Plant Archives Vol. 22, No. 1, 2022 pp. 238-245 e-ISSN:2581-6063 (online), ISSN:0972-5210

Plant Archives

Journal homepage: http://www.plantarchives.org
DOI Url : https://doi.org/10.51470/PLANTARCHIVES.2022.v22.no1.037

EFFECT OF SUPPLEMENTATION OF WHEAT BRAN ON NUTRITIONAL, FUNCTIONAL AND SENSORY QUALITY OF BREAD

Nidhi, Devinder Kaur*, Vinti Singh and Rahul Agrawal
Centre of Food Technology, University of Allahabad. Prayagraj U.P. India
*Email:devi_sonu@yahoo.com

(Date of Receiving : 10-11-2021; Date of Acceptance : 19-02-2022)

ABSTRACT

In present study supplementation of wheat bran with refined wheat flour was done at various levels (control to 30%) for preparation of bread which were subjected to morphological, sensory and nutritional analysis to obtain the best formulation. Nine-point hedonic test was used for sensory evaluation of prepared bread which showed increasing trends on increasing the level of wheat bran. Bread obtained by incorporating 25% wheat bran showed highest scores in terms of overall acceptability. Nutritional evaluation of supplemented bread shows the increasing trends that were ranged from 3.46-8.4% moisture, 2.5–6 % crude fat, 1.13–9.81 % crude fibre, and 0.66–4.1% ash content, while protein content (10.5–4%) was decreased with increasing of wheat bran. The functional properties of the bread were analyzed in terms of water holding capacity (3.14–5.59 ml/g), oil holding capacity (2.20–4.34 ml/g), and swelling capacity (0.68–2.45ml/g) were increase from control to 30 % supplementation of bran while bulk density (13.34-9.57 g/ml) was reduced with increasing of bran. Antioxidant compounds and their properties (Total Phenolic content, metal ion chelating activity, free radical activity and reducing power) of prepared bread were also increased as the supplementation of wheat bran was increased. Texture analyses of bread were analyzed by texture analyzer TA-XT2i in terms of hardness, cohesiveness, chewiness, gumminess and springiness. Thus supplementation of wheat bran up to 20% level would improve the nutritional quality of bread without adversely affecting the sensory parameters.

Keywords **:** Wheat bran, Supplementation, Texture analyzer, Antioxidant properties, Metal ion chelating activity.

Introduction

Wheat (*Triticum aestivum* L.) is an important raw material for traditional and commercial available food in many countries. The grain is consists of mainly three layers-inner part, the starchy endosperm, and surrounded by numerous layers of pericep, testa and nucellar epidermis. Processing of wheat separates starchy endosperm from embryo and bran layers into flour and this flour is the base of cereal products. The bran which is obtained as a byproduct of milling is significant source of dietary fiber, tocopherol and bioactive compounds. As a byproduct derived from roller milling of wheat flour production, wheat bran has high dietary fiber content, which contains 44-50% of fiber and can be incorporated into food products to alter the nutritional quality of foods (Onipe *et al*., 2015). Several health benefits such as reduced risk of diabetes, degenerative and chronic diseases are well established with the consumption of wheat bran (Wu *et al*., 2015; Mellen *et al*., 2009). Bakery products can be supplemented with wheat bran for improving its nutritional and functional value. The starchy portion of the wheat, endosperm is milled to refined flour and forms an essential ingredient for cereal based formulated products such as bread, cake or cookies where the bran, in contrast, is mainly used for animal feed. However, it would be further improve the nutritionally and physiologically effects on supplemented backed and extrudates products. Indeed, fibre rich foods has been associated with a curing of major chronic diseases (Wu *et al.,* 2015) and it is also known as induce several positive health effects. Some reducing compounds such as glutathione and bound ferulic acid are present abundantly in aleurone layer compared to the starchy endosperm (Noort *et al*., 2010).

In terms of health, epidemiological (as well as experimental) studies is accumulating to show that dietary fibre may cure the risk of certain chronic diseases, in particular cardiovascular disease (CVD), metabolic syndrome, type 2 diabetes and certain cancers (Fung 2002; Koh-Banerjee *et al.,* 2004; Sayhoun *et al*. 2006; Seal 2006; De, Munter *et al.,* 2007; Schatzkin *et al.,* 2007; Mellen *et al*., 2009). Nutritionally, bran fractions obtained by milling are rich in fibre, minerals, pyrodoxine, thiamine, folate and tocopherol and some phytochemicals, in particular antioxidants such as phenolic compounds (Shewry, 2009). The phenolic compounds present in wheat bran have been shown to inhibit LDL oxidation, possibly by binding with apolipoprotein-B (Yu *et al*., 2005; Liyana-Pathirana and Shahidi, 2007).

