ORIGINAL RESEARCH

Nutrafoods (2017) 16:175-183
DOI 10.17470/NF-017-1002-3

Received: February 07, 2017
Accepted: May 25, 2017

Effect of sprouting on barley flour and cookie quality of wheat–barley flour blends

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ABSTRACT

Different proportions of raw and sprouted barley flour were separately blended with wheat flour and the mixtures analyzed for their physicochemical and pasting properties. Blending with barley flour resulted in an increase in protein, fat and ash content. An increase in barley fraction significantly decreased the bulk, true density and colour (L* and b* values) of blends, while the angle of repose increased significantly. Incorporation of sprouted barley flour compared with raw barley flour led to a greater percentage change in the parameters studied: peak viscosity and setback increased, while peak time and pasting temperature decreased. Blending of wheat flour with sprouted barley flour in comparison with raw barley flour showed lower increases in peak viscosity and setback values. Flour blends were then used for the production of cookies that were analyzed for their physicochemical, textural and sensory attributes. Cookies containing raw barley flour had a higher spread ratio than cookies made from sprouted barley flour blends. Blending with barley decreased the L* value of cookies, with incorporation of sprouted barley showing a smaller decrease in L* value than raw barley. Hence, incorporation of sprouted barley resulted in flour blends with improved pasting properties and better quality gluten-free cookies.

Keywords
Sprouting
Barley
Pasting properties
Gluten-free cookies

Introduction

Barley (*Hordeum vulgare* L.) is considered a functional grain because it contains β-glucan, B-complex vitamins, tocotrienols and tocopherols, and has significant antioxidant potential [1, 2] as compared with more widely consumed cereals such as wheat and rice [3]. The risk posed by free radicals and oxidation products generated during cellular metabolism could be lowered by consuming foods rich in phenolics which include barley [4]. The Food and Drug Administration recommends a daily intake of 3 g of β-glucan to reduce the risk of coronary heart disease by lowering blood cholesterol. In light of the health benefits of barley β-glucan, barley consumption should be encouraged [5]. Although mainly utilized for malting, brewing and animal feed, barley is gaining popularity as an ingredient in different baked and extruded foods [6]. The supply of barley bioactive compounds in baked products such as cookies may be an effective way to increase consumption [7]. In addition, replacing wheat flour with barley flour is a viable option for the production of gluten-free products [8]. Some people are allergic or intolerant to the gluten protein present in wheat, a problem being studied by researchers worldwide [9].

To improve the nutritional and nutraceutical potential of cereals, pretreatments and different minimal processing techniques, such as sprouting, roasting and fermentation, have shown promising results [6, 10]. However, different processing conditions may either increase or decrease the nutritional and nutraceutical properties of food components. Sprouting of grains is considered a good method to improve the nutritional and nutraceutical quality of cereals since the technique is simple, inexpensive, improves the availability of various nutrients and significantly reduces anti-nutritional factors [8].

Although wheat–barley blends have been used for the production of bakery items, very few data are available on the minimal processing of barley for incorporation into baked products. Hence, the present investigation studied the effect of blending raw and sprouted barley flour with wheat flour on the physicochemical properties, pasting properties and cookie-making behaviour of wheat–barley flour blends.

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Materials and methods

Procurement of raw materials

Barley grain (*Hordeum vulgare* L.) was procured from the Ladakh region. Refined wheat flour (WF) was obtained from a local market in Srinagar, Kashmir.

Preparation of barley flour and sprouted barley flour

For the preparation of barley flour (BF), barley grains were thoroughly cleaned and then milled using a Brabender Quadrumat Junior rolling mill. The flour obtained was placed in an air-tight plastic container until use.

For the preparation of sprouted barley flour (SBF), barley grains were cleaned and washed in distilled water. The grains were then soaked in a volume of water three times the weight of the grains (3:1) for 12 h in a container at ambient temperature. The steeping water was then drained off and the soaked grains were washed twice with distilled water to prevent microbial growth during sprouting.

The soaked grains were next wrapped in a damp muslin cloth in ambient conditions and watered two to three times a day for 48 h to encourage sprouting. The sprouted grains were dried overnight at room temperature under a fan and then milled (Brabender Quadrumat Junior rolling mill).

The flour obtained was placed in an air-tight plastic container until use.

Formulation of flour blends

Blends were prepared by mixing BF and SBF separately with WF in the ratios 0:100, 5:95, 10:90, 15:85, 20:80 and 25:75. Refined wheat flour was treated as control.

Cooking, preparation and evaluation

Cookies were prepared using 64 g flour, 18 g sugar, 20 g fat, 1.5 g dried skimmed milk, 0.38 g baking powder and 6 ml water. The ingredients were mixed using a GF-101 food mixer (Good Friends, Taichung, Taiwan) and the dough rolled into a sheet 3 mm thick using a dough sheeter. Circular biscuits were cut with a cookie cutter (5.5 cm) and baked at 150°C for 15 min in an electric oven. The baked cookies were cooled to room temperature and packed in air-tight containers for further analysis.

