

Exploring microstructural integrity and functional attributes of cookies enriched with a composite blend of water chestnut and barley flour

Majid Hussain¹, Taha Rababah^{2*}, Tayyaba Akram¹, Ahmad Alsaad³, Muhammad Azam^{4*}, Mohammed I. Saleh⁵, Ali Almajwal⁶, Numan AL-Rayyan⁷, Bandar N. Hamadneh⁸

¹Faculty of Food Science & Nutrition, Bahauddin Zakariya University, Multan, Pakistan; ²Department of Nutrition and Food Technology, Jordan University of Science and Technology, Irbid, Jordan; ³Physics Department, College of Science, Imam Mohammad Ibn Saud Islamic University (IMSIU), Riyadh 11623, Saudi Arabia; ⁴National Institute of Food Science and Technology, University of Agriculture, Faisalabad, Pakistan; ⁵Department of Nutrition and Food Technology, School of Agriculture, The University of Jordan, Amman, Jordan; ⁶College of Applied Medical Sciences, King Saud University, Saudi Arabia; ⁷National Agricultural Research Center, Al Baqa'a, Jordan; ⁸Department of Nutrition and Dietetics, Faculty of Agriculture, Ajloun National University, Ajloun, Jordan

*Corresponding Authors: Muhammad Azam, National Institute of Food Science and Technology, University of Agriculture, Faisalabad 38000, Pakistan. Email: muhammadazamfst@yahoo.com; Taha Rababah, Department of Nutrition and Food Technology, Jordan University of Science and Technology, P.O. Box 3030, Irbid 22110. Email: trababah@just.edu.jo

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Abstract

This study developed functional cookies using composite flours of raw/commercial water chestnut (RWCF/CWCF) and barley flour (BF). Compositional analysis showed CWCF with higher protein (6.12%) and BF with elevated fiber (4.99%) and ash (3%). Functional attributes revealed CWCF's superior water absorption (166.67%) and swelling (276%) capacities. The texture analysis for the cookies showed increased hardness values (N) 841, 935, 1128, 1208, 1214 for WBF-B, WBF-A, CWCF, WBF-C, BF, respectively, when compared with control (410 N), while cookies with WBF-B blend (CWCF + BF) maintained standard color parameters and achieved the optimal sensory scores in color, texture, crispiness, and taste, alongside a homogenized, smooth, and fluffy microstructure through scanning electron microscopy (SEM). Infra-red spectrum revealed C-H, C=O, and C-N stretch in all treatments while little variation was found at nitro compound and alkynes. Conclusively, the results declared WBF-B cookies as a nutritious, palatable option with enhanced antioxidant potential from natural starch, flavonoids, and vitamins to manage various health disorders.

Keywords: Barley; Chestnut; Functional properties; Cookies formation; SEM; Infra-red spectrum; Sensory evaluation

Introduction

Cookies are an important product of the baking industry in the human diet, usually consumed with tea, and also used as weaning foods for infants (Soni *et al.*, 2018). Soft wheat flour (WF) has been the major ingredient used in the production of cookies, but due to natural processes as well as anthropogenic factors, global warming and climate change are the major and important environmental issues that are widely affecting crop production, especially wheat (Paymard *et al.*, 2018).

It is now widely acknowledged that the global climate is changing at an unprecedented rate, and one of the sectors most at risk from this change is agriculture (Demirhan & Bayraktar, 2025). Globally, 60% of wheat growing areas are seriously affected by global warming. Currently, 15% of wheat productivity is affected by drought conditions (Elahi *et al.*, 2022). Climate change has also impacted productivity of wheat in Australia and Mexico, resulting in 2.85 billion dollars of wheat loss every year. Similarly, climate change adversely affected the wheat productivity in Pakistan and reduced the crop yield up to 7.4% with an increase of 1°C during sowing time (Ahmad *et al.*, 2014). As wheat is a staple food, it is important for Pakistan's food security. A decline in its production in any crop year may lead to the food insecurity (Ali *et al.*, 2017). Global warming poses a serious concern for the bakery industry. To address this, non-conventional resources must be explored to develop healthier food options and promote sustainability. Global warming poses a serious concern for the bakery industry. To address this, non-conventional resources must be explored to develop healthier food options and promote sustainability (Ahsan *et al.*, 2023). For this reason, composite flours are better utilized for cookie production, relatively rich in essential nutritional and functional components, resulting in good eating quality and prolonged shelf-life (Ahmed *et al.*, 2025; Chopra *et al.*, 2018; Ikuomola *et al.*, 2017). The incorporation of other food ingredients in addition to flours significantly increased essential amino acids, dietary fiber (Lu *et al.*, 2025), and enhanced the flavonoid and phenolic contents of the cookies making them more appealing to consumers (Saeed *et al.*, 2023; Krajewska & Dziki, 2025).

Water chestnut (*Trapa natans*) is not only rich in starch and protein but also contains vitamins B, E, A, and ascorbic acid. Water chestnut possesses strong antioxidant, antimicrobial, and anticancer activities, which have been attributed to their bioactive components, such as polyphenols, flavonoids, and alkaloids. In addition, the cookies prepared from water chestnut flour (WCF) blend received positive feedback from the sensory panel, having good-quality features (Jabeen *et al.*, 2021; Shafi *et al.*, 2016).

Barley (*Hordeum vulgare* L) is the world's most important cereal, enriched with starch, protein, β -glucan, dietary fiber, antioxidants, and B vitamins, and can be better used for cookie development in combination with WF (Aly *et al.*, 2021; Hammes *et al.*, 2005).

Keeping in view the climatic impact on lowered wheat yield, the scope of the current study was to enhance the utility of water chest nut and barley flour (BF) in bakery products and further subjected to evaluate physicochemical, functional, dimensional, morphological (SEM, FTIR) and sensory properties of developed cookies. The research helped to understand their potential and effectiveness as substitutes for WF and optimizing alternative composite flour utilization in bakery products.

Material and Methods

Procurement of raw material

Water chestnut and barley grains were procured from the local market of Khanewal and Multan, Punjab, Pakistan and ground to prepare fine flour. All the other chemicals and reagents were of analytical grade.

Preparation of water chestnut and BF

The kernels of water chestnut and barley were cleaned and washed to get rid of impurities like dust particles, injured nuts/seeds, other crop seeds, weed seed, and metals. Any diseased and bruised fruits were also discarded. The kernel of water chestnut was peeled off to obtain the internal white nut. The samples of water chestnut were spread homogeneously on perforated trays (stainless steel). The sample was dried in an oven at 70°C till the constant weight (Singh *et al.*, 2008). The flour of water chestnut and barley as a whole were ground to fine flour, packed in poly bags, and stored at room temperature until use.

Determination of chemical composition

Compositional analysis of CWCF and BF were determined by their respective methods (AOAC, 2000). Moisture and ash were determined placing the samples at 105°C in hot air oven and at 550°C in muffle furnace. The fat and protein were quantified using Soxhlet and Kjeldahl digestion methods. The fiber contents of flours were evaluated through acid and base digestion method.

