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RESEARCH ARTICLE

ASSESSMENT OF NUTRITIONAL QUALITIES AND ACCEPTABILITY OF BREADS PRODUCED WITH MORINGA OLEIFERA POD FLOUR.

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Abstract

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An in-vitro assay was carried out to ascertain the effect of addition of dry *Moringa oleifera* pod flour on the quality of composite bread, which hitherto had constituted an important dietary intake of many people in Nigeria. *M. oleifera* pod was processed into flour and mixed with composite flour for bread production. Four samples of bread were produced and labelled Samples 20 g, 40 g, 60 g and Control. The control contained 100 % wheat flour, while Sample 20 g to 60 g had dry *M. oleifera* pod flours added in an increasing order of 20 %, 40 % and 60 %. The nutritional properties of the bread loaves were evaluated and the result varied significantly with increasing levels of *M. oleifera* pod flours. The proximate composition of bread fortified with *M. oleifera* pod flours showed that the moisture content decreased significantly ($p<0.05$) with increased *M. oleifera* pod flours varying from 22.4 % to 32.6 % while that of protein contents increased from 2.84 % to 3.06 %. The fat contents of the bread samples did not differ significantly ($p<0.05$) with increase in *M. oleifera* pod flours while the carbohydrate contents of the bread samples increased significantly ($p<0.05$) with increase in *M. oleifera* pod flours. The mineral, phytochemical and vitamin contents increased with the increase in the addition of *M. oleifera* pod flours. The result of the sensory properties showed that the flavour, texture, mouth feel, colour and overall acceptability of each product was significantly ($p<0.05$) enhanced by the addition of *M. oleifera* pod flour. The 100 % wheat bread recorded the lowest scores in all the major parameters evaluated while all the other samples were well accepted. Overall, there was an increase in the values of acceptability, appearance and flavour of the bread samples with increasing levels of *M. oleifera* pod flours.

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Introduction:-

Bread is a prominent staple food prepared from dough of flour and water. In many parts of the world, bread is made from the flour of wheat species. The wheat-flour dough is normally, cultured with yeast, which is the rising agent (Dewettinck *et al*., 2008). In Nigeria, however, bread is produced from other flour sources such as maize and

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cassava because of the paucity of wheat flour. These flours used in Nigeria for bread production have poor nutrient compositions and have affected negatively the nutritional status of a great number of Nigerians. This is because bread is highly consumed in Nigeria by many people owing to its quick way of production and the low cost at which it is sold to the consumer (David, 2006). Thus, in Nigeria, a good number of people who consume bread are deficient in one micronutrient or the other. These micronutrient deficiencies cause intellectual and developmental disabilities in many people and can be prevented (Hettiarachchi *et al*., 2006).

One of the ways of preventing these deficiencies and disabilities is to develop a bread production protocol with available plant materials that are rich in micronutrients. At present, plant like *Moringa oleifera* that is sustainable and has been found to be rich in both macro and micro-nutrients (Anwar *et al*., 2007), could be harnessed for this mission. Kalibbala *et al*. (2009) reported *M. oleifera* to be widely cultivated, rich in nutrients and even phytochemicals. The presence of phytochemicals such as flavonoids and phenol in *M. oleifera* can produce both antimicrobial and antioxidant effects (Kalibbala *et al*., 2009). The antioxidant effects of *M. oleifera* will take care of some of the additives used in commercial breads production as bread improvers and dough conditioners to improve the texture and volume but facilitate oxidation in human cells (Tenbergen, 1999). Thus, the use of *M. oleifera* pod in bread production done in this research will not only place millions of bread consumers on adequate nutrition but also on safe and sustainable diet.

Materials and Methods:-

Sample Sources

The moringa pod was obtained from the Department of Crop Science, University of Nigeria, Nsukka in Enugu State, Nigeria. The freshly harvested moringa pod was oven dried at a temperature of 60 ^oC for two days. This was ground to a fine particle size using Thomas-Wily Laboratory Mill (USA, Model no: 4).