Analysis of flour samples

Proximate composition of flour

The proximate composition of the flours including moisture, crude ash, crude fat and crude protein was determined according to AOAC methods as described by Shafi *et al* [9]: crude protein by the micro-Kjeldahl method (AOAC Method 960.52), crude fat by the Soxhlet extraction method (AOAC Method 963.15) and ash content by the dry ashing method (AOAC Method 923.03). The carbohydrate content (%) was calculated by subtracting crude ash, fat and protein content from 100% dry matter. The energy value per 100 g was calculated and expressed in kilocalories using the Atwater system where 1 g carbohydrate provides 4 kcal, 1 g protein provides 4 kcal and 1 g fat provides 9 kcal.

Physical properties of flour samples

Loose and packed bulk density

Bulk density was determined as described by Shafi *et al* [9]. To determine loose bulk density, an empty 50 ml measuring flask was weighed and then filled with gentle tapping up to the mark with flour and weighed again. For packed bulk density, the sample in the measuring flask was tapped down and more flour was added up to the mark before weighing. The results were reported as g/ml.

Angle of repose

To determine the angle of repose, an acrylic box with a wooden bottom (100×100 mm) and a removable front panel (especially designed for the experiment) was used. The box was filled with barley grain and placed on a horizontal surface. The front panel was then quickly removed, allowing the grains to slide down and take their natural slope. The angle of repose was calculated by measuring the grain bed depth (height of the natural slope) at two points.

Colour characteristics of flour

The colour characteristics of flour samples were measured using a MiniScan XE Plus 45/0 colour analyzer (HunterLab, Reston, VA, USA). L*, a* and b* were measured, with each value being the average of four determinations.

Pasting properties

The pasting properties of WF, BF and SBF were evaluated using the RVA-TecMaster (Perten Instruments, Macquarie Park, NSW, Australia). A programmed heating and cooling cycle was employed. The samples were held at 50°C for 1 min and then heated to 95°C at 12°C/min. Samples were held at 95°C for 2.5 min and then cooled to 50°C at 12°C/min and held at 50°C for 2 min. The following were measured: pasting temperature, peak viscosity, trough viscosity (minimum viscosity at 95°C), final viscosity (viscosity at 50°C), breakdown viscosity (peak–trough viscosity) and setback viscosity (final–trough viscosity).

Analysis of cookies

Proximate composition of cookies

The proximate composition of the cookies including moisture, crude ash, crude fat and crude protein was determined according to AOAC 2000 methods as described by Bazaz *et al* [10]. The carbohydrate content (%) was calculated by subtracting the crude ash, fat and protein content from 100% dry matter. The energy value per 100 g was calculated and expressed in kilocalories using the Atwater system where 1 g carbohydrate provides 4 kcal, 1 g protein provides 4 kcal and 1 g fat provides 9 kcal.

Physical characteristics of cookies

The thickness (T) of the cookies was measured using a Vernier caliper and the diameter (W) was measured. The spread ratio (W/T) was then calculated. Cookies were weighed using a Kern EMB 1000-2 electronic precision weighing scale. The colour characteristics of the cookies were measured using a MiniScan XE Plus 45/0 colour analyzer (HunterLab). L*, a* and b* were measured, with each value being the average of four determinations.

Texture analysis of cookies

The textural properties of the final products were investigated using a TA.XP plus texture analyzer (Stable Micro Systems, Haslemere, UK). The fracture strength was measured using a three-point bending rig and a 5 kg load cell. The distance between the two beams was 60 mm. A third identical beam was lowered from above (pre-test speed of 1.0 mm/s, test speed of 3.0 mm/s, post-test speed of 10.0 mm/s, distance 5 mm) on to the cookie until the cookie broke. The peak force was reported as fracture strength. Ten samples from each formulation were tested.

Sensory evaluation of cookies

Cookies prepared from WF and its blends with BF and SBF were subjected to sensory evaluation by a semi-trained panel of 15 individuals aged 25–30 years. Before sensory evaluation, the panel members were trained using commercial cookies so that they were familiar with the rating method. The attributes and sensory characteristics of each sample were evaluated on a nine-point hedonic rating scale where 9 was 'like extremely', 8 'like very much', 7 'like moderately', 6 'like slightly', 5 'neither like nor dislike', 4 'dislike slightly', 3 'dislike moderately', 2 'dislike very much' and 1 'dislike extremely'. The colour, appearance, flavour, texture, taste and overall acceptability of the cookies were evaluated. The panel members were instructed to cleanse their palates with cold, filtered tap water before tasting. Testing was conducted under daylight illumination.

Statistical analysis

The results are expressed as the mean±SD of three independent replications. One-way analysis of variance (ANOVA) and Duncan's test were used to determine statistical significance, which was set at $p \leq 0.05$. SPSS Statistics V21 software was used.