Bread is a fermented bakery product made primarily from white wheat flour, water, yeast, and salt through a series of mixing, kneading, proofing, shaping, and baking processes (Dewettinck *et al.,* 2008; Banu *et al.,* 2012). Bread is an important staple food and the most popular bakery item (Aini

and Maimon, 1996; Abdelghafor *et al.,* 2011). White flour is generated from the processing of whole wheat grain and is used to improve the aesthetic value of white bread, but it has a lower nutritious value than bread made from whole grain cereals (Maneju *et al.,* 2011). Today, bread plays a significant role in the baking business, accounting for more than half of the overall Indian bakery market. It is consumed by the majority of the world's population as part of their daily diet, and it is equally popular in urban areas. The current bread production in India is 9.58 lakh tonnes, with a projected annual growth rate of 9.7% (Khateeb & Kumar, 2019). So the goal of this research was to investigate the effect of the addition of wheat bran on nutritional, functional and sensory quality of bread.

Materials and Methods

PBW-154 variety was procured from seed store Alopibag Chungi, Allahabad U.P., and other ingredients such as sugar, oil, active dry yeast and refined flour etc. were purchased from local market of Allahabad.

Fermentation of Wheat Bran

Wheat bran fraction 710µm was mixed with per gm dry yeast with water, then covered with aluminium foil, and placed in an incubator at 30^{o}C for 24 h. The bran was spread on a wide tray then placed in an oven at 70^{o} C until dry. Each of the samples was packed in polyethylene bags and stored in a deep freezer until used.

Blend Preparation

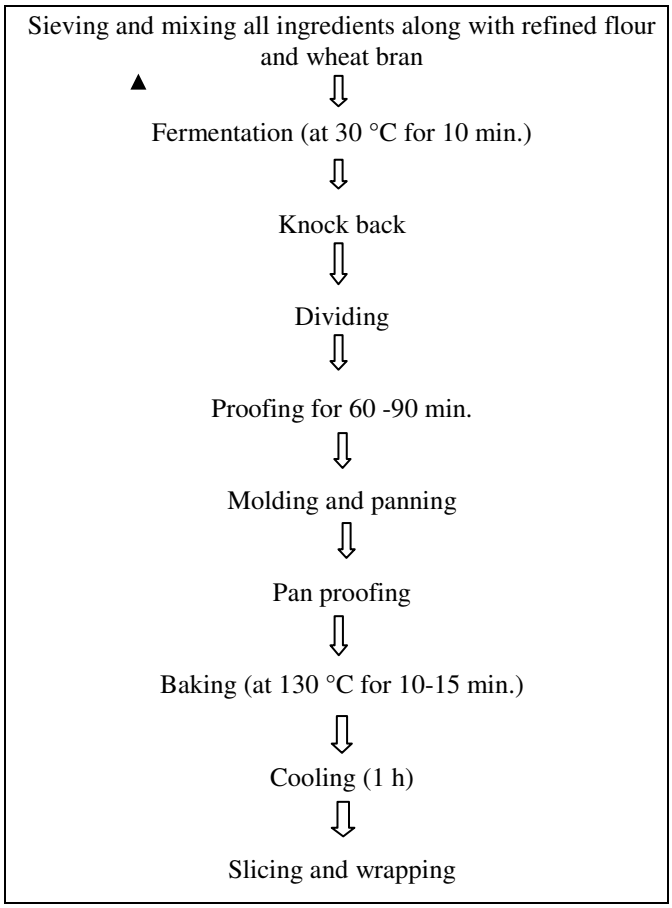
The bread were prepared from blends of wheat refined flour and fermented wheat bran at various levels (95:5, 90:10, 85:15, 80:20, 75:25 and 70:30). The traditional creamery method was used for bread preparation. Other ingredients added were active dry yeast (10), milk powder (5 g), oil (1.5), sugar (10g) and salt (1.5 g).

Processing of bread

The water was used in this process. All ingredients were then weighed and mixed for 5 min. to form a dough. The dough was allowed to keep for 10 minutes at room temperature (30^{o}C) and then scaled to seven portions of 100 g each. The dough portions were made as round balls and allowed to keep for another 15 minutes then molded and put into a pan and placed in the fermentation cabinet for final proof between 60 and 90 minutes. Baking was done in oven at 130^{o}C with steam saturation for 10-15 minutes. Different types of bread were prepared by using refined wheat flour with fermented wheat bran. Refined wheat flour bread was prepared as a control sample. The loaves were sliced with an electric knife.

