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Nutritional and Sensory Characteristics of Bread Produced from Wheat and Cassava Flour, Fortified with Sorrel Seed Protein Isolate

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Abstract- The possibility of making bread from composite flour containing 60-80% wheat flour, 10-25% cassava flour and 5-15% Sorrel seeds protein isolate was investigated. The proximate composition, mineral constituents, functional and sensory evaluation were analysed using standard methods. The results of the analysis showed that crude protein (8.80±0.36 to 18.70±0.35) and crude fibre (0.77±0.02 to 1.58±0.04) contents of the composite breads increased significantly with increased incorporation of cassava flour and sorrel seed protein isolate flours. The moisture (34.00±1.00 to 32.04±1.00), ash (1.22±0.03 to 0.66±0.03), carbohydrate(54.99±0.25 to 46.83±0.77) and fat (0.24±0.01 to 0.20±0.02) contents were observed to decrease significantly with corresponding increase in the percentage of the composite flours from 5-25% for both cassava flour and sorrel seed protein isolate flour. The results of the mineral contents showed that calcium element increased as the level of composite flour increased, while sodium, potassium and magnesium decreased as the level of inclusion increased. The functional properties, water and oil absorption, and swelling index of the composite flour showed varying degrees of variation from the control sample (100% wheat flour). The results of the sensory evaluation showed that there were no significant differences ($P>0.05$) in taste, texture, colour, flavour, appearance and overall acceptability, however, the mean sensory scores decreased with increased addition of cassava flour and sorrel seed protein isolate in the composite flour. The outcome of the research showed that, nutritious bread could be produced from the composite flours of wheat, cassava and sorrel seeds protein isolate.

Keywords- Bread, Wheat, cassava, Protein Isolate

1 INTRODUCTION

Over the years, much research aimed at incorporating non wheat materials of local origin to bread and other flour products has been undertaken to reduce wheat importation in countries where wheat are not grown. Such non- wheat flours are produced from other cereals, tubers, and root crops such as maize, sorghum, cassava, potato and plantain (Badifu and Aka, 2001). The use of blends of wheat and non-wheat flour, known as composite flours, have been used for producing bread, biscuits and other snacks for ages (Oladumoye *et al.*, 2010). Bread consumption has increased substantially in many developing countries due to changes in eating habits and a steadily growing population. However, the wheat flour used for making bread had to be imported in many tropical countries, as the climatic conditions and soil are not suited for wheat to be grown locally (Julianti et al., 2015). Flours from maize, barley, cassava and chickpea are among the most predominantly studied crops for the production of composite flour breads (Ali *et al.*, 2000). Fewer works have been done on some underutilized but highly nutritious oil seeds.

Cassava is a tropical crop with high content of carbohydrate. It does not contain the gluten protein which makes wheat preferred major baking flour. Physiochemical properties of cassava starch are suitable for supplementation of wheat flour in bread-making without compromising its sensory attributes (Eduardo *et al.*, 2013). Protein malnutrition is real in most developing countries because of the low animal proteins intake. Meat, fish, milk and eggs provide proteins with satisfactory amino acids pattern as well as bioavailable micronutrients such as iron, zinc, calcium and vitamin A which many malnourished people are deficient.

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However, these animal products are expensive, and most populations in developing countries cannot afford them. Thus, there is a need to look for locally available and cheap sources of food ingredients, particularly those that do not attract competition between humans and livestock, and one of such feed ingredients that can be used as protein supplement with little or no cost is Hibiscus sabdariffa L. (sorrel) seeds. Sorrel seeds are bi-products of calyxes which is cultivated in many countries such as Egypt, India, Mali, Malaysia, Nigeria, and Sudan, and have been found to contain high amount of protein, dietary fiber, lipids, and minerals. The usefulness of sorrel seeds as source of protein has been established by many authors (Hainida *et al.*, 2008; Sanni *et al.*, 2016). The present study was undertaken to examine the effect of the inclusion of high-quality cassava flour and sorrel seeds protein isolate to wheat flour on the functional properties of the composite flour and the nutritional and sensorial attributes of bread, thus producing inexpensive and nutritionally balanced food.