Minerals quantification

The wet digestion of dried and defatted CWCF, RWCF, and BF composite was performed with HNO_3 and HClO_4 till clear solution. The solution was diluted and subjected to atomic absorption spectrophotometer and flame photometer to measure out the macro and micro elements (AOAC, 2000).

Functional analysis of flour

The water absorption capacity (WAC) of CWCF, RWCF, and BF was examined by using the method of Enidiok *et al.* (2008). Flour (0.5 g) was dissolved in 10 mL distilled water using centrifuge tubes and vortexed for a small period of 30 s. It was allowed to stand for 30 min at room temperature and centrifuged for 25 min at 3000 rpm. After centrifugation, it was filtered and the volume recovered was precisely computed. The difference in initial volume added and final volume recovered was determined, and the results were described as milliliters of water absorbed per gram of sample. The swelling index (SI) of CWCF, RWCF, and BF was examined by using 25 g of flour in a measuring cylinder of 210 mL. Add distilled water (150 mL) and let it to stand for about 4 h before detecting swelling level. The SI was calculated by the following formula:

$$SI \% = \frac{\text{Volume after soaking} - \text{Volume before soaking}}{\text{Weight of sample}} \times 100 \quad (\text{Eqn. 1})$$

The gel acquired from SI was used to reckon the SC with the help of the given formula:

$$SC \% = \frac{\text{Weight of wet gel}}{\text{Weight of sample}} \times 100 \quad (\text{Eqn. 2})$$

Product development

The development of widely/commonly accessible “cookies” was accomplished with the supplementation of commercial water chestnut and BF. The proposed samples of cookies were made by adopting a standard recipe with some modifications in the method No. 10-50D (AOAC, 2000). The composite flour of WCF and BF was used to prepare cookies with different concentrations: WF, Wheat flour 100%; BF, Barley flour 100%; CWCF, Commercial water chestnut flour; WBF-A, Water chestnut flour 50%, Barley flour 50%; WBF-B, Commercial water chestnut flour 75%, Barley flour 25%; WBF-C, Commercial water chestnut flour 25%, and Barley flour 75%.

Dimensional characteristics of cookies

The weight (W) of each batch of cookies was measured by an electronic weighing balance in the analysis lab of the Faculty of Food Science and Nutrition BZ, University Multan, Pakistan. The method used was in accordance with Shafi *et al.* (2016). The thickness (T) of cookies of different concentrations of composite flour were measured by a digital vernier caliper with the accuracy of 0.01 mm. The diameter (D) of each batch of cookies was measured with the aid of a measuring scale by placing it edge to edge from each side, and the data were calculated (Singh *et al.*, 2011). The spread ratio of the samples “cookies” was computed by using the formula D/T .

Color analysis

Color characteristics of cookies made up of commercial water chestnut and BF blends were measured using a Color Tec-PCM SN 3001378 U.S.A. Color values, L^* , a^* , and b^* , were recorded, with each value being the average of three measurements (Mamat *et al.*, 2010).

Texture analysis of cookies (hardness)

Textural properties of the final products were investigated using Texture analyzer (TA X2 plus Exponent Stable Micro System, Haslemere, UK) (Singh *et al.*, 2011). The hardness was measured using a three-point Bending Rig and 5 kg load cell. The distance between two beams was 60 mm. Another identical beam was brought down from above (pretest speed of 1.0 mm/s, test speed of 3.0 mm/s, posttest speed of 10.0 mm/s, and distance of 5 mm) to contact the cookie. The downward movement was continued until the cookie broke. Peak force was reported as hardness (g). Three representative samples were analyzed from each formulation and the average was calculated.

SEM analysis

SEM analysis of baked cookies was carried out using a JSM-6400 scanning electron microscope (JEOL, Tokyo, Japan). Prior to examination, samples were sputter coated with gold-palladium to render them electrically conductive using HUMMLE VII Sputter Coating Device (Anatech Electronics, Garfield, N.J., USA). The micrographs were taken at magnification of 200×, 250×, 500×, and 1000× for the baked cookie samples (Singh *et al.*, 2011).

Fourier transform infrared spectroscopy (FTIR) Analysis

The FTIR spectra of the samples were obtained using a FTIR spectrophotometer [Thermo Scientific Smart iTR, (Attenuated Total Reflectance), Thermo Fisher Scientific Inc. USA]. Background spectra of the instrument were collected before samples (0.5 g of each cookies/flour) were mounted on the mirror of the instrument, and the spectra were recorded with characteristic peaks in wave numbers from 600 to 4000 cm^{-1} at 16 runs per scan (Wetzel & LeVine, 1999).

Sensory analysis

A panel of 20 experts consisting of faculty members and senior students (age 25–40 years) in the Faculty of Food Science and Nutrition, BZU, Multan. Sensory attributes like texture, taste, crispiness, color, and overall acceptability of the cookies of composite flour (water chestnut and barley) was tested on a – nine-point Hedonic Scale (Sarkar *et al.*, 2025) where 1 denoted extreme dislike, and 9 denoted extreme like. Testing of sensory characteristics was performed in a panel room that was completely free from mixing of daylight, noise, and chemical/food odor (McWatters *et al.*, 2003).

Statistical analysis

The data were subjected to statistical analysis and presented in the form of means. Data were analyzed based on a two-factor factorial design, analysis of variance of different parameters were calculated, and significance of the data were measured ($p < 0.05$). All statistical procedures were performed using the statistical software (Statistix 8.1, USA) (Montgomery, 2017).

Results and Discussion

The research investigated the chemical composition, mineral content, functional properties, dimensional characteristics, texture, sensory attributes, and chemical/morphological analysis of cookies made from composite flour with varying water chestnut and barley concentrations. Results reveal significant variations in all parameters, indicating potential impacts on nutritional profile, baking properties, and sensory appeal of the cookies. These findings offer insights for optimizing composite flour formulations to enhance both nutritional value and consumer acceptance of baked products.

Chemical composition of flour

The chemical composition analysis of the composite flour (water chestnut and barley) (Figure 1) indicated varying moisture content, with the highest observed in RWCF (8.27%), followed by CWCF (7.97%), and the lowest in BF (7.87%) (Table 1). Similar moisture content findings align with previous studies on water chestnut flour (Mir *et al.*, 2015a; Singh *et al.*, 2011).

Ash content was the highest in RWCF (3.33%) and BF (3.00%), while CWCF exhibited the lowest ash content (2.33%), consistent with prior research on cassava and WCF (Bala *et al.*, 2015; Shafi *et al.*, 2016). BF demonstrated the highest fat content (1.80%), followed by RWCF (1.50%), and CWCF with the lowest fat content (1.36%), corroborating findings in biochemical composition studies (Alfasane *et al.*, 2011; Sarabhai & Prabhasankar, 2015). Protein content was the highest in RWCF (6.78%), followed by CWCF (6.12%), and the lowest in BF (4.85%), consistent with protein content variations observed in WCF blends (Shafi *et al.*, 2016, 2017). BF exhibited the highest fiber content (4.99%), followed by RWCF (1.94%), with CWCF having the lowest fiber content (1.39%), aligning with previous studies on CWCF's compositional and functional properties (Ahmed *et al.*, 2016). The comprehensive analysis underscores the importance of understanding the compositional variations in composite flour, providing valuable insights for nutritional profiling and product development. These findings contribute to the body of knowledge regarding the utilization and optimization of water chestnut and barley in composite flour formulations for enhanced food quality and functionality.

