ISSN: 2350-0328

International Journal of Advanced Research in Science, Engineering and Technology

Vol. 6, Issue 5 , May 2019

Production, Proximate, Phytochemical and Sensory Evaluation of *Moringa Oleifera* Enriched Bread at Various Levels of Enrichment

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ABSTRACT: This study investigated the effect of *moringa oleifera* leaf powder on enriched bread samples which were produced at various enrichment levels. Wheat flour and moringa powder were formulated at different percent (1%, 2%, 3% and 4%), to determine the best enrichment ratio. Proximate, phytochemical and sensory evaluation of the bread samples were determined using bread made from 100% wheat flour (0 % moringa) as control. The result showed a progressive drop ($p<0.05$) in the fibre content of enriched bread samples with decreasing level of enrichment. The energy value of the bread samples ranged from 392.98% to 394.73%. The sample with 1% moringa enrichment recorded the highest value of 394.73% and was significantly different ($p<0.05$) from the other samples. Result of the sensory evaluation using a nine-point Hedonic scale showed that bread samples had 5.10 to 7.65 in appearance, 5.45 to 6.85 for aroma, 5.35 to 7.75 in texture, 5.55 to 7.70 in mouth feel and 5.55 to 7.60 in general acceptability. Bread sample containing 3% moringa enrichment was most acceptable to the panelists.

KEY WORDS: Bread, Enrichment, Moringa, Proximate and Unenriched.

I.INTRODUCTION

This research focuses on the enrichment of bread using processed *Moringa oleifera* leaves in bid to improve the quality and functionality of breads produced in Nigeria. The enrichment became necessary due to the fact that bread which is an important staple in the country has as its main dietary principle starch, with little or no other diet to make it a nutritionally balanced food product. Therefore the use of *Moringa oleifera* as an enrichment agent will significantly improve the nutritional quality of the food product as well as enhance its functionality in prevention and management of some chronic health conditions. In other words, the dynamic product of this research will combine the nutritional characteristics and biological activities of *Moringa oleifera* to deliver optimum result as a functional food. Food enrichment is a process of adding vitamins or minerals to a processed food at levels specified by the law. 'Enriched' denotes that extra nutrients have been added to the food. These nutrients are added to replace the original vitamins and minerals that were lost during refining process [1]. Enrichment is also carried out to act as public health measure, to ensure appropriate vitamins and mineral composition of foods are adequate for special dietary purposes, and also prevent nutritional associated ailments.

II. RELATED WORK

Moringa oleifera is a rapidly growing food plant from the family moringaceae native to the sub-Himalayan area of India and Pakistan [2]. The plant which is highly nutritious is considered a complete food as all parts of the tree are useful and contains all the essential amino acids required for a healthy body. *Moringa oleifera* leaves have been used as an alternative food source to combat malnutrition, especially among children and infants [3]. It is a significant source of fats, proteins, beta-carotenes, vitamin C, Iron, potassium etc [4].Studies have shown its various biological activities to include anti-cardiovascular disease, anti-atherosclerotic, immune boosting, anti-oxidant, anti-microbial, anti-inflammatory properties, tumor suppressing effects and anti-cancer [4]. *Moringa oleifera* leaves can be eaten fresh, cooked or stored as dried powder for many months without refrigeration and reportedly without loss of nutritional value [5]. It is especially promising as a food source in the tropic because the tree is in full leaf at all seasons i.e. it is readily available. In African countries like Ghana and Nigeria, both fresh and dried leaves are included in meals. The dried leaves are processed into powder and added in food systems such as weaning foods such as cereal gruel (*Ogi*),