2 MATERIALS AND METHODS

2.1 PREPARATION OF THE FLOURS AND FORMULATIONS OF COMPOSITE FLOUR FOR BREAD PRODUCTION

Wheat flour and other ingredients such as granulated sugar and salt, baking fat, and milk were purchased from Ikole-Ekiti local market, in Ekiti State. High quality cassava flour of the yellow flesh variety TMS/01/1412 was obtained from the Federal University of Agriculture Abeokuta, Ogun State, Nigeria. The sorrel seeds were purchased from Kaduna Central market in Kaduna State of Northern Nigeria, and was processed into protein isolate, following the methods of Tounkara *et al.* (2013). The flour was packed separately into waterproof polyethylene film and kept at 27±2°C, until used. The composite flour is prepared and baked according to the method specified by Edwardo *et al*. (2013). Four different samples of bread were produced and coded as W_{100}, for 100% wheat flour, W_{80}, for 80% wheat, 15% cassava and 5% sorrel seed protein isolate (SPI): W_{70}, for 70% wheat, 20% cassava and 10% (SPI) and W_{60}, for 60% wheat, 25% cassava and 15% (SPI). The bread recipe consisted of 100g of each blend, 6.2g sugar,1.7g salt, 3.9g margarine, 3.3g yeast, 0.02g ascorbic acid and 56ml of warm water (43°c).

The dough was prepared, proofed in pre-oiled baking pans. The loaves were baked in a charcoal oven at 180°C for 25 min. The loaves of bread were de-panned and cooled at room temperature for 2 and packed in imperforated low-density polythene bags. All samples were stored at 27°C prior to analysis within 24 h.

2.2 Proximate Analysis

Moisture content, crude protein, crude fat, crude fibre and ash were determined by the standard methods of AOAC (2000). Carbohydrate was expressed as a percentage of the difference between the addition of other proximate chemical components and 100%.

Carbohydrate = 100 - (protein crude fat + ash + fibre + moisture)

2.3 Functional Properties

Water and oil absorption capacity of the blends were determined by modification of the method described by Lin and Zayas (1987), respectively. Swelling power was evaluated based on modified method of Leach *et al.* (1959)

2.4 Mineral Composition

The analyses for essential mineral elements were investigated using atomic absorption spectrophotometric method (Fashakin *et al.,* 1991). 0.5g of the sample was weighed into a digestion flask and 10 ml of nitric acid and HCl (10 ml) was added. The mixture was digested for 10 min. The digested mixture was filtered using No 1 Whatman filter paper. The filtrate was made up to 50ml with distilled water. An aliquot was transferred to the Auto-analyser for total phosphorus analysis at 420nm. The left-over digest was used to determine the other elements (calcium, sodium, magnesium) using the Atomic Absorption Spectrophotometer (Perkin Elmer, model 402) while sodium and potassium were determined using flame photometry

2.5 Sensory Evaluation

The test bread samples were subjected to sensory analysis. Three (3) samples of composite bread and the control were served to 10 semi-trained panellists, who were the Students of Federal University, Oye-Ekiti, Nigeria, and are familiar with bread. The panellists tasted the bread and scored each sample using a 9-point Hedonic scale (Iwe, 2010) where 1 = extremely unacceptable and 9 = extremely acceptable. Attributes evaluated include; bread appearance, crust colour, texture, taste, flavour and overall acceptability.

2.6 Statistical Analysis

Data were analysed using Analysis of Variance ANOVA and mean separated by New Duncan Multiple range test using SPSS 21 computer programme. Significance was accepted at 5 % level of probability.

3 Results and Discussion

3.1 Proximate Composition Of The Bread Samples

The results of the proximate compositions are presented in Table 1. The control sample W_{100} had significantly ($P>0.05$) high moisture content than the composite flour, followed closely by sample W_{80}, W_{60} and W_{70}, respectively. There was no significant difference ($P<0.05$) in the moisture content of sample W_{80} and W_{60}. These values are relatively high, though are within the range reported by other researchers (Xiao *et al*., 2016) and an indication of their short shelf life. The inclusion of sorrel seeds protein isolate produced significantly ($P>0.05$) higher protein 18.70±0.035% in $W_{60,}$ 15.03±0.05 in W_{70} and 12.01±0.36% in W_{80} than the control W_{100} which is 8.80±0.36.