Mineral composition of flours

The mineral nutrition is categorized into two groups, "Macro" and "Micro," based on their requirement in the human body (Newmark *et al.*, 2004). The kernel of water chestnut is an excellent source of amino acids, starch, vitamins, and minerals, emphasizing its use in the preparation of healthy food (Puste, 2004). The observed data in Table 2 showed a wide difference in mineral content among CWCF, RWCF, and BF.

The analysis revealed that the maximum sodium (Na) content was found in CWCF, followed by RWCF with an average value of 166.67 mg/L and 126.67 mg/L, respectively. Calcium (Ca) contents, i.e., 79223 mg/L, were found to be higher in raw water chestnut flour (RWCF), while the minimum calcium (Ca) content was found in BF with an average value of 27000 mg/L. Regarding potassium (K) content, it was found to be higher in BF with an average value of 169667 mg/L, while the minimum



Figure 1. Flour samples of (A) barley and (B) water chestnut flour.

Table 1. Moisture, ash, fat, protein and fiber contents (%) of water chestnut and barley flour.

Treatments	Moisture (%)	Ash (%)	Fat (%)	Protein (%)	Fiber (%)
CWCF	7.96 ± 0.25 ^a	2.33 ± 0.28 ^b	1.36 ± 0.06 ^b	6.12 ± 0.29 ^b	1.94 ± 0.08 ^b
RWCF	8.26 ± 0.57 ^a	3.33 ± 0.28 ^a	1.50 ± 0.10 ^b	6.78 ± 0.18 ^a	1.94 ± 0.08 ^b
BF	7.86 ± 0.32 ^a	3.00 ± 0.50 ^{ab}	1.80 ± 0.10 ^a	4.85 ± 0.21 ^c	4.99 ± 0.33 ^a

RWCF, Raw water chestnut flour; CWCF, Commercial water chestnut flour; BF, Barley Flour.

potassium (K) content was observed in CWCF with an average value of 33013 mg/L. The findings of the present study are related to Gupta *et al.* (2011), who analyzed the effect of BF and freeze–thaw cycles on the nutritional and functional properties of cookies. Mineral elements must be supplied from food because the body cannot synthesize them (White & Broadley, 2007). The analysis of observed data regarding micro mineral contents like Iron, Zinc (Zn), Copper (Cu), and (Pb) of CWCF, RWCF, and BF is presented in Table 2. There is a great variation in the micro mineral content of CWCF, RWCF, and BF. The results pertaining to iron content showed the

maximum value, i.e., 3724.7 mg/L in RWCF, while the minimum iron content was observed in CWCF with an average value of 2366.7 mg/L. Maximum zinc content was observed in BF, i.e., 1516.7 mg/L, while the minimum zinc content, i.e., 590 mg/L, was observed in CWCF. In the case of copper content, the higher value, i.e., 203.33 mg/L, was observed in BF, while the flour of commercial water chestnut was found to be poor in copper content with an average value of 43.59 mg/L. In the case of Pb content, only BF was tested, which was 5083.3 mg/L. The results of the current study are in accordance with Škrbić & Cvejanov (2011), who studied the impact of BF on the

nutritional composition content of heavy elements and physical properties. The present research results are in line with Shafi *et al.* (2017), who observed the influence of particle size of WCF on nutraceutical potential and quality of Indian flatbreads.

Functional analysis of composite flour

The results of the functional analysis of composite flour are depicted in Table 3. It demonstrated that RWCF and CWCF have the same WAC with an average value of 166.67%, which is higher than the value of BF, which was 60.00%.

The high observed value of WAC may have been due to the nature of starch and possible contributions to water absorption by the cell wall materials, which were not removed completely. Variation in the WAC between the flours might be due to differences in protein structures and the presence of hydrophilic carbohydrates (Sarabhai & Prabhasankar, 2015). The current study results are in line with Yadav *et al.* (2014), who reported a WAC value of 131% in their studies on the suitability of WF blend with WCF for noodle making. Present research results are in accordance with the research findings of Jan *et al.* (2015). The observed values in Table 3 reveal that the maximum values of swelling capacity were observed in CWCF followed by RWCF with an average value of swelling capacity of 276.00% and 251.33%, respectively. The minimum

value of swelling capacity was observed in BF with an average value of 232.67%. The maximum values of SI are observed in BF with an average value of SI of 1.64%. Barley has a high value because it is rich in fiber content. The minimum value of SI was observed in RWCF with an average value of 0.44%, and CWCF has a mean value of 0.46%. The current study results are in accordance with Awolu (2017), whose values are similar to the current values that he declared in the optimization of the functional characteristics, pasting, and rheological properties of pearl millet-based composite flour. The functional analysis provides crucial insights into the water absorption and swelling properties of composite flour, which are essential for various food applications. These findings underscore the potential of water chestnut and barley-based composite flours in enhancing the texture and overall quality of food products.

Dimensional characteristics of cookies

Cookies prepared from water chestnut, barley, and WF (Figure 2) exhibited significantly different observations in terms of weight, thickness, diameter, and spread ratio, as shown in Table 4.

Cookies prepared from water chestnut, barley, and WF exhibited significantly different weights, with the highest recorded in WBF-C (13.92g) and the lowest in WBF-A (11.67g). Cookies made with 100% BF and 100% CWCF

Table 2. Analysis of variance of micro minerals' contents (Fe, Zinc, Cu., Pb, and Ni) of water chestnut and barley flour.

Treatments	Iron (mg/L)	Zn (mg/L)	Cu (mg/L)	Pb (mg/L)	Ni (mg/L)
CWCF	2366.7 ^b	590.00 ^b	43.59 ^b	00.00	00.00
RWCF	3714.7 ^a	790.00 ^b	50.82 ^b	00.00	00.00
BF	2583.3 ^b	1516.7 ^a	203.33 ^a	5083.3 ^a	00.00
LSD ($p < 0.05$)	937.10	391.22	29.19	7109.5	00.00
Significance	*	*	*	**	–

RWCF, Raw water chestnut flour; CWCF, Commercial water chestnut flour; BF, Barley Flour.

Table 3. Water absorption capacity, swelling capacity, and swelling index (%) of water chestnut and barley flour.

Treatments	WAC (%)	SC (%)	SI (%)
CWCF	166.67 ± 57.73 ^a	276.00 ± 4.00 ^a	0.47 ± 0.02 ^b
RWCF	161.39 ± 51.23 ^a	251.33 ± 1.15 ^b	0.44 ± 0.04 ^b
BF	60.00 ± 20.00 ^b	232.67 ± 1.15 ^c	1.64 ± 0.04 ^a

RWCF, Raw water chestnut flour; CWCF, Commercial water chestnut flour; BF, Barley flour.