Proximate analysis

The proximate composition of wheat bran bread viz. moisture, fat, protein, ash, and crude fiber content was done by using standard methods AOAC, (2016) for all the bread samples. Protein content was calculated by multiplying a factor of 6.25 to the percent of nitrogen found in the sample. Carbohydrate content was determined by subtracting the sum of moisture, protein, ash, fiber, and fat, crude fiber content of the sample from 100. All the experiments were done in triplicate and analyzed statistically.



Flow chart for preparation of wheat bran supplemented bread

Color value

Color value of the bread sample was measured using X-rite (Grandville, MI, USA). The color attributes i.e. Hunter lightness (L^{*}), redness (a^{*}), and yellowness (b^{*}) were recorded three times for each sample (n=3) according to the method of Chen *et al.* (1997).

Functional properties

Bulk density

Bulk density of wheat bran supplemented bread was estimated by Chau and Huang (2003). A 5g sample of bread was taken in a 10 ml graduated measuring cylinder. The measuring cylinder was tapped on until the level gets stabilized leaving no molecular space in each molecule of sample. Then, the volume occupied by the sample was determined and the bulk density was representing as weight per unit volume (g/ml).

Water and Oil holding capacity

Water and oil absorption capacity wheat bran supplemented bread was done according to the method given by Chau and Huang (2003) with some modification. 1g wheat bran sample (control & treated) was taken in centrifuge tube and mixed with 10ml water / vegetable oil and stirred at room temperature using a rotary shaker for 10 min. The sample was then centrifuged at 3500 rpm for 20 min. and the clean supernatant was collected in measuring cylinder. The tube was weight along with the sample and water / oil absorbed. The retention of water / oil in the

control and treated wheat bran sample was compared as water absorption capacity or oil absorption capacity (g/g).

Swelling capacity (SC)

Swelling capacity of bread sample was determined as described by Robertson *et al.* (2000). In a graduated test tube 10 ml deionised water added to 0.5g of bread and allowed to hydrate for 18 h, and the final volume attained by bread was measured.

$$SC = \frac{\text{Final Volume (V2)} - \text{Initial Volume (V1)}}{\text{Weight of Sample(g)}} \times 100$$

Antioxidant properties

Extraction method of antioxidants

Bread sample were extracted by using methanol at 50% conc. One gram of bread sample was extracted with 5 ml of methanol in a screw-capped tube in the dark condition at room temperature for 24 hrs. The tubes were centrifuged at 2000 rpm for 5 min. The supernatant was collected and kept in refrigerator for further analysis (Moore *et al*., 2006).

Total phenolic content

The total polyphenolic content of the aqueous methanolic extract of bread sample was determined according to the 'Folin ciocalteu method'. 1 ml aliquot of the sample extract was taken in a test tube. There after 5 ml. of diluted folin ciocalteu reagent (1:10 with distilled water) and 4 ml sodium carbonate solution (7.5 %, w/v) were added sequentially to each tube. Soon after mixing, the test tubes were placed in the dark for 60 minutes at room temperature and the absorbance was monitored by UV-VIS spectrophotometer (modelEvolution600) at 765 nm against blank as standard. A standard curve was prepared with "Gallic acid" and results were expressed in terms of mg/100g of polyphenol present in the sample. Samples were analyzed in triplicates and mean was calculated (Matthaus, 2002).

Percent Free Radical Scavenging Activity (DPPH activity)

The DPPH (2,2-diphenyl 1-pycril hydrazil) radical scavenging activity of bread sample extracts was measured according to the method given by Sanja *et al.* (2009) with slight modification. As per method 150 µl of DPPH solution (4.3 mg in 3.3ml. of methanol) was mixed with 3 ml acidified methanolic extract of selected cereals varieties. The mixture was shaken and decrease in absorbance was measured at 515 nm with the help of UV/VIS spectrophotometer after 15 min. incubation at room temperature. DPPH solution was used as control.

$$\% \text{ free radical scavenging activity} = \frac{(A_{control} - A_{sample})}{A_{control}} \times 100$$

Reducing capacity of bread

Determination of reducing capacity of bread sample extracts was measured according to the method given by (Oktay *et al,* 2003), with slight modification. Accordingly, as per method phosphate buffer (2.5 ml, pH, 6.6) and potassium ferricyanide (2.5ml, 1% w/v) mixture was incubated at 50 ºC for 20 min. A portion (2.5 ml) of trichloroacetic acid solution (10%) was added to the mixture, which are centrifuged at 10,000g for 10 min. The upper layer of solution (2.5 ml) was mixed with deionised water (2.5 ml) and Ferric chloride solution (0.5 ml, 0.1%, w/v) and the absorbance of the mixture was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power. A standard curve was prepared with ascorbic acid (AA) solution, and the final results were expressed as micromoles of ascorbic acid equivalents (AAE) per gram of flour.

