

Biochemical and Structural Characterization of Rice and Oat Bran Hydrolysates and Their Functional Implications in Food Systems

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ABSTRACT

Cereal bran, a by-product of grain milling, is a nutrient-rich material with significant potential for use in functional food applications. This paper examined the biochemical structure, antioxidant properties, and functionality of rice and oat bran hydrolysates obtained via hydrothermal treatment. Proximal analysis revealed significant compositional differences between raw brans and their hydrolysates, indicating increased nutrient extractability after hydrolysis. Antioxidant activity was evaluated using DPPH (% inhibition), FRAP ($\mu\text{mol Fe}^{2+}/\text{g}$), ABTS (mmol TE/g), total phenolic content (mg GAE/g), and total flavonoid content (mg RE/g). Oat bran hydrolysates showed higher antioxidant activity in DPPH, FRAP, and ABTS assays. Rice bran hydrolysates exhibited slightly higher total phenolic content, while oat bran hydrolysates showed superior antioxidant activity. The difference was explained by the differences in phenolic composition and synergistic interactions with β -glucan in oat bran. FTIR and SEM analyses were used to confirm structural changes, which showed that functional groups and the microstructure changed after incorporation of hydrolysates. Bread systems' functional analysis revealed that hydrolysates had a considerable impact on texture, color, and sensory characteristics. Rice bran hydrolysates raised firmness and flavor and oat bran hydrolysates raised structural balance and acceptability. These results indicate the promise of rice and oat bran hydrolysates as value-added functional ingredients to enhance the creation of nutritionally enriched and structurally enhanced food items.

Keywords: Oat bran, Rice bran, Antioxidants, Flavonoids, Enzymatic hydrolysates, Phenolic compounds.

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Introduction

Cereal bran is one of the main by-products of grain powdering that attracts more and more attention because

of its high nutritional value and possible use in the development of functional foods [1]. Rice bran and oat bran are some of the cereal brans that are characterized

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by a high level of dietary fiber, bioactive compounds, essential fatty acids, and antioxidants [2, 3]. Rice bran, the outer layers of *Oryza sativa*, is a rich source of phenolic compounds, γ -oryzanol, tocopherols, and tocotrienols that add to its high antioxidant and health-promoting properties. Oat bran, which is the product of *Avena sativa*, is characterized by a high level of β -glucan (soluble dietary fiber) with its cholesterol-lowering, glycemic-regulating, and immunomodulatory properties [4]. Although rich in nutrients, the direct inclusion of raw bran in food substances is usually restricted by low bioavailability of nutrients, poor sensory qualities, and the existence of anti-nutritional compounds. Research has been conducted on different processing methods to improve both functional and nutritional characteristics of cereal brans [5, 6]. Hydrothermal and enzymatic treatments have become an effective method of altering the structural and biochemical properties of bran. Hydrothermal hydrolysis helps break down complex macromolecules like polysaccharides, proteins and lignocellulosic components into smaller and more bioavailable molecules. The process not only enhances the release of bound phenolic compounds but also increases the antioxidant activity and functional properties like water-holding capacity, emulsification, and solubility. Bran hydrolysates have shown considerable promise as value-added ingredients in the formulation of functional foods [7]. Recent research has indicated that the physicochemical and structural properties of cereal brans are different when hydrolysis is applied to them. Such changes can be characterized using FTIR and SEM which determine functional groups and chemical bonds, and identify changes in the morphology and microstructure of the surface [8, 9]. These structural changes are directly linked to the enhancement of functional properties and the overall functionality of bran-derived ingredients in food systems. The extent and nature of these changes vary depending on the type of bran and the processing conditions applied. Considering the compositional differences between rice and oat bran, i.e., the higher content of phenols in rice bran and the higher content of β -glucans in oat bran [10,11]. It is necessary to evaluate the effect of hydrolysis on the respective properties and possible use in each case. Understanding these differences is essential for optimizing their use in the development of nutritionally enriched and functionally improved food products. Further enhancing the nutritional quality, the incorporation of bran hydrolysates into food systems,

particularly bakery products, has been reported to affect the textural, sensory, and structural properties. The reaction of hydrolysates with gluten and other macromolecules has the capability of altering the rheology of dough, crumb structure, and the overall acceptability of the product [12,13,14]. Assessing the functional characteristics of rice and oat bran hydrolysates in model food systems is an important step towards their application in the food industry.

