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Analysis of Wheat Bran after Ultrasonication

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Abstract: With its enormous and significant health benefits, dietary fibre is a crucial component of the global diet. However, its average human food consumption remains lower than recommended. This study investigates the effect of ultrasonication on wheat bran (WB) chemical composition and structure. The WB, sourced from a new wheat variety, "Leningradskaya 6", was subjected to ultrasonic waves with the frequency of 19.3 kHz at 50°C for 10 minutes. The treated and untreated (control) samples were meticulously examined using a scanning electron microscope (SEM) and infrared spectroscopy to discern the changes in their structure. Both samples were analysed for their water-soluble protein concentration, free amino nitrogen (FAN) and cellulose concentration. The results revealed that the cellulose concentration was statistically the same in both samples (20.2 ± 0.013 and 19.8 ± 0.003 g $100g^{-1}$ in the control and treated samples, respectively). Water-soluble protein (WSP) and Free Amino Nitrogen (FAN) concentrations were higher in treated samples (1.65 ± 0.07 mg ml^{-1} < 2.07 ± 0.08 mg ml^{-1} and 0.116 ± 0.00003 mg ml^{-1} < 0.132 ± 0.00013 mg ml^{-1} for WASP and FAN, respectively). Peaks were located from 3429.43 to 599.86 cm-1 for treated WB and from 3367.71 to 528.50 cm-1 for control. The ultrasonic treatment enhanced the structure of the WB fibers manifested by the scanning electron microscope.

Keywords: Wheat bran, ultrasonication, SEM, FTIR

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1. Introduction

While dietary fibre exhibits enormous and significant health benefits to the global populace (Huang et al., 2018), its average share in human food remains lower than recommended (Zhu et al., 2014). Some of the benefits attributed to the consumption of dietary fibre include the promotion of general good health, such as the control of blood glucose and lipid levels, as well as the reduction of colorectal and cardiovascular diseases (Anderson & Jhaveri, 2012; Zhu et al., 2015).

However, Huang et al. (2018) asserted that the stated benefits depended on the physicochemical properties of the dietary fibre. Thus, the physicochemical properties of the dietary fibre, particularly the wheat bran, remain the focus of this study.

Wheat is nutritious and rich in fibre diet, minerals, B Vitamins, and proteins (Reisinger et al., 2013). Moreover, Onipe et al. (2015) have argued that wheat remains the second most consumed cereal after rice and is a staple food for more than one-third of the global population. Whereas previous studies have reaffirmed the high content of dietary fibre in wheat bran (Babu et al., 2018; Hu et al., 2015), its undesirable physical and chemical properties, such as gritty texture and rough mouthfeel, have limited the incorporation of the dietary fibre in food products (Hu et al., 2015). Furthermore, Foschia et al. (2013) argue that there is an occurrence of undesirable sensory changes, including texture and colour, because of the inclusion of insoluble dietary fibre in most food products. Hence, this study seeks to ascertain the effect of WB ultrasonication on its physiochemical compounds.

Although extensive research exists on various food storage and preservation technologies, there are considerable challenges in developing new technologies that can simultaneously ensure high-quality properties and long storage life of food products (Li & Niu, 2023; Palumbo et al., 2022). This calls for adopting and embracing contemporary technology to improve the physical and chemical properties of food products, thereby ensuring high quality and storage. Particularly, ultrasound technology may be applied in the food industry mostly as a processing aid for cleaning, disinfecting, and improving the chemical and physical properties of food products (Gallo et al., 2018). The reduction of particle size alters the structure, surface area, as well as functional properties of the wheat bran dietary fibres.

In previous studies, ultrasonication has contributed to changing the physical and chemical composition. In the study of Liu et al. (2022), the physicochemical properties of myofibrillar proteins (MPs) were affected by ultrasonication.

"Leningradskaya 6" is the variety of spring wheat that was created by the method of individual selection from the hybrid of the second generation 9 Ralle (Germany) x Leningradskaya 88 (Russia). Leningradskaya 6 is the new wheat variety for the North-Western region. A distinctive feature of the variety is the high content of crude gluten in the grain, more than 18%. The experiments on the study of the quality of gluten showed that the quality of gluten points to the" quality group I "with good characteristics (Lebedev & Barakova, 2018).

Therefore, this research was performed to investigate the effect of ultrasonic treatment on the WB chemical composition and structure.

