | 519.35 ± 0.56^{d} |
| C-02 | $3.41\pm0.14^{\rm d}$ | $10.83\pm0.03^{\rm f}$ | 2.18 ± 0.04^{ab} | 28.62 ± 0.35^{d} | $1.63\pm0.06^{\rm a}$ | $53.33 \pm 0.47 {}^{cd}$ | $514.22 \pm 0.99^{\ d}$ |
| C-03 | $3.21\pm0.15^{\rm d}$ | 11.05 ± 0.14^{d} | 1.69 ± 0.08^{cd} | 31.75 ± 0.41^{e} | $1.40\pm0.01^{\text{d}}$ | $50.90\pm0.86^{\rm d}$ | 533.55 ± 0.93^{e} |
| C-04 | $2.42\pm0.05^{\rm c}$ | $9.65\pm0.03^{\rm f}$ | 1.57 ±0.12 ^{cd} | 31.25 ± 0.10^{de} | $1.73\pm0.11~^{ab}$ | $53.38\pm1.09^{\ cd}$ | 533.37 ± 0.05^{e} |
| C-05 | $2.22\pm0.03^{\rm c}$ | $10.05\pm0.02^{\text{d}}$ | 1.89 ± 0.70^{cd} | 30.75 ± 0.10^{de} | $1.69\pm0.02^{\rm a}$ | 53.40 ± 1.23 ^{cd} | 530.55 ± 1.05^{e} |
| C-06 | $2.81\pm0.01^{\rm c}$ | 10.77 ± 0.01^{bc} | 1.78 ± 0.15^{cd} | 29.62 ± 0.23^{de} | $1.57\pm0.08^{\text{e}}$ | $53.45 \pm 0.68 ^{cd}$ | 523.46 ± 0.08^{de} |
| C-07 | $3.22\pm0.01^{\text{d}}$ | 9.80 ± 0.12^{b} | 2.04 ±0.01 ^e | 30.75 ± 0.08^{de} | $1.73\pm0.04^{\ ab}$ | $52.46\pm0.87^{\ cd}$ | 525.79 ± 0.97^{de} |
| C-08 | $2.81\pm0.04^{\rm c}$ | 9.40 ± 0.16^{de} | 1.38 ± 0.07^{cd} | 29.75 ± 0.25^{de} | 1.63 ± 0.07^{aa} | $55.03 \pm 0.12 ^{cd}$ | 525.47 ± 0.67^{de} |
| C-09 | $2.62\pm0.12^{\rm c}$ | $10.85 \ \pm 0.04^{bc}$ | 1.46 ± 0.04^{cd} | 30.25 ± 0.17^{de} | 1.71 ± 0.1 ^{ab} | $53.11\pm1.04^{\ cd}$ | 528.09 ± 0.85^{de} |
| C-10 | $3.21\pm0.01^{\text{d}}$ | 9.81 ± 0.06^{bc} | 1.95 ±0.13 ^{de} | 30.75 ± 0.03^{de} | $1.72\pm0.01~^{ab}$ | $52.56\pm0.85^{\ cd}$ | 526.23 ± 0.49^{de} |

Table 6. Proximate analysis of raw flour blends and baked biscuit samples (dry basis)

^a100%AF: 100% Amaranth Flour, 100% SF: 100% Sorghum Flour, 100% FMF: 100% Finger Millet Flour, 100% WF: 100% Wheat Flour and C-00: biscuit prepared from 100% wheat flour as a control. Samples coded with C-01 up to C-10. Represents the biscuit samples prepared from composite flour blends which were developed by using D-optimal Design as presented in Table 1. a-e All data are means of three replicates \pm SD. Means with the same superscripts in a column do not differ significantly (p<0.05).