The current research seeks to examine the biochemical structure, structural, and functional qualities of rice and oat bran hydrolysates. The research aims to assess their possible use in food systems by assessing their effects on product quality characteristics. Having offered a detailed insight into the connection between structural changes and functional effectiveness, the study will also help to make cereal by-products more valuable and create new, health-promoting food components.

Materials and Methods

Purchase of Raw Materials

The rice and oat brans were obtained through source of agriculture. The local markets were also procured with other ingredients like wheat flour, butter, sugar, salt, baking powder, and eggs. All chemicals and reagents used in this study were of analytical grade and were procured from certified commercial suppliers. The raw materials were kept in tight polyethylene bags at ambient temperature ($25\pm 2^\circ\text{C}$) without absorbing moisture or contaminating it with microorganisms before being subjected to further treatment.

Proximate Composition of Rice and Oat Bran

The proximate analysis of rice and oat bran was done to determine the moisture, ash, crude fat, crude protein, and crude fiber as well as nitrogen-free extract (NFE). They were analyzed by typical American Association of Cereal Chemists (AACC, latest approved methods) procedures. Each determination was done 3 times ($n = 3$) to verify the accuracy and reproducibility.

Preparation of Bran Hydrolysate by Hydrothermal Treatment

Hydrothermal processing was used to prepare rice bran and oat bran hydrolysates. Samples of bran were suspended in distilled water in a ratio of 1:8w/v. The suspension was maintained at neutral pH (7.0 ± 0.2) without the addition of chemical buffers or enzymes. The sample was then autoclaved at 121°C under 15psi for 45 minutes to facilitate hydrothermal degradation of macromolecules, including polysaccharides and proteins. The slurry was filtered through muslin cloth, then

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Whatman No. 1 filter paper in that order, after letting it cool to room temperature to remove insoluble residues. The filtrate was centrifuged at 10,000 rpm ($\approx 9000 \times g$) for 15 minutes at 4°C using a refrigerated centrifuge (rotor radius as specified by the manufacturer). The supernatant obtained was collected as crude hydrolysate. The hydrolysate was subjected to controlled spray drying under the following conditions: inlet temperature 200°C, outlet temperature 95°C, feed rate 5 mL/min, nozzle diameter 0.7mm, and compressed air pressure 8 bar, to obtain powdered hydrolysates. The resulting powders were stored in airtight containers at 4°C until further analysis.

Bran Hydrolysates Composition

Standard AACC methods were used to determine the chemical composition of rice and oat bran hydrolysates, including moisture, ash, crude fat, crude protein, and crude fiber.

The calculation of NFE was achieved by using the following equation:

$$\text{NFE (\%)} = 100 - (\text{crude protein} + \text{crude fat} + \text{crude ash} + \text{crude fiber})$$

Antioxidant Activity

Diphenyl Picrylhydrazyl assay

A common spectrophotometric procedure was used to determine the DPPH radical scavenging activity. A fresh stock of DPPH (0.025g/L) was prepared in 99% ethanol and kept in the dark.

To analyze, 0.1mL of sample extract was added to 3.9mL of DPPH solution. The mixture of the reaction was vortexed and kept dark at room temperature for 30 minutes to avoid the photo-degradation of DPPH radicals. The absorbance of the samples at 517nm was recorded using a UV-Visible spectrophotometer against a blank sample (ethanol in the absence of DPPH). The radical scavenging activity was determined as a percentage of inhibition:

$$\text{Scavenging activity (\%)} = \frac{(A^0 - A_s)}{A^0} \times 100$$

Where the sample absorbance value is A_s , and the blank absorbance value is A^0 .

