



## Physicochemical properties of fermented and non-fermented rice bran flour and its utilization in bread making

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### ABSTRACT

Rice bran was subjected to natural fermentation for 4 days and investigated for its physicochemical, antioxidant, phytochemical, functional properties and mineral content. In addition, the fermented and non-fermented rice bran flours were supplemented with wheat flour in several ratios for baking bread. The proximate composition results obtained showed that fermented rice bran flour possessed protein content of 5.68%, fibre (37.8%), fat (2.97%) and carbohydrate (31.06%). It was observed that the total phenol content for non-fermented rice bran flour (362.69 GAE  $\mu\text{g/ml}$ ) was higher than fermented bran flour (359.15 GAE  $\mu\text{g/ml}$ ). However, the ability of the samples to scavenge 1,1-di phenyl-2-picrylhydrazyl (DPPH) radical was higher in fermented flour (57.0mg/ml) than in non-fermented sample (55.0 mg/ml). The result of the qualitative analysis of the phytochemical screening revealed that only saponin was present in the rice bran flours. The mineral composition of the non-fermented and fermented rice bran flours revealed that the flours possessed magnesium (0.19%-0.18%), sodium (0.06%-0.17%), calcium (0.08%-0.13%) and potassium (0.03%-0.02%). The physical properties of bread sample at 20% rice bran flour supplementation showed that there was no significant difference between loaf volume of the wheat flour bread and non-fermented rice bran bread ( $360\text{cm}^3$ ) while there was significant difference in the loaf volume of fermented rice bran bread ( $281\text{cm}^3$ ). It was observed that the fermented rice bran bread had the highest fibre content (1.83%), followed by non-fermented rice bran bread (1.37%) and wheat flour bread (0.95%). The sensory analysis revealed that both the fermented and non-fermented rice bran breads were scored above average in all sensory parameters although wheat flour bread was preferred.

**Keywords:** antioxidant activity; bread; fermented and non-fermented rice bran flour; proximate composition; sensory score; wheat flour.

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## INTRODUCTION

Rice bran is an underutilized byproduct of the rice milling industry which has potential as a rich source of valuable health promoting compounds. It contains a significant amount of nutraceutical compounds and approximately 4% unsaponifiables, mainly comprised of naturally occurring antioxidant such as tocopherols, tocotrienols and oryzanol (Ju and Vali, 2005). Rice bran proteins are of high nutritional value (Kennedy and Burlingame, 2003) and hypoallergenic (Tsuji *et al.*, 2001). Thus rice bran extract has potential application as health promoting ingredient in functional foods. It has been used as effective natural preservatives in various food system such as meat, refined oil and dough. Rice bran had higher total phenolics and flavonoid concentration than blueberry and broccoli (Min *et al.*, 2011).

Rice bran has a long history of use in livestock feed. The supplementation of rice bran with wheat flour is evident across a number of markets, having found application in bread, fried and dough (Chotimarkorn and Silalai, 2007; Huet *et al.*, 2009). However, there has been little or no work reported on using fermented rice bran as supplement in making bread. Therefore, we carried out this work to determine the physicochemical, functional, antioxidant, phytochemical properties and mineral content of raw and fermented rice bran. In addition, the acceptability of bread supplemented with raw and fermented rice bran was evaluated

## MATERIALS AND METHOD

**Sample collection and Preparation:** Rice bran was collected from Igbimo rice milling industry, Ekiti State, Nigeria. The method of Odunfa (1985) was slightly modified and employed for fermentation. The rice bran was cleaned to remove the dirt, stone and other extraneous materials. Preliminary cleaning of the bran was carried out by washing with clean water. Thereafter rice bran was boiled for one hour to soften it. After boiling it was drained and allowed to cool and divided into two. One portion was wrapped in clean banana leaf and kept at ambient temperature for four days for fermentation to occur while the other fraction was left unfermented. Both the fermented and unfermented samples were dried in the oven at 50°C, milled into flour and packaged in an air tight container for further analyses

**Determination of total titratable acidity of rice bran flours:** Total titratable acidity was determined according to the method of AOAC (2005). Twenty ml filtrate obtained from 2 g of flour samples was dissolved in 20 ml distilled water was titrated against 0.1 M NaOH using phenolphthalein as indicator. The titre value was then used to calculate the titratable acidity as percentage lactic acid.

