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Preparation of Steamed Cupcakes Using Wheat, Barley, and Water Chestnut Flour

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ABSTRACT

Cupcakes are the most commonly used bakery products. The present research was planned to evaluate the effect of wheat flour replacement with barley and water chestnut flour on cupcake's shelf-life and nutritional profile. Cupcakes were prepared with different ratios of wheat, barley, and water chestnut flour and were evaluated for their proximate composition, minerals, and organoleptic properties. Moisture content in different combinations of wheat, barley and water chestnut cupcakes ranged from 24.53±0.38-29.52±0.29% with variation observed among treatments. Similarly, crude protein, crude ash, crude fiber, crude fat, and NEF values ranged from 13.44±0.23-17.67±0.01%, 1.38±0.01-2.35±0.01%, 1.31±0.002-2.62±0.005%, 25.28±0.10-28.80±0.07%, 23.12±0.27-32.69±14.56% respectively. In mineral analysis magnesium, iron, potassium, and zinc values ranged from 1.07±0.055-2.10±0.04%, 1.58±0.009-2.38±0.004%, 22.19±0.17-36.47±0.11%, 0.49±0.029-2.66±0.03%, respectively. Beta-glucan values among treatments ranged from 0.34±0.04-3.15±0.03%. Total sugar, reducing and non-reducing sugar values ranged from 4.59±0.01-8.33±0.04%, 1.006±0.006-1.84±0.03%, 3.26±0.05-7.12±0.02%. Texture analysis over a 14-day storage period revealed a decrease in hardness, suggesting improved moisture retention and shelf-life stability. Sensory evaluation by a trained panel identified that formulations with balanced proportions of barley and water chestnut flour achieved high scores in appearance, flavor, texture, and overall acceptability, indicating that the modified cupcakes maintained desirable sensory qualities throughout storage. These findings suggest that incorporating barley and water chestnut flour in cupcake formulations enhances nutritional value and consumer appeal, providing a promising strategy for producing nutritious baked goods. Further studies are recommended to explore industrial-scale applications of these formulations.

Keywords: Cupcakes, Steaming, Barley, Water chestnut, Wheat.

INTRODUCTION

Cereal-based products are part of the daily human diet since they contain carbohydrates, proteins, and fiber. Wheat is the most commonly grown and consumed cereal in the world, and it serves approximately 20 % of the total dietary energy requirement globally, making it a key staple food commodity (Grote et al., 2021). Cereals contribute significantly to global health by serving as the primary energy source. Cereals include important

elements such as carbs, minerals, vitamins, proteins and lipids (Deshwal et al., 2021). Barley has significant levels of iron, zinc, magnesium and phosphorus, making this grain a valuable food for the human diet. Barley proteins also help to produce gluten, which gives dough elasticity and is important in the baking sector (Panizo-Casado et al., 2020). Moreover, barley was found to enhance glycemic control, and therefore it is recommended for use in functional food production, especially for

diabetics (Geng et al., 2022).

Water chestnut is an aquatic vegetable used in Asian dishes, a good source of starch and dietary fiber. It also has significant amounts of potassium, phosphorus, and vitamin B6, as well as phenolic compounds, which are the source of the food's functional health properties. Water chestnut flour (WCNF) is high in starch and hence provides a strong energy source. Furthermore, vitamins, phenolics, tannins, antioxidants, omega-3 fatty acids and other nutrient-dense substances are found in the fruit (Rajput & Singh, 2023). Among flour made from fruits, water chestnuts (Siddiqui and Leghari, 2007, 2008) are attracting more attention. Incorporating water chestnut flour into wheat flour creates bakery products with enhanced nutritional value (Din et al., 2024). Increasing consumer demand for healthier and more nutritious options is drawing attention to the diversification of ingredients in bakery products. Generally, the key ingredient is wheat flour as it plays a crucial functional role by providing volume and structure (Guiné and Florença, 2024). However, cupcakes made only from wheat are high in carbohydrates but lack nutritional qualities like fiber and essential minerals. Using composite flour in bakery products enhances their nutritional profile by increasing the content of protein, fiber and phenolic compounds. Furthermore, it ensures that these products have good sensory qualities (Amadeu et al., 2024). Cupcakes can be either baked or steamed. Steaming is a moist-heat cooking method that uses hot steam to transfer heat to the food. Compared to baking, steaming usually results in a cake with a lighter, softer, and moister crumb (Ho et al., 2018). The basic purpose of this study is to investigate the effect of barley and water chestnut flour composition on steamed cupcakes' nutritional profile and storage study.