ISSN: 2350-0328
International Journal of Advanced Research in Science, Engineering and Technology
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soups, cake, yoghurt and cheese, biscuits and other pastry product. Bread as mentioned earlier is an important staple in the country and has as its main dietary principle starch, with little or no other diet to make it a nutritionally balanced food product. As a developing country, Nigeria is faced with numerous challenges such as under nutrition and prevalence of some chronic ailments. Tackling these challenges becomes imperative using raw materials that are readily available locally. The use of *Moringa oleifera* in the enrichment of bread is borne out of the need to create a product capable of improving the nutritional and well-being of a greater number of Nigerians that consume bread daily, by utilizing the nutritional characteristics and biological activities of the well-known food plant *Moringa oleifera.* The objectives of this study are to produce bread samples enriched with various percentages of processed *Moringa oleifera* leaves and to determine the best enrichment ratio. To carry out other analysis on the bread sample like proximate, phytochemical and sensory evaluation.

III. MATERIALS AND METHODS

This study was undertaken between the months of February and October 2018 in the Dry Food Workshop of the Food Technology Department Akanu Ibiam Federal Polytechnic Unwana, Afikpo, Ebonyi State; Biochemistry Laboratory of the National Root Crops Research Institute Umudike, and the Michael Okpara University Umudike both in Abia State. Nigeria. The materials used for the production of bread samples namely wheat flour, margarine, yeast, sugar, salt, nutmeg and improver, were procured from Eke market, Afikpo, Ebonyi State.

VI. COLLECTION OF PLANT MATERIAL AND PROCESSING OF *MORINGA OLEIFERA*

The leaves of *Moringa oleifera* were collected from the botanical gardens of Michael Okpara University Umudike, Umuahia, Abia State. Plant material was authenticated at Michael Okpara University Umudike, Umuahia, Abia State. The *Moringa oleifera* leaves were first sorted to remove dirt and other foreign materials. The leaflets were striped and washed properly with clean portable water and then spread on a wooden tray to drip dry. The leaflets were further blanched at 100°C using steam for about 15 seconds. The leaves were then dried in a drying cabinet maintained at a temperature of 50°C. The dried *Moringa oleifera* leave samples were then milled and sieved at least two times using 0.5mm sized mesh screen till fine flour was obtained.

V. PRODUCTION OF BREAD ENRICHED WITH PROCESSED *MORINGA OLEIFERA* LEAVES

Production of bread samples enriched with various percentages of processed *Moringa oleifera* leaves was carried out at the Dry Food Workshop of the Food Technology Department Akanu Ibiam Federal Polytechnic Unwana. The raw materials namely wheat flour, margarine, yeast, sugar, salt, nutmeg, processed *Moringa oleifera* flour, bread improver and water were all measured as shown in table 1 and mixed. For the production of 1% *Moringa oleifera* bread sample, 2.5g of *Moringa* powder to 247.5g of wheat flour was used to produced enriched bread, while for 2%, 3% and 4% enriched bread samples, 5g, 7.5g and 10g of *Moringa* powder to 245g, 242.5g and 240g respectively were used. The other raw materials were left constant for each of the bread samples as indicated in table 1. The no time dough method [6] was employed, in preparing the various dough. The dough was then baked in an oven reheated at 200^{o}C for 15minutes and afterwards maintained at a temperature of 160^{o}C. The total baking time was 1 hour, 15 minutes. The bread produced was allowed to cool, packaged and labelled.

Table 1: Level of enrichment of bread sample

Level of Enrichment	Wheat flour	*Moringa oleifera* flour	Yeast	Margarine	Sugar	Salt	Nutmeg	Improver	Water
5%	245.5g	2.5g	5g	12g	25g	0.5g	0.3g	0.5g	275ml
10%	245g	5g	5g	12g	25g	05g	0.3g	0.5g	275ml
15%	242.5g	7.5g	5g	12g	25g	0.5g	0.3g	0.5g	275ml
20%	240g	10g	5g	12g	25g	0.5g	0.3g	0.5g	275ml

ISSN: 2350-0328
International Journal of Advanced Research in Science, Engineering and Technology
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A. PROXIMATE ANALYSIS

a) Protein content determination

Protein content was determined by adopting kjedahl method of [7]. About 2g of the sample was used for the analysis. Total nitrogen was calculated and multiplied by a conversion factor of 6.25 to obtain the crude protein.