Ferric Reducing Antioxidant Power (FRAP) Assay

FRAP was done as per the routine protocol with minor adjustments. The FRAP reagent was freshly prepared by mixing acetate buffer (300 mmol/L, pH 3.6), TPTZ solution (10 mmol/L in 40 mmol/L HCl), and FeCl_3 solution (20mmol/L) in a ratio of 10:1:1 (v/v/v). The reagent was pre-incubated at 37°C and no post-preparation pH adjustment was done to ensure stability

of the assays. To conduct the analysis, 100 μ L of the sample extract was incubated with FRAP reagent (3mL), and incubation was done after 10 minutes at room temperature. The absorbance was observed at 593nm. The results were expressed as $\mu\text{mol Fe}^{2+}$ equivalents per gram of sample ($\mu\text{mol Fe}^{2+}/\text{g}$) using a calibration curve prepared with $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$.

2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) assay

The ABTS assay was done on the ABTS radical cation decolorization method. ABTS^+ solution was produced by mixing 7 mM stock solution of ABTS with 2.45 mM stock solution of potassium persulfate in equal portions and incubating in the dark for 12-16 hours at room temperature until full radicalization was produced. The ABTS^+ solution was diluted to 0.70 ± 0.02 at 734nm before analysis. For the assay, 0.1mL of sample extract was added to 3.9mL of diluted ABTS solution and incubated for 6-10 minutes. The decrease in absorbance at 734nm was measured. The results were reported as mmol Trolox equivalents per gram (mmol TE/g).

Preparation of Samples used in Antioxidant Assays

The antioxidant assays were all conducted with hydrolysate extracts made under the same conditions. The samples were centrifuged at 10,000 rpm ($\approx 9,000 \times g$) at 4 °C for 10–15 minutes to remove insoluble particles. All assays were done with the clear supernatant. Measurements were done three times.

Total Phenolic Content

The extraction was done by using 80% methanol (v/v) with a solid to solvent ratio of 1:10 (w/v). The samples were shaken at 300rpm and incubated at room temperature, followed by centrifugation at 10,000rpm ($\approx 9,000 \times g$) at 4 °C. The supernatant was taken and a Folin-Ciocalteu assay was performed with it. The results were presented in the form of mg gallic acid equivalents per gram (mg GAE/g).

Total Flavonoid Content

A standard spectrophotometric procedure was used to measure total flavonoid content and it was represented as mg rutin equivalents per gram (mg RE/g). All the analyses were done in three folds.

Product Development

A typical baking formulation of wheat flour, yeast, sugar, salt, fat, and water was used to prepare bread. Sugar was put into lukewarm water and yeast was put into it and left to rest till foaming. Rice bran and oat bran hydrolysates were incorporated according to the treatment design (T_0 , T_1 , and T_2) (Table 1) and mixed using a laboratory dough

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mixer for 10 minutes to obtain a homogeneous dough. The dough was then rested for 10–15 minutes and fermented at 30–32°C for 1–1.5 hours until it doubled in volume. Following the fermentation process, the dough was rolled into loaves and put in greased pans. The baking was done at 180°C within a time range of 30–35 minutes. Bread samples were cooled down to room temperature and put in polypropylene to continue with the analysis.

Table 1. Formulation of Bread Treatments with Rice and Oat Bran Hydrolysates

Treatments	Wheat Flour (%)	Rice Bran Hydrolysates (%)	Oat Bran Hydrolysates (%)
T ₀	100	0	0
T ₁	90	10	0
T ₂	90	0	10

Structural Analysis

Fourier Transform Infrared Spectroscopy

FTIR spectroscopy was done on a Vertex 70 ATR-FTIR spectrometer (4000–400cm⁻¹ with a resolution of 4cm⁻¹). The presence of functional groups and structural changes caused by the incorporation of the bran hydrolysate was determined by spectra.

Scanning Electron Microscopy Analysis

Scanning electron microscopy was done to analyze microstructural features of bread samples. Samples were put on the aluminum stubs and sputtered with gold to improve the conductivity. To assess the pore structure and morphology of the gluten network, images were taken at the suitable accelerating voltage.

Textural Analysis

Texture profile analysis was performed using a texture analyzer (TA.XT Plus, Stable Micro Systems, UK). To measure crumb hardness, the samples were subjected to a double compression cycle up to 50% deformation. Triple measurements were carried out.