Results and discussion

Proximate composition of flour samples

Blending with BF and SBF had a significant effect on the proximate composition of flour blends and increased their moisture content (Table 1). Barley has a higher moisture content than wheat due to its greater fibre content [11]. Replacement of WF with BF non-significantly increased the moisture content to 11.25–11.96%. An increase in moisture

Blend		**Moisture** (%)	**Ash** (%)	**Crude**	**Crude fat** (%)	**Carbohydrates** (%)	**Energy** (kcal/100 g)
Control		10.55^{a}±0.59	0.53^{a}±0.11	11.73^{a}±0.33	1.20^{a}±0.01	75.98^{a}±0.05	361.64^{a}±0.01
WF/BF	5%	11.25^{b}±0.23	0.66^{a}±0.02	12.43^{b}±0.90	1.80^{b}±0.01	73.86^{b}±0.05	361.36^{b}±0.01
	10%	11.43^{b}±0.01	0.81^{a}±0.07	12.57^{b}±1.00	1.93^{b}±0.02	73.56^{c}±0.02	316.89^{c}±0.02
	15%	11.69^{b}±0.01	1.03^{a}±0.05	13.51^{c}±0.34	2.67^{c}±0.01	71.10^{d}±0.03	362.47^{d}±0.01
	20%	11.87^{c}±0.01	1.05^{a}±0.01	13.60^{c}±0.15	2.83^{c}±0.02	71.19^{e}±0.02	364.63^{e}±0.02
	25%	11.96^{c}±0.01	1.08^{a}±0.01	13.92^{d}±0.29	3.20^{d}±0.03	69.84^{d}±0.05	363.84^{f}±0.01
WF/SBF	5%	11.87^{c}±0.01	1.42^{b}±0.01	13.47^{c}±0.3	1.56^{e}±0.40	71.68^{e}±0.02	354.64^{b}±0.01
	10%	11.93^{c}±0.02	1.49^{b}±0.02	13.83^{d}±0.03	1.77^{b}±0.11	70.98^{f}±0.03	355.17^{c}±0.01
	15%	12.23^{d}±0.02	1.53^{c}±0.01	14.25^{e}±0.01	2.28^{f}±0.17	69.71^{g}±0.05	356.36^{c}±0.01
	20%	12.45^{e}±0.02	1.61^{d}±0.01	14.43^{e}±0.05	2.69^{g}±0.05	68.82^{h}±0.01	357.21^{d}±0.01
	25%	12.62^{f}±0.01	1.72^{e}±0.03	14.77f±0.02	2.97h±0.05	67.92 i±0.01	357.49e±0.01

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 1 - Effect of sprouting on the proximate composition of wheat–barley flour blends

content with blending has also been reported by Ndife *et al* [12]. However, blending with SBF significantly increased moisture content to 11.88–12.62% compared with control (10.55%) as well as with WF–BF (11.25-11.96%) blends.

These results are in agreement with those reported by Hooda and Jood [13]. The greater increase in moisture content with incorporation of SBF compared with BF is due to moisture uptake during sprouting. Sprouting results in hydrolysis and the solubilization of complex carbohydrates and proteins that affects their water binding properties and hence moisture content.

Compared with control (11.73%), blending with SBF significantly ($p \leq 0.05$) increased the protein content of blends to 12.43–14.77%, while blending with BF increased it to 12.43–13.92%. Bazaz *et al* [10] also reported a higher protein content in diets containing sprouted green gram flour compared with raw green gram flour. The greater increase in protein content following the addition of SBF might be due to compositional change following the degradation of other constituents [14]. The enzymes produced during sprouting may lead to the hydrolysis of some components such as starch, resulting in an increase in the percentage of the protein fraction [10].

Blending with different proportions of BF and SBF significantly increased the fat content of blends in comparison with control (1.20%). Similar results were reported following fortification with soybean [15] and green gram flour [10]. Replacement of WF with BF and SBF significantly increased the fat content to 1.80–3.20% and 1.56–2.97%, respectively. The slightly lower fat content of SBF–WF blends compared with BF–WF blends might be due to the biosynthesis of new compounds from fats present in barley during sprouting [16].

Compared with control (75.98%), replacement of WF with BF and SBF significantly decreased the carbohydrate content to 73.86–69.84% and 71.68–67.92%, respectively. The greater reduction in the carbohydrate content of SBF–WF blends compared with BF–WF blends may be due to the increase in endogenous α- and β-amylases during sprouting and consequent hydrolysis of starch, resulting in a decrease in carbohydrate content.

The ash content of food indicates mineral content and increased with barley flour incorporation. In comparison with control (0.533%), blending with different proportions of BF and SBF significantly increased the ash content of flour blends to 0.66–1.08% and 1.42–1.72%, respectively. The slight increase in the ash content of SBF–WF blends compared with BF–WF blends might be due to phytase enzyme activity during sprouting.

Compared with control (361.64 kcal), blending with different proportions of BF and SBF significantly increased the energy value of blends to 361.36–363.84 kcal and 354.64–357.49 kcal, respectively. The greater increase in energy content following blending with SBF is due to the greater increase in the fat and protein content of the blends.

Physical properties of BF/SBF–WH blends

The physical properties of flour blends are shown in Table 2. Bulk density depends on sample particle size. It is a measure of the heaviness of a flour sample and is important for determining packaging requirements, material handling and wet processing [17]. The bulk density of WF (0.47 g/ml) was significantly higher than that of BF–WF and SBF–WF blends (0.40–0.19 g/ml and 0.18–0.12 g/ml, respectively). Sprouting resulted in a decrease in the bulk density of barley flour.