b) Determination of Carbohydrate content

The method for carbohydrate determination was carried out as described by [8]. The difference was calculated as the Nitrogen Free Extract (NFE).

c) Determination of Moisture content

Moisture content was determined by adopting the method of [7]. 2g of bread sample was used in the analysis while the gravimetric method was adopted.

d) Determination of Ash content

The ash content determination was carried out on the bread samples according to the method of [7].

e) Determination of Crude fibre

Crude fibre was determined on the bread samples by adopting the method of [7].

f) Determination of Fat content

The fat content was determined by using the continuous solvent extraction method according to [7].

g) Determination of Dry matter

The dry matter content (in %) of bread samples was determined by subtracting the moisture content from 100% according to the method of [7].

h) Energy value

Energy value was determined by calculations according to the method of [8].

B. PHYTOCHEMICAL ANALYSIS

Determination of Phytate

Phytate content of the bread samples were determined by adopting the method of [9].

a) Determination of Tannin

The tannin content was determined on 1g of sample using the Folin Denis spectrophotometric method described by [9].

b) Determination of Flavonoid

The flavonoid content was determined on 5g of the bread sample using the acid hydrolysis gravimetric method described by [10].

c) Determination of Saponin

The saponin content was determined on 5g of bread sample using the method described by [11].

d) Determination of Oxalate

The oxalate content was determined on 2g of the bread sample using a method adopted by [10].

e) Determination of HCN Hydrogen Cyanide

The Hydrogen cyanide content was determined on 5g of bread sample using a method adopted by [10].

C. SENSORY EVALUATION OF ENRICHED BREAD SAMPLES

Sensory evaluation of coded bread samples namely A, B, C, D, E with various levels of enrichment (except sample E which was unenriched) was carried out using 20 semi trained panelist drawn from staff of National Roots Crop Research Institute Umudike. Parameters evaluated include Appearance, Aroma, Texture, Mouth feel and General acceptability. A 9 point hedonic scale ranging from 9=extremely liked to 1= extremely disliked was used for the scoring. Also the mean scores were subjected to Analysis of variance (ANOVA) and mean separated using Turkey test at 95% confidence level.

ISSN: 2350-0328

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D. STATISTICAL ANALYSIS

Replicate determinations were carried out on every parameter and the results presented as mean and standard error of mean.

VI. RESULTS AND DISCUSSION

The result of the proximate analysis of the bread samples enriched with moringa is presented in Table 2. Dry matter of bread samples ranged between 87.28 to 88.05%. Bread sample with 4% moringer powder recorded the highest at 88.05% and was significantly different (P<0.05) from the other samples. The dry matter of the samples showed a progressive drop (P<0.05) with decreasing level of enrichment. However the unenriched bread sample (0% moringa) had the lowest dry matter at 87.28. The dry matter of any food product provides a measure of the amount of the particular food that is required to supply a given amount of nutrients. Increase or decrease in the value of dry matter is directly proportional to the availability of nutrients. The moisture content of the bread samples ranged between 11.95 to 12.72%. The unenriched bread sample (0% moringa) had a significant high moisture content than the other samples. The result was within the values considered acceptable for dried foods and other dried samples which is in accordance with the findings of [5].