Metal chelating (Fe^{+2}) activity of bread

Metal chelating (Fe^{+2}) activity of bread sample extracts was measured according to the method given by Sharma and Gujral, (2009) with slight modification. The metal ion chelating ability measures how effectively the compounds can compete with ferrozine for ferrous ion. Ferrozine can quantitatively form complexes with Fe^{+2}. The presence of antioxidant compounds in sample extracts can disrupt the formation of ferrozine- Fe^{+2} complex resulting in decolourisation of red or purple color of the complex. Measurement of color reduction, therefore, allowed the estimation of the metal ion chelating activity.

Texture Profile Analysis

Texture parameters in terms of hardness, springiness, cohesiveness, gumminess and chewiness of wheat bran bread samples were measured objectively by using a texture analyzer TA-XT2i (TAHDI, Stable Micro system, UK) as adopted by the standard method by (AOAC, 2000). All samples were prepared and baked on the day of test. The probe was calibrated according to the instruction before conducting the test. A cube sample (2cm *2cm* 2cm) was cut from the middle of sample (bread) and it was placed centrally beneath the probe [(p/36 cylinder probe (36mm)] in order to meet with a consistent flat surface. The compression test was selected in texture analysis using a 5 kg load cell and sample was compressed to 45% of its original height. The strain required for 45%compression was recorded using the following conditions: pretest speed: 1.0 mm/s, test speed: 1.7 mm/s, post test speed: 10 m/s, compression distance: 25% and trigger type: auto 5 g. the values reported were the average of three readings. Data was analyzed using Texture expert Version 1.05 (Stable Micro system Ltd) software.

Sensory evaluation

All prepared bread sample from different formulations were organoleptically evaluated after 4 h. by a selected panel of 15 trained judges, by using 9-pointhedonic scale (Amerin *et al*., 1965), for the assessment of all sensory parameters (quality crust colour, crumb colour, crumb taste, crumb aromas, crumb elasticity, over acceptability characteristics) of bread samples . Water was provided for rinsing the mouth between the samples. The baking experiments were repeated twice to substantiate the results. The results are expressed in terms of average acceptability scores.

Statistical Analysis

The data collected from the proximate composition, antioxidant, texture, functional properties and sensory analysis were presented as means of triplicates. Bread sample data were subjected to one-way analysis of variance using SPSS statistical software version 20.0. The mean was separated by applying the Duncan Multiple Range test at 95% confidence level (p <.05).

Results and Discussion

Proximate composition of bread

The nutritional composition of bread prepared by incorporation of wheat bran at different levels (5-30%) is presented in Table 1. The results showed that moisture (3.46 to 8.4 %), fat 2.5 to 6.0%), crude fibre (1.13 to 9.8%) and ash contents (0.66 to 4.1%) were increased significantly (p-0.05) with increasing of wheat bran while protein content (10.5 to 4.0%) were decreased. Moisture content were observed higher in 30% incorporated wheat bran bread (8.4%) followed by 20% incorporated wheat bran (7.3%) and significantly lowest were found in control (3.46%) bread. As the wheat bran contains more cellulose and other polysaccharides that hold moisture several times higher to its weight therefore, with the increases of wheat bran level there was increased moisture content of bread. Similar results were obtained for ash and crude fiber content. The ash and crude fiber was found to be highest (0.66 and 4.16% respectively) in 30%wheat bran incorporated bread because wheat bran contains higher amount of nutrient like dietary fibre, antioxidants properties and some micro nutrients (vitamins and minarals), so it would be better than control sample. Ronhotra *et al.* (1994) reported the ash content of wheat bran ranged from 5.2 to 5.5%. Halverson and Zeleny (1988) found the protein content of wheat bran to be ranged from 14 to 16%. Tian *et al.* (2020) reported that the proximate composition (protein, fat, ash) of wheat bran that was in range from 17.66 to 17.81; 2.89 to 4.16%; and 17.58 to 24.74%. The ash content of selected wheat bran cultivars was similar to values reported by D'hoe *et al.* (2018). Similar results were reported for fat content of wheat bran by Curti *et al.,* 2013. Elawad, *et al.* (2016) reported moisture, fat, fiber, and carbohydrate content which was 7.8%, 5.6%, 9.6%, 61.3% in wheat bran respectively; these compositions were found close to the existing value. The variation in each nutritional content among all cultivars of wheat bran is due to environmental related factors like maturity period, climate, location, temperature, fertility, diseases, pest exposure, climate, soil condition, etc. (Zheng and Wang, 2001). (Khateeb & Kumar, 2019) reported the ash, fat, and fibre content (0.93-2.62, 1.7-2.66 and 2.5-4.46%) in supplemented fermented wheat bran sample bread these compositions were found closer to this study. According to Kouidri *et al.,* (2015) chemical composition (fat, ash, moisture content) of value added products was increased with increasing of addition of wheat bran. Sangle *et al.,* (2017) reported the moisture, crude fat, crude fibre and ash contents were increased, and these nutrients were also found to be closer to our existing value.