Preparation of Bread Samples

The preparation of bread with dry *Moringa* pod flour was carried out as described by Campbell *et al*. (2001), except that the dry moringa pod flour were used to replace milk. The recipes used were 500 g wheat flour, 125 g butter, 12 g yeast, 60 g sugar, 125 g liquid, one teaspoonful of salt and dry moringa pod flour at 0 g, 20 g, 40 g and 60 g. The baking of the Moringa pod bread was carried out in the Bakery Unit of the Department of Home Economics Education, University of Nigeria, Nsukka using a hot air oven at a temperature of 220 ^oC for 20 minutes.

Proximate Analysis

Proximate analysis involved the determination of ash, crude protein, crude fat, crude fibre, moisture and carbohydrate contents. These were carried out using the standard method of analysis by the Association of Official Analytical Chemists (AOAC, 2000).

Determination of Vitamins

The thiamin and the riboflavincontents of the bread were determined as described by Onwuka (2005) while the pro-vitamin A was determined as described by Jakkutowicz *et al*. (1977). The analysis of vitamin C and vitamin E were carried outas described by Olokodona (2005) and Pearson (1976), respectively.

Mineral Determination

Phosphorus, calcium copper, zinc and iron were determined by directly reading in the Atomic absorption spectrophotometer (Model number AA-7000, ROM Version 1.01 and Serial number A30664700709) as described by Pearson (1976).

Determination of the Phytochemical Constituents

The phytochemicals determined in the bread samples produced were alkaloids, flavonoids, tannin, tepernoid and saponnins.

Determination of alkaloids

The alkaloids present in the bread were determined using the method described by Harbone (1973). Five gram of bread was measured into 100 ml beaker then 50 ml of 10 % acetic acid in ethanol was introduced into it and covered to stand for 4 hours after which it was filtered. Thereafter, 10 ml of ammonium hydroxide was introduced into the filtrate to precipitate the alkaloids. The precipitate was filtered, weighed, dried and the percentage alkaloid calculated as shown below.

$$\text{Alkaloid (\%)} = \frac{(\text{weight of the filter paper+Alkaloid}) - (\text{weight of filter paper only})}{\text{Weight of ground sample used}} \times \frac{100}{1}$$

Determination of Flavonoids

Flavonoids were determined using the method described by Boham and Kocipal (1974). Five gram of the bread was measured into flask and the flavonoids were extracted repeatedly with 100 ml of 80 % aqueous methanol at room temperature. Thereafter, it was filtered with Whatman filter paper No 43 (125 mm) and the filtrate transferred into weighed beaker and evaporated to dryness to get the weight of the flavonoids. The percentage flavonoids were calculated thus:

$$\text{Flavonoid (\%)} = \frac{(\text{weight of beaker+flavonoid}) - (\text{weight of beaker only})}{\text{Weight of sample used}} \times \frac{100}{1}$$

Determination of Tannins

Tannin was determined as described by Pearson (1976). One gram of the sample was weighed and 10.0 ml of distilled water added. This was shaken at 5 min interval for 30 min. The solution of the sample was centrifuged or filtered to get the extract. Then, 2.5 ml of the supernatant was transferred into a test-tube. This was added 1.0 ml Folin-Denis reagent, followed by 2.5 ml of saturated Na_2CO_3 solution. About 2.5 ml of standard tannic acid solution and blank were also prepared alongside with the test sample and the absorbance read after 90 min of incubation at room temperature. The percentage tannin was calculated thus:

$$\%\ \text{Tannin} = \text{An/As x C x } \frac{100}{W} x \frac{vf}{va}$$

where An = absorbance of test sample

As = absorbance of standard solution

C = Conc of standard solution

W = weight of sample used

Vf = total volume of extract

Va = volume of extract analyzed.