Color Analysis

Color measurements were performed using a colorimeter (ST-CP60, Stalwart, China). The instrument was calibrated using a standard white tile (L* = 93.5, a* = 1.0, b* = 0.8). At three points per slice, measurements were made in the CIE L, a, b system.

Sensory Evaluation

A 9-point hedonic scale was used to analyze sensory traits such as color, texture, taste, aroma, and overall

acceptability at a controlled setting through the evaluation of a trained panel.

Statistical Analysis

Each experiment was done in triplicate (n = 3). The data were presented as mean ± standard deviation. Statistical significance was determined using one-way ANOVA followed by Tukey's post hoc test at p ≤ 0.05.

Results and Discussion

A growing interest in rice and oat bran hydrolysates has been due to their potential to improve nutritional functionality and sensory qualities of food products and phenolic compounds, proteins, and bioactive constituents that are abundant in rice and oat bran hydrolysates. The current research was aimed at assessing the outcomes of rice and oat bran hydrolysates addition in bread recipes and paying special attention to how they influence the chemical composition, antioxidant capacity, bread structure, texture, and sensory qualities.

Chemical Composition of Rice and Oat Bran

Physicochemical properties of food materials are important in influencing the quality, stability, and nutritional content of developed products. Changes in chemical, physical, and sensory properties of food systems can be caused by any alteration of the processing conditions [15]. Proximate composition analysis is one of the critical steps in the analysis of the raw materials with the aim of developing functional food. The immediate analysis of the composition of rice and oat bran in the current research was performed as per standard techniques of the AACC [16]. The mean values of moisture, ash, protein, fat, fiber, and NFE are presented in Table 2.

Table 2. Chemical Composition of Rice and Oat Bran

Component	Rice Bran	Oat Bran
Moisture	10.82±0.21	11.09±0.18
Protein	11.85±0.28	13.42±0.22
Ash	2.98±0.04	3.25±0.06
Fat	4.10±0.11	6.92±0.09
Fiber	22.64±0.30	16.08±0.25
NFE	48.61±1.12	50.24±0.95

(Values are expressed as Mean ± SD, n = 3)

Moisture content is an important factor affecting storage stability and microbial susceptibility of cereal brans. In this study, the moisture content of rice and oat bran was found to be 10.82±0.21% and 11.09±0.18%,

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respectively. The protein, ash, fat, fiber, and NFE contents of rice and oat bran were recorded as (11.85±0.28, 13.42±0.22%), (2.98±0.04, 3.25±0.06%), (4.10±0.11, 6.92±0.09%), (22.64±0.30, 16.08±0.25%), and (48.61±1.12, 50.24±0.95%), respectively. Rice bran, especially, is high in phenolic acids and lipids, but oat bran has more soluble fiber content, predominantly β -glucan and is one of the reasons oat bran has functional properties. Fiber in food is of great importance in the maintenance of the digestive system and in the regulation of lipid and glucose metabolism [17]. The differences in proximate composition can be explained by genetic differences, environmental factors, and processing.

Chemical Composition of Rice and Oat Bran Hydrolysates

Hydrolyzed samples differed significantly in terms of their chemical composition in comparison to raw bran because of structural breakdown during the hydrothermal treatment. Table 3 shows the proximate composition of rice and oat bran hydrolysates.

Table 3. Chemical Composition of Rice and Oat Bran Hydrolysates

Component	Rice Bran Hydrolysates	Oat Bran Hydrolysates
Moisture	12.40±0.19	11.87±0.16
Protein	12.18±0.21	13.56±0.17
Ash	3.12±0.05	3.44±0.06
Fat	5.18±0.12	6.73±0.10
Fiber	11.45±0.09	12.01±0.11
NFE	55.36±1.18	53.82±1.05

(Values are expressed as Mean \pm SD, n = 3)

Rice and oat bran hydrolysates were found to have moisture contents of 12.40±0.19% and 11.87±0.16%, respectively. The protein contents of rice bran hydrolysates and oat bran hydrolysates were recorded as 12.18±0.21% and 13.56±0.17%, respectively. There were also differences in the Ash content, fat content, fiber, and NFE of the 2 hydrolysates, which proved structural alteration and partial degradation of the complex macromolecules. These modifications indicate that hydrothermal treatment increased the extractability of nutrients and functional potential of the two types of bran. These findings have been reported in earlier studies, whereby hydrolysis enhanced the release of bioactive and nutritional properties in cereal-based by-products [18,19,20]. DPPH, FRAP, ABTS, TPC, and TFC tests

were used to measure the antioxidant activity of rice and oat bran hydrolysates (Table 4).