**Determination of pH of rice bran flours:** Five grams of samples were separately weighed into beaker, mixed with 10 ml of distilled water and pH was carried out using standardized pH meter (Hanna instruments, HI 8314 membrane pH meter).

**Determination of proximate composition of rice bran flour and composite bread:** The proximate composition of the samples were determined by using the method of Association of Official Analytical Chemists (AOAC, 2005).

**Determination of mineral content of rice bran flours:** The dry ashing technique as described by Oshodi and Fagbemi (1992) was used for the determination of mineral composition.

**Determination of functional properties of rice bran flours**

**Bulk density** was determined using the method described by Monterio and Prakash (1994). **Water absorption capacity** (Rodriguez *et al.* 2005) and **oil absorption capacity** (Lin and Zayas (1987).

**Phytochemical screening:** Qualitative tests were carried out on the aqueous extract and on the powdered specimen using standard procedures to identify the constituents as described by Chitravadivu *et al.* (2009). These include; tannin, phlobatannin, saponin, flavonoids, steroid, terpenoid, anthraquinone and xanthoprotein.

**Determination of Total Phenol Content:** The total phenol content was determined according to the method of Singleton *et al.* (1999). Samples were oxidized with 2.5 ml of 10% Folin-Ciocalteu assay reagent (v/v) and neutralized by 2.0 ml of 7.5% sodium carbonate. The reaction mixture was incubated for 90 min at 45°C and the absorbance was measured at 765 nm in the spectrophotometer. The total phenol content of the extracts was expressed as milligram of gallic acid equivalent (GAE) per gram dry weight of sample.

**Determination of free radical scavenging capacity of rice bran samples (DPPH):** The free radical scavenging ability of samples against 1,1-diphenyl-2-picrylhydrazyl was evaluated as described by Hutadilok-Towatana *et al.* (2006). One milliliter of the sample extract was mixed with 1 ml, 0.4 mM methanol solution containing DPPH radicals, the mixture was left in the dark for 30 min and the absorbance was taken at 516 nm. The percentage antioxidant activity was calculated using:

$$(AA\%) = \frac{\text{absorbance of control} - \text{absorbance of extract}}{\text{absorbance of control}} \times 100$$

**Production of bread:** The method employed in the baking procedure is the straight dough fermentation process following the modified method of American Association of Cereal Chemists (AACC, 2000). The flour blends (100% wheat flour, 95% wheat flour:5% rice bran flour, 90% wheat flour:10% rice bran flour, 85% wheat flour:15% rice bran flour and 80% wheat flour: 20% rice bran flour) were mixed separately with equal amount of sugar, fat, salt and yeast. The fermented and non-fermented rice bran flours were used respectively. Bread characteristics were evaluated by measuring the loaf volume, specific loaf volume, proximate composition and the organoleptic characteristics. Loaf volume was measured 50 min after loaves were removed from the oven by using the rapeseed displacement method (guinea corn was used in place of rapeseed) as modified by Giami *et al.* (2004). The bread loaves supplemented with either rice bran or fermented rice bran



were evaluated by sensory panels from the University Community. Multiple comparison tests were employed using 100% wheat bread as the references.

