Food Analytical Methods (2022) 15:2505–2512
https://doi.org/10.1007/s12161-022-02311-5

Germinated Barley Cultivars: Effect on Physicochemical and Bioactive Properties

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Received: 19 April 2022 / Accepted: 20 April 2022 / Published online: 30 April 2022

Abstract

Germination is an excellent green food development process for improving the nutritional content of grains. Baking with sprouted barley flour is highly recommended to get the most out of barley nutrition. Soaking and sprouting whole grains helps to release their nutrients, and then the body can absorb and use the various vitamins and minerals contained in the grain, making the grains more health-beneficial. This study was conducted to investigate the effect of germination of different barley cultivars on the improved health-benefit quality of flours. Germination of different barley cultivars resulted in a significant increase in ash, protein and fiber content, antioxidant properties (total flavonoids content, total phenolic content, antioxidant activity, metal chelating activity, and $ABTS^{+}$ scavenging activity), and a*, b*, and redness intensity values related to color parameters, compared to the L* value of germinated flours which decreased. Several studies have shown that digestibility and nutrient absorption improve when grains are processed, soaked, and sprouted and that the levels of vitamins, minerals, proteins, and antioxidants also increase. The present study results are in line with highlighting the benefit of germination in helping for a better valorization and increasing potential applications of barley.

Keywords Barley · Germination · Flour · Antioxidant · Bioactive compound · Flavonoid · Phenolic · Protein · Fiber

Introduction

People recognized and appreciated the fact that germinated grain is something special and extremely healthy from an early age. The germ is the grain's heart. The grain begins to germinate after being exposed to light, water, and warmth (Benincasa et al. 2019; Lemmens et al. 2019). Many enzymes are now being formed. The metabolically active enzymes improve the grain by breaking down and converting nutrients and forming new ones. Not only are many vitamins produced during germination, but large macromolecules such as gluten and other proteins are enzymatically broken down (Canlı et al. 2021). This significantly improves digestibility. Therefore, sprouted grain is something special, and the sprout contains the maximum amount of vital substances (Pagand et al. 2017). The germinated grain is a treasure trove of vitamins, whole grain energy, trace elements, secondary plant substances, and high-quality protein packed into the tiniest space (Cho and Lim 2016; Dietrich et al. 2016).

Barley (*Hordeum vulgare*), considered the oldest cereal cultivated by humans, is one of the most enzyme-rich plants known, and even in its resting state, it is "a bundle full of enzymes" (FAO 2017). Barley contains less gluten

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than wheat and is therefore not as suitable for baking. Germinated barley is naturally rich in maltose, a type of sugar that is used for various purposes (Andriotis et al. 2016). Barley malt is barley that has been briefly germinated and dried again. Therefore, barley malt syrup made from barley maltose can be used as a natural sweetener (Neylon et al. 2020). Barley flour (or barley meal) has also been used for centuries to make traditional "porridge," representing its basic ingredient of it. Barley bread is brown bread made from barley flour, which dates back to the Iron Age (Revilla et al. 2022).

Sprouted barley flour for baking is highly recommended to get the most benefit from barley nutrition (Rico et al. 2020). Sprouting whole grains helps to release their nutrients, and then the body can absorb and use the various vitamins and minerals contained in the grain. Whole grain products contain certain anti-nutrients, such as phytic acid and oxalates, which bind to nutrients and make them very difficult to absorb (Călinoiu and Vodnar 2018). Germination is an excellent green food development process for improving the nutritional content of grains and seeds (Ikram et al. 2021). Soaking and sprouting grains, such as uncooked barley in the husk, can reduce anti-nutrient levels, making the grains more nutritious and easier to digest (Sinha and Khare 2017). In this way can be also reduced the amount of gluten present in barley to some extent. Several studies have shown that digestibility and nutrient absorption improve when grains are processed, soaked, and sprouted and that the levels of vitamins, minerals, proteins, and antioxidants also increase (Pradeep and Guha 2011; Chinma et al. 2015). Barley flour has been demonstrated to possess different antioxidant capacities playing an important role in health benefits (Bangar et al. 2022; Zanoaga et al. 2019; Punia Bangar et al. 2022).

The present research has been driving to find out how germination affects different characteristics and physical qualities of six different barley cultivars' flours. Therefore, the present study aimed to formulate germinated barley flours and analyze the effect of germination on ash, protein and fiber content, antioxidant properties, and related color parameters of barley flour obtained after germination.

Materials and Methods

Materials

Six barley cultivars (BH-393, BH-932, BH-902, BH-885, DWR-52, and PL-172) were procured from Haryana Agriculture University, Hisar, India, as described in the previous studies (Punia Bangar et al. 2022; Bangar et al. 2022).

Germination of Barley

According to the method described by Sharma and Gujral (2010), grains were germinated. Samples of barley grains were steeped for 24 h at 25 °C, with water changes every 2 h. After soaking, barley seeds were placed in a BOD incubator (Calton, Delhi) and allowed to germinate for 24 and 48 h at 25 °C and 100% relative humidity (RH). The germinated barley was dried to a moisture content (mc) of 10% in a dryer at 40 °C. The dried samples were then sealed in airtight bags and kept for further examination.

Dehusking and Milling of Barley Grains

Dehusking of barley samples was carried out using a rice miller (Khera, Delhi). Barley cultivars were conditioned to 