Table 4. Antioxidant Potential of Rice and Oat Bran Hydrolysates

Parameter	Unit	Rice Bran Hydrolysates	Oat Bran Hydrolysates
DPPH	% inhibition	49.37±0.22	56.42±0.18
FRAP	$\mu\text{mol Fe}^{2+}/\text{g}$	59.44±0.98	68.15±1.03
ABTS	$\mu\text{mol TE}/\text{g}$	61.28±1.14	66.73±1.09
TPC	mg GAE/g	195.40±2.31	178.26±2.12
TFC	mg RE/g	185.62±1.68	198.45±1.76

(Values are expressed as Mean \pm SD, n = 3)

In the present study, oat bran hydrolysates exhibited higher DPPH radical scavenging activity than rice bran hydrolysates, with values of 56.42±0.18% and 49.37±0.22%, respectively. Similarly, FRAP values were higher in oat bran hydrolysates (68.15±1.03 $\mu\text{mol Fe}^{2+}/\text{g}$) compared to rice bran hydrolysates (59.44±0.98 $\mu\text{mol Fe}^{2+}/\text{g}$). A similar trend was observed for ABTS radical scavenging activity, with oat bran hydrolysates showing greater antioxidant capacity (66.73±1.09 mmol TE/g vs. 61.28±1.14 mmol TE/g for rice bran hydrolysates). This apparent discrepancy in the observation of increased TPC of rice bran compared with increased antioxidant activity of oat bran may be attributed to variations in phenolic content, structure, and interaction with dietary fibers. Rice bran contains high levels of phenolic acids, including ferulic acid, p-coumaric acid and caffeic acid, γ -oryzanol and tocopherols [21]. However, antioxidant activity was comparatively higher in oat bran hydrolysates, which may be due to the higher bioactivity of avenanthramides and synergistic interactions with β -glucans. These compounds exhibit strong electron-donating capacity and efficient radical scavenging potential, which enhances performance in DPPH, ABTS, and FRAP assays despite relatively lower total phenolic content [22]. Therefore, antioxidant capacity is not only dependent on total phenolic concentration but is strongly influenced by phenolic composition, structural configuration, and interactions with dietary fiber components. Oat bran hydrolysates exhibit higher antioxidant activity due to the presence of more potent

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phenolic subclasses, particularly avenanthramides, as well as synergistic interactions with β -glucans, which enhance radical scavenging and reducing power [23].

Product Development

The findings have made it clear that both the rice and oat bran hydrolysates have both high nutritional and antioxidant potential and could be considered suitable products in the development of functional foods. Despite rice bran having a higher phenolic content, oat bran had better overall antioxidant activity, indicating complementary functional properties between the two materials.