The moisture content of food sample is an index of stability and the result obeserved in the moisture content may be due to the difference in the processing method. Results obtained from the crude protein of the bread samples showed that the 4% moringa enriched sample recorded 12.89% of protein, which was significantly different (P<0.05) from the other samples. This indicates that this level of moringa powder can be used in food formulation. The unenriched (0% moringa) bread sample recorded the lowest protein at 12.52%. The result was in accordance with the findings of [12;5] The unenriched (0% moringa) bread sample recorded the highest value interms of fat content recorded at 12.16% and was significantly different (P<0.05) from the enriched samples. The fat content of the enriched bread samples showed a progressive drop (P<0.05) with increasing level of enrichment leading to speculations that increasing levels of moringa powder in the bread samples lowered their fat content. High fat content in meals promotes high plasma cholesterol concentration which increases the risk of thrombosis [13]. How ever the levels obtained in the research was within acceptable safe limits. There was significant difference (P<0.05) in the crude fibre content of the bread samples. The result showed a progressive drop (P<0.05) in the fibre content of enriched bread samples with decreasing level of enrichment. The unenriched (0% moringa) bread sample had the lowest crude fibre content of 1.68%. The consumption of dietary fibre such as those contained in moringa may aid in reducing blood cholesterol which in turn reduces the risk of coronary heart disease [13].

The ash content of the samples showed that the 4% moringa enriched bread sample recorded the highest ash content of 2.67% and was significantly different (P<0.05) from the other samples.The result showed a progressive drop (P<0.05) with decreasing level of enrichment and were within reported values. The result showed there was no significat (P<0.05) difference in the carbohydrate content of the bread samples and was within reported values.

The energy value of the bread samples ranged from 392.98% to 394.73%. The sample with 1% moringa enrichment recorded the highest value of 394.73% and was significantly different (P<0.05) from the other samples. There was no significant difference (P<0.05) in energy content of bread samples containing 2% and 0% moringa enrichment, however they were significantly different from bread samples with 3% and 4% moringa enrichment. The results were within reported values.

Table 2: Result of proximate analysis of enriched bread samples

Parameter %	Samples				
	A (1% moringa)	**B (2% moringa)**	**C (3% moringa)**	**D (4% moringa)**	**E(control)**
DM	87.67 ± 0.01^{d}	87.79 ± 0.01^{c}	87.93 ± 0.01^{b}	88.05 ± 0.01^{a}	87.28 ± 0.02^{e}
MC	12.33 ± 0.01^{b}	12.21 ± 0.01^{c}	12.07 ± 0.01^{d}	11.95 ± 0.01^{e}	12.72 ± 0.02^{a}
CP	12.62 ± 0.01^{d}	12.68 ± 0.01^{c}	12.74 ± 0.01^{b}	12.89 ± 0.01^{a}	12.52 ± 0.01^{e}
FAT	12.09 ± 0.01^{b}	12.05 ± 0.01^{c}	11.96 ± 0.01^{d}	11.90 ± 0.01^{e}	12.16 ± 0.02^{a}
CF	1.77 ± 0.01^{d}	1.84 ± 0.01^{c}	1.91 ± 0.01^{b}	2.01 ± 0.01^{a}	1.68 ± 0.02^{e}
ASH	2.32 ± 0.01^{d}	2.41 ± 0.01^{c}	2.53 ± 0.01^{b}	2.67 ± 0.01^{a}	2.22 ± 0.02^{e}
CHO	58.87 ± 0.01^{a}	58.81 ± 0.01^{a}	58.79 ± 0.01^{a}	58.58 ± 0.01^{a}	56.19 ± 2.49^{a}
EV	394.73 ± 0.13^{a}	394.41 ± 0.09^{b}	393.76 ± 0.01^{c}	392.98 ± 0.01^{d}	394.32 ± 0.02^{a}

Mean scores with the same super scripts in the same row are not significantly different (p<0.05)

ISSN: 2350-0328

International Journal of Advanced Research in Science, Engineering and Technology

Vol. 6, Issue 5 , May 2019

Key DM= Dry matter; MC=Moisture; CP=Crude protein; FAT; CF=Crude fiber; ASH; CHO=Carbohydrate; EV= Energy value. All measured as mg/100g

VII. PHYTOCHEMICALS ANALYSIS

The result of the phytochemical content of bread sample is shown in table 3.