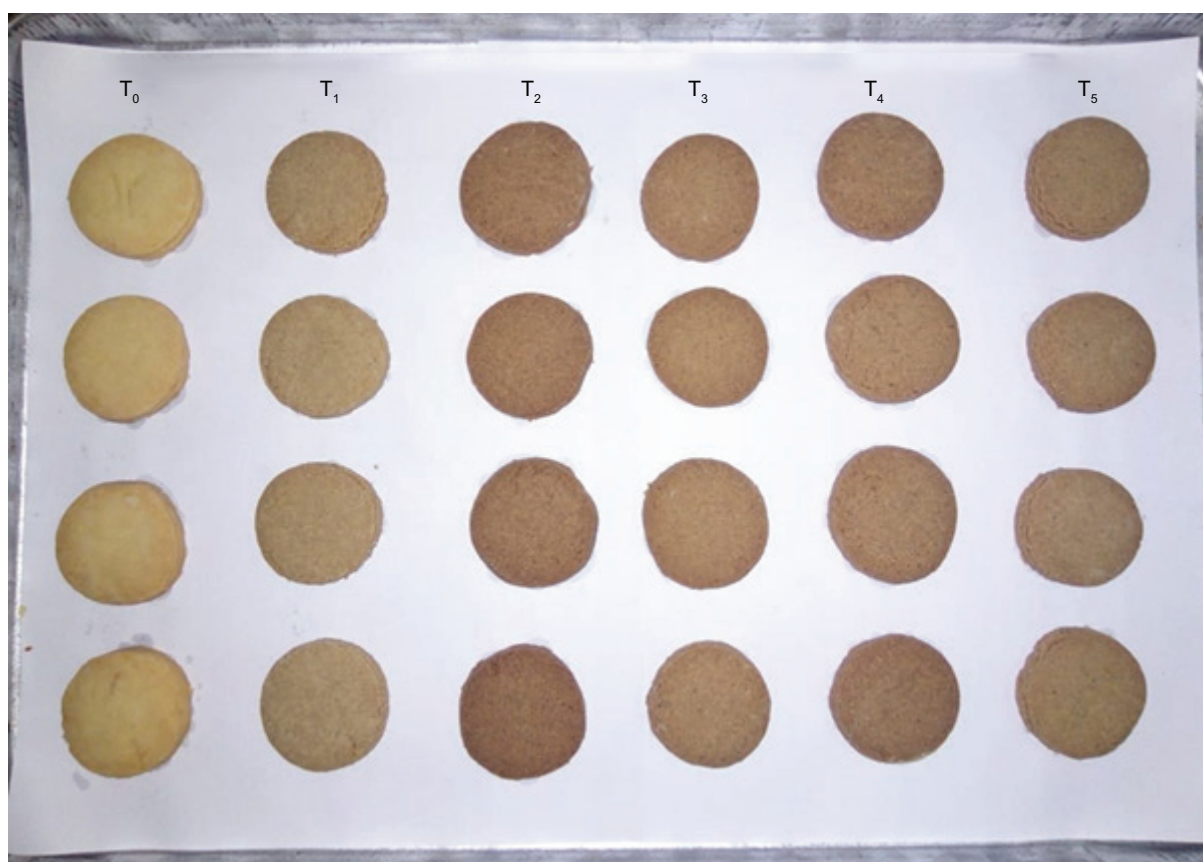


Figure 2. Baked cookies prepared with various treatments, namely, (T_0) WF = wheat flour 100%; (T_1) BF = Barley flour 100%; (T_2) WCF = Water chest nut flour 100%; (T_3) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T_4) WBF-B = Water chestnut flour 75%, Barley flour 25%; and (T_5) WBF-C = Water chestnut flour 25%, Barley flour 75%.

Table 4. Weight, thickness, diameter, and spread ratio contents of water chestnut and barley flour cookies.

Treatments	Weight (g)	Thickness (mm)	Diameter (mm)	Spread ratio	Hardness (N)
T_0	11.98 ± 0.55 ^b	10.40 ± 0.94 ^{bc}	91.65 ± 1.27 ^a	8.800 ± 0.85 ^a	410.0 ± 14.14 ^e
T_1	13.66 ± 0.35 ^a	9.82 ± 0.17 ^c	86.70 ± 2.73 ^{bc}	7.975 ± 0.49 ^{abc}	1214.0 ± 5.65 ^a
T_2	13.32 ± 0.35 ^a	12.70 ± 0.21 ^a	85.30 ± 2.51 ^c	6.950 ± 0.53 ^c	1128.0 ± 8.48 ^b
T_3	11.67 ± 0.38 ^b	10.97 ± 0.26 ^b	89.78 ± 1.618 ^{ab}	8.475 ± 0.70 ^{ab}	935 ± 2.12 ^c
T_4	11.92 ± 0.68 ^b	12.05 ± 0.42 ^a	88.35 ± 2.22 ^{bc}	7.700 ± 0.93 ^{bc}	841 ± 2.82 ^d
T_5	13.92 ± 0.62 ^a	12.50 ± 0.77 ^a	87.80 ± 2.49 ^{bc}	7.050 ± 0.76 ^c	1208.5 ± 2.12 ^a

(T_0) WF = wheat flour 100%; (T_1) BF = Barley flour 100%; (T_2) WCF = Water chest nut flour 100%; (T_3) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T_4) WBF-B = Water chestnut flour 75%, Barley flour 25%; (T_5) WBF-C = Water chestnut flour 25%, Barley flour 75%.

had average weights of 13.66 g and 13.32 g, respectively. Cookies made with 100% WF and WBF-B had average weights of 11.98 g and 11.92 g, respectively. These findings align with the work of Arshad *et al.* (2007), who studied the nutritional assessment of cookies supplemented with defatted wheat, observing variations in cookie weight with different flour percentage levels.

Cookies prepared from water chestnut, barley, and WF also exhibited significantly different scores in terms of thickness, as mentioned in Table 4. The maximum score for thickness (12.70) was recorded in treatment (CWCF), where cookies were prepared with 100% CWCF. In treatment WBF-C, cookies were treated with 25% CWCF and 75% BF, with an average thickness

value of 12.50 mm. In treatment WBF-B, cookies were treated with 75% CWCF and 25% BF, with an average thickness value of 12.05 mm. The lowest thickness value (9.83 mm) was recorded in treatment BF, where cookies were prepared with 100% BF. Treatment WBF-A, with cookies prepared with 50% CWCF and 50% BF, had a thickness score of 10.98 mm. The average thickness values of treatments WBF-A and WF were 10.97 mm and 10.40 mm, respectively. These results match with Sarabhai & Prabhasankar (2015), whose results were 12.4, similar to the results of the CWCF treatment. The findings of this project also align with Gupta *et al.* (2011), who studied the effect of BF on textural, nutritional, and functional properties of cookies.