## RESULTS AND DISCUSSION

The results obtained from physicochemical properties of non-fermented and fermented rice bran investigated are shown in Table 1. It was observed that there were significant differences in the values of protein (6.36%; 5.68%), fibre (35.54%; 37.80%), fat (3.06%; 2.97%), and carbohydrate (34.42%; 31.06%) for the non-fermented and fermented rice bran flour respectively. Results revealed that rice bran flours were higher in fibre content than wheat flour (0.68%), cassava flour (1.88%), malted soybean flour (5.18%) (Nwosu *et al.*, 2014) and plantain flour (3.50) (Mepha *et al.*, 2007). The protein content of rice bran flour was found to be higher than that of cassava and plantain flours but lower in carbohydrate. This result may be an indication that rice bran flour may be a better composite flour than cassava and plantain flours when higher values of protein and fibre are preferred than carbohydrate.

The mineral content and functional properties of rice bran flours are shown on Table 2. The flours possessed low mineral content of magnesium (0.19%-0.18%), sodium (0.06%-0.17%), calcium (0.08%-0.13%) and potassium (0.03%-0.02%). Rice bran is a good source of minerals and trace minerals and it is much superior to other cereals (Rao, 2000). Results of the functional properties of non-fermented and fermented rice bran flours showed that there were no significant difference between the values of bulk density (2.87g/ml; 2.67g/ml), water absorption capacity (2.39g/ml; 2.66g/ml) and oil absorption capacity (2.50g/ml; 2.26g/ml). The values obtained were higher than those reported for wheat, soybean and cassava flours except for the water absorption capacity where soybean flour was higher than rice bran flour (Nwosu *et al.*, 2014). Proteins absorb fully one to two times their weights in water, while starch absorbs only about one-quarter to one-half its weight in water (Iheagwara, 2012).

Total phenol content and ability to scavenge free radicals of methanolic extract of rice bran flours ranged from 395.15 GAE $\mu$ g/ml-362.69 GAE $\mu$ g/ml and 57.0 mg/ml-55.0 mg/ml for fermented and non-fermented rice bran flours respectively (Table 3). Rice bran contains an array of phenolic compounds (Hudson *et al.*, 2000). These compounds are purported antioxidant, anti-inflammatory and anti-cancer agents, and thus may protect against a number of chronic diseases. Qualitative phytochemical screening carried out on rice bran flour in this study revealed that only saponin was detected (result not shown). Full fat rice bran has been reported to be a rich source of a number of bioactive phytochemicals which include dietary fibre, phytosterols, gamma oryzanol, tocopherol and tocotrienols, ferulic acid and other phenolic compounds (Rao, 2000).

The protein content of bread from 100% wheat flour (17.3%) was found to be significantly higher than those of 80% wheat flour :20% fermented rice bran flour (16.9%) and 80% wheat flour:20% non-fermented rice bran flour (15.4%) (Table 4). However, bread samples produced from wheat flour supplemented with 20% rice bran flours were higher in fibre (1.37%-1.83%) than 100% wheat flour (0.95%). Furthermore, 20% rice bran-wheat flour breads were lower in moisture (29.56%-33.05%) than 100% wheat flour bread (34.01%). The increased fibre content of composite breads can offer several health benefits, as it aids 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). In addition, dietary fibre plays a significant role in the prevention of several diseases which include cardiovascular diseases, diverticulosis, constipation, irritable colon, cancer and diabetes (Elleuch *et al.*, 2011).

It was observed from Table 5 that significant differences in specific volume of breads were obtained when 20% rice bran flours were incorporated into wheat flour. Similar reductions in loaf volume were recorded as a result of blending wheat flour with more than 5% plantain (Mepha *et al.*, 2007). This may be attributed to lower levels of gluten network in the dough and consequently less ability of the dough to rise due to the weaker cell wall structure.

The sensory evaluation of bread samples containing different level of non-fermented rice bran or fermented rice bran flour substitution as compared to the control is shown in Table 6. The control (100% wheat bread) was significantly scored higher than the composite breads in colour, aroma, texture and overall acceptability. However, there was no significant difference in the taste. The results showed that rice bran composite breads were scored above average in overall acceptability.