2. Materials and Methods

This study was performed at the National Research ITMO University in the Russian Federation. The object of this study was wheat bran extracted from wheat "Leningradskaya 6" wheat variety. The developer of this variety and patent holder is the Leningrad Scientific Research Institute of Agriculture "Belogorka." Since 2010, the variety has been included in the state register and recommended for cultivation in the Leningrad region, Russia. Wheat was bought from a market in Saint Petersburg, Russian Federation.

2.1. Wheat bran extraction

The wheat bran (WB) was extracted by mixing wheat grains, water, and a liquid of Cellolux-A enzymes. The mixture was kept at 50 °C for 2 hours for the enzyme to hydrolyse the cellulose. After that, water was removed, and grains were dried in a hot air oven at a temperature of 105 °C. After getting dry, wheat grains were cooled down, and wheat bran was separated from the endosperm using the gain separator. Finally, sieves with different diameters were used to separate the bran from the endosperm.

2.2. Sonication of Wheat Bran

Ultrasonic processor UIP1000hdT (Hielscher Ultrasonics, Germany) was used during the ultrasonication of wheat bran. 5 g of wheat bran were weighed and transferred into a glass of 250 ml, and 100 ml of distilled water was added and mixed by a mixer. The sample was exposed to ultrasound waves using an ultrasonic processor UIP1000hdT at a frequency of 19.3 kHz and an amplitude of 60%. The glass containing the solution was placed into a plastic container with ice. Then, the tip of the sonotrode was inserted into the solution. During the treatment, the maximum temperature was 50℃, which was controlled by cooling down the solution and adding ice to the plastic container. The treated sample was exposed to ultrasound waves for 10 minutes. The power increased over time, and the final power was 185 W.

Distilled water (100 ml) was added to the control sample (5 g) of wheat bran, and the solution was mixed with a mixer for 10 minutes. Both solutions were centrifuged at 6000 rpm for 10 minutes, and the supernatant was separated from the filtrate. The filtrate was dried in the drier cabinet at 50 ℃ for 4 hours. After that, the treated and untreated (control) samples were ground using a coffee grinder Sinbo SCM-2929 (Sinbo Electronic Company, Turkey). To get 0.5 mm of the powder, the stainless-steel sieve (mesh size: 0.5 mm) was used to sift the powder.

2.3. Analysis of wheat bran

A scanning electron microscope (SEM) VEGA 3 SBH TESCAN (Tescan, Czech Republic) was used for image acquisition to study the surface, composition and particle size of both WB types. Different resolutions (100 and 200μm) and magnifications (1.10 to 5.00 kx) were applied.

The water-soluble protein concentration in WB was determined using the Bradford method (Bradford, 1976). The cellulose concentration was determined using a semi-micro method (Updegraff, 1969). Free amino nitrogen (FAN) was measured after the Ninhydrin method (Lie, 1973). Fourier transform infrared (FT-IR) spectroscopy was used to analyze wheat bran (control and treated). For the operational parameters, 400–4000 cm−1 spectral range and 60 scans in transmittance mode with a resolution of 4 cm−1 were used.

2.4. Statistical analysis

The experimental research design was used during this study. The data generated were subjected to analysis of variance (ANOVA) using Origin statistical software (version 8.1). The means of the data related to the studied traits in the treatments were compared following a t-test at an alpha (type I error) rate of 5%. The experiment was conducted in three replicates for the control and treated samples. Results were reported as means ± standard errors (mean ± SE).

3. Results

Wheat bran under Scanning Electron Microscope (SEM). The images of wheat bran under a scanning electron microscope (SEM) of both untreated (control) and treated (ultrasonically treated) samples are shown in Figures 1 and 2.