The maximum diameter (91.65 mm) was recorded in the cookies treated with 100% WF due to the standard of cookies. According to different treatments, the maximum diameter (89.78 mm) was recorded in the cookies prepared with 50% CWCF and 50% BF. The cookies prepared with treatment WBF-B showed a diameter score nearly identical to WBF-A and WF. The lowest cookie diameter score (85.30 mm) was recorded when treated with CWCF. Cookies with 100% BF and 25% CWCF and 75% BF (WBF-C) had average diameters of 87.80 mm and 86.70 mm, respectively. These results align with Gupta *et al.* (2011). The spread ratio of the cookies ranged between 8.80 and 6.95; the maximum spread ratio score (8.80) was recorded in the cookies treated with 100% WF, while the lowest spread ratio (6.95) was observed when treated with CWCF, prepared with 100% WCF, whose starch showed higher amylose content than wheat starch (Tran *et al.*, 2013). Hence, the greater hydrophilic nature of WC starches limits the available water, decreasing the cookie spread. An SR of 8.47 was recorded in the cookies prepared with 50% BF and 50% WCF. The cookies prepared with treatment BF and WBF-B showed a middle value of spread ratio. Cookies with 25% and 75% WCF and BF flour had a spread ratio of 7.05. These results align with Chinma & Gernah (2007) and Bala *et al.* (2015).

Texture evaluation is a crucial step in the development of new food products, significantly impacting their sensory acceptability (Adebisi *et al.*, 2016). Among various textural attributes, hardness holds particular importance as it influences consumer perception and overall quality. The physical properties of enriched cookies showed positive effects, including decreased height, increased diameter, higher spread ratio, and reduced hardness, resulting in softer eating characteristics desirable in cookies. WF represents 100% WF (Table 4). The minimum hardness score (410 N) was recorded for cookies made from 100% WF, while the average hardness score (935 N) was noted in treatment WBF-A, comprising 50% barley and 50% WCF. The maximum hardness score (1214 N) was observed

in cookies made with 100% BF, statistically comparable to those treated with 75% barley and 25% WCF, with an average value of 1208 N. Cookies with higher percentages of WCF exhibited lower hardness scores compared to WBF-A and WBF-C. Treatments such as CWCF, with cookies made from 100% WCF, showed a hardness score of 1128 N, while cookies made with 75% WCF exhibited a score of 841 N. The decrease in hardness and fracturability with an increase in water chestnut and BF levels may be attributed to the lower fat content of CWCF and BF compared to wheat. High fat content increases brittleness due to moisture migration from the center to the surface, causing breakage, but CWCF and BF contain lower fat content. Similar results were reported by Shafi *et al.* (2016). Understanding how different flour compositions influence cookie texture aids in optimizing their sensory attributes for consumer satisfaction and market acceptance.

Color analysis of cookies

The surface color of a baked product, alongside texture and taste, plays a crucial role in its initial acceptability by consumers (Sarabhai & Prabhasankar, 2015). Cookies were assessed for lightness (L^*), redness (a^*), and yellowness (b^*), revealing significantly lower L^* values (46.45–52.39) in cookies prepared from various flour concentrations compared to the control (59.97) (Table 5).

The highest L^* value (52.39) was found in cookies made with 100% BF, while the lowest L^* value (46.45) was observed in WBF-B treatment, prepared with 75% commercial water chestnut and 25% BF. The a^* values, indicating red or green color (positive for red and negative for green), did not significantly differ between composite cookies made with CWCF and BF and the control treatment. The maximum a^* value was observed in cookies containing 100% WCF, attributed to the presence of red pigments in water chestnut, which may offer protection against certain oxidative stress-related diseases. The b^* color value, indicating yellow or blue colors (positive for yellow and negative for blue), showed significantly different crust yellowness values across various cookie formulations (8.14–12.65), compared to the control (18.99). The yellowness of composite cookies decreased, possibly due to the degradation of unstable yellow compounds during baking. The highest b^* value (12.65) was observed in cookies made with 100% BF in BF treatment, while the lowest value (8.14) was found in cookies made with 100% water chestnut flour. These findings were aligned with those of Shafi *et al.* (2016). Understanding how different flours affect the color attributes of cookies is essential for optimizing their visual appeal and overall sensory experience.

Table 5. Color contents of water chestnut and barley flour cookies.

Treatments	L*	a*	b*
T ₀	59.97 ± 2.93 ^a	5.80 ± 1.29 ^a	18.99 ± 2.17 ^a
T ₁	52.39 ± 0.59 ^b	4.60 ± 0.69 ^a	12.65 ± 0.40 ^b
T ₂	46.71 ± 0.07 ^{cd}	6.03 ± 0.19 ^a	8.145 ± 0.20 ^c
T ₃	48.38 ± 1.41 ^{cd}	6.01 ± 0.34 ^a	9.90 ± 0.21 ^{bc}
T ₄	46.45 ± 1.08 ^d	4.24 ± 0.90 ^a	9.80 ± 1.20 ^{bc}
T ₅	50.09 ± 0.84 ^{bc}	4.96 ± 0.63 ^a	11.81 ± 1.56 ^b

n = 6, Standard Error for Comparison 1.464, 0.76, and 1.21; Critical Value for Comparison 3.58, 1.87, and 2.97; (T₀) WF = wheat flour 100%; (T₁) BF = Barley flour 100%; (T₂) WCF = Water chest nut flour 100%; (T₃) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T₄) WBF-B = Water chestnut flour 75%, Barley flour 25%; and (T₅) WBF-C = Water chestnut flour 25%, Barley flour 75%.

SEM analysis

SEM serves as a valuable tool for elucidating sample surface topography and composition (El Fray *et al.*, 2007). The SEM results, illustrated in Figure 3, provide insights into the distinctive characteristics of cookies examined at both low (200×) and high (500×) magnifications (Figure 3 A, B).

Cookies made from 100% WF and 100% BF displayed a granular structure with uniform distribution, contrasting with those made from 100% WCF, which exhibited a smooth texture and fluffiness due to higher starch content and lower fiber content (Correia *et al.*, 2009). The micrograph of WBF-A indicated a less smooth structure compared to CWCF, reflecting the blend of 50% WCF and 50% BF. Conversely, cookies prepared with a 75% WCF and 25% BF ratio (WBF-B) showed a homogenized structure with increased smoothness and fluffiness relative to WBF-A. The presence of fiber, which absorbs and releases water during baking, contributes to increased porosity and cavity formation, potentially reducing breaking strength. SEM images of CWCF and WBF-B exhibited similarities due to their high WCF content. In contrast, cookies from treatment WBF-C displayed greater granule distribution, dominating over smoothness, owing to the 25% WCF and 75% BF composition. These findings align with those of Adebisi *et al.* (2016), who observed a regular and smooth structural network in their study on the effect of fermentation on the microstructure of pearl millet flour and biscuits. Understanding the microstructure of cookies aids in optimizing their texture and overall quality, ensuring consumer satisfaction.

Spectral analysis of cookies

Infrared spectroscopy serves as a complementary technique for chemically characterizing absorbing molecules.