Colour measurement of Bread

The color of the bread was measured by the X-rite color lab using 'L', 'a' and 'b' values. 'L' value decreased significantly with the increase in the levels of wheat bran. In control bread highest brightness (78.35) was observed as compared to wheat bran supplemented bread (Table 2). The effect of the wheat bran on bread color showed significant changes in all the evaluated parameters ($p<0.05$). Redness (a) and yellowness (b) values increased significantly with the addition of wheat bran in all the substitution levels. The 'a' value of control bread was +0.93 where as it increased significantly ($P<0.05$) +10.63 for the 30% wheat bran supplemented bread. Similar trend was observed for 'b' value at all the levels of incorporation of wheat bran. It was concluded that when the wheat bran as the substitution increases, the 'L' value was decreases while 'a' and 'b' value were increases.

Functional Properties of bread

The present study showed functional properties of bread (Table 3). There were water holding (3.14 to 5.59ml/g), oil holding (2.2 to4.34 ml/g) and swelling capacity (0.68 to 2.45 ml/g) increases with increasing of wheat bran while bulk density (13.3 to 9.5 g/ml) are decreased due to porous matrix structure of the insoluble fiber chain which can hold large amounts of water through hydrogen bonds (Kethireddipalli *et al*., 2002).

Functional properties were the intrinsic physiochemical characteristics which may affect the behavior of food systems during processing and storage. These properties will decide the acceptability of the product. The water holding capacity (WHC) is the first functional properties to determine the bread sample. 15% & 20% bread show higher WHC than the control, which can be attributed to high fiber and starch content in the bread. More amount of water is needed for 5%-30% dough formation than in the refined wheat dough formation. Oil holding capacity (OHC) of control refined wheat flour (RWF) was more than composite flours. Water holding capacity (WHC), represents the amount of water that flour can absorb to obtain a pre-set dough consistency to produce a torque at room temperature (Blandino *et al*., 2015). According to Sattanathan *et al.* (2010) oil holding capacity also affects the flavor, texture and taste of the products. The increase of oil absorption may be attributed to the presence of more hydrophobic groups or polar amino acid on the surface of protein tends to decrease oil absorption capacity (Sathe *et al.,* 1982).

Antioxidant Activities of Bread

Cereals contain variety of compounds showing antioxidant properties. Different methods have been developed to determine the antioxidant property of different cultivars of wheat bran (Moore *et al.,* 2006). In present study, four different methods have been used for the evaluation of the antioxidant capacity of the wheat bran extracts namely total phenolic content, DPPH free radical scavenging assay, metal ion chelating activity and reducing power. The polyphenolic compounds in plant extracts are more often associated with other molecules like protein, polysaccharides, terpenes, chlorophyll and inorganic compounds. Hence, it requires suitable solvent for maximum extraction of polyphenols (Michalak *et al.*, 2017).

The total phenolic content of wheat bran substituted bread was expressed as mg Gallic acid per gram of dry weight. As summarized in Table 4. Antioxidants properties (total phenolic content (TPC), metal ion chelating activity, free radical scavenging activity and reducing power) of wheat bran substituted bread varied significantly between the substitution levels. Incorporation of wheat bran in bread resulted in an increase in the total phenolic content. The bread containing 30% wheat bran resulted in highest TPC, free radical scavenging activity, metal ion chelating activities and reducing power as compared to other samples and control bread, which showed an increase (23.07 to 88.60 mg GAE/100g, 23% to 59.6%, 45.1 to 81.5 % and 2.9 to 12.4mg AAE/g) as the level of wheat bran was increased from 5 to 30%. The above results were supported by Safa *et al*. (2014),

who found maximum radical DPPH activity of wheat bran in 70% ethanol extract as compared to methanol and acetone extract. Previous study determined the free radicals scavenge activity in wheat bran that was similar to current study (Brewer *et al.,* 2014). Similar to our study the free radical scavenging activity of seven cultivars of wheat bran from different countries and found that at 50% acetone extracts have higher value antiradical activity (Zhou *et al*., 2004). According to Vaher *et al.* (2010) bran layers have the highest content of total phenolics content, when stated that the Phenolic compounds and the antioxidant activity of the bran, flour and whole grain of different wheat varieties.