Structure Analysis

Fourier-Transform Infrared Spectroscopy

FTIR spectra of the bread samples (T_0 , T_1 , and T_2) were recorded using a Vertex 70 ATR-FTIR spectrometer at a resolution of 4 cm^{-1} over the range of $4000\text{--}400\text{ cm}^{-1}$. The spectra of all samples are presented in Figure 1. The control sample (T_0) exhibited characteristic peaks at approximately 3275 cm^{-1} (O–H stretching), $2920\text{--}2850\text{ cm}^{-1}$ (C–H stretching of aliphatic compounds), and 1740 cm^{-1} (C=O stretching), indicating the presence of native polysaccharides, proteins, and lipids associated with the wheat flour-based matrix. In T_1 (rice bran hydrolysates), a broader O–H stretching band was observed at $3268\text{--}3385\text{ cm}^{-1}$ with higher intensity than the control, suggesting stronger hydrogen bonding interactions due to the increased release of phenolic compounds and soluble oligosaccharides from rice bran hydrolysates. The absorption bands in the region of $1628\text{--}1435\text{ cm}^{-1}$ were attributed to aromatic C=C stretching and N–O functional groups, confirming the presence of phenolic structures. In addition, strong carbohydrate-related vibrations were observed at $1095\text{--}1015\text{ cm}^{-1}$, indicating enrichment of polysaccharide-derived functional groups. A slight shift and decrease in the intensity of the OH band ($3200\text{--}3350\text{ cm}^{-1}$) with more pronounced C–O stretching vibrations in the range of $1150\text{--}1020\text{ cm}^{-1}$ were observed in T_2 (oat bran hydrolysates). This tendency is connected to the increased β -glucan level of oat bran, which adds to the enhanced carbohydrate-related interrelationships of the bread matrix. Moreover, a strong peak at $1650\text{--}1655\text{ cm}^{-1}$, corresponding to the amide I (C=O stretching), indicated increased protein-polysaccharide interactions and enhanced matrix stability. C–H bending vibrations and aromatic out-of-plane bending modes were attributed to the minor absorption bands at $700\text{--}820\text{ cm}^{-1}$ observed in all the samples. On the whole, T_1 and T_2 showed

significant structural changes as compared to T_0 . T_1 exhibited more phenolic-related absorption characteristics, and T_2 exhibited more polysaccharide and β -glucan-related vibrations. These results indicate that incorporating rice and oat bran hydrolysates modifies the molecular architecture of bread during hydrothermal processing. This modification leads to the formation of bread enriched with bioactive functional groups. The resulting product can be considered a healthier bread with enhanced nutritional value and improved antioxidant potential. The findings are also consistent with the previously documented FTIR alterations in cereal functional foods [24,25].

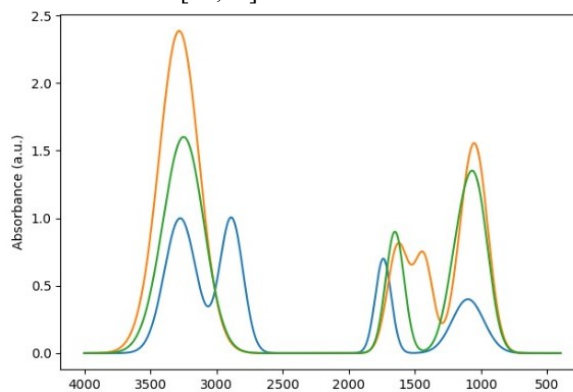


Figure 1. FTIR spectra of bread samples (T_0 , T_1 , T_2) showing changes in functional groups across $4000\text{--}500\text{ cm}^{-1}$.

Scanning Electron Microscopy

SEM is a high-resolution imaging technique used to determine the surface morphology, internal structure, and absorbency of food matrices. SEM was utilized in the current experiment to explore the difference in microstructure of the bread samples contaminated with rice and oat bran hydrolysates at various magnifications (1 , 3 and $5\mu\text{m}$). The control bread (T_0) had a well-formed and continuous gluten network with uniformly distributed gas cells which is a requirement to keep bread soft and elastic (Figure 2). This is a sign of optimum gluten formation and no interference of fibers. In T_1 (rice bran hydrolysates), a more compact and denser microstructure was found that had partially disrupted the gluten matrix. The appearance of rice bran hydrolysates resulted in the formation of irregular pore structure and surface roughness which might be attributed to intense interactions between rice-derived dietary fiber, phenolic compounds, and gluten proteins. These reactions decreased the capacity to retain gases and helped to create a more solid structural matrix. A more heterogeneous

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structure of T₂ (oat bran hydrolysates), where the pores were moderately enlarged, and the aggregation of bran particles was visible, was observed. The oat-enriched sample maintained a more uniform structure than T₁ due to the presence of soluble β -glucan, which enhanced water-binding capacity and contributed to partial stabilization of the gluten network. This resulted in improved structural integrity despite the incorporation of dietary fiber. Both treatments of hydrolysates caused observable changes in the microstructure of bread relative to the control. The hydrolysates of rice bran were more disruptive of gluten continuity and the oat bran hydrolysates aided in a more balanced structural change owing to their mixed soluble/insoluble fiber structure. These data are in line with the prior reports, indicating that the implementation of cereal bran changes the gluten network structure by interacting with fibers and protein, affecting the ultimate bread structure and quality characteristics [26, 27].