MATERIALS AND METHODS

Procurement of raw materials

Wheat and barley grains were obtained from the Department of Plant Breeding and Biotechnology (IPBB), MNS-University of Agriculture Multan, Pakistan. The water chestnut flour used in this study was purchased from a certified local supplier. All chemicals and reagents were obtained from Sigma Aldrich and Duksan Pure Chemicals of Kyung Kido, South Korea.

Milling process

Flour of wheat and barley was made in a lab by using the mill (Culatti Type MFC) by following the procedure of

Yaqoob et al. (2018).

Analysis of raw material

Moisture content

Flour was analyzed for moisture by using method No.44-15A of AACC (2000). Moisture content was evaluated by a drying oven (SLN-53-STD, POL-Eko-Appratus). A sample of 2g was placed in a china dish for drying at 105°C for 24 h. Moisture content was calculated using equation 1.

$$\text{Moisture} = \frac{W_2 - W_3}{W_1} \times 100 \text{ ----- (1)}$$

Where

W_1 = weight of sample

W_2 = China dish + Sample weight.

W_3 = after oven dry weight.

Crude protein

Crude protein content was measured by using Kjeldahl's apparatus following the procedure outlined in method (46-10) in AACC (2000). A 2-g sample was weighed and digested with sulphuric acid along with digestion tablets in a digestion chamber. After digestion was complete, the digestion tubes were transferred to a volumetric flask and made volume up to 250 ml with distilled water. For distillation 10 ml of the diluted sample was transferred to the distillation unit. A solution containing 4% boric acid and a few drops of methyl red was used. After distillation was complete, the distilled boric acid was titrated using a 0.1N sulphuric acid solution.

$$\text{N\%} = \frac{\text{Vol of sulphuric acid} \times 0.005 \times 250}{\text{sample wt}} \times 100 \text{ ----- (2)}$$

$$\text{Protein \%} = \text{Nitrogen\%} + 6.25$$

Fat content

To determine the crude fat content of wheat, barley and water chestnut flour Soxhlet apparatus and solvent extraction method were used, following method No. 30-25 AACC (2000). First, a thimble was made using filter paper and weighed. Then, a moisture-free sample was weighed with the thimble and placed in the tube. Petroleum ether was used for extraction in the receiving flask and after 3-4 washings the fat content was removed. Fat content was determined using the following formula (3).

$$\text{Fat} = \frac{\text{dried sample wt} + \text{thimble wt} - \text{Dried fat free sample} + \text{thimble wt}}{\text{Sample wt}} \times 100 \text{ --- (3)}$$

Ash content

Ash content of the sample was measured using AACC (2000) method No. (08-01). In this method sample was taken in the crucible and burned on a spirit lamp. After charring, the samples were placed in the muffle furnace

at 550°C for 5 hours until ash appeared. After 5 hours muffle furnace was turned off and waited until it cooled down. After that samples were carefully removed from the muffle furnace or placed in the desiccator and allowed to cool. Using the following formula, the ash concentration was calculated.

$$\text{Ash} = \frac{\text{Weight of Ash (g)}}{\text{Sample wt (g)}} \times 100 \quad \text{--- (4)}$$

Crude fiber

Fiber content was determined using the AACC (2000) method No. (33-10). A 5g dried fat-free sample was digested for 25–30 minutes in 200ml sulphuric acid (1.25%). The sample was filtered using filter paper and washed. The residue sample was again digested with 200 ml sodium hydroxide (1.25%) for 25- 30 minutes. Filtered the sample using filter paper and washed it with distilled water. Placed the sample in a hot air oven at 100±05°C for 24 hours. The sample was placed in a muffle furnace at 550 °C for 5 hours. After that, the sample was weighed. Fiber content was determined by using this formula.

$$\text{Crude Fiber \%} = \frac{\text{wt. of oven-dried sample-wt. of the sample after ashing wt. of sample}}{\text{wt. of sample}} \times 100 \quad \text{--- (5)}$$

Nitrogen-free extract

The following formula was used to calculate NEF content.

$$\text{NFE \%} = 100 - (\text{Crude Protein \%} + \text{Ash Content \%} + \text{Crude Fiber \%} + \text{Moisture Content \%} + \text{Crude Fat \%}) \quad \text{--- (6)}$$

Mineral profile

The mineral content of composite flour was determined by a Unit Atomic Absorption Spectrophotometer (AA240 Varian K, Australia) and flame photometer by following method No. 3.014-016 described in AOAC (2006). Sample (1g) was taken in a clean and moisture-free conical flask. Nitric and perchloric acid were added in the flask with a ratio of (3:1). The flask was placed on a hot plate until white fumes appeared. A solution of 100 ml was prepared for further analysis Omar et al, (2024).

Rheological properties of composite flour

The rheological parameters of the dough were determined using the Brabender Farinograph-E and (AACC, 2000) method No.54-21.