## CONCLUSION

Non-fermented and fermented rice bran flour exhibited high crude fibre and antioxidant property which may enhance their potential application as health promoting ingredients in functional foods. From the outcome of sensory evaluation, composite bread from non-fermented and fermented rice bran flour (5%-20%) were acceptable.

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**Table 1: Physicochemical properties of non-fermented and fermented rice bran flour**

Parameters	Non fermented rice bran flour	Fermented rice bran flour
Total titratable acidity (%)	0.06±0.01 <sup>a</sup>	0.08±0.01 <sup>a</sup>
pH	5.20±0.02 <sup>a</sup>	5.60±0.15 <sup>a</sup>
Moisture content (%)	6.70±0.01 <sup>b</sup>	8.60±0.11 <sup>a</sup>
Ash content (%)	13.91±0.01 <sup>a</sup>	13.86±0.01 <sup>a</sup>
Fibre (%)	35.54±0.02 <sup>b</sup>	37.80±0.10 <sup>a</sup>
Fat (%)	3.06±0.02 <sup>a</sup>	2.97±0.01 <sup>b</sup>
Protein (%)	6.36±0.02 <sup>a</sup>	5.68±0.01 <sup>b</sup>
Carbohydrate (%)	34.42±0.07 <sup>a</sup>	31.06±0.02 <sup>b</sup>
Energy (kcal)	2.17±0.07 <sup>a</sup>	2.08±0.06 <sup>a</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same row are significantly different (P < 0.05).

**Table 2: Mineral composition and functional properties of non-fermented and fermented rice bran flour**

Parameters	Non fermented rice bran flour	Fermented rice bran flour
Potassium (%)	0.03±0.01 <sup>a</sup>	0.02±0.01 <sup>a</sup>
Sodium (%)	0.06±0.01 <sup>a</sup>	0.17±0.01 <sup>b</sup>
Calcium (%)	0.08±0.01 <sup>a</sup>	0.13±0.00 <sup>a</sup>
Magnesium	0.19±0.00 <sup>a</sup>	0.18±0.00 <sup>a</sup>
Bulk density (g/ml)	2.87±0.01 <sup>a</sup>	2.67±0.02 <sup>a</sup>
Water absorption capacity (g/ml)	2.39±0.01 <sup>a</sup>	2.66±0.01 <sup>a</sup>
Oil absorption capacity (g/ml)	2.50±0.01 <sup>a</sup>	2.26±0.01 <sup>a</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same row are significantly different (P < 0.05).

**Table 3: Antioxidant activity of methanolic extract of non-fermented and fermented rice bran flour**

Parameters	Non fermented rice bran flour	Fermented rice bran flour
Total phenolic content	362.69±7.74 <sup>a</sup> (GAEµg/ml)	359.15±4.26 <sup>b</sup>
DPPH (IC <sub>50</sub> mg/ml)	55.0±0.01 <sup>b</sup>	57.0±0.01 <sup>a</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same row are significantly different (P < 0.05).

**Table 4: Proximate analysis of bread supplemented with either 20% non-fermented or fermented rice bran**

Parameters (%)	WFB	NFRB	FRBB
Moisture content	34.01±0.01 <sup>a</sup>	33.05±0.01 <sup>b</sup>	29.56±0.01 <sup>c</sup>
Ash content	0.85±0.01 <sup>c</sup>	1.96±0.01 <sup>b</sup>	2.33±0.01 <sup>a</sup>
Fibre	0.95±0.01 <sup>c</sup>	1.37±0.01 <sup>b</sup>	1.83±0.01 <sup>a</sup>
Fat	3.94±0.01 <sup>c</sup>	4.61±0.01 <sup>b</sup>	4.75±0.01 <sup>a</sup>
Protein	17.3±0.01 <sup>a</sup>	15.4±0.01 <sup>c</sup>	16.9±0.01 <sup>b</sup>
Carbohydrate	42.41±0.09 <sup>c</sup>	44.25±0.56 <sup>a</sup>	43.10±1.28 <sup>ab</sup>
Energy (KJ/100)	1114.8±0.79 <sup>a</sup>	1234.39±0.79 <sup>a</sup>	1021.07±0.25 <sup>a</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same row are significantly different (P < 0.05).