| Samples | Mineral Composition (mg/100 g) | | | | | | | |
|---------|--------------------------------|------------------------------|------------------------------|--------------------------------|--|--|--|--|
| | Fe | Zn | Р | Ca | | | | |
| 100%AF | $13.96\pm0.98^{\text{a}}$ | $2.67\pm0.23^{\rm a}$ | $273.80\pm0.52^{\mathrm{a}}$ | $25.53\pm0.74^{\rm a}$ | | | | |
| 100%SF | 24.19 ± 0.42^{b} | $2.73\pm0.35^{\rm a}$ | $283.03\pm0.32^{\rm a}$ | 24.16 ± 1.04^{a} | | | | |
| 100%FMF | $22.97 \pm 0.38^{\ b}$ | $3.05\pm0.85^{\rm b}$ | $281.01 \pm 0.47^{\rm a}$ | $24.93{\pm}~1.25^{\mathrm{a}}$ | | | | |
| 100%WF | 10.96 ± 0.15 a | $3.74\pm1.02^{\mathrm{b}}$ | $34.56\pm0.65^{\mathrm{b}}$ | 57.19 ± 0.63^{b} | | | | |
| C-00 | $16.88 \pm 1.04^{\rm c}$ | $2.92 \pm 1.09^{\mathrm{a}}$ | $26.87\pm0.74^{\mathrm{b}}$ | $22.71\pm0.54^{\rm a}$ | | | | |
| C-01 | $18.14\pm2.14^{\rm c}$ | $2.86\pm0.94^{\rm a}$ | $252.76 \pm 0.68^{\circ}$ | $18.84\pm0.41^{\rm c}$ | | | | |
| C-02 | $17.62\pm0.90^{\circ}$ | 2.44 ± 1.21^{a} | $269.77 \pm 0.64^{\circ}$ | $12.75 \pm 0.30^{\circ}$ | | | | |
| C-03 | $16.41 \pm 1.05^{\circ}$ | $2.82 \pm 1.41^{\mathrm{a}}$ | $255.65 \pm 0.91^{\circ}$ | $27.54 \pm 1.25^{\mathrm{a}}$ | | | | |
| C-04 | 20.05 ± 0.68^{bc} | $2.69\pm0.56^{\rm a}$ | $261.70\pm1.02^{\text{c}}$ | $20.20\pm1.42^{\mathrm{ac}}$ | | | | |
| C-05 | 19.15 ± 0.26^{bc} | $2.71\pm0.32^{\rm a}$ | $277.39\pm0.54^{\rm a}$ | $25.02\pm0.57^{\rm a}$ | | | | |
| C-06 | $17.29\pm0.87^{\rm c}$ | $2.75\pm0.47^{\rm a}$ | $276.63 \pm 0.97^{\rm a}$ | 25.20 ± 1.36^{a} | | | | |
| C-07 | 23.21 ± 1.42^{b} | $2.49\pm0.36^{\rm a}$ | $260.84\pm1.03^{\circ}$ | 22.32 ± 0.74^{ac} | | | | |
| C-08 | $15.79\pm1.03^{\rm a}$ | $2.68\pm0.85^{\rm a}$ | $276.11 \pm 0.92^{\rm a}$ | 26.46 ± 0.95^{a} | | | | |
| C-09 | $14.21\pm0.45^{\rm a}$ | $2.60\pm0.96^{\rm a}$ | $260.55 \pm 0.90^{\rm c}$ | $15.65 \pm 1.45^{\circ}$ | | | | |
| C-10 | 14.86 ± 0.91 a | 2.70 ± 1.03 a | 274.52 ± 0.46^{a} | $16.5\pm0.85^{\rm a}$ | | | | |

 Table 7. Mineral analysis of raw flour blends and baked biscuit samples (dry basis)

^a100%AF: 100% Amaranth Flour, 100%SF: 100% Sorghum Flour, 100% FMF: 100% Finger Millet Flour, 100% WF: 100% Wheat Flour and C-00: biscuit prepared from 100% wheat flour as a control. Samples coded with C-01 up to C-10. Represents the biscuit samples prepared from composite flour blends which were developed by using D-optimal Design as presented in Table 1.