Cupcake preparation

The method of Aniemena et al. (2024) was used for the preparation of cupcakes formulation and the method of Hou (2020) was used for the steaming of cupcakes. After steaming the cupcakes were allowed to cool for 30 min and then transferred to an airtight container for analysis.

Formulation of composite flour of wheat, barley and water chestnut

Treatment	Wheat (%)	Barley (%)	Water Chestnut (%)
T ₀	100	-	-
T ₁	85	-	15
T ₂	85	15	-
T ₃	70	15	15
T ₄	60	-	40
T ₅	60	40	-
T ₆	60	20	20

Compositional analysis of cupcake

Proximate analysis of baked cupcakes was carried out by using the method described by (AACC, 2000).

Mineral analysis of cupcake

Mineral analysis of cupcakes was carried out by using the same procedure as described earlier.

Color analysis of cupcakes

Color analysis was carried out using a Chroma meter (Chroma Meter CR-400 by Konica Minolta) by using the method described in (AACC, 2000).

Beta-glucan content of cupcake

Using a wet extraction approach based on alcohol, the sample beta-glucan content was measured. The slurry was created using 50 g of flour and (300ml: 50%) ethanol. A filter was used to remove the solution, and the residue was then mixed with a 50% ethanol solution that included bromelain and amylase. To get β-glucan, the filter process was done again after 20 hours at 35°C. In an oven set to 55°C for 24 hours, the moist residue was dried. A percentage of β-glucan content was calculated from the dried sample weight (Hussain et al., 2019).

Total sugar content of cupcakes

The technique as described by Behlil et al. (2014) was used for measuring the total sugar content of cupcakes

Texture analysis

Texture analysis was performed by following the criteria outlined by Omar et al. (2024). A universal testing Brookfield Engineering Lan. Inc., Middleboro, MA 02346-1031, USA) device was used to identify texture. A cylindrical probe with a diameter of 25mm was used to compress the cupcake sample at a speed of 2mm/s.

Sensory evaluation

A sensory evaluation was performed to measure organoleptic properties. Ten trained panelists from MNS University of Agriculture Multan participated in the

sensory evaluation. Each panelist received a single slice of each cupcake. Samples were presented on white odorless and disposable plates. The panelists evaluated the cupcakes using a 9-point hedonic scale of the following format: 1 (extremely dislike) to 9 (extremely like) (Ngan et al., 2023).

Statistical analysis

RESULTS AND DISCUSSION

The proximal composition of wheat flour, barley flour and water chestnut flour used in this study is presented in Table-2. It was observed that moisture, crude ash and crude fat were higher in water chestnut powder compared to wheat and barley. In the earlier study conducted by Shafi et al. (2016), the protein content was found to be 3.18% in water chestnut flour and 10.94% in wheat flour. In the present research work, it was observed that crude protein content was higher in wheat flour $12.32 \pm 0.035\%$ compared to water chestnut flour $3.26 \pm 0.309\%$ and barley flour $6.34 \pm 0.08\%$. Crude ash, crude fat and nitrogen-free extract (NFE) content were observed to be higher in water chestnut flour $2.37 \pm 0.09\%$, $1.43 \pm 0.015\%$ and $80.70 \pm 0.76\%$ compared to wheat $1.41 \pm 0.07\%$, $1.30 \pm 0.005\%$, $72.72 \pm 0.105\%$ and barley flour $2.18 \pm 0.06\%$, $1.42 \pm 0.04\%$, $75.55 \pm 0.36\%$ respectively. The crude fiber was observed to be higher in barley flour $1.53 \pm 0.13\%$, compared to wheat and water chestnut flour $1.37 \pm 0.02\%$ and $0.705 \pm 0.014\%$, respectively. The present findings for wheat and water chestnut flour were consistent with Din et al. (2024) who reported similar proximal composition of wheat and water chestnut flour in their research. The composition of barley flour was consistent with that of Gupta et al. (2011), who observed a similar barley flour composition in their study. Table 3 presents the mineral composition of wheat flour, barley flour and water chestnut flour. Data revealed that barley flour was relatively higher in potassium ($54.38 \pm 0.09\text{mg}/100\text{g}$), zinc ($1.34 \pm 0.51\text{mg}/100\text{g}$) and iron ($2.18 \pm 0.01\text{mg}/100\text{g}$). Water chestnut flour was found to be higher in sodium ($18.34 \pm 0.05\text{mg}/100\text{g}$) and magnesium ($2.41 \pm 0.31\text{mg}/100\text{g}$) as compared to wheat and barley flour. These findings were consistent with (Dobhal et al., 2021; Wotango et al., 2024; Shafi et al., 2016). The rheological qualities of dough may provide information regarding its behavior during mixing, extension, and fermentation, as well as the influence of the ingredients added to the chemical composition, which can hinder or stimulate molecular interactions in the dough matrix.