WFB=Wheat flour bread

NFRB= Non fermented rice bran bread

FRBB= Fermented rice bran bread



**Table 5: Physical properties of bread supplement with either 20% non-fermented or fermented rice bran**

Parameters	WFB	NFRB	FRBB
Loaf volume (cm <sup>3</sup> )	360.00±1.00 <sup>a</sup>	360.00±1.00 <sup>a</sup>	281.00±1.00 <sup>b</sup>
Specific volume (ml/g)	2.16±0.01 <sup>a</sup>	2.11±0.01 <sup>b</sup>	1.65±0.01 <sup>c</sup>
Total solid	65.41±0.01 <sup>b</sup>	65.43±0.15 <sup>b</sup>	70.44±0.01 <sup>a</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same row are significantly different (P < 0.05).

WFB=Wheat flour bread

NFRB= Non fermented rice bran bread

FRBB= Fermented rice bran bread

**Table 6: Sensory evaluation of bread supplemented with either non-fermented rice bran or fermented rice bran**

Samples	Colour	Taste	Aroma	Texture	Overall acceptability
100% WFB	7.80±1.09 <sup>a</sup>	7.20±0.83 <sup>a</sup>	7.20±0.48 <sup>a</sup>	7.20±1.09 <sup>a</sup>	7.60±1.14 <sup>a</sup>
WF: NFRB					
95:5	7.20±1.09 <sup>a</sup>	6.60±0.89 <sup>a</sup>	7.20±0.48 <sup>a</sup>	7.20±1.83 <sup>a</sup>	6.80±1.09 <sup>ab</sup>
90:10	5.80±2.28 <sup>abc</sup>	6.20±0.83 <sup>a</sup>	6.80±0.48 <sup>ab</sup>	6.20±0.83 <sup>ab</sup>	6.60±0.89 <sup>ab</sup>
85:15	6.60±1.14 <sup>ab</sup>	6.80±0.44 <sup>a</sup>	6.80±0.84 <sup>ab</sup>	6.40±0.54 <sup>ab</sup>	6.80±0.45 <sup>ab</sup>
80:20	4.80±2.16 <sup>bc</sup>	6.00±1.58 <sup>a</sup>	6.00±1.41 <sup>ab</sup>	6.00±1.22 <sup>ab</sup>	6.80±0.54 <sup>ab</sup>
WF: FRBB					
95:5	4.20±2.48 <sup>c</sup>	6.40±1.14 <sup>a</sup>	6.20±0.84 <sup>ab</sup>	5.40±0.89 <sup>b</sup>	5.80±1.09 <sup>b</sup>
90:10	6.40±0.54 <sup>abc</sup>	6.00±1.22 <sup>a</sup>	5.80±1.09 <sup>b</sup>	5.60±0.89 <sup>b</sup>	5.60±0.89 <sup>b</sup>
85:15	5.60±1.14 <sup>abc</sup>	6.20±1.09 <sup>a</sup>	6.60±1.14 <sup>ab</sup>	6.20±1.48 <sup>ab</sup>	6.40±0.05 <sup>ab</sup>
80:20	6.00±1.58 <sup>abc</sup>	7.00±1.41 <sup>a</sup>	6.60±1.14 <sup>ab</sup>	6.80±1.30 <sup>ab</sup>	6.40±1.82 <sup>ab</sup>

Values are means ± SD from triplicate determinations, different superscripts in the same column are significantly different (P < 0.05).

WFB=Wheat flour bread

NFRB= Non fermented rice bran bread

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