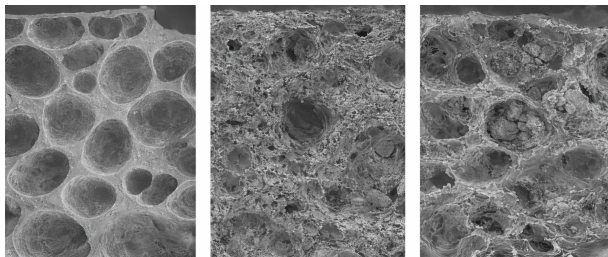


Figure 2. SEM micrographs of bread samples: (A) T₀ (control), (B) T₁ (10% rice bran hydrolysates), and (C) T₂ (10% oat bran hydrolysates).

Textual Analysis of Bread

The texture is a key quality attribute that defines consumer acceptability, structural integrity and overall eating quality of bread. A texture analyzer (Model TA.XT.Plus, Stable Micro Systems, UK) was used to determine textural properties in the current study. Measurements have been made in triplicate following storage at room temperature without considering the outermost slices to provide a consistent sample. The findings showed that there was a significant difference between treatments. The hardness of control bread (T₀) was recorded as 147.12±0.13 N, while a significant increase was observed in bread supplemented with rice bran hydrolysates (T₁), which exhibited the highest hardness value of 230.87±0.20 N (Table 5). This increase in firmness may be attributed to stronger interactions between phenolic compounds, gluten proteins, and rice-derived dietary fibers, resulting in a more compact and rigid crumb structure. The hardness of oat bran

hydrolysate-enriched bread (T₂) was 218.52±0.28 N, which, although lower than T₁, was substantially higher than that of the control. This reduction in hardness relative to T₁ is moderate, and it is possible that this level of solubility of β -glucan in oat bran enhances water retention and helps to partially soften the gluten network without affecting structural stability. There were increases in the firmness of bread with both hydrolysate treatments relative to the control, and oat bran hydrolysates had a more significant effect on the changes in the texture than rice bran hydrolysates. These findings suggest that the kind of bran hydrolysate is important in controlling the rheology of the dough and the ultimate bread texture.

Table 5. Textural Analysis of Bread

Treatments	Hardness (N)
T ₀	147.12±0.13
T ₁ (Rice bran hydrolysates)	230.87±0.20
T ₂ (Oat bran hydrolysates)	218.52±0.28

Values are expressed as Mean ± Standard Deviation (n = 3).

Color Analysis

Bread color is one of the most important quality attributes that plays a major role in the perception of consumers and their acceptability and general market worth of bakery products. Color assessment of the crust and crumb is regarded as a valuable quality control parameter in baking science because it is a measure of the degree of the Maillard reaction, pigment transformation, and efficiency of the process. In the present study, the effect of rice and oat bran hydrolysates on bread color was evaluated using the CIE L*, a*, and b* color space, as presented in Table 6. The control bread (T₀) exhibited lightness and yellow-red hue typical of wheat-based bread, with L* = 65.32 ± 0.07, a* = 8.08 ± 0.03, b* = 30.81 ± 0.05. Incorporation of rice bran hydrolysates (T₁) resulted in a significant decrease in lightness (L* = 63.45±0.02) and a concomitant increase in redness (a* = 8.45±0.04) and yellowness (b* = 33.92±0.02). This darker appearance may be associated with enhanced Maillard reaction activity and the higher phenolic content

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of rice bran hydrolysates, which promotes pigment formation during baking and intensifies crust coloration. Oat bran hydrolysate-enriched bread (T₂) showed higher lightness ($L^* = 72.85 \pm 0.05$) and a lower b* value (20.45 ± 0.07), indicating a lighter and less yellow appearance compared to both the control and rice-based formulations. This effect may be attributed to the higher β -glucan content of oat bran, which can dilute pigment concentration and modify heat-induced browning reactions during baking. The findings indicate that rice and oat bran hydrolysate have significant effects on the bread color parameters by changing the pigment interactions and thermal reaction mechanisms. These transformations eventually have a visual impact and can impact consumer acceptance of the end product.