Blends		Bulk density (g/ml)	True density	Angle of repose	Colour		
					L*	a*	b*
Control		$0.47^{a}\pm0.02$	$0.71^{a}\pm0.02$	$27.1^{a}\pm0.01$	$86.53^{a}\pm0.01$	$1.32^{a}\pm0.02$	$19.00^{a}\pm0.02$
WF/BF	5%	$0.40^{b}\pm0.01$	$0.68^{b}\pm0.01$	$28.4^{b}\pm0.02$	$86.40^{b}\pm0.02$	$1.33^{b}\pm0.01$	$18.09^{b}\pm0.02$
	10%	$0.31^{c}\pm0.01$	$0.65^{c}\pm0.01$	$30.10^{c}\pm0.05$	$86.31^{c}\pm0.59$	$1.37^{c}\pm0.03$	$17.08^{c}\pm0.01$
	15%	$0.25^{d}\pm0.01$	$0.60^{d}\pm0.05$	$31.10^{d}\pm0.01$	$86.21^{d}\pm0.02$	$1.40^{d}\pm0.01$	$16.03^{d}\pm0.05$
	20%	$0.20^{e}\pm0.05$	$0.58^{e}\pm0.02$	$31.23^{e}\pm0.02$	$86.11^{e}\pm0.01$	$1.49^{e}\pm0.02$	$15.09^{e}\pm0.01$
	25%	$0.19^{f}\pm0.03$	$0.55^{f}\pm0.02$	$31.36^{f}\pm0.03$	$86.10^{f}\pm0.01$	$1.57^{f}\pm0.05$	$14.04^{f}\pm0.01$
WF/SBF	5%	$0.18^{g}\pm0.02$	$0.50^{g}\pm0.01$	$32.00^{g}\pm0.01$	$85.90^{g}\pm0.02$	$1.69^{g}\pm0.05$	$13.80^{g}\pm0.59$
	10%	$0.16^{h}\pm0.59$	$0.48^{h}\pm0.02$	$32.55^{h}\pm0.01$	$85.88^{h}\pm0.03$	$1.72^{h}\pm0.01$	$12.00^{h}\pm0.02$
	15%	$0.15^{i}\pm0.01$	$0.45^{i}\pm0.02$	$32.73^{i}\pm0.03$	$85.75^{i}\pm0.01$	$1.81^{i}\pm0.02$	$11.00^{i}\pm0.01$
	20%	$0.14^{j}\pm0.02$	$0.42^{j}\pm0.05$	$32.97^{j}\pm0.59$	$85.63^{i}\pm0.03$	$1.89^{j}\pm0.05$	$10.62^{j}\pm0.02$
	25%	$0.12^{k}\pm0.01$	$0.40^{k}\pm0.01$	$33.20^{k}\pm0.05$	$84.91^{j}\pm0.02$	$1.96^{k}\pm0.01$	$10.00^{k}\pm0.59$

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 2 - Physical properties of wheat–barley flour blends

Victor *et al* [18] also reported a decrease in bulk density due to sprouting and attributed it to the increased activity of α-amylase which converts starch to dextrin. Chauhan *et al* [19] also reported a lower bulk density in sprouted amaranth flour as compared with raw amaranth flour. The true density of WF (0.71 g/ml) was significantly higher than that of BF–WF and SBF–WF blends (0.68–0.55 g/ml and 0.50–0.40 g/ml, respectively). The angle of repose of WF (27.1) also significantly increased following blending with BF and SBF to 28.4–31.36 and 32.00–33.20, respectively. Sprouting results in an increase in moisture content and an increase in moisture content is usually associated with an increase in the angle of repose. As the angle of repose is very important in the design of hopper openings, pending side walls and storage structures, the moisture content of materials should also be taken into account when designing equipment and structures.

Compared to control (86.53), blending with BF and SBF resulted in significant decreases in L* values to 86.40–86.10 and 85.90–84.91, respectively. Chauhan *et al* [19] also reported a decrease in the L* value of cookies with added amaranth flour compared with wheat flour cookies. The authors reported a negative correlation between protein content and lightness of flour, so the decrease in the lightness of SBF could be due to the increased total protein content. Compared with control (1.32), blending with BF and SBF resulted in significant increases in a* values to 1.33–1.96. The b* values of the blends were also significantly lower than control (19.00), with values of 18.09–10.00.