a).Phytate

The phytate content of bread samples varied between 0.02 to 1.15 mg/100g. The result indicated that there was a progressive drop ($P<0.05$) in the phytate content of the sample with decreasing level of moringa enrichment. The 4% moringa enriched bread sample at 1.15mg/100g was significantly different ($P<0.05$) from the other samples. There was no significant difference ($P<0.05$) in the phytate content of bread samples containing 1% and 0% (unenriched) moringa enrichment. Phytate in food products are well known for the metal thelation which affects their bioavailiability [14]. However the values obtained in this study were within acceptable limits.

b). Tannin

Result of the tannin content of bread samples showed a progressive drop ($P<0.05$) from the other samples. The unenriched sample (0% moringa) at 0.03 mg/100g recorded the lowest tannin content. Tannins in food are often responsible for colour changes which accounted for the dark brown coloration during processing of bread samples with high level of moringa enrichment. Also tannin is a good antioxidant and may inhibit carcinogen activation and cancer promotion [15].

c). Flavonoids

The flavonoid content of the bread samples varied between 0.02 to 0.03gm/100g. The 4% moringa enriched sample at 0.13gm/100g recorded the highest value and was significantly different ($P<0.05$) from the other samples. There was no significant different ($P<0.05$) in the flavonoid content of bread samples containing 1%, 2%, and 0% (unenriched) moringa enrichment. The result showed that the moringa enriched bread samples are good source of flavonoid which performs several important functions in the body such as antioxidants, scavenging carcinogens, inhibiting cell proliferation and binding to nitrates in the stomach, thereby preventing their conversion to nitrosamines [15].

d).Saponin

The saponin content of bread samples showed a progressive drop ($P<0.05$) with decreasing level of moringa enrichment. However there was no significant difference ($P<0.05$) between bread samples with 1% and 0% (unenriched) moringa enrichment. It was reported that saponins may prevent cancer cells from multiplying by interfering with DNA replication and also stimulate immune response [13].

e).Oxalate

The result obtained from the oxalate content of bread samples indicated that the 4% moringa enriched sample recorded the highest value at 0.19mg/100g and was significantly different ($P < 0.05$). There was no significant different ($P<0.05$) between bread samples containing 2% and 3% moringa on one hand and bread sample containing 1% and 0% (unenriched) moringa on the other hand. Oxalates in food tend to bind minerals affecting their bioavailability [15].

f).Hydrogen cyanide (HCN)

The hydrogen cyanide content of bread samples ranged between 0.2 to 0.5. The result showed no significant difference ($P<0.05$) between the bread samples. The values obtained for the bread samples were within FAO acceptable safe limits [16].

Table 3: Result of phytochemical content of enriched bread samples

Parameter %	Samples				
	A (1% moringa)	**B (2% moringa)**	**C (3% moringa)**	**D (4% moringa)**	**E(control)**
Phytate	0.03 ± 0.01^{d}	0.07 ± 0.01^{c}	0.11 ± 0.01^{b}	0.15 ± 0.01^{a}	0.02 ± 0.01^{e}
Tan	0.07 ± 0.01^{d}	0.10 ± 0.01^{c}	0.15 ± 0.01^{b}	0.19 ± 0.01^{a}	0.03 ± 0.01^{e}
Fla	0.03 ± 0.01^{c}	0.04 ± 0.02^{c}	0.09 ± 0.01^{b}	0.13 ± 0.01^{a}	0.02 ± 0.01^{c}
Sap	0.38 ± 0.01^{d}	0.42 ± 0.01^{c}	0.47 ± 0.01^{b}	0.51 ± 0.01^{a}	0.36 ± 0.01^{e}
Ox	0.09 ± 0.01^{c}	0.13 ± 0.01^{b}	0.14 ± 0.02^{b}	0.19 ± 0.01^{a}	0.08 ± 0.01^{e}
HCN	0.02 ± 0.01^{a}	0.02 ± 0.01^{a}	0.04 ± 0.01^{a}	0.05 ± 0.01^{a}	0.02 ± 0.01^{a}