This result is in contrast with a previous report by Nwosu et al. (2014) where increased cassava ratio in wheat/cassava/ soybean bread was observed to significantly decreased the protein content of the composite flour bread. This could be attributed to protein isolate with high protein content used in the fortification as compared with the malted soybean flour used by the investigators (Nwosu *et al.,* 2014). Also the reason for the low protein content, as the cassava flour substitution was increased can also be attributed to the low protein content of cassava flour since it has low protein content. Mashayekh *et al.* (2008) also reported increase in protein content of bread as a result of the addition of soy flour to wheat flour. Other studies also reported similar increase in protein content of sorghum-soy composite flours (Awadel kareem *et al.,* 2008).

The ash contents of the bread samples decreased as the level of composite flour inclusion increased. The control sample (W_{100}) had the highest ash content (1.22 %) while sample W60 had the least ash content (0.66%). The values recorded in this study is in line with what Olapade and Oluwole, (2013) found in Wheat-acha, cowpea composite flour bread. The crude fibre contents of the bread samples also increased as the substitution of cassava flour increased in the composite blend. Sample W_{100} (100% wheat bread) had the least crude fibre content (0.77%) while sample W_{60} (60% Wheat flour, 15% Sorrel Protein Isolate, 25% cassava flour) had the highest crude fibre content (1.58%). All values of crude fibre obtained were significantly different ($P > 0.05$) from each other. The increased fibre content observed with increase cassava flour substitution might be due to high fibre content in cassava (Nwosu *et al*., 2014). The fat contents of the formulated bread samples were low and no significant difference ($P > 0.05$) in the fat content was observed between W_{80} and $W_{100.}$ Sample W_{60} had the lowest fat content and this may be attributed to the high content of cassava flour (25%) used in for the sample.

The increased fibre and lower carbohydrate content of the composite breads have several health benefits, as it will aid in the digestion of the bread in the colon and reduce constipation often associated with bread produced from refined wheat flour (Elleuch *et al*., 2011). According to well documented studies, it is now accepted that dietary fibre plays a significant role in the prevention of several diseases such as; cardiovascular diseases, constipation, irritable colon, cancer and diabetes (Elleuch et *al*., 2011).

Table 1. Proximate composition of formulated bread

Samples	Moisture (%)	Fat (%)	Ash (%)	Fibre (%)	Protein (%)	Carbohydrate (%)
W100	34.00±0.04 ᵃ	0.24±0.01 ᵃ	1.22±0.03 ᵃ	0.77±0.02 ᵈ	8.80±0.36 ᵈ	54.99±0.25 ᵃ
W80	32.80±0.25ᵇ	0.24±0.01ᵃ	1.15±0.01 ᵃ	1.08±0.03 ᶜ	12.01±0.04 ᶜ	52.73±0.23 ᵇ
W70	27.59±0.37ᶜ	0.21±0.01ᵃᵇ	0.95±0.01 ᵇ	1.27±0.02 ᵇ	15.03±0.05 ᵇ	54.97±0.32 ᵃ
W60	32.04±1.00ᵇ	0.20±0.02 ᵇ	0.66±0.03 ᶜ	1.58±0.04 ᵃ	18.70±0.35 ᵃ	46.83±0.77 ᶜ

The values are mean ±standard deviations and those in the same column not sharing the same superscript letter are significantly different from each other (P<0.05).

3.2 Functional Properties of Samples

The result of the water absorption capacity from Fig.1 showed that, sample W_{80} had the highest water absorption capacity (WAC). The water absorption capacity for W_{80} was 4.88 g/g which was closely followed by W_{70}. The water absorption capacity was observed to be higher in bread formulation with Sorrel protein isolate fortification when compare to the control sample made from 100% wheat flour. The composite flour showed higher affinity for water than wheat flour, except where the level of inclusion with cassava flour reached 25%. Similar results were observed by Juliant *et al*. (2017).