Pasting properties of BF/SBF–WF blends

Incorporation of BF and SBF significantly increased the pasting properties of flour blends (Table 3) due to the higher pasting properties of barley starch compared with wheat starch [20] and the presence of β-glucan in barley. Lazaridou *et al* [21] also reported that a solution containing β-glucan is more viscous than a starch solution of the same concentration. Hence, the increased β-glucan content in flour blends with an increased barley fraction will result in increased pasting properties. Peak viscosity is an indicator of the ease with which starch granules disintegrate and is often correlated with final product quality. Compared with control (1212 cP), incorporation of varying proportions of BF and SBF increased the peak viscosity to 1224–1312 cP and 1210–1258 cP, respectively. Similar results were reported by Sullivan *et al* [20] and Sharma and Gujral [22]. Likewise, the trough viscosity of flour blends significantly ($p \leq 0.05$) increased with increased proportions of blended BF. Incorporation of different proportions of BF and SBF with WF increased the trough viscosity to 516–571 cP and 513–559 cP, respectively. Blending with BF and SBF also significantly ($p \leq 0.05$) increased the breakdown value of blends to 361–415 cP and 355–395 cP compared with control (352 cP) and increased final viscosity to 1977–2027 cP and 1967–2012 cP, respectively. Incorporation of SBF resulted in a slightly lower increase in the pasting properties of flour blends compared with incorporation of BF, as mentioned above. Sprouting results in starch disintegration due to enzyme activity and so the peak viscosity of SBF–WF blends is slightly lower than that of BF–WF blends. Also sprouting causes a decrease in the molecular chain length of β-glucan which further decreases the viscosity imparted by SBF compared with BF. Incorporation of BF and SBF also increased the setback of blends to 832–873 cP and 820–851 cP, respectively in com-

Blends		Peak viscosity (cP)	Trough viscosity (cP)	Break down (viscosity)	Final viscosity	Setback	Peak time	Pasting temperature
Control		$1212^{a}\pm0.05$	$507^{a}\pm0.01$	$352^{a}\pm0.05$	$1960^{a}\pm0.02$	$820^{a}\pm0.03$	$6.97^{a}\pm0.04$	$92.54^{a}\pm0.02$
WF/BF	5%	$1224^{b}\pm 0.01$	$516^{b}\pm0.03$	$361^{b}\pm0.01$	$1977^{b}\pm0.01$	$832^{b}\pm0.01$	$6.78^{a}\pm0.07$	$92.41^{b}\pm0.01$
	10%	$1236^{c}\pm 0.01$	$532^{c}\pm0.05$	$376^{e}\pm0.03$	$1989^{c}\pm0.57$	$840^{c}\pm0.01$	$6.69^{a}\pm0.02$	$92.33^{b}\pm0.05$
	15%	$1247^{d}\pm 0.02$	$549^{d}\pm0.02$	$388^{f}\pm0.01$	$1996^{c}\pm0.01$	$851^{d}\pm0.05$	$6.56^{a}\pm0.01$	$92.18^{b}\pm0.02$
	20%	$1267^{e}\pm0.02$	$560^{e}\pm0.01$	$397.3^{g}\pm0.03$	$2012^{d}\pm0.93$	$862^{e}\pm0.03$	$6.42^{a}\pm0.02$	$92.10^{b}\pm0.02$
	25%	$1312^{f}\pm0.03$	$571^{f}\pm0.02$	$415^{h}\pm0.01$	$2027^{e}\pm0.05$	$873^{f}\pm0.05$	$6.31^{a}\pm0.01$	$91.96^{b}\pm0.01$
WF/SBF	5%	$1210^{g}\pm0.03$	$513^{e}\pm0.93$	$355^{i}\pm0.02$	$1967^{f}\pm0.01$	$820^{a}\pm0.04$	$6.29^{b}\pm0.04$	$92.21^{b}\pm0.01$
	10%	$1221^{h}\pm0.02$	$523^{f}\pm0.01$	$367^{j}\pm0.02$	$1971^{g}\pm0.02$	$829^{a}\pm0.01$	$6.14^{c}\pm0.01$	$92.13^{c}\pm0.01$
	15%	$1239^{i}\pm0.01$	$537^{g}\pm0.05$	$379^{k}\pm0.05$	$1987^{g}\pm0.01$	$832^{b}\pm0.06$	$6.14^{d}\pm0.01$	$92.00^{c}\pm0.01$
	20%	$1242^{i}\pm0.05$	$549^{h}\pm0.02$	$389^{k}\pm0.02$	$1996^{g}\pm0.03$	$847^{c}\pm0.05$	$5.98^{e}\pm0.01$	$91.98^{e}\pm0.06$
	25%	$1258^{k}\pm0.57$	$559^{i}\pm0.01$	$395^{k}\pm0.01$	$2012^{h}\pm0.01$	$851^{d}\pm0.02$	$5.82^{f}\pm0.02$	$91.79^{f}\pm0.0.05$

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 3 - Effect of sprouting on the pasting properties of wheat–barley flour blends

parison to control (820 cP). An increase in β-glucan content should decrease the setback value as reported previously by Brennan and Cleary [23] in wheat flour incorporated with β-glucan. However, setback is also affected by barley starch (a major component of barley flour) which seems to counter the setback lowering effect of β-glucan. The addition of BF led to a greater increase in the setback values of blends than the addition of SBF. Sprouting also decreases the chain length of the β-glucan molecule which impedes reassociation of starch molecules (steric hindrance).

Proximate composition of cookies

The proximate composition of cookies prepared from BF/SBF–WF blends is presented in Table 4. Moisture content non-significantly increased in cookies made of different proportions of BF (1.95–2.69%) and SBF (2.90–3.56%) in comparison with control (1.74%). Differences in the moisture content of cookies containing BF compared with SBF may result from their different moisture-holding capacities.