Data collected from various analyses were analyzed using the SPSS 20.0 software program. Means and Standard deviations were determined using descriptive statistics. Comparisons between samples were determined using analysis of one-way variance (ANOVA) and the Tuckey test. Statistical significance was defined as $P \leq 0.05$ (Omar et al., 2024).

Water absorption was defined as the amount of water required to center the highest point of the mixing curve at an arbitrary 500 BU (Brabender units) (Ungureanu-luga et al., 2021). The rheological properties of composite flour blends are represented in Table 4. During research, it was observed that treatment T4 (T4=60% wheat + 40% water chestnut) showed more water absorption compared to other treatments. Water chestnut flour's lower protein and fat content could be the possible reason for this increase. Singh et al. (2011), during their research observed that water absorption values increased linearly as the amount of water chestnut flour increased. The results of the present research align with Abdullah et al. (2022), who observed that dough stability time (min) showed a direct proportional relationship with increased barley flour in the blends, indicating that stability time increased with additional sprouted barley in the dough blend when compared to the control treatment. Table 5 represents the chemical composition of cupcakes formulated with barley and water chestnut flour. The proximate composition of the various formulations (T0 to T6) reveals differences in moisture, protein, fat, fiber, and ash, which is due to the incorporation of barley and water chestnut flour. It was observed that the T0 formulation had the highest protein content ($17.67 \pm 0.01\%$) and reduced in the subsequent formulations as the barley and water chestnut inclusion levels increased. This reduction in protein can be explained by the fact that water chestnut and barley contain less protein than wheat. Moisture content in T0 (control treatment) had the highest value ($29.52 \pm 0.29\%$) and T4 had the least ($24.53 \pm 0.38\%$). The fiber content increased with the increase in the level of barley and water chestnut flour in the formulations with T1 (75% wheat and 15% water chestnut) having the highest fiber content of $2.62 \pm 0.005\%$. Din et al. (2024) observed cookies that contain water chestnut flour contain higher ash, fiber and mineral content while Gupta et al. (2009) noted that as the proportion of barley

flour increased there was a significant rise in beta-glucan content from 0.10% to 1.68% which is considered an important dietary fiber for nutrition. The fat content of the cupcakes did not change across the formulations, although some slight differences were noted this may be a result of the (Siddiqui and Muniza. 2024, 2025) shortening and other ingredients that are added in the formulation of cupcakes. The highest fat content of $28.80 \pm 0.07\%$ was observed in the T3 formulation, which contained wheat flour (70%), barley flour (15%), and water chestnut flour (15%). The ash content which depicts the mineral content of the cupcakes, rose as the proportion of barley and water chestnut flour was raised. The ash content was highest in T3 which was $2.35 \pm 0.01\%$. The result of the present research is aligned with Sharma & Devi (2021) who observed similar changes in cookies' chemical composition which were prepared with the incorporation of soya and water chestnut flour into wheat flour. Table 6 represents the mineral analysis of cupcakes. The mineral content of magnesium, iron, potassium, zinc and calcium in cupcakes showed a significant increase by the addition of barley and water chestnut flour compared to a control sample. Results revealed that the Mg content varied between 1.07 ± 0.055 mg/100g to 2.10 ± 0.04 mg/100g, with a very significant difference ($p < 0.01$). Treatment T4 cupcakes had a higher iron level of 2.38 ± 0.004 mg/100g, whereas T0 cupcakes had the lowest iron content of 1.58 ± 0.009 mg/100g. The zinc concentration was found to be higher in treatment T5 2.66 ± 0.03 mg/100g. Cupcakes' potassium (K) content ranged from 22.19 ± 0.17 mg/100g to 26.71 ± 0.32 mg/100g. The result of the present study was found to align with Shafi et al. (2016) who found an increase in the mineral content of cookies prepared with a wheat and water chestnut flour blend. On the other hand Gupta et al. (2009) also found that the mineral content of barley-rich cakes was higher compared to cakes with normal wheat flour. Beta-glucan, a form of soluble fiber found in cereal grain, is gaining popularity due to its multiple functional and bioactive qualities. Extensive studies have been undertaken on its beneficial benefits on disorders such as insulin resistance, dyslipidemia, hypertension and obesity. The fermentability of beta-glucans in the human gut and their ability to form very viscous solutions may contribute to their health-promoting properties (El Khoury et al., 2012). Treatment T5 had the higher beta-glucan concentration