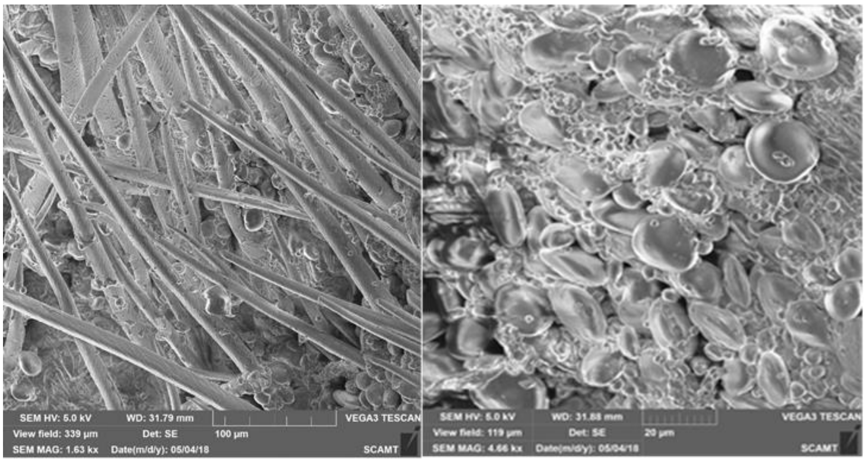

Figure 1. Image of wheat bran under scanning electron microscope (SEM) untreated ultrasonically (control sample)

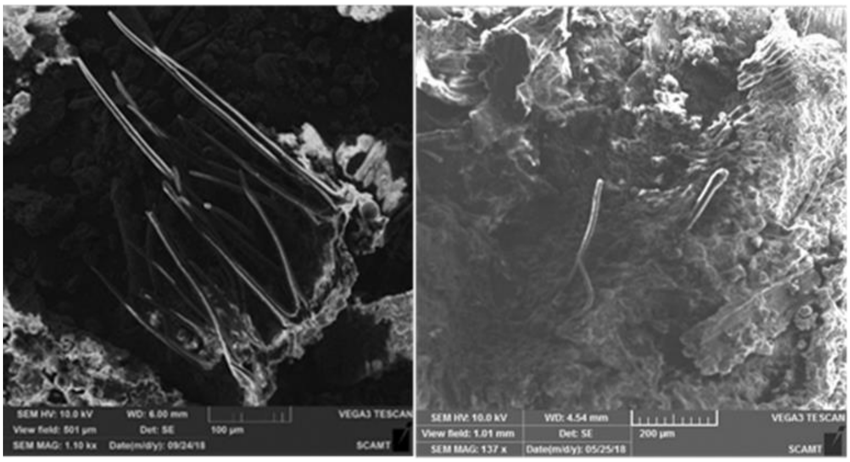

Figure 2. Image of wheat bran under scanning electron microscope (SEM) treated ultrasonically (treated sample)

Water-soluble protein fractions of wheat bran. The concentration of water-soluble protein was lesser in control (1.65 ± 0.07 mg ml^{-1}) than treated WB (2.07 ± 0.08 mg ml^{-1}) (Table 1). The means of water-soluble proteins of the two samples (untreated and treated) were significantly different ($p < 0.05$).

Table 1. Concentration of water-soluble proteins of wheat bran after and without sonication

Samples	Concentration of protein (mg ml^{-1})
Untreated wheat bran	1.65 ± 0.07 ^{a}
Treated wheat bran	2.07 ± 0.08 ^{b}

"The results represent the means of wheat bran untreated and treated with ultrasonication treatment, and their standard error values (±SE). Data in the same column with different letters are significantly different ($p < 0.05$)."

Free Amino Nitrogen (FAN) content of Wheat bran. The concentration of free amino nitrogen in untreated WB was lower (0.116 ± 0.00003 mg ml^{-1}) than in the treated WB (0.132 ± 0.00013 mg ml^{-1}). The means of FAN of the two samples were significantly different ($p < 0.05$). (Table 2).

Table 2. The concentrations of free amino nitrogen content in wheat bran

Samples	Concentration of FAN (mg ml^{-1})
Untreated wheat bran	0.116 ± 0.00003 [a]
Treated wheat bran	0.132 ± 0.00013 [b]

"The results represent the mean of wheat bran untreated and treated with ultrasonication treatment and their standard error values (±SE). Data in the same column with different letters are significantly different ($p < 0.05$)."

Cellulose Concentration in Wheat Bran. The concentration of cellulose in the control sample was 20.2 ± 0.013 and 19.8 ± 0.003 g 100g-1 in the treated sample. The difference in cellulose concentration of both WBs was not statistically significant ($p > 0.05$); however, the ultrasonic treatment had a slight effect on decreasing the cellulose concentration by 0.4 g 100g-1 compared to the control WB sample (Table 3).

Table 3. The concentration of cellulose in control and ultrasonic-treated wheat bran samples

Samples	The concentration of cellulose (g 100g^{-1})
Untreated wheat bran	20.2 ± 0.013 [a]
Treated wheat bran	19.8 ± 0.003 [a]

"The results represent the mean of wheat bran untreated and treated with ultrasonication treatment and their standard error values (±SE). Data in the same column with different letters are significantly different ($p < 0.05$)."