^{a-e} All data are means of three replicates \pm SD. Means with the same superscripts in a column do not differ significantly (p<0.05).

Physical properties of biscuit samples

The physical characteristics of biscuit samples prepared from composite flour, as well as a control (100% wheat flour), are presented in Table 8. The diameter (D), thickness (T) and weight (W) of biscuit samples were ranged from 51.48 to 62.00 mm, 8.43 to 11.95 mm and 11.12 to 11.87 g, respectively. In terms of D, there was a significant (p<0.05) difference among the biscuit samples, while there were no significant (p≥0.05) differences in biscuit weight.

The result showed that, as the level of sorghum flour increases, the decrease in biscuit T was more remarkable and the increase in finger millet reduced the W and T of biscuit samples. According to Ragaee and Abdel-Aal (2006), T is affected by the quantity and quality of protein in the flour, whereas weight is basically determined by the quantity of dough baked and the amount of moisture and carbon dioxide diffused out of the biscuit samples prepared from the composite flour had relatively higher weight, which may be due to low retention of carbon dioxide gas in the blended dough, hence providing dense biscuit texture (Haridas and Malini, 1991).

The spread factor (SF), which is the ratio of D and T, has been generally adopted as a more reliable measure of biscuit size (Shittu et al., 2007) and it ranged from 5.01 to 6.28. Comparing to the control sample, the most of the biscuit samples formulated from the composite flour exhibited a lower water activity (a_w) (Table 8). It could be associated with the higher WAC of wheat flours (Table 5). According to Labuza et al. (1972), reducing a_w below 0.7 prevents microbial spoilage and to successfully preserve a food product, water activity would have to be lowered to a range where the rate of deteriorative reactions is minimized. The maximum force (hardness) required to break the biscuit sample was recorded along with the thickness, where this maximum force applied is presented in Table 8. The texture analysis showed a higher firmness as it was required a relatively higher force in biscuit made from composite flours when compared with biscuit made from a control with a confidence level (p<0.05).

The colour of the biscuit samples was related to the colour of the corresponding grain materials composited to prepare the samples. Sample C-08 had higher b* due to the colour derived from the amaranth flour and no differences were observed in lightness (L*). Thus, sample C-05 had the lowest lightness (L=45.51 \pm 0.12), which could be due to the increased amount of sorghum flour and decreased value of finger millet flour proportions. The hue green (a*) varied from 10.07 for C-04 to 13.55 for C-01. Lightness of ingredients plays an important role in bakery products due to consumer preferences.

Sensory qualities attributes of baked biscuit samples

The mean sensory scores of biscuit samples prepared from a composite flour blends at different level of ration as compared to the control (100% wheat flour)