Water absorption has been reported to have important implication for baking application, viscosity, bulking and consistency of flour products (Niba *et al.,* 2001). So also, the Oil absorption capacity follow the same trend, as with the Water absorption capacity. W_{80} (3.94 g/g) was significantly higher (p>0.05) than sample W_{70} and W_{60} but not significantly different from the control sample (W_{100}). The low cassava flour substitution in W_{80} compared to other samples might have been a reason for this observation. This observation agrees with previous reports by Gbadamosi *et al*. (2012) where increased in protein concentration and decrease in carbohydrate concentration were observed to increase oil absorption capacity of the tested samples.

The swelling capacity showed similar trend as of the WAC. But sample with 25% cassava (W60) inclusion showed a higher swelling capacity than the 20% (W70). This may be because cassava flour had higher swelling capacity than wheat. Swelling index and Water absorption index were reported to contribute to dough formation and stability, and is part of the criteria for good quality product (Olapade and Oluwole, 2013).

3.3 Minerals Composition of the Bread Samples

Table 2 showed the results for minerals content of different wheat-cassava-sorrel protein isolate substitution products bread. The results of the mineral elements evaluated showed that there were significant reductions in all minerals except calcium, when compared to the control, the 100% wheat flour bread.

Low content of this mineral element in the substituted products (cassava flour and sorrel protein isolate) might be responsible for this observation. Generally, the same trend was reported by Salama et al. (1992) and Nwosu et al. (2014) in cassava incorporated bread. Minerals are essential nutrients that are needed in the body to facilitate proper functioning of certain organs.

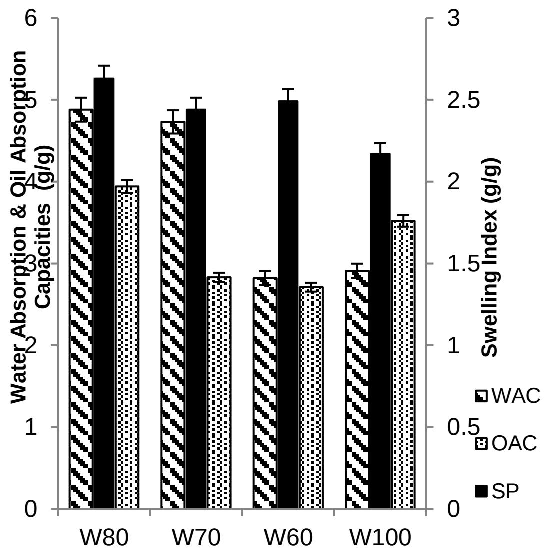

Fig.1: Functional Properties of Samples

Table 2. Minerals content of formulated bread

Samples	Na (mg/ 100g)	K (mg/ 100g)	Ca (mg/ 100g)	Mg (mg/ 100g)
W_{100}	12.08±0.03ᵃ	4.10 ±0.01 ᵃ	1.24 ±0.01 ᶜ	0.98 ±0.01 ᵃ
W_{80}	11.03±0.02 ᵇ	4.10 ±0.02 ᵃ	1.29 ±0.01 ᶜ	0.85 ±0.01 ᵇ
W_{70}	10.03±0.02ᶜ	4.02 ±0.01 ᵇ	1.46 ±0.03 ᵇ	0.82 ±0.01 ᵇ
W_{60}	10.03±0.02ᶜ	3.98 ±0.02 ᵇ	1.87 ±0.01 ᵃ	0.66 ±0.02 ᶜ

The values are mean ±standard deviations and those in the same column not sharing the same superscript letter are significantly different from each other (P<0.05).

3.4 Sensory Evaluation of the Bread Samples

Sample W_{100} from Table 3 had the best score in terms of the mean score, and is closely followed by sample W80. There was no significant difference (P > 0.05) between tastes of W100 (control sample) and W80 (80% Wheat flour, 5% Sorrel Protein Isolate, 15% cassava flour). Likewise, there were no significant difference (P > 0.05) between sample W_{70} and sample W_{60}. Thus, at 80% and 100 % wheat substitution, there was no difference in the tastes of the bread sample but at wheat substitution of 60% and 70%, there were changes in the tastes of the bread sample. The mean score for sample W_{80} and W_{100} indicates that these samples were liked very much while sample W_{60} and W_{70} were liked slightly.