It facilitates both quantitative and qualitative analysis (Dutta, 2017). FTIR spectra of cookies prepared from different flours (water chestnut & barley) in various proportions were compared to elucidate their chemical structure and characteristics (Figure 4). The IR spectra results for cookies revealed peaks ranging from 2920.35 to 2922.53 cm⁻¹, indicating the presence of alkanes (C-H stretch) due to tightly bound water in the form of moisture (Table 6). Control treatment cookies exhibited 70% transmittance at 2921.44 cm⁻¹ wavelength, while other treatments such as BE, CWCF, WBF-A, WBF-B, and WBF-C showed higher transmittance levels ranging from 70% to 94%, 93.5%, 83%, 84%, and 91%, respectively. Sharp peaks for esters and saturated aliphatic compounds (C=O stretch) were observed in the spectra of control treatment cookies, with a value of 1743.65 cm⁻¹ at 67% transmittance (Figure 4). Among different treatments, the maximum peak appeared at 1745.50 cm⁻¹ with 92% transmittance in BE, while minimum peaks were observed at 1742.80 cm⁻¹ with 91% transmittance in CWCF, indicating changes in flour nature.

Higher transmittance levels suggest a reduction in carbonyl peaks, correlating with a decrease in total lipids in the samples, as reported in similar studies (Reller *et al.*, 2008). Peaks for nitro compounds (N-O asymmetric stretch) appeared only at 1541.29 cm⁻¹ in cookies treated with 100% BF at 92% transmittance. Peaks for aromatic compounds (C-C stretch in ring) were observed at 1457.12 cm⁻¹ with 97% transmittance only in cookies treated with 100% BF, while the C-C stretch bond was absent in all other treatments. In the 6th section, control treatment cookies exhibited a peak at 1159.06 cm⁻¹ with 85% transmittance, while among different treatments, IR spectra values ranged from 1159.06 cm⁻¹ to 1160.53 cm⁻¹, indicating the presence of aliphatic amines (C-N stretch). The intensity decreased due to changes in crystallinity in the sample. Peaks for alkanes compound (C-H rock) appeared at 723.49 cm⁻¹ with 99% transmittance

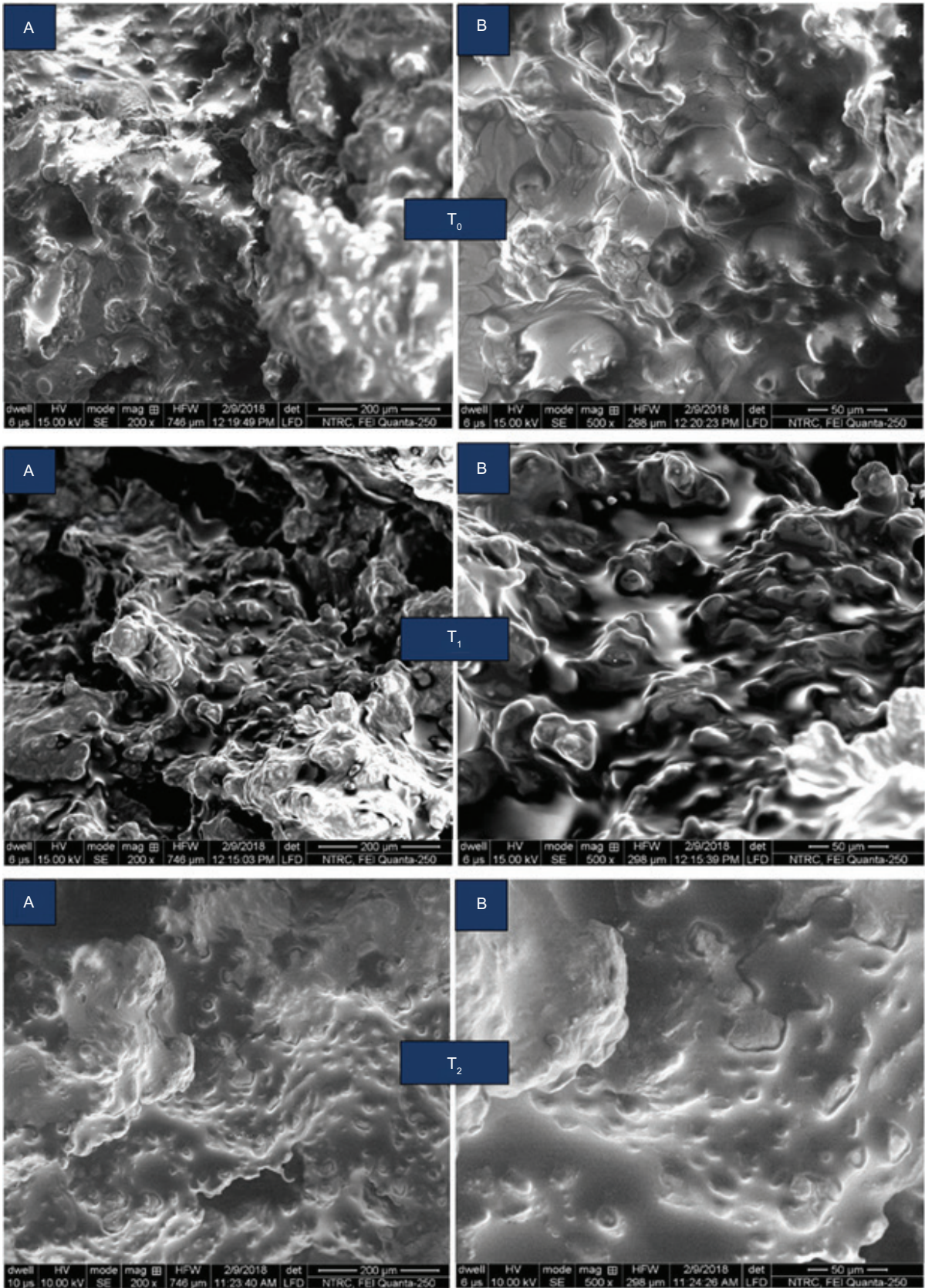


Figure 3. SEM micrographs of controlled cookies at magnifications. (A) 200× and (B) 500×. (T₀) WF = wheat flour 100%; (T₁) BF = Barley flour 100%; (T₂) WCF = Water chest nut flour 100%; (T₃) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T₄) WBF-B = Water chestnut flour 75%, Barley flour 25%; and (T₅) WBF-C = Water chestnut flour 25%, Barley flour 75%.