ISSN: 2350-0328

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Key

Phytate; Tan=Tannin; Fla=Flavonoid; Sap=Saponin; Ox=Oxalate; HCN=Hydrogen cyanide. All measured as mg/100g

VIII. RESULTS OF SENSORY EVALUATION

The result of the sensory evaluation of bread samples is shown in table 4. From the table it was seen that the appearance of freshly baked bread samples ranged from 5.10 to 7.65. Sample E (unenriched bread) scored highest at 7.65 and was significantly different (P<0.05) from the other samples. This could be attributed to the presence of Moringa powder which tainted the bright colour of the wheat flour during baking. In terms of aroma, the samples were all accepted and were not significantly different ((P<0.05). Analysis of texture indicated that the samples were fairly accepted with scores ranging from 5.35 to 7.75. However sample E (unenriched bread) which scored highest at 7.75, was different (P<0.05) from the other samples. The texture of the enriched bread samples looked a bit spongy which may be as a result of the method employed in mixing the bread dough. Evaluation of mouth feel, indicated significant difference (P<0.05) among the bread samples with sample E (unenriched bread) recording the highest score at 7.70. Also sample E (unenriched bread) was significantly different (P<0.05) from the other sample. The result on the general acceptability showed that the bread samples were fairly acceptable to the panelists as shown by their scores. Sample E scored highest at 7.60 and was significantly different (P<0.05) from other samples. Also bread samples A and C containing 1% and 3% moringa enrichment respectively were slightly different (P<0.05) in their acceptability from samples B and D, containing 2% and 4% moringa enrichment. This result shows that with little improvement in the method of preparation, the enriched bread samples can compete favorably with their commercial counterparts.

Table 4: Result of mean sensory scores of enriched bread samples

Sample	Appearance	Aroma	Texture	Mouth feel	General Acceptability
A (1% moringa)	6.60 ± 1.43^{ab}	6.65 ± 1.31^{a}	6.00 ± 1.65^{b}	5.60 ± 1.76^{b}	6.75 ± 1.33^{ab}
B (2% moringa)	6.10 ± 1.80^{b}	5.95 ± 1.57^{a}	5.35 ± 1.42^{b}	5.25 ± 2.05^{b}	5.70 ± 1.30^{b}
C (3% moringa)	5.60 ± 1.67^{b}	6.10 ± 171^{a}	6.65 ± 1.66^{ab}	6.10 ± 1.65^{ab}	6.30 ± 1.72^{ab}
D (1% moringa)	5.10 ± 1.94^{b}	5.45 ± 1.70^{a}	5.95 ± 1.82^{b}	5.55 ± 2.04^{b}	5.55 ± 1.73^{b}
E (1% moringa)	7.65 ± 1.66^{a}	6.85 ± 1.84^{a}	7.75 ± 1.16^{a}	7.70 ± 1.59^{a}	7.60 ± 1.82^{a}

Mean scores with the same super scripts in the same column are not significantly different (p<0.05)

IX. CONCLUSION

Results show that processed moringa leave powder (*moringa oleifera*) could be used in the production of acceptable and palatable bread products. The enrichment of bread products could be best achieved at moringa levels of 2% to 4% also it was shown that the use of moringa leave powder as an enrichment agent at these levels significantly improved the nutritional quality of the bread samples as well as their functionality in providing the essential nutrients such as some phytochemicals needed for the prevention and management of some health conditions. Therefore the use of moringa oleifera in bread enrichment is effective in producing a nutritionally balanced food product.

X. ACKNOWLEDGEMENT

We want to acknowledge the sole sponsorship of this work, Tertiary Education Trust Fund towards the successful completion. Also the Akanu Ibiam Federal Polytechnic, Unwana Institute Based Research Committee for their effort.

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ISSN: 2350-0328

International Journal of Advanced Research in Science, Engineering and Technology

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