| Samples | | | | | Physical | Parameters | | | | |
|---------|---------------------------|-----------------------|------------------------|-----------------------|-------------------------|-------------------------|------------------------|------------------------|-----------------------------|--------------------------|
| | | Color | | | Texture F | rofile | D(mm) | T(mm) | W (g) | SF (D/T) |
| | \mathbf{a}_{w} | L* | a* | b* | Force (N) | Thickness (mm) | | | | |
| C-00 | $0.31\pm1.0^{\rm a}$ | 56.94± 0.1ª | $10.19\pm0.09^{\rm a}$ | $28.28\pm0.4^{\rm a}$ | $5.50\pm1.0^{\rm a}$ | $3.20\pm1.0^{\rm a}$ | $62.00\pm0.02^{\rm a}$ | $10.63\pm0.14^{\rm a}$ | $11.12\pm0.03^{\rm a}$ | $5.83\pm0.10^{\rm a}$ |
| C-01 | $0.28\pm1.0^{\rm b}$ | 45.46 ± 0.7^{b} | $13.55\pm1.04^{\rm b}$ | $29.10\pm1.1^{\rm a}$ | $4.00\pm1.0^{\rm b}$ | $2.50\pm1.0^{\rm b}$ | $56.82\pm0.01^{\rm b}$ | $09.04\pm0.02^{\rm b}$ | 11.41 ± 0.20^{a} | $6.28\pm0.21^{\text{b}}$ |
| C-02 | $0.34\pm1.0^{\rm a}$ | 46.46 ± 0.3^{b} | $12.72\pm0.23^{\rm b}$ | 27.95 ± 0.6^{ab} | $3.05\pm1.0^{\rm c}$ | $3.30\pm1.0^{\rm a}$ | $58.23\pm0.12^{\rm b}$ | 09.54 ± 0.42^{b} | $11.14\pm0.12^{\rm a}$ | 6.10 ± 0.32^{b} |
| C-03 | $0.26 \pm 1.0^{\rm c}$ | 47.77 ± 1.1^{b} | $12.06\pm1.45^{\rm b}$ | $26.94\pm0.4^{\rm a}$ | 4.50 ± 1.0^{ab} | $5.01\pm1.0^{\rm c}$ | 54.22 ± 0.41^{b} | 09.11 ± 0.21^{b} | $11.82\pm0.02^{\mathrm{a}}$ | 5.95 ± 0.02^{b} |
| C-04 | 0.28 ± 1.0^{b} | $53.16\pm0.6^{\rm a}$ | $10.07\pm0.02^{\rm a}$ | $28.81\pm0.6^{\rm a}$ | $3.00\pm1.0^{\rm c}$ | $5.00\pm1.0^{\rm c}$ | $58.93 \pm 0.2b \\$ | $10.63\pm0.01^{\rm a}$ | 11.81 ± 0.06^{a} | 5.54 ± 0.04^{b} |
| C-05 | $0.28\pm1.0^{\rm b}$ | 45.51 ± 0.1^{b} | $12.75\pm0.45^{\rm b}$ | 27.83 ± 0.2^{ab} | $3.00\pm1.0^{\rm c}$ | $4.40\pm1.0^{\rm d}$ | $57.99\pm0.15^{\rm b}$ | 09.35 ± 0.02^{b} | $11.17\pm0.04^{\rm a}$ | 6.20 ± 0.18^{b} |
| C-06 | 0.29 ± 1.0^{b} | 48.62 ± 0.8^{b} | 12.25 ± 0.68^{b} | 26.74 ± 1.0^{b} | $6.00\pm1.0^{\rm a}$ | $3.0\pm1.0a$ | $51.48\pm0.10^{\rm c}$ | $08.43\pm0.05^{\rm c}$ | $11.47\pm0.14^{\rm a}$ | 6.11 ± 0.05^{b} |
| C-07 | $0.25\pm1.0^{\rm c}$ | 45.92 ± 0.1^{b} | $10.56\pm0.22^{\rm a}$ | $26.27\pm0.8^{\rm b}$ | 4.00 ± 1.0^{b} | $3.05\pm1.0^{\rm a}$ | $59.83\pm0.24^{\rm a}$ | $11.95\pm0.14^{\rm a}$ | $11.14\pm0.25^{\rm a}$ | $5.01\pm0.14^{\rm a}$ |
| C-08 | $0.26\pm1.0^{\rm c}$ | 48.01 ± 1.2^{b} | 12.50 ± 0.39^{b} | $30.06\pm1.1^{\rm c}$ | $4.00\pm1.0^{\rm b}$ | $3.81\pm1.0^{\rm a}$ | 58.71 ± 0.32^{b} | $10.08\pm0.06^{\rm a}$ | $11.62\pm0.14^{\rm a}$ | 5.82 ± 0.02^{b} |
| C-09 | 0.29 ± 1.0^{b} | 51.15 ± 1.2^{ab} | 12.83 ± 0.57^{b} | $29.81\pm0.8^{\rm c}$ | 5.00 ± 1.0^{ab} | $3.98 \pm 1.0^{\rm d}$ | $59.88\pm0.04^{\rm a}$ | 09.61 ± 0.05^{b} | $11.87\pm0.05^{\mathrm{a}}$ | 6.23 ± 0.07^{b} |
| C-10 | $0.28\pm1.0^{\rm b}$ | 48.39 ± 0.2^{bc} | 13.11 ± 1.0^{cd} | 26.86 ± 0.2^{ab} | $6.00 \pm 1.0^{\rm cc}$ | $4.40\pm1.0^{\text{e}}$ | $54.89\pm0.08^{\rm b}$ | $10.74\pm0.16^{\rm a}$ | $11.36\pm0.02^{\rm a}$ | $5.11\pm0.05^{\rm a}$ |