The maillard reaction products formed during the production of bread could also act as the antioxidants and scavenge free radical, which consequently contribute to better antioxidant activity of the bread (Jing and Kitts 2000). Epidemiological studies showed that consumption of phenolic rich foods is associated with low risk of several chronic diseases such as ageing, cancer, cardiovascular disease and Alzheimer disease.

Texture profile analysis of bread

The results of texture profile analysis are shown in Table 5. Addition of wheat bran in formulation of bread samples significantly affected the textural properties of the product. Hardness of bread samples were significantly ($p<0.05$) increased by increasing the wheat bran. Bread samples substituted with 30% wheat bran were not significantly ($p<0.05$) harder than those with 5% wheat bran. Hardness is mainly attributed to the amylose and amylopectin matrix which contribute to overall bread texture (Schiraldi and Fessas, 2000). Gomez *et al.* (2013) reported that bread hardness was due to interactions between gluten and fibrous materials. Springiness of the bread samples significantly were reduced by addition of WB in their formulation. The springiness of bread samples containing 5% to 30% wheat bran were not significantly different ($p>0.05$). According to a report by Hoseney *et al*. (1994) interaction between gelatinized starch and gluten dough which cause dough to be more elastic can form continuous sponge structure of bread after baking. Therefore, the high springiness could be attributed to dilution of the gluten structure in composite breads (5-30%). Lower amount of gluten causes lower ability to hold gases which caused an elasticity reduction in breads (Pyler, 1973). This reduction indicates that the breads formulated with WB have low ability to resist before the bread structure deformed under the teeth. Bread samples substituted with wheat bran showed significantly ($p<0.05$) higher values of gumminess and chewiness, it's due to higher amount of fibre content in wheat bran. A report by Wang *et al*., (2002) also showed similar trend for breads with addition of fibers since they caused an increase in gumminess and chewiness of tested breads.

Sensory evaluation

Sensory analysis summarized the mean scores of hedonic sensory evaluation for color, flavour, taste, texture, appearance and overall acceptability of bread samples. As can be seen, substitution of 5-30% wheat bran has a significant ($p<0.05$) effect on all sensory parameters of the bread samples. According to ANOVA, all sensory parameters of bread were significantly ($p<0.05$) different. Generally, addition of wheat bran had significant effects on sensory attributes and overall acceptability of bread samples. Addition of wheat bran caused darker color and denser texture, which at level of 20% seem acceptable for consumers (Table 6). However, increasing incorporation of wheat bran to 25% or 30% seems to have negative effect on consumer's overall acceptability. For consumers, color of the bread is one of the important factors in sensory evaluation depending on their perception of bread type. Average scores of bread flavor, which can be determined by the sense of smell, was significantly ($p<0.05$) lower in 25% and 30% compared to control bread sample, which showed addition of wheat bran more than 5% has a negative effect on final product in terms of flavor. As shown in Table 6, same trend observed in taste, flavor, texture, and appearance. Scores of softness attribute were in accordance with the results of texture analysis which showed increase in wheat bran formulation can cause harder breads. Bread samples which received higher scores than 4 (neither like nor dislike) were considered as acceptable. Moreover, the evaluation of taste showed that addition of wheat bran increased the bitter taste of cookies, which was due to phenolic acids and tannins (Heinio *et al*., 2016). Measurements of colour of the biscuits showed that the biscuit became darker with increasing level of wheat bran (Gamal *et al*., 2012). Khateeb and Kumar, (2019) reported the sensory analysis of wheat bran formulated bread in terms of color, texture, Flavor, taste and overall acceptability of bread, which were also decreased with increasing of wheat bran, which was also closer to our study. Therefore, Overall acceptability was determined on the basis of bread quality scores obtained from the evaluation by the experts of colour, flavour, taste and texture.