Inyang and Zakari [24] reported that germination and fermentation increase mineral content due to an increase in phytase enzyme activity during germination that increases the availability of minerals. Replacement of WF with BF and SBF non-significantly increased the ash content of cookies to values of 0.96–1.39% and 1.46–1.93%, respectively, in comparison with control cookies (0.76%). Abayomi *et al* [25] also reported a non-significant increase in cookies prepared from fermented soybean flour. This increase in ash content after sprouting might be due to variation in the ash content of the flour.

Blending with different proportions of barley flour significantly increased the protein content of cookies. In comparison with control (9.21%), replacing WF with BF and SBF significantly increased the protein content of cookies to 9.43–9.82% and 9.90–10.30%, respectively. This trend of increase in protein content in the treatments as compared with control was supported by several studies [26, 27, 28]. Soybean is a high-protein legume so if it is added to wheat flour it increases the protein content and complements lysine-limited cereal protein. Hence, soy flour is used as an economical protein supplement in biscuits, breads, pasta and other cereal products [29]. Bazaz *et al* [10] also reported a significant increase in the protein content of complementary diets. The increase in the protein content of cereals after sprouting might be due to enzymatic changes, phytohormone changes or a compositional change following the degradation of other constituents [16].

The fat content of food can affect its shelf stability because fat can undergo oxidative deterioration, which leads to rancidification and spoilage. Hence food with a high fat content is more likely to spoil. Compared with control (11.76), replacement of WF with BF and SBF significantly ($p \leq 0.05$) increased the fat content of cookies to 11.93–13.00% and 13.23–13.96%, respectively, which can be attributed to the higher fat content of barley flour. These results are in agreement with earlier results for soybean [16] and sprouted brown rice [29]. Compared with control (76.53%), blending with different proportions of BF and SBF significantly ($p \leq 0.05$) decreased the carbohydrate content of cookies to 75.73–73.10% and 72.51–70.25%, respectively. A reduction in carbohydrate content after sprouting can be attributed to an increase in endogenous α- and β-amylases during sprouting and consequent hydrolysis of starch [30]. Blending with SBF significantly decreased the carbohydrate content of blends as the

Blends		Moisture (%)	Ash (%)	Crude protein (%)	Crude fat (%)	Carbohydrates (%)	Energy (kcal/100 g)
Control		$1.74^a \pm 0.01$	$0.76^a \pm 0.03$	$9.21^a \pm 0.01$	$11.76^a \pm 0.01$	$76.53^a \pm 0.05$	$448.8^a \pm 0.01$
WF/BF	5%	$1.95^b \pm 0.02$	$0.96^b \pm 0.01$	$9.43^b \pm 0.02$	$11.93^b \pm 0.02$	$75.73^b \pm 0.03$	$448.01^a \pm 0.03$
	10%	$2.09^c \pm 0.03$	$1.12^c \pm 0.02$	$9.57^c \pm 0.03$	$12.40^c \pm 0.03$	$74.82^c \pm 0.01$	$449.16^a \pm 0.04$
	15%	$2.32^d \pm 0.01$	$1.20^d \pm 0.02$	$9.66^d \pm 0.04$	$12.44^c \pm 0.01$	$74.38^d \pm 0.01$	$448.12^a \pm 0.01$
	20%	$2.42^d \pm 0.01$	$1.26^d \pm 0.01$	$9.75^e \pm 0.01$	$12.48^c \pm 0.02$	$74.09^e \pm 0.02$	$447.64^a \pm 0.01$
	25%	$2.69^e \pm 0.01$	$1.39^e \pm 0.02$	$9.82^f \pm 0.05$	$13.00^d \pm 0.03$	$73.10^f \pm 0.05$	$448.68^a \pm 0.01$
WF/SBF	5%	$2.90^f \pm 0.01$	$1.46^f \pm 0.02$	$9.90^f \pm 0.03$	$13.23^e \pm 0.01$	$72.51^g \pm 0.94$	$448.71^a \pm 0.02$
	10%	$3.09^f \pm 0.02$	$1.60^g \pm 0.93$	$10.0^g \pm 0.02$	$13.47^f \pm 0.01$	$71.84^h \pm 0.01$	$448.59^a \pm 0.02$
	15%	$3.26^g \pm 0.05$	$1.74^g \pm 0.94$	$10.12^h \pm 0.01$	$13.62^g \pm 0.94$	71.26i±0.01	448.1a±0.05
	20%	$3.38^g \pm 0.03$	$1.80^h \pm 0.01$	$10.21^i \pm 0.01$	$13.82^h \pm 0.04$	70.79j±0.02	448.38a±0.01
	25%	$3.56^h \pm 0.93$	$1.93^h \pm 0.05$	10.30±0.01	$13.96^i \pm 0.01$	70.25k±0.02	447.84a±0.02

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 4 - Effect of sprouting on proximate composition of wheat–barley flour cookies

barley proportion increased compared with control as well as with blends prepared from BF.

Compared with control (448.8 kcal/100 g), replacement of WF with BF and SBF non-significantly increased the energy content of cookies to 448.01–448.68 kcal/100 g and 448.71–447.84 kcal/100 g, respectively.