Fourier Transform Infrared Spectroscopy (FTIR). The interactions between the molecules of WB (ultrasonic treated and control/untreated) were studied. Peaks were located from 3429.43 to 599.86 cm-1 for treated WB and from 3367.71 to 528.50 cm-1 for control (Fig. 3). Many peaks were observed between 1600 and 400 cm-1 for both samples.

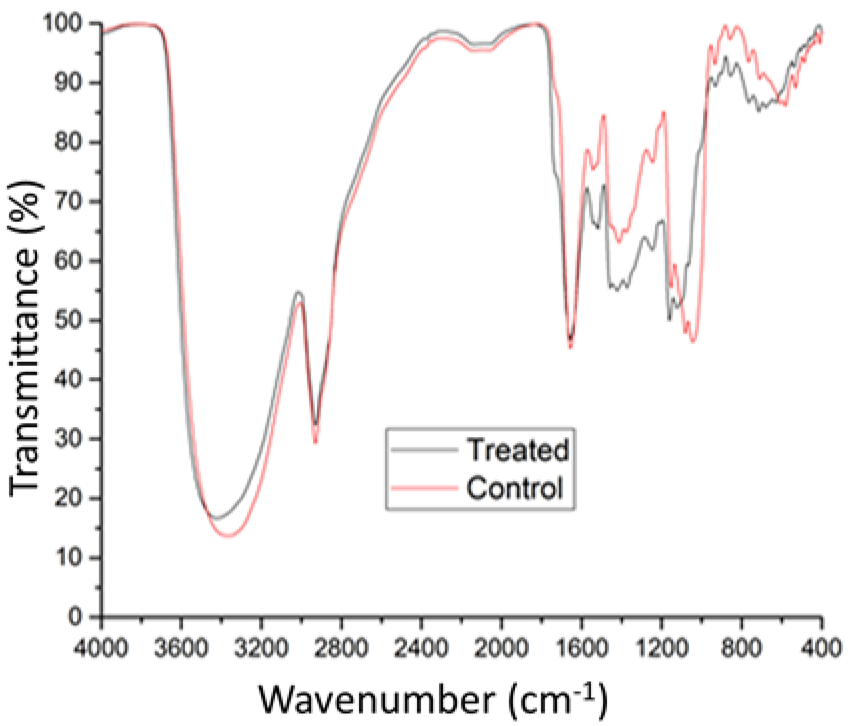

Figure 3. FTIR spectrum of wheat bran treated with ultrasonication and untreated wheat bran (control)

4. Discussion

4.1. Wheat bran under Scanning Electron Microscope (SEM)

The bran layers of the wheat grain consist of three separate layers: viztesta, aleurone and pericarp. The innermost layer of the bran is the aleurone layer, which contains a rich stock of lignans and proteins with a balanced amino acid content, bioactive compounds, phytic acid, antioxidants, vitamins and minerals. The middle layer is the viztesta layer. Dietary fibres are located in the pericarp; the latter has inner and the outer part (Onipe et al., 2015)

Referring to Figures 1 and 2, the ultrasonic treatment has contributed to the reduction of particle size. The reduction of particle size alters the structure, surface area, as well as functional properties of the WB dietary fibres. Particle size plays a vital role in controlling several events occurring in the digestive tract, i.e. transit time, fermentation, and faecal excretion. The particle size range depends on the type of cell walls present in the foods and degree of their processing. The particle size of fibre may vary during transit in the digestive tract as a result of chewing, grinding and bacterial degradation in the large intestine (Dhingra et al., 2012).

4.2. Water-soluble protein fractions of wheat bran

Ultrasonication affects proteins. The concentration of proteins in untreated WB was less than that in treated WB. Treating WB with ultrasound may increase protein solubility.

WB contains 13–18% of proteins, which can be considered a possible source for protein extraction. Those proteins are surrounded by a polysaccharide matrix, which prevents their solubility (Balandrán-Quintana, 2015). Osborne classification shows that proteins from the outer and intermediate layers of WB, called albumins, are water-soluble (Luna-Valdez et al., 2017). According to the study of Chaquilla-Quilca et al. (2018), the aleurone layer comprises 57% of globulin-type storage proteins, while 16.2% are involved in carbohydrate metabolism and 17.6% in defense pathways. Based on the results, the ultrasound has increased the water solubility of WB proteins (albumins). This can increase the presence and accessibility of proteins to probiotics.