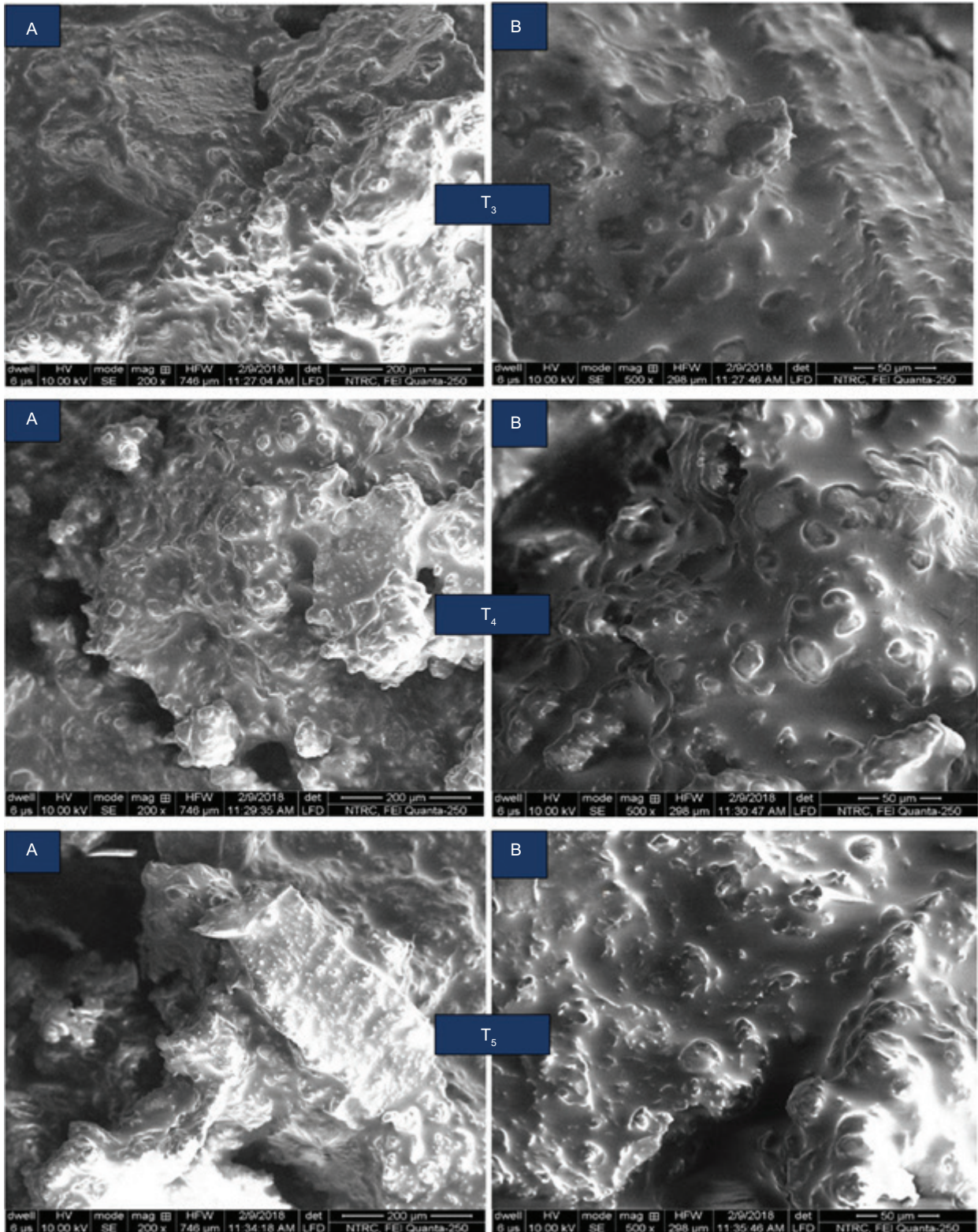


Figure 3. (Continued) SEM micrographs of controlled cookies at magnifications. (A) 200× and (B) 500×. (T₀) WF = wheat flour 100%; (T₁) BF = Barley flour 100%; (T₂) WCF = Water chest nut flour 100%; (T₃) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T₄) WBF-B = Water chestnut flour 75%, Barley flour 25%; and (T₅) WBF-C = Water chestnut flour 25%, Barley flour 75%.

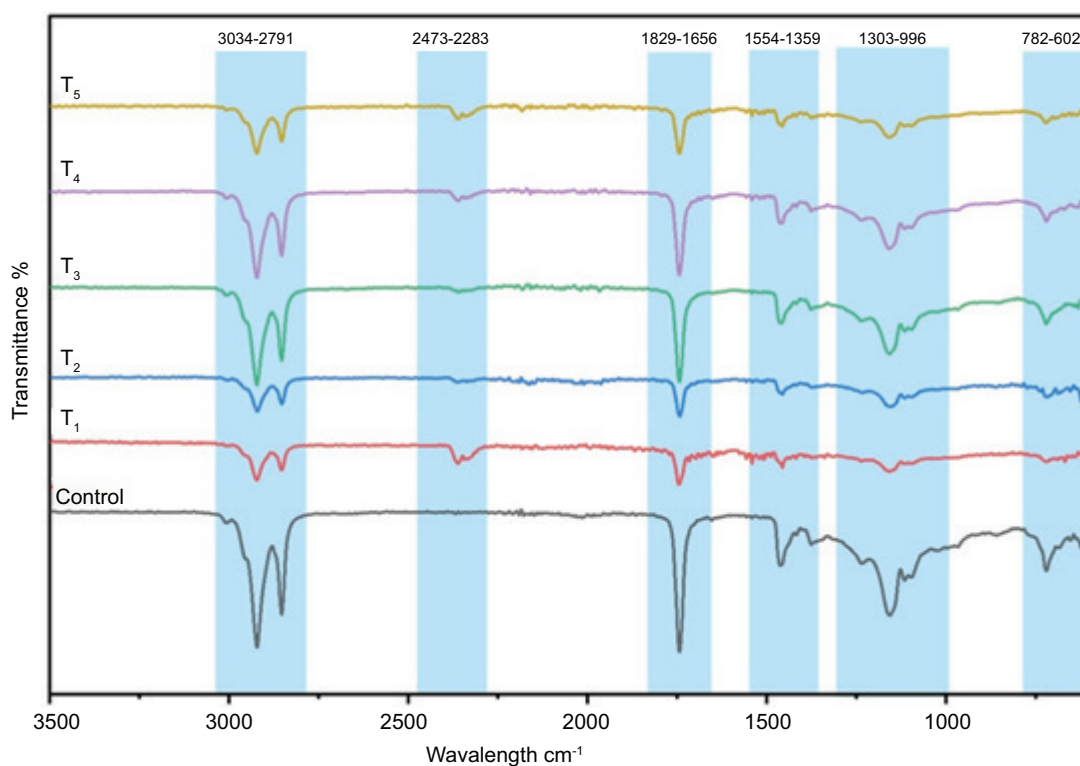


Figure 4. Fourier Transform Infra-red Spectrophotometric analysis, (T_0) WF = wheat flour 100%; (T_1) BF = Barley flour 100%; (T_2) WCF = Water chest nut flour 100%; (T_3) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T_4) WBF-B = Water chestnut flour 75%, Barley flour 25%; and (T_5) WBF-C = Water chestnut flour 25%, Barley flour 75%.

Table 6. Functional groups' evaluation of various treatments of cookies at specific wave number (cm^{-1}) in the infra-red spectral region.