Table 6. Color Analysis of Bread

Treatments	L (Lightness)	a (Redness)	b (Yellowness)
T ₀	65.32±0.07	8.08±0.03	30.81±0.05
T ₁ (Rice bran hydrolysates)	63.45±0.02	8.45±0.04	33.92±0.02
T ₂ (Oat bran hydrolysates)	72.85±0.05	8.24±0.07	20.45±0.07

Values are expressed as Mean ± Standard Deviation (n = 3).

Sensory Evaluation

Sensory evaluation is a scientific method of evaluating food quality properties on human perception. Sensory attributes such as color, flavor, taste, texture, aroma, and overall acceptability are used by trained panelists to assess products in terms of their visual, olfactory, gustatory, and tactile senses. To determine the sensory attributes of bread made of rice and oat bran hydrolysates, the 9-point hedonic scale was used, and the data are listed in Table 7. The control bread (T₀) had relatively lower sensory scores in all the parameters examined, where the mean scores were 7.80 ± 0.13 in color, 7.62 ± 0.26 in flavor, 7.60 ± 0.33 in taste, 7.49 ± 0.68 in texture, and 7.46 ± 0.15 in overall acceptability. Incorporation of rice bran hydrolysates (T₁) significantly improved sensory attributes, with values of 8.20 ± 0.13 (color), 8.63 ± 0.12 (flavor), 8.34 ± 0.14 (taste), 8.37 ± 0.11 (texture), and 8.58 ± 0.25 (overall acceptability).

The increased flavour and taste markers can be linked to the liberation of phenolic compounds, amino acids and volatile precursors during hydrolysis, which promote aroma formation and palatability [28].

Table 7. Sensory Evaluation of Bread (9-point hedonic scale)

Parameter	T ₀ (Control)	T ₁ (Rice bran hydrolysates)	T ₂ (Oat bran hydrolysates)
Color	7.80 ± 0.13	8.20 ± 0.13	7.99 ± 0.12
Flavor	7.62 ± 0.26	8.63 ± 0.12	7.95 ± 0.15
Taste	7.60 ± 0.33	8.34 ± 0.14	8.29 ± 0.23
Texture	7.49 ± 0.68	8.37 ± 0.11	8.33 ± 0.17
Overall Acceptability	7.46 ± 0.15	8.58 ± 0.25	8.20 ± 0.18

Values are expressed as Mean ± Standard Deviation (n = 3).

Oat bran hydrolysate-enriched bread (T₂) also exhibited better sensory performance than control with a color of 7.99 ± 0.12 , flavor of 7.95 ± 0.15 , taste of 8.29 ± 0.23 , texture of 8.33 ± 0.17 and overall acceptability of 8. The moderately balanced sensory character of T₂ can be explained by the fact that soluble β -glucan is present and enhances the feel and texture of the product, preserving the mouthfeel and texture of the product to be acceptable. Rice and oat bran hydrolysates had a positive effect on the sensory acceptability of bread, but rice bran had a better effect on improving flavor intensity, and oat bran had a better effect on improving the textural balance and the overall product acceptability.

Conclusion

The current research indicates that hydrothermal treatment is a successful method to increase the functional and nutritional characteristics of rice and oat bran by generating bioactive hydrolysates with greater food systems application. Rice bran hydrolysates had a greater total phenolic content, whereas oat bran hydrolysates had a better antioxidant activity based on the presence of more active phenolic compounds and synergistic activity of β -glucan. Structural determinations validated massive alterations in both molecular interactions and microstructure which had a

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direct impact on functional behavior in bread formulations. Addition of hydrolysates led to bread hardness, color changes, and enhanced sensory properties, and rice bran led to greater flavor and oat bran to improved texture. Both hydrolysates demonstrated potential as functional food ingredients. The research will be useful in understanding the connection between biochemical composition, structural modification, and functionality when utilizing cereal bran hydrolysates to create health-promoting and value-added food products.

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