Determination of the Organoleptic Properties of the Formulated Breads

The sensory evaluation was carried out as described by Ihekoronye and Ngoddy (1985) using a twenty-man panel from the Department of Home Economics Education, University of Nigeria Nsukka. The members of the panel were instructed to indicate their preference of the test products. A nine-point hedonic scale, where 9 was the highest score while 1 was the lowest score for each characteristics such as flavour, texture, mouth feel, colour and overall acceptability of each product was selected for sensory evaluation of the products as described by Iwe (2002). The interpretation of the panelists' response showed the level of acceptability of the products.

Statistical Analysis:-

The data obtained were subjected to analysis of variance (ANOVA) using GenStat Release 10.3 DE (2011) statistical software.

Results and Discussion:-

The proximate compositions of the formulated breads are shown in Table 1. The moisture content of the bread produced with *M. oleifera* pod flours significantly ($p<0.05$) decreased with increase in the concentration of the Moringa pod flour at 20 g, 40 g and 60 g. This could be attributed to capability of the dry *M. oleifera* pod flours to absorb the free moisture in the bread. Similar result was observed by Mbaeyi and Anyanwu (2010) where dry mango pulp decreased moisture content of yoghurt formulated with solar dried bush mango pulp. The moisture content of the control sample which had no *M. oleifera* pod flour was therefore significantly ($p<0.05$) higher. The protein content of the bread produced with *M. oleifera* pod flours significantly ($p<0.05$) increased with increase in the concentration of the *Moringa* pod flour. This shows that the protein of bread can be complemented by that of *Moringa* pod flour. Harnessing complementary sources of protein in recipe development in meal preparation have been highly recommended for handling protein malnutrition (Healthy Water Living, 2007)

The ash content of the bread samples increased with the increase in the concentration of the *M. oleifera* pod flours used in bread formulation. This showed that the mineral composition of the bread increased with the addition of the *M. oleifera* pod flours since ash content is a measure of the total amount of minerals present within a food. There was no difference in fibre contents of the bread samples produced with *M. oleifera* pod flours. This agreed with the findings of Awaziem (2007) that fibre contents of locally produced foods hardly differ from their counterpart sample with varying compositions. The fat content of the bread produced with *M. oleifera* pod flours significantly ($p<0.05$) decreased with increase in the concentration of the *Moringa* pod flour. This shows that *Moringa* pod flour can lower the fat contents of bread samples. The bread produced with the highest concentration of *M. oleifera* pod flours is therefore low in calories and fat. Foods that contain low fat and oil are therefore good source of food supplement for patients with cardiac problems or at risks with lipid induced disorder (Egwim *et al*., 2011).

Table 1:-The effect of the *M. oleifera* pod flours on the proximate compositions of the formulated bread

Samples	**Moisture%**	**Ash %**	**Fat %**	**Fibre %**	**Protein %**	**Carbohydrate %**
20 g	32.6	1.0	2.70	0.5	2.84	61.36
40 g	32.0	1.2	1.95	0.5	2.91	62.44
60 g	22.4	1.2	1.75	0.5	3.06	71.09
Control	33.5	0.95	1.65	0.1	2.63	60.06
LSD(p<0.05)	1.88	0.24	0.34	0.23	1.88	1.90