Compound	Functional group	T_0	T_1	T_2	T_3	T_4	T_5
C-H stretch	Alkanes	2921.44	2922.35	2920.53	2921.68	2921.78	2921.93
C-H stretch	Alkanes	2852.52	2852.72	2852.15	2852.61	2852.64	2852.63
C=O stretch	Esters, saturated aliphatic	1743.65	1745.50	1742.80	1743.96	1744.28	1744.21
N-O asymmetric stretch	Nitro compound	-	1541.29	-	-	-	-
C-C stretch in ring	Aromatics	-	1457.12	-	-	-	-
C-N stretch	Aliphatic amines	1158.06	1159.19	1153.86	1159.06	1158.87	1160.53
C-H rock	Alkanes	-	723.49	-	-	-	-
$-\text{C}\equiv\text{C}-\text{H}$: C-H	Alkynes	-	668.57	623.03	-	-	-

(T_0) WF = wheat flour 100%; (T_1) BF = Barley flour 100%; (T_2) WCF = Water chest nut flour 100%; (T_3) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T_4) WBF-B = Water chestnut flour 75%, Barley flour 25%; (T_5) WBF-C = Water chestnut flour 25%, Barley flour 75%.

only in cookies treated with 100% BF. In addition, peaks for alkanes compound ($-\text{C}\equiv\text{C}-\text{H}$: C-H) were higher at 658.57 cm^{-1} in cookies treated with 100% BF, followed by peaks at 623.03 cm^{-1} in cookies treated with 100% WCF at transmittance levels of 99% and 94%,

respectively. These findings align with those of Adebisi *et al.* (2016). Understanding these spectral patterns aids in elucidating the chemical composition and properties of the cookies, thereby contributing to the improvement of their quality and nutritional value.

Table 7. Sensory analysis of water chestnut and barley flour cookies.

Treatments	Color	Texture	Crispiness	Taste	Overall acceptability
T ₀	7.55 ± 1.23 ^a	7.14 ± 1.03 ^a	6.92 ± 1.47 ^a	7.47 ± 1.36 ^a	7.46 ± 1.11 ^a
T ₁	6.59 ± 1.22 ^b	6.27 ± 1.15 ^c	6.04 ± 1.62 ^b	5.97 ± 1.57 ^{cd}	6.15 ± 1.42 ^{cd}
T ₂	6.28 ± 1.37 ^b	6.27 ± 1.19 ^c	6.41 ± 1.38 ^{ab}	5.97 ± 1.82 ^{cd}	6.10 ± 1.37 ^d
T ₃	6.60 ± 1.13 ^b	6.65 ± 1.13 ^{bc}	6.70 ± 1.33 ^a	6.55 ± 1.44 ^{bc}	6.65 ± 1.23 ^{bc}
T ₄	6.78 ± 1.11 ^b	7.01 ± 0.09 ^{ab}	6.86 ± 1.18 ^a	6.53 ± 1.29 ^{bc}	6.72 ± 1.22 ^b
T ₅	6.65 ± 0.95 ^b	6.62 ± 0.99 ^{bc}	6.50 ± 1.15 ^{ab}	6.66 ± 1.08 ^b	6.65 ± 0.95 ^{bc}

n=6, Standard Error for Comparison 1.464, 0.76, and 1.21; Critical Value for Comparison 3.58, 1.87, and 2.97, (T₀) WF = wheat flour 100%; (T₁) BF = Barley flour 100%; (T₂) WCF = Water chest nut flour 100%; (T₃) WBF-A = Water chestnut flour 50%, Barley flour 50%; (T₄) WBF-B = Water chestnut flour 75%, Barley flour 25%; (T₅) WBF-C = Water chestnut flour 25%, Barley flour 75%.

Sensory analysis of cookies

The sensory assessment results of cookies varied significantly ($p < 0.05$) produced from water chestnut and barley flours at concentrations of 100%, 75%, 50%, and 25% for color, taste, texture, crispiness, and overall acceptability are presented in Table 7.

The highest scores were observed for color, texture, crispiness, taste, and overall acceptability in cookies made from 100% WF, which served as the standard. The average color score ranged between 6.28 and 7.55. Among the treatments, WBF-B had the highest color score (6.78), with cookies prepared from 75% WCF and 25% BF, while CWCF treatment cookies had the lowest color score (6.28). The color scores for treatments BF, WBF-A, and WBF-C were recorded as 6.59, 6.60, and 6.65, respectively. These findings are consistent with previous results (Mir *et al.*, 2015b), who obtained a color score of 6.7 in their study. Regarding texture, the scores ranged from 6.27 to 7.24. After standardization, the maximum texture score (7.01) was noted in the cookies from WBF-B treatment, while the lowest texture score (6.27) was recorded in CWCF treatment with 100% WCF. Treatments BF, WBF-A, and WBF-C had texture scores of 6.2791, 6.65, and 6.62, respectively. These results align with Frost *et al.* (2011) and Sharma & Gujral (2014), who observed similar texture scores in their studies. The maximum crispiness was noted in WBF-B treatment cookies (6.86), while the lowest crispiness value (6.04) was recorded in cookies treated with 100% BF. These findings are consistent with Sarabhai & Prabhasankar (2015), who found a crispiness score of 6.8 in their study. In addition, the maximum taste score (7.48) was recorded for WF, while the highest taste score among different treatments (6.66) was noted in the WBF-C treatment. The lowest taste score (5.97) was recorded in cookies treated with 100% WCF, statistically at par with cookies treated with 100% BF (5.97). Treatments WBF-A and WBF-B showed intermediate taste scores, with WBF-A having a score of 6.55 and WBF-B having a score of 6.53. The overall acceptability scores varied significantly among

cookies prepared from water chestnut, barley, and WF. The maximum score for acceptability (7.46) was recorded for cookies prepared from 100% WF. Among different treatments, the maximum overall acceptability (6.72) was noted in WBF-B treatment with 75% water chestnut and 25% BF. These results are also consistent with Shafi *et al.* (2016), who reported similar overall acceptability scores in their study.

Conclusion

The current study's goal was to draw attention to the nutritionally beneficial qualities of composite flour, which includes barley and water chestnut and can be readily combined with other flours to prepare rich, healthful food products. It was declared that cookies developed with a blend of 75% WCF and 25% BF possessed better physical and microstructural properties. In addition, sensory evaluation favored this blend, resulting in an overall acceptability score of 6.7. These findings suggested that adopting the proposed blending of water chestnut and BF can produce balanced, nutritious functional cookies with favorable qualitative features with the possibility to affect human health positively. Furthermore, enhancing consumption of barley and WCF in cereal baked goods can bridge up the food gap caused by the low wheat production and eliminate the need to import wheat grains from other countries.

Data Availability

Data will be provided by the corresponding author on demand.

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Mandatory AI Disclosure

The authors declare that no AI-assisted tools were used in the preparation of this manuscript.

Authors' Contributions

Conceptualization was done by M.H. and M.A.; methodology was the concern of M.A. T.A., and T.R.; software was the responsibility of A.A., A.A., and M.I.S.; validation was done by M.H. and B.N.H.; formal analysis was looked into by T.A. and M.A.; investigation was done by M.H. and M.I.S.; resources were the responsibility of T.R., M.S.S., and A.A.; data curation was done by M.N.H. and M.S.S.; writing—original draft preparation was the concern of T.A., A.A., and M.A.; writing—review and editing were done by A.A., A.A., M.S.S., N.A.R., and B.H.; visualization was done by N.A.R., A.A., and B.N.H.; supervision was taken care of by M.S.S and A.A.; project administration was done by M.H.; funding acquisition was done by M.I.S., A.A., A.A., T.R., and N.A.R. All authors have read and agreed to the published version of the manuscript.

Conflict of Interest

The authors have declared no conflicts of interest for this article.

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