3.15 ± 0.03 , while T1 had the lowest at 0.34 ± 0.04 . The results of the present research were aligned with Hussain et al. (2019), who found higher beta-glucan content in muffins prepared with barley and water chestnut flour. Who further stated that barley flour played a key role in boosting the beta-glucan content of the muffins due to its naturally high level of fiber. Table 7 represents the beta-glucan content of cupcakes. Total sugar and non-reducing sugar were found to be higher in treatment T2 8.33 ± 0.04 % and $7.12 \pm 0.02\%$, respectively. Reducing sugar was higher in the treatment T5 $1.84 \pm 0.03\%$. The rise in total sugar content can be attributed to the formation of maltose during baking (Chen et al., 2012). The results of the present research were consistent with (Hussain et al., 2019). Sugar concentration may vary due to a (Siddiqui, 2021, 2022) variety of reasons, including recipe variances, baker preferences, ingredient substitutions, culinary innovation, and cultural and regional preferences (Siddiqui et al., 2009, 2011, 2013, 2016, 2019, 2022, 2023) . These variables impact the amount and kind of sugar used in the recipe, resulting in changes in the final sugar level of the cupcake. Table 8 represents the mean values of the total sugar of cupcakes. Texture is an important quality attribute in bakery items since it determines how long they can be stored. This suggests that even in situations when moisture loss is prevented, the cake may lose its freshness during storage, particularly at 15 to 20°C. This is most likely caused by water migration, starch retrogradation, and the reaction between starch and protein. Cake tissue is affected by a number of variables, including the quantity of liquid, sugar, and fat (Ali et al., 2023). The effect of adding barley and water chestnut flour on the 14-day storage time on the hardness of cupcakes is presented in table 9. Treatment T5 showed the highest mean value of hardness during the 1st, 7th and 14th day of storage 7.15 ± 0.05 , 7.14 ± 0.05 and 7.9 ± 0.04 , respectively. It was observed that the hardness of cupcakes decreased over time of storage. On 1st day of storage T4 showed the lowest hardness value 6.37 ± 0.04 . While on the 7th and 14th day T1 showed the lowest value 6.21 ± 0.02 and 6.2 ± 0.01 respectively. The amount of barley and water chestnut flour used in cupcake recipes greatly impacts how hard they get over time. Fiber content in barley affects the hardness of cupcakes (Mirani & Goli, 2021; Hussien, 2023). This may be the possible reason why T5 containing (60% wheat + 40% barley flour) showed

more hardness during storage. Lee and Lin (2008) also noted that incorporating any kind of fiber into baked goods makes them harder. The result of the present study is aligned with Hussain et al. (2019) who prepared low glycemic index muffins using water chestnut and barley flour. The crust color of cupcakes is mainly affected by ingredients used in preparation. It was observed that the L^* value showed a non-significant relation while a^* and b^* values showed a significant relation ($p < 0.01$) between treatments. Treatment T6 showed a higher L^* value of 21.33 ± 3.20 while T2 showed the lowest L^* value among treatments 14.39 ± 3.41 . Control treatment (T0) showed a higher a^* and b^* value of 25.02 ± 0.83 and 47.78 ± 6.63 respectively while T6 showed a lower a^* value of 7.02 ± 1.55 and T5 showed a lower b^* value of 26.63 ± 5.06 among treatments. The results of the present research are similar to (Batista et al. 2018; Chuango et al. 2019; Cakmak et al. 2021). Table no 10 represents color values of cupcakes during storage. The shelf life and storage stability of baked goods might be described as preserving the sensory and physical qualities related to freshness such as crumb tenderness, compressibility and moistness by preventing change linked to staling during storage. However sensory methods are the only means to evaluate consumer acceptance. Consumers look for a

product with a soft, spongy tender crumb that also offers some resistance and does not crumble easily. However these qualities tend to deteriorate over time in storage (Baixauli et al., 2008). On the evaluation day freshly prepared cupcakes were placed on clean white plates covered with transparent sheets and labeled accordingly. On the first day T5 showed more values of appearance 8.26 ± 0.59 while on the 7th and 14th day T6 showed more values of appearance 8.3 ± 0.61 and 8.26 ± 0.59 respectively. Treatment T2 showed higher scores for aroma 8.13 ± 0.91 on the first day of storage while treatments T6 and T5 showed higher scores on the 7th and 14th days $8.5 \pm 0.51a$ and $8.5 \pm 0.62abc$ respectively. Treatment T6 showed higher scores of texture, taste and overall acceptability during all days of storage. The current study's findings are consistent with those of Nazir et al. (2022), who observed similar scores in their research on the qualitative and sensory characteristics of muffins prepared with chestnut flour substitution. Hussain et al. (2019), who prepared low glycemic index muffins with water chestnut and barley flour, observed similar sensory assessment scores during their research. Sensory evaluation of cupcakes with different flour compositions on the 1st, 7th, and 14th day of storage.