Table 1: Proximate analysis of bread supplemented with wheat bran

Wheat bran: wheat flour (%)	Moisture (%)	Crude Fat (%)	Crude Fiber (%)	Ash (%)	Protein (%)	CHO (%)
Control	3.46 ± 0.33^{e}	2.5 ± 0.4^{d}	1.13 ± 0.27^{f}	$0.66\pm.23^{g}$	10.5 ± 1.4^{a}	81.75 ± 5.7^{a}
05:95	4.8 ± 0.12^{d}	3.6 ± 0.8^{cd}	2.93 ± 0.33^{e}	1.3 ± 0.2^{f}	8.5 ± 1.0^{b}	78.8 ± 5.0^{a}
10:90	5.4 ± 0.32^{d}	4.0 ± 0.4^{bcd}	3.8 ± 0.21^{e}	2.2 ± 0.2^{e}	7.2 ± 1.0^{b}	77.4 ± 5.0^{a}
15:85	6.6 ± 0.57^{c}	4.16 ± 0.4^{bc}	4.85 ± 0.22^{d}	2.6 ± 0.2^{d}	6.9 ± 0.8^{bc}	74.9 ± 5.0^{a}
20:80	7.3 ± 0.49^{bc}	5.0 ± 1.8^{abc}	6.37 ± 1.19^{c}	3.0 ± 0.2^{c}	5.5 ± 0.5^{cd}	72.8 ± 4.2^{a}
25:75	7.8 ± 0.12^{ab}	5.6 ± 0.8^{ab}	7.93 ± 0.33^{b}	3.4 ± 0.2^{b}	4.8 ± 0.5^{d}	70.5 ± 4.0^{a}
30:70	8.4 ± 0.32^{a}	6.0 ± 0.4^{a}	9.81 ± 0.21^{a}	4.1 ± 0.3^{a}	4.0 ± 0.5^{d}	67.7 ± 3.5^{a}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a - g) in the same column indicate significant difference ($P<0.05$) between all treatments of bread.

Table 2: Colour measurement of Bread prepared by incorporation of wheat bran

Wheat Bran: Wheat bran: wheat flour (%)	L*	a*	b*
Control	78.35±7.44^{a}	+0.936±.09^{e}	+12.91±2.19^{e}
5:95	69.08±6.68^{ab}	+3.776±0.1^{d}	+18.72±3.16^{d}
10:90	62.52±5.69^{bc}	+4.206±0.1^{d}	+21.82±3.15^{cd}
15:85	59.09±5.2^{cd}	+5.53±0.3^{c}	+23.07±4.11^{cd}
20:80	55.43±4.7^{cd}	+6.63±0.9^{c}	+26.41±4.94^{bc}
25:75	49.43±4.37^{de}	+8.43±0.37^{b}	+29.43±5.37^{b}
30:70	42.43±3.37^{e}	+10.63±1.37^{a}	+35.12±5.37^{a}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a-e) in the same column indicate significant difference (P<0.05) between all treatments of bread.

Table 3: Functional properties of wheat bran supplemented bread

Wheat Bran: Wheat bran: wheat flour (%)	Bulk Density	Water Holding Capacity	Oil Holding Capacity	Swelling Capacity
Control	13.34±1.5^{a}	3.14±0.4^{d}	2.20±0.1^{c}	0.68±0.10^{e}
5:95	13.19±1.2^{a}	3.69±0.6^{cd}	2.60±0.2^{bc}	0.98±0.1^{de}
10:90	12.47±1.30^{ab}	4.04±0.3^{cd}	2.90±0.2^{bc}	1.20±0.4^{de}
15:85	12.19±1.7^{ab}	4.13±0.8^{cd}	3.02±0.2^{b}	1.50±0.5^{cd}
20:80	11.26±1.20^{ab}	4.45±0.4^{bc}	3.20±0.5^{b}	1.82±0.2^{bc}
25:75	10.24±1.5^{b}	5.31±0.8^{ab}	4.10±0.8^{a}	2.15±0.5^{ab}
30:70	9.51±0.8^{b}	5.59±0.3^{a}	4.34±0.34^{a}	2.45±0.2^{a}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a - e) in the same row indicate significant difference (P<0.05) between all treatments of bread.