| Table 8. Physical | dimensions a | ind properties | of biscuit samples |
|-------------------|--------------|----------------|--------------------|
| | | | |

^{*a*}C-00: biscuit prepared from 100% wheat as a control and Samples coded with C-01 up to C-10, represents the biscuit samples prepared from a composite flour blends which was developed by using D-optimal Design as presented in Table 1.; L^* : lightness; a^* : greenness; b^* yellowness. ^{*a*-d} All data are means of three replicates \pm SD. Means with the same superscripts in a column do not differ significantly (p<0.05).

Table 9. Sensory quality attributes evaluation of biscuit samples biscuit samples

| | Sensory Evaluation Parameters | | | | | | | |
|--------|-------------------------------|------------------------------|----------------------------|----------------------------|-------------------------|----------------------------|--------------------------|--|
| Sample | Appearanc | Color | Crispiness | Taste | Flavor | Texture | Overall Accept. | |
| C-00 | $7.85 \pm 1.45^{\rm a}$ | $7.80 \pm 1.60^{\mathrm{a}}$ | 7.75 ± 1.11^{a} | $7.20\pm2.04^{\rm a}$ | $7.40 \pm 1.50^{\rm a}$ | $7.90 \pm 1.48^{\rm a}$ | 7.80 ± 1.67^{a} | |
| C-01 | 6.90 ± 1.37^{b} | $6.80\pm1.67^{\rm b}$ | 6.70 ± 1.78^{b} | 6.40 ± 1.56^{b} | $5.60\pm1.81^{\rm b}$ | $5.95 \pm 1.84^{\text{b}}$ | $6.45\pm1.46^{\text{b}}$ | |
| C-02 | 6.40 ± 1.71^{b} | 6.40 ± 1.50^{b} | $6.70\pm1.41^{\text{b}}$ | $6.20\pm1.64^{\text{b}}$ | $6.00\pm1.58^{\rm c}$ | $6.30\pm1.80^{\circ}$ | 6.20 ± 1.31^{b} | |
| C-03 | 5.80 ± 1.85^{b} | $5.30 \pm 1.83^{\rm c}$ | $6.50\pm1.63^{\text{b}}$ | $5.85\pm2.10^{\rm c}$ | 5.20 ± 1.85^{b} | $6.10\pm1.68^{\text{b}}$ | $5.85 \pm 1.30^{\rm c}$ | |
| C-04 | 5.90 ± 1.71^{b} | $5.75 \pm 1.65^{\rm c}$ | $6.95\pm1.31^{\text{b}}$ | $6.10\pm1.33^{\rm c}$ | $5.95 \pm 1.35^{\rm c}$ | $5.95\pm1.50^{\text{b}}$ | 6.20 ± 1.28^{b} | |
| C-05 | 6.50 ± 1.71^{b} | $6.15\pm1.03^{\text{b}}$ | $6.85\pm1.13^{\text{b}}$ | $6.65 \pm 1.49^{\text{b}}$ | $6.20\pm1.36^{\rm c}$ | $6.35\pm1.46^{\circ}$ | 6.30 ± 0.97^{b} | |
| C-06 | 5.50 ± 1.14^{b} | $5.25 \pm 1.48^{\rm c}$ | $6.20\pm1.60^{\rm c}$ | $5.70 \pm 1.89^{\rm c}$ | 5.20 ± 1.54^{b} | 5.90 ± 0.96^{b} | $5.65 \pm 1.53^{\circ}$ | |
| C-07 | 5.80 ± 1.27^{b} | $5.70\pm1.86^{\rm c}$ | 6.60 ± 1.87^{b} | $5.35\pm1.75^{\rm c}$ | 5.10 ± 1.77^{b} | 5.90 ± 1.07^{b} | $5.41\pm2.08^{\rm c}$ | |
| C-08 | $6.25\pm1.74^{\rm a}$ | 6.25 ± 1.71^{b} | $6.85 \pm 1.26^{\text{b}}$ | $6.10 \pm 1.41^{\circ}$ | 5.90 ± 1.58^{b} | 5.80 ± 1.57^{b} | 6.15 ± 1.26^{b} | |
| C-09 | $7.05\pm1.31^{\rm a}$ | $6.95 \pm 1.05^{\text{b}}$ | $6.90 \pm 1.25^{\text{b}}$ | 6.50 ± 1.23^{b} | $6.15 \pm 1.53^{\circ}$ | $6.90\pm1.33^{\circ}$ | $6.65\pm1.26^{\text{b}}$ | |
| C-10 | 5.8 ± 1.73^{b} | 5.90 ±1.33c | 6.60 ± 1.39^{b} | $5.55 \pm 1.23^{\circ}$ | $5.55\pm1.53^{\rm b}$ | 6.00 ± 1.52^{b} | $5.70 \pm 1.26^{\circ}$ | |