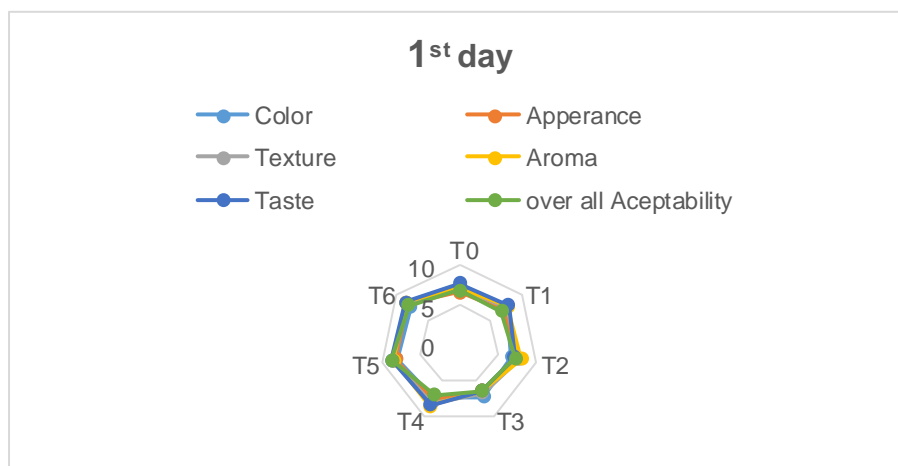


Figure 1. Sensory evaluation of cupcakes on the first day of storage.

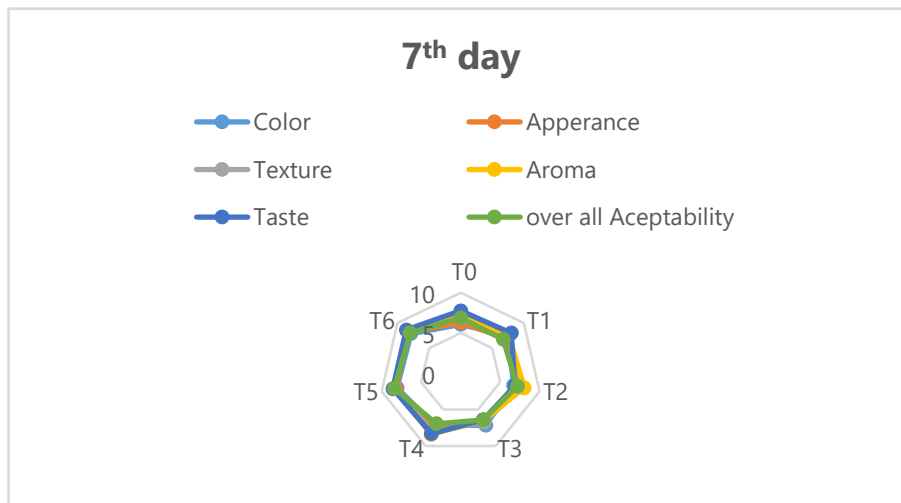


Figure 2. Sensory evaluation of cupcakes on the seventh day of storage.

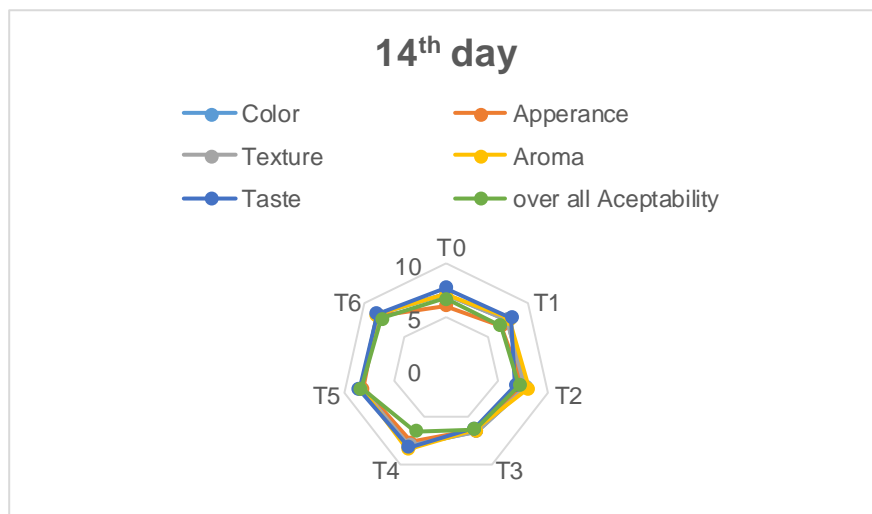


Figure 3. Sensory evaluation of cupcakes on the 14th day of storage.