Table 4: Antioxidant properties of wheat bran supplemented bread

Wheat bran: wheat flour (%)	DPPH (%)	Metal Chelating Activity (%)	Total Phenolic Content (mg gallic acid/100)	Reducing Power (mgAAE/g)
Control	23.7±2.5^{e}	45.1±3.4^{d}	19.0±1.4^{d}	2.92±0.3^{e}
5:95	30.7±3.4^{d}	49.1±4.0^{cd}	24.1±2.1^{d}	4.44±0.9^{de}
10:90	34.8±4.4^{d}	53.6±5.6^{cd}	36.3±3.2^{c}	6.45±1.2^{cd}
15:85	41.2±4.9^{cd}	58.4±5.9^{bc}	48.1±3.1^{b}	7.55±1.5^{bc}
20:80	45.7±5.9^{bc}	67.6±6.6^{b}	56.7±4.2^{b}	9.50±1.2^{b}
25:75	51.4±6.7^{ab}	79.6±7.3^{a}	64.7±5.3^{a}	10.1±2.5^{ab}
30:70	59.6±7.8^{a}	81.5±7.7^{a}	71.3±5.5^{a}	12.3±2.0^{a}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a - e) in the same row indicate significant difference (P<0.05) between both cultivars of wheat bran.

Table 5: Texture parameters of different bread samples

Wheat bran: wheat flour (%)	Hardness(N)	Cohesiveness	Chewiness	Gumminess	Springiness
Control	444 ±22^{g}	1.00 ±0.12^{a}	516 ±10^{g}	445 ±57^{g}	1.5 ±0.21^{a}
5:95	931 ±23^{f}	0.97 ±0.1^{a}	969 ±34^{f}	906 ±45^{f}	1.4 ±0.2^{a}
10:90	1307 ±30^{e}	0.88 ±0.1^{ab}	1219±25^{e}	1147 ±42^{e}	1.16 ±0.1^{a}
15:85	1717 ±40^{d}	0.91 ±0.1^{ab}	1700±25^{d}	1566 ±52^{d}	1.13 ±0.3^{a}
20:80	2039 ±50^{c}	0.81±0.1^{ab}	2536±82^{c}	1663 ±46^{c}	1.08 ±0.3^{a}
25:75	3460 ±45^{b}	0.73±0.1^{b}	3537 ±74^{b}	2527 ±48^{b}	1.07 ±0.2^{a}
30:70	4115±75^{a}	0.92 ±0.1^{ab}	5126 ±105^{a}	3806 ±71^{a}	1.06 ±0.2^{a}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a - g) in the same row indicate significant difference (P<0.05) between both cultivars of wheat bran.

Table 6: Sensory Analysis of wheat bran Supplemented Bread

Wheat bran: wheat flour (%)	Colour	Flavour	Taste	Texture	Appearance	Overall quality
Control	8.0±0.9^{a}	8.21±1.2^{a}	7.97±1.0^{a}	7.4±1.0^{a}	7.32±0.8^{a}	7.79±1.0^{a}
5:95	7.8±0.5^{ab}	7.98±0.9^{a}	7.54±1.0^{a}	7.2±0.9^{a}	6.98±.54^{a}	7.28±0.8^{a}
10:90	7.26±0.5^{abc}	7.65±0.9^{a}	7.26±0.9^{a}	6.88±0.8^{a}	6.82±0.6^{a}	7.17±0.9^{a}
15:85	7.46±0.5^{abc}	7.89±0.8^{a}	7.67±0.9^{a}	7.29±0.9^{a}	7.04±0.8^{a}	7.47 ±0.7^{a}
20:80	6.90±0.4^{bc}	7.31±0.6^{a}	7.11±0.7^{a}	6.65±0.9^{a}	7.11±0.5^{a}	7.01±0.5^{a}
25:75	6.72±0.4^{c}	6.74±0.5^{a}	6.83±0.6^{a}	7.21±0.5^{a}	7.21±0.5^{a}	6.94±0.5^{a}
30:70	5.02±0.3^{d}	5.04±0.5^{b}	5.23±0.5^{b}	5.21±0.5^{b}	5.01±0.4^{b}	5.14±0.5^{b}

All values are means ± standard deviations of data from three independent experiments. Different superscripts (a - b) in the same row indicate significant difference (P<0.05) between both cultivars of wheat bran.

Conclusion

By addition the different level of Wheat Bran into wheat flour, the hardness and darkness of bread samples were significantly ($p<0.05$) increased while their volume significantly ($p<0.05$) was reduced compared to the control sample (white wheat bread). The incorporation of fermented wheat bran in bread formulation reduces the bread specific volume slightly at 10% level but significantly at the higher levels (30%). Therefore, the addition of fermented wheat bran affects the physical and sensory properties of the baked bread. Bread samples substituted with 20% wheat bran had the highest mean scores of overall acceptance among samples. Dietary fiber is a common and important ingredient of new generation of healthy food products and increased the nutritional value of bread. Finally, the quality of the high fiber bread was noticeable when higher level of bran was added to the whole wheat flour. Therefore, this knowledge can be used to make commercial products. The introduction of such new technology, would increase overall economic, society health and well-being.

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