^aC-00 stands for biscuit sample used as a control prepared from 100% wheat flou blends; Samples coded with C-01 up to C-10, represents the biscuit samples prepared from composite flour which was developed by using D-optimal Design as presented in Table 1.

a-c All data are means of three replicates \pm SD. Means with the same superscripts in a column do not differ significantly (p<0.05).

are presented in Table 9. The statistical analysis of the data showed that the biscuit samples prepared from the composite flour blends were significantly (p < 0.05)different from the control with the exception of C-09 which had comparably good sensory score with the control in appearance, colour, crispness, texture and overall acceptability with a sensory score of 7.05, 6.95, 6.90, 6.90 and 6.65, respectively, and sample C-05 in terms of flavour with a sensory score of 6.20. However, there were no significant (p>0.05) differences with respect to biscuit texture, appearance and crispness among the composite biscuit samples. It is evident from the results that the control had the highest overall acceptability in sensory score followed by biscuit prepared from C-09 composite flour blends, while sample C-06 ranked the least in most of the sensory attributes, which is comparably in agreement with the results of Karki et al. (2016). The low rating recorded may be due to the low level of sorghum and finger millet flour which resulted in poor colour and it might have affected the overall sensory attribute of the biscuit. This shows that using gluten free cereals alone to prepare biscuits will have very low accpetability as compared to wheat. The low sensory score for texture, flavour and acceptability for the composite flour blend biscuits may possibly be improved by addition of non-gluten proteins, such as egg and milk protein or soybean protein, or hydrocolloids and natural emulsifying agents that could mimic the viscoelastic properties of gluten (Alvarez-Jubete et al., 2010; Lazaridou and Biliaderis, 2009; Renzettia et al., 2008).

Conclusion

From the results, it was found that the incorporation of amaranthus flour improved the quality of composite flour in terms of nutrient density and pasting properties.

The reduced BDV and FQN in composite flour reasulted in poor physical characteristics like texture and apperance in the formulated biscuit samples, which can possibly be improved by using natural emulsifing and non-gluten proteins. The indicative optimal proportion of composite flours was amaranth flour 50%, sorghum 40%, and 10% finger millet flour. At this proportion the biscuits had a comparably better sensory quality attributes. Thus, these flours can be used in gluten-free biscuit formulations as replacement for wheat flour as they increase the nutritional value.

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