Table 2. Proximal analysis of wheat flour, Barley flour and water chestnut

Analysis	Wheat flour	Barley flour	Water chestnut powder
Moisture	10.21±0.01	12.21±0.36	11.48±0.43
Crude Protein	12.32±0.035	6.34±0.08	3.26±0.309
Crude Ash	1.41±0.07	2.18±0.06	2.37±0.09
Crude Fiber	1.37±0.02	1.53 ±0.13	0.705±0.014
Crude Fat	1.30±0.005	1.42±0.04	1.43±0.015
Nitrogen Free Extract	72.72±0.105	75.55±0.36	80.70±0.76

Table 3. Mineral analysis of raw material

Mineral analysis			
Minerals	Wheat	Barley	Water chestnut
Mg mg/100g	2.08±0.062	2.15±0.03	2.41±0.31
K mg/100g	50.8±0.1	54.38±0.09	23.31±0.15
Zn mg/100g	0.86±0.007	1.34±0.51	0.58±0.07
Cu mg/100g	0.078±0.003	0.019±0.0009	0.03±0.006
Fe mg/100g	2.07±0.01	2.18±0.01	1.48±0.009
Na mg/100g	17.13±0.12	16.46±0.22	18.34±0.05

Table 4. Rheological characteristics of different flour blends

Treatments	Water Absorption (WA) (%)	Dough Development Time (DDT) (min)	Dough Stability (min)
T ₀	61.93±2.04 ^a	1.60±0.42 ^b	6.81±0.15 ^c
T ₁	58.56±6.00 ^a	1.46±0.09 ^b	5.37±0.15 ^d
T ₂	57.7±6.03 ^a	1.4±0.12 ^b	10.47±0.16 ^b
T ₃	59.5±3.5 ^a	1.32±0.13 ^b	7.37±0.16 ^c
T ₄	65.03±2.25 ^a	1.72±0.23 ^{ab}	4.62±0.26 ^e
T ₅	54.03±0.07 ^a	2.46±0.20 ^a	11.52±0.33 ^a
T ₆	61.88±1.77 ^a	1.33±0.49 ^b	5.36±0.24 ^d

T₀=100% wheat, T₁=85% wheat + 15% water chestnut, T₂=85% wheat + 15% barley flour, T₃=70% wheat + 15% barley + 15% water chestnut, T₄=60% wheat + 40% water chestnut, T₅=60% wheat + 40% barley flour, and T₆=60% wheat + 20% barley + 20% water chestnut flour (p<0.01)

Table 5. Chemical composition of different formulations for cupcakes.

Treatments	Moisture	Crude protein	Crude ash	Crude fiber	Crude fat	NFE
T ₀	29.52±0.29 ^a	17.67±0.01 ^a	1.38±0.01 ^f	1.88±0.002 ^b	25.28±0.10 ^e	32.69±14.56 ^a
T ₁	29.26±0.05 ^a	16.73±0.06 ^b	1.53±0.002 ^d	2.62±0.005 ^a	25.88±0.05 ^d	23.95±0.12 ^a
T ₂	25.75±0.11 ^d	16.32±0.10 ^{bc}	1.47±0.016 ^e	1.57±0.003 ^d	25.81±0.059 ^d	29.05±0.02 ^a
T ₃	28.50±0.31 ^b	15.805±0.13 ^c	2.35±0.01 ^a	1.41±0.003 ^f	28.80±0.07 ^a	23.12±0.27 ^a
T ₄	24.53±0.38 ^e	13.44±0.23 ^e	1.97±0.007 ^b	1.31±0.002 ^g	26.30±0.06 ^c	32.42±0.56 ^a
T ₅	26.31±0.27 ^d	14.47±0.42 ^d	1.99±0.011 ^b	1.55±0.004 ^e	27.14±0.03 ^b	28.508±0.67 ^a
T ₆	27.65±0.11 ^c	14.45±0.18 ^d	1.78±0.005 ^c	1.63±0.004 ^c	26.40±0.039 ^c	28.07±0.28 ^a

T₀=100% wheat, T₁=85% wheat + 15% water chestnut, T₂=85% wheat + 15% barley flour, T₃=70% wheat + 15% barley + 15% water chestnut, T₄=60% wheat + 40% water chestnut, T₅=60% wheat + 40% barley flour, and T₆=60% wheat + 20% barley + 20% water chestnut flour Mean± S.D (p<0.01)