The mineral contents of the bread samples are presented in Table 2. The bread contained variable amount of macro-nutrients (Ca, Mg and P) and trace elements (Fe, Zn and Cu). Bread samples containing 40 g and 60 g of *M. oleifera* pod flour were significantly ($p<0.05$) higher than the control (sample without *Moringa* pod flour) in magnesium and calcium compositions. Consumption of this type of bread containing adequate magnesium and calcium will be beneficial to both infant and aging population because of the utilization of these minerals in bone formation (Touyz, 2003). Breads rich in phosphorus are of importance in the synthesis of adenosine triphosphate (ATP). All the bread samples formulated with *Moringa* pod flour were significantly ($p<0.05$) higher than the control in iron concentration. The high contents of iron can be attributed to high level of iron in *Moringa* pod. Aslam *et al*. (2005) had reported moringa pod to be rich in iron content. Iron is very useful in haemoglobin synthesis in the body (American Society of Hematology, 2019). Since iron deficiency is very common among people who consume foods low in iron such as bread, fortifying breads with *Moringa* pod flour could serve as an alternative to iron supplements which are costly and even scarce in many Africa settings like Nigeria. Thus, this type of breads high in iron can be harnessed for treatment of iron deficiency anemia. National Heart, Lung, and Blood Institute (2019) also reported that eating iron supplements or intravenous iron therapy could be effective in the treatment of moderate iron-deficiency anemia. The bread sample formulated with 60 g moringa pod flour was significantly ($p<0.05$) higher than the control in copper concentration. This shows that bread formulation with 60 g of *Moringa* pod flour could serve well as iron supplements, since the association of copper and iron in foods had always been useful in enhancing iron metabolism and absorption in human body system (Collins *et al*., 2010). The concentration of zinc in the bread samples increased significantly ($p<0.05$) with the addition of the Moringa pod flour. The high contents of zinc in the formulated breads could be attributed to high level zinc in Moringa pod (Aslam *et al*., 2005). Since zinc deficiency is one of the serious problems of micronutrient deficiencies in the developing countries, production of cheap and commonly consumed foods such as breads rich in zinc will go a long way to ameliorate the negative effects in the lives of millions of people.

Table 2:-The effect of the *M. oleifera* pod flours on the mineral compositions of the formulated bread in g/Kg

Samples	Fe	Mg	Ca	Zn	Cu	P
20 g	115.8	0.5	0.06	4.18	1.93	1.00
40 g	231.6	1.02	1.34	8.36	3.86	2.01
60 g	347.4	1.52	2.0	12.54	5.80	3.04
Control	20.06	0.1	0.03	1.61	0.05	0.25
LSD(p<0.05)	1.88	0.54	1.00	1.67	1.33	1.34

The phytochemical contents of the formulated bread samples increased with the addition of the dry *M. oleifera* pod flour on the composite bread as represented in Table 3. The percentage alkaloids present in the bread samples increased proportionally with the increase in the *M. oleifera* pod flour. Plant alkaloids, like that of *M. oleifera* pod, have high medicinal values and can produce sedative and analgesic effects. Eze and Orjioke (2010) reported that plant alkaloids are useful raw material for the production of pain relieving drugs. Breads produced from *M. oleifera* pod flour can therefore double as a source of nutrients and a pain reliever when eaten. This will be beneficial to thousands of rural and urban poor dwellers in Nigeria who can only afford to eat breads with water after engaging in hard manual labours.The percentage flavonoid contents of the bread samples fortified with *M. oleifera* pod flour also increased with the increase in the concentration of the pod flour. Flavonoids are highly rich in antioxidants (Pietta, 2000) and could play a beneficial role in the reduction of oxidative stress (Botella *et al*., 2004). Since oxidative stress is the primary cause of aging and several degenerative diseases, such as atherosclerosis, cardiovascular disease, type II diabetes and cancer (Tsao, 2010), bread produced from *M. oleifera* pod flour can salvage millions of Nigerians who eat bread with carbonated drinks and other junk foods by its antioxidant actions**.** The tannin contents of all theformulated bread samples were low**.** Although tannins can inhibit protein absorption in the gastric system (Osagie, 1998), moderate levels of tannins in foods have healing effects in man. Tsao (2010) reported that little quantity of tannins in foods could have a therapeutic effect in human beings. Besides, the values of tannins in the bread samples are within the safe level for human consumption (Okonwu and Ugiomoh, 2015) and will, therefore, have no influence on the protein absorption in the gastric system**.** Therefore, the presence of the medicinal phytochemicals such as alkaloid, flavonoid and saponnin in the formulated bread samples might confer high medicinal values to the bread samples.