Furthermore, the mean scores for the crust colour of the bred samples also decreased as the cassava flour substitution increased in the samples (W100-W60). Increased addition of sorrel protein isolate did not improve the mean score of the sensory evaluation of crust colour. The crust colour of the control sample (W100) was significantly different from all other samples. Mean score of samples W80, W70 and sample W60 indicates that there was no significant difference (P> 0.05) in their sensory evaluation in terms of crust colour. This results showed clearly that the crust colours of the bread were comparable and almost the same. This could be attributed to Maillard reaction caused by about by the amino acid proteins of the protein isolate of the Sorrel seeds and the carbohydrate of the wheat flour which reacted with the added sugar to form the brown crust of the bread. The mean score for colour indicates that sample W80, W70 and W60 were liked moderately while sample W100 was liked very much. Likewise, there was no significant difference (P > 0.05) in the aroma of bread sample W80 and W100.

Same observation was also recorded for sample W70 and W60 where no significant difference was observed. Sample W100 had the highest mean score for aroma (7.67±0.33) closely followed by W80 (7.11±0.35) and the reason why there was no marked in the aroma of the bread samples could be that the sorrel protein isolate used in blend, could have masked the odour of cassava flour in each of the various blends. Also, there was no significant difference in the textures of the bread samples (W80-W60) except sample W100, the control product. A marked difference existed between sample W100 (the control sample) and the experimenter products (W60-W80). These results showed that the Sorrel protein isolates used as improver might have had same effect on the texture consistency of sample W60-W80, with exception to W100 which was not formulated with it. The mean scores showed that the textures of sample W60-W80 were liked moderately while sample W100 was liked very much.

Table 3. Sensory attributes of formulated bread

Samples	Taste	Colour	Flavour	Texture	Appearance	Overall Acceptability
W_{100}	$8.22±0.22^{a}$	$8.22±0.22^{a}$	$7.67±0.33^{a}$	$7.78±0.22^{a}$	$8.00±0.24^{a}$	$8.22±0.22^{a}$
W_{80}	$7.67±0.17^{a}$	$7.44±0.29^{b}$	$7.11±0.35^{a}$	$7.11±0.42^{b}$	$7.22±0.28^{ab}$	$7.11±0.31^{ab}$
W_{70}	$6.00±0.33^{b}$	$6.33±0.41^{b}$	$6.22±0.28^{b}$	$6.67±0.55^{ab}$	$6.22±0.40^{b}$	$6.33±0.37^{b}$
W_{60}	$5.78±0.46^{b}$	$6.22±0.62^{ab}$	$6.22±0.46^{b}$	$6.11±0.54^{ab}$	$6.00±0.60^{b}$	$6.00±0.62^{b}$

The values are mean ±standard deviations and those in the same column not sharing the same superscript letter are significantly different from each other (P<0.05).

Furthermore, the mean scores for overall acceptability showed that there were no significant differences in the acceptability of sample W_{80} and the control sample (W_{100}) made from 100% wheat flour. Sample W_{100} had the highest mean score followed closely by W_{80}. Bread sample with up to 25% cassava substitution were acceptable and their characteristics were similar to the control sample. This observation is in agreement with previous report by Nwosu *et al.* (2014) where quality parameters of bread produced from substitution of wheat flour with cassava flour using soybean as an improver.
These results were similar to those obtained by Eddy *et al.* (2007) who reported that breads baked with 10 and 20 % cassava-wheat composite flour were not significantly different in any sensory attributes, and also in consumers' readiness to buy compared to the control (100% wheat).

4 Conclusion

This study highly nutritious bread rich in protein and mineral content could be produced from composite flours of wheat, cassava and sorrel seeds protein isolate. The most preferred of all the samples, based on all parameter assessed is the sample with 75% wheat, 15% cassava and 10 sorrel isolates.

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