Physical properties of cookies

The physical parameters of cookies prepared from BF/SBF–WF blends are given in Table 5. Physical changes during baking include alterations in dimension, loss of moisture and the development of colour and flavour. Compared with control (48.15 mm), blending with different proportions of BF and SBF significantly ($p \leq 0.05$) decreased the width of cookies to 46.19–40.20 mm and 44.02–39.54 mm, respectively. Similar results were found for raw rice and germinated rice flour cookies [31]. During germination, enzymes degrade macromolecules such as starch and protein to smaller sugars and peptides [32]. As a result, the hydrophilic nature of the cookies is increased. Hoojjat *et al* reported that cookie spread was decreased with increasing amount of hydrophilic additives in cookie dough [33].

Compared with control (5.47 mm), blending with different proportions of BF and SBF significantly ($p \leq 0.05$) increased the thickness of cookies to 6.21–7.39 mm and 6.57–7.78 mm. This can be attributed to the hydrophilic nature of the flour which caused a reduction in spread and thus an increase in cookie thickness.

The most desirable biscuits are those with high spread ratios. Compared with control (8.80), blending with different proportions of BF and SBF significantly ($p \leq 0.05$) decreased the spread ratio of cookies to 7.43–5.43 and 6.70–5.08, respectively. Singh *et al* [34] and Sudha *et al* [35] reported that incorporation of barley bran reduced the spread ratio of cookies.

Compared with control (95.10 g), the snap force of cookies also increased to 96.37–105.62 g and 95.12–101.29 g with incorporation of BF and SBF, respectively. Similar results were reported in cookies by Sharma and Gujral [22]. An increase in breaking strength was also reported by Frost *et al* [7] and Sudha *et al* [35] with incorporation of barley flour. However, the increase in snap force was lower in SBF–WF blended cookies. Chauhan *et al* [19] showed a similar trend in the snap force values of cookies when sprouted and raw amaranth flours were blended with wheat flour. Sprouting usually causes degradation of macromolecules which might affect the cookie matrix causing a decrease in snap force values.

The colour of cookies prepared from BF/SBF–WF blends are given in Table 5. Colour develops during the later stages of baking and is very important for the initial acceptability of baked products by consumers. Blending and sprouting significantly affected colour parameters. Compared with control (62.46), replacement of WF with BF and SBF significantly ($p \leq 0.05$) decreased the lightness of cookies to 61.15–53.2 and 52.1–48.0, respectively. The decrease in L* values with increased BF and SBF may be due to the higher protein content of barley flour; a previous study reported a decrease in L* values with increased protein content [36]. Sprouting results in release of sugars and proteins that promote the Maillard reaction resulting in an increase in the formation of brown pigments (melanoidins) which darken the cookies. Compared with control (5.16), blending with BF and SBF significantly ($p \leq 0.05$) increased the a* values of blends to 5.6–8.26 and 5.0–6.9, respectively. Frost *et al* [7] reported similar results in barley cookies with increase proportions of

Blends		Width (mm)	Thickness (mm)	Spread ratio	Snap force	Colour		
						L*	a*	b*
Control		48.15^{a}±0.01	5.47^{a}±0.01	8.80^{a}±0.01	95.10^{a}±0.01	62.46^{a}±0.05	5.16^{a}±0.01	28.99^{a}±1.0
WF/BF	5%	46.19^{b}±0.01	6.21^{b}±0.01	7.43^{b}±0.01	96.37^{b}±0.02	61.15^{b}±0.01	5.6^{a}±0.02	30.21^{b}±1.0
	10%	45.12^{c}±0.01	6.44^{c}±0.03	7.00^{c}±0.01	98.29^{c}±0.01	59.1^{c}±0.01	6.11^{b}±0.01	33.12^{c}±0.95
	15%	44.01^{d}±0.02	6.71^{d}±0.02	6.55^{d}±0.01	100.54^{d}±0.05	57.10^{d}±0.06	7.21^{c}±0.01	36.04^{d}±0.01
	20%	42.02^{e}±0.02	7.06^{e}±0.01	5.95^{e}±0.01	103.39^{e}±0.01	55.0^{e}±0.01	8.72^{d}±0.26	36.98^{e}±1.0
	25%	40.20^{f}±0.01	7.39^{f}±0.02	5.43^{f}±0.01	105.62^{f}±0.02	53.2^{f}±0.05	8.26^{d}±0.30	38.61^{f}±1.0
WF/SBF	5%	44.02^{d}±0.01	6.57^{g}±0.01	6.70^{g}±0.01	95.12^{a}±0.01	52.1^{f}±0.05	5.0^{e}±0.04	36.01^{e}±1.0
	10%	43.09^{g}±0.02	6.78^{d}±0.005	6.35^{h}±0.01	95.75^{a}±0.05	50.10^{g}±0.03	5.6^{f}±0.10	38.82^{f}±0.97
	15%	41.62^{h}±0.02	7.01^{e}±0.01	5.93^{e}±0.01	97.35^{g}±0.01	49.1^{h}±0.03	6.1^{g}±0.01	40.01^{g}±0.95
	20%	41.13^{h}±0.01	7.41^{f}±0.01	5.55^{f}±0.03	99.19^{h}±0.02	48.1^{i}±0.05	6.7^{h}±0.02	42.05^{h}±1.0
	25%	39.54^{i}±0.03	7.78^{h}±0.03	5.08^{i}±0.01	101.29^{i}±0.01	48.0^{i}±0.05	6.9^{i}±0.02	42.04^{h}±1.0

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 5 - Physical characteristics of cookies made from wheat–barley flour blends

barley flour. The Maillard reaction is influenced by many factors such as water activity, pH, temperature, sugars, and type and ratio of amino acids. Compared with control (28.99), incorporation of BF and SBF significantly ($p \leq 0.05$) increased b* values to 30.21–38.61 and 36.1–42.04, respectively.