Table 3:-The effect of the *M. oleifera* pod flours on the percentage phytochemical compositions of the formulated bread

Samples	Alkaloid	Flavonoid	Tannin	Tepernoid	Saponin
20 g	0.62	1.04	0.007	0.94	0.29
40 g	1.24	2.08	0.009	1.88	0.61
60 g	1.86	3.12	0.011	2.82	0.89
Control	0.01	0.13	0.001	0.03	0.07
LSD ($p<0.05$)	0.14	1.33	0.004	0.034	0.13

The vitamin composition of the formulated bread samples increased with the addition of the dry *M. oleifera* pod flour on the composite bread except vitamin B_2 (Table 4). The bread samples fortified with *Moringa oleifera* pod flour contained high levels of B-carotene, vitamins B_1, C and E than the control. This shows that addition of the dry *M. oleifera* pod flour to bread can supplement its vitamin contents and make it delicious. Stanley *et al*. (2011) observed that foods with multivitamins are always delicious and provide good tastes, flavor and texture. Since, the bread samples produced with *M. oleifera* pod flours were significantly ($p<0.05$) higher than the control in all the vitamins analyzed, except in vitamin B_2, it could be deduced that the recipe for conventional bread production contain vitamin B_2 which was depleted with the addition of *M. oleifera* pod flours. Vitamins C and E as well as the B-carotene were significantly ($p<0.05$) higher in the fortified bread samples than the control and they have antioxidant actions. Okwulehie and Odunze (2004) reported foods high in these vitamins to have high antioxidative activity in human body system.

Table 4:-The effect of the *M. oleifera* pod flours on the vitamin compositions of the formulated bread

Samples	Vitamin B_1 (mg/100g)	Vitamin B_2 (mg/100g)	Vitamin C (mg/100g)	Vitamin E (mg/100g)	B-Carotene (IU)
20 g	0.7	5.25	26.25	22.50	500.00
40 g	0.7	3.75	26.88	24.50	444.45
60 g	1.0	0.75	29.37	27.0	722.23
Control	0.6	2.25	4.00	24.0	466.68
LSD ($p<0.05$)	0.30	0.42	1.88	2.49	3.77

The organoleptic scores of the formulated bread samples were shown in Table 5. The flavour of the formulated breads containing 60 g and 40 g were the most desirable with the sensory scores of 9.0. The organoleptic parameters scores of the formulated bread sample containing 60 g were statistically ($p<0.05$) higher than those of the control sample. Bread samples containing 60 g was therefore the most preferred of all the samples with the score of 9.0 for colour, aftertaste and overall acceptability. Meanwhile, the main quality factors for bread are the colour, taste and mouth feel (Fellows, 1997).

Table 5:-The Effect of the *M. oleifera* pod Flours on the Organoleptic Properties of the Formulated Bread

Samples	**Colour**	**Flavour**	**Mouth feel**	**Aftertaste**	**Overall Acceptability**
20 g	6.0	6.0	8.0	7.0	6.0

40 g	9.0	9.0	8.0	8.0	8.0
60 g	9.0	9.0	9.0	9.0	9.0
Control	5.0	4.0	7.0	6.0	4.0
LSD ($p<0.05$)	2.49	2.50	2.98	2.49	2.50

Values are mean of replicate treatment samples

Conclusion:-

The results of this study have shown that high nutrient containing breads could be developed from fortification of composite bread with *Moringa oleifera* pod flour. The breads contained reasonable amounts of macro-elements and trace elements that would be highly beneficial to human body. The *Moringa oleifera* pod flour also incorporated considerable amounts of medicinal phytochemicals into the bread at non-toxic levels for consumption. It is therefore evident that breads fortified with *M. oleifera* pod flour have great potentials of reducing food insecurity and malnutrition as well as providing prophylactic actions for the consumers, especially in developing nations. Fortification of composite breads with 60 g of *M. oleifera* pod flour for commercial consumption by both urban and rural dwellers, who eat bread daily, is highly recommended by this research.

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