4.3. Free Amino Nitrogen (FAN) content of Wheat bran

FAN is the general measure of these essential nutrients, constituting the nitrogen that microorganisms can assimilate during fermentation. Free Amino Nitrogen (FAN) is defined as the sum of the individual amino acids, ammonium ions, and small peptides (di- and tripeptides). FAN is the degradation product of the proteins in raw materials such as barley, malted barley, sorghum, and wheat. The quantity of basic amino acids in the globulin, particularly arginine and lysine, exceeds that of most other vegetable proteins which have been studied.

The bran prolamin contains about twice as much basic nitrogen as that recorded for gliadin, although its total nitrogen content was about 2% lower. The free amino nitrogen of the bran prolamin was found to be practically three times that of gliadin. It has been shown that the free amino nitrogen of many proteins corresponds closely to one-half of the lysine nitrogen. It is noted that the ratio of free amino nitrogen to total nitrogen in the case of bran prolamin is nearly three times that found in gliadin (Hartley, 1915).

4.4. Cellulose Concentration in Wheat Bran

WB is a promising source of dietary fibre rich in dietary fibre and unsaturated fatty acids. WB contains 36.5–52.4% total dietary fibre (TDF), and 35.0–48.4% is insoluble DF (cellulose, hemicellulose, and lignin), whereas 1.5–4.0% is soluble DF (SDF) (Vitaglione et al. 2008). Cellulose is the major component of dietary fibre; its mechanical strength, resistance to biological degradation, low aqueous solubility and resistance to acid hydrolysis result from hydrogen bonding within the microfibrils (Dhingra et al. 2012). Insoluble dietary fibres, including cellulose, are characterised by their porosity, low density, and ability to increase faecal bulk and reduce intestinal transit time (Elleuch et al., 2011).

4.5. Fourier transform infrared spectroscopy

In wheat bran (control/untreated and ultrasonic treated), O–H (hydroxy compound) at 3400 cm^{-1}, N–H (amino group) at 1716 cm^{-1}, C –H (acetylenic compound) at 2940 cm^{-1}, C≡C (alkyne group), C=C (aromatic ring) at 1645 cm^{-1}, C=N (imino) at 1658 cm^{-1}, C=O (carbonyl compound) at 1737 cm^{-1} and others are present (Ahmad et al. 2010; Limberger-Bayer et al. 2014). The O–H, N–H, C–H, C≡C, C=C (aromatic ring) and C=N spectral bands above 1600 cm^{-1} were relatively similar for both samples. However, there was a difference between control and ultrasonically treated WB in the spectral bands below 1600 cm^{-1}. For spectral bands between 1600 and 1000 cm^{-1}, the transmittance of control was higher than that of treated WB.

A big difference was observed (Figure 3) in the range of 1250–1050 cm^{-1} (C=O, carbonyl compound) (Smith 1999, Li-Chan et al. 2002). Vibration between 1100 and 1200 cm^{-1} is attributed to hemicellulose and cellulose. The interactions between the molecule and molecular structure of the component affect the quality of food and agricultural products (Zhang, 2023).

5. Conclusions

Based on this study's results, ultrasound treatment affected the physicochemical parameters of WB. This statement is emphasized by the images taken using a scanning electron microscope and the effects of spectroscopy showing the functional groups of two samples.

Based on the previous results of this study, intensive ultrasonication can have a promising effect on improving WB physicochemical attributes. The ultrasonic treatment enhanced the structure of the WB fibers manifested by the scanning electron microscope. Additionally, water-soluble protein and free amino nitrogen concentration increased significantly in the WB treated ultrasonically, compared to the control sample. The latest structural and chemical alterations in the WB can promote the growth of probiotics by providing optimal environmental growth conditions (pH and essential nutrients). Thus, the WB treated with ultrasonic can be considered an ideal matrix for fermented dairy analogues production. However, further studies are required to effectively evaluate the effect of ultrasonic treatment on WB physicochemical parameters, probiotics growth rates and viability. Further studies on the impact of ultrasonication on other types of proteins in WB are also needed.

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Data availability statement: the raw data supporting the conclusions of this article will be made available by the authors upon reasonable request

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