Sensory analysis of cookies

The results of sensory analysis of cookies prepared from BF/SBF–WF blends are presented in Table 6. Compared with control (7.89), the colour scores of cookies prepared from BF/SBF–WF blends were 8.20–8.96 and 7.46–8.52, respectively. The appearance score increased as the percentage of barley flour increased. Taste and flavour are important parameters when evaluating the sensory attributes of food as the product might be appealing and have a high energy density but without a good taste and flavour it is unlikely to be accepted [7]. The flavour score increased with increasing proportions of BF but decreased with increasing proportions of SBF [37]. Skrbic *et al* [37] also reported that cookies prepared with 70% barley flour were acceptable to consumers. Sudha *et al* [35] also similarly reported that incorporation of up to 20% barley bran into wheat flour did not affect the sensory quality of biscuits but that further increases did. In our study, the taste score decreased as SBF levels increased. Variation in overall acceptability scores due to varying amounts of natural additives has also been reported [9, 10, 38].

Conclusions

Barley can be used for the production of gluten-free products with improved protein and fibre content. Sprouting can be a useful technique to improve the quality of gluten-free cookies made from wheat–barley blends. Sprouting both improved the pasting property of flour blends and also enhanced cookie characteristics such as snap force, but negatively affected the colour of cookies. Further studies, for instance on barley sprouting time and conditions, are needed so final product characteristics can be improved.

Acknowledgements

The authors thank the Department of Biotechnology, Government of India.

Conflict of interest

None of the authors has a conflict of interest.

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Blends		Colour	Appearance	Texture	Flavour	Taste	Overall acceptability
Control		$7.89^{a}\pm0.01$	$7.1^{a}\pm0.17$	$7.43^{a}\pm0.25$	$5.36^{a}\pm0.20$	$7.36^{a}\pm0.37$	$6.61^{a}\pm0.28$
WF/BF	5%	$8.20^{b}\pm0.05$	$8.14^{b}\pm0.01$	$8.33^{b}\pm0.40$	$6.46^{b}\pm0.20$	$7.66^{b}\pm0.34$	$7.62^{b}\pm0.23$
	10%	$8.42^{c}\pm0.01$	$8.26^{c}\pm0.01$	$8.24^{c}\pm0.02$	$7.18^{c}\pm0.04$	$8.00^{c}\pm0.25$	$8.41^{c}\pm0.10$
	15%	$8.53^{d}\pm0.01$	$8.31^{d}\pm0.36$	$8.15^{d}\pm0.04$	$8.30^{d}\pm0.04$	$8.36^{d}\pm0.30$	$8.41^{c}\pm0.10$
	20%	$8.63^{e}\pm0.02$	$8.42^{e}\pm0.32$	$8.00^{e}\pm0.02$	$8.52^{e}\pm0.04$	$8.62^{e}\pm0.11$	$8.53^{d}\pm0.25$
	25%	$8.96^{f}\pm0.05$	$8.53^{f}\pm0.32$	$8.00^{e}\pm0.03$	$8.82^{f}\pm0.28$	$8.90^{f}\pm0.02$	$8.59^{d}\pm0.32$
WF/SBF	5%	$7.46^{g}\pm0.16$	$8.13^{b}\pm0.04$	$8.29^{c}\pm0.02$	$7.11^{g}\pm0.04$	$7.62^{g}\pm0.02$	$7.12^{e}\pm0.03$
	10%	$8.08^{h}\pm0.09$	$8.27^{c}\pm0.20$	$8.22^{c}\pm0.04$	$7.22^{h}\pm0.02$	$7.51^{h}\pm0.01$	$7.00^{e}\pm0.25$
	15%	$8.27^{i}\pm0.08$	$8.38^{d}\pm0.15$	$8.12^{d}\pm0.04$	$6.40^{i}\pm0.04$	$7.11^{i}\pm0.02$	$6.97^{f}\pm0.30$
	20%	$8.35^{j}\pm0.12$	$8.46^{e}\pm0.15$	$7.89^{e}\pm0.28$	$6.91^{j}\pm0.02$	$7.00^{i}\pm0.03$	$5.89^{g}\pm0.01$
	25%	$8.52^{k}\pm0.08$	$8.56^{f}\pm0.02$	$7.80^{f}\pm0.04$	$6.78^{k}\pm0.03$	$6.97^{j}\pm 0.01$	$5.80^{g}\pm0.02$

Mean±SD. Values followed by different superscript letters in a column are significantly different ($p \leq 0.05$)
BF barley flour, *SBF* sprouted barley flour, *WF* wheat flour.

Table 6 - Sensory analysis of cookies made from wheat–barley flour blends

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