Table 6. Mineral analysis of cupcakes

Treatments	Mg (mg/100g)	Fe (mg/100g)	K (mg/100g)	Zn (mg/100g)
T ₀	1.4±0.03 ^c	1.58±0.009 ^f	22.19±0.17 ^e	0.49±0.029 ^d
T ₁	1.63±0.03 ^b	1.85±0.002 ^e	26.71±0.32 ^b	0.525±0.0039 ^{cd}
T ₂	1.32±0.007 ^d	2.09±0.02 ^d	23.31±0.15 ^d	1.13±0.004 ^b
T ₃	1.07±0.055 ^e	2.22±0.003 ^c	36.47±0.11 ^a	0.509±0.002 ^{cd}
T ₄	2.10±0.04 ^a	2.38±0.004 ^a	23.43±0.28 ^d	0.56±0.03 ^c
T ₅	1.58±0.04 ^b	2.33±0.0055 ^b	23.73±0.15 ^d	2.66±0.03 ^a
T ₆	1.29±0.055 ^d	2.09±0.02 ^d	24.5±0.1 ^e	0.51±0.001 ^{cd}

Mean± S.D (p<0.01)

Table 7. Beta-glucan of cupcakes

Treatments	Beta-Glucan
T ₀	0.96±0.03 ^e
T ₁	0.34±0.04 ^g
T ₂	2.12±0.01 ^c
T ₃	1.95±0.02 ^d
T ₄	0.55±0.01 ^f
T ₅	3.15±0.03 ^a
T ₆	2.82±0.04 ^b

Mean± S.D (p<0.01)

Table 8. The total sugar content of Cupcakes

Treatment	Sugar of Cupcake		
	Total Sugar	Non-Reducing	Reducing Sugar
T ₀	6.22±0.02 ^d	5.15±0.04 ^c	1.05±0.03 ^{bc}
T ₁	4.59±0.01 ^f	3.26±0.05 ^e	1.4±0.005 ^{abc}
T ₂	8.33±0.04 ^a	7.12±0.02 ^a	1.12±0.01 ^{bc}
T ₃	5.91±0.04 ^e	4.70±0.01 ^d	1.48±0.43 ^{ab}
T ₄	6.39±0.01 ^d	5.11±0.02 ^c	1.25±0.03 ^{bc}
T ₅	7.78±0.38 ^b	6.03±0.04 ^b	1.84±0.03 ^a
T ₆	7.10±0.01 ^c	6.11±0.009 ^b	1.006±0.006 ^c

Mean± S.D (p<0.01)

Table 9. Hardness of Cupcakes during storage study

Treatment	Hardness of Cupcake		
	1 st Day	7 th Day	14 th Day
T ₀	6.87±0.01 ^{cd}	6.50±0.04 ^{ef}	7.8±0.03 ^{efg}
T ₁	6.45±0.01 ^{efg}	6.21±0.02 ^{ij}	6.2±0.01 ^j
T ₂	7.05±0.03 ^{ab}	7.00±0.01 ^{abc}	6.88±0.10 ^{cd}
T ₃	6.96±0.06 ^{bc}	6.87±0.02 ^{cd}	6.73±0.03 ^d
T ₄	6.37±0.04 ^{fghi}	6.29±0.01 ^{ghij}	6.22±0.04 ^{hij}
T ₅	7.15±0.05 ^a	7.14±0.05 ^a	7.9±0.04 ^a
T ₆	6.56±0.10 ^e	6.51±0.01 ^{ef}	6.38±0.13 ^{fgh}

Mean± S.D (p<0.01)

Table 10. Color analysis of Cupcakes

Treatment	l value	a value	b value
T ₀	20.94±0.64 ^a	25.02±0.83 ^a	47.78±6.63 ^a
T ₁	19.71±3.08 ^a	21.87±0.63 ^a	43.66±11.64 ^{ab}
T ₂	14.39±3.41 ^a	21.53±0.37 ^a	43.33±5.88 ^{ab}
T ₃	18.72±3.60 ^a	7.50±2.25 ^c	36.68±8.64 ^{ab}
T ₄	19.25±2.77 ^a	20.57±1.32 ^a	31.59±3.69 ^{ab}
T ₅	15.48±2.46 ^a	15.46±3.04 ^b	26.63±5.06 ^b
T ₆	21.33±3.20 ^a	7.02±1.55 ^c	27.91±1.29 ^b

a and b values shows Mean± S.D (p<0.01) while l-value shows non-significant value

CONCLUSION

In conclusion, adding barley and water chestnut flour improved the nutritional quality of steamed cupcakes. Using composite flour with barley and water chestnut increases dietary fiber, minerals, and other nutritional properties of cupcakes. As the study was conducted using a small number of formulations, and the findings may not be generalizable to a broader range of bakery products. The study focuses on a 14-day storage period, but long-term stability, microbial growth, and potential spoilage beyond this timeframe remain unexamined. More research is needed on wheat, barley, and water chestnut flour composition on the industrial scale to enhance the nutritional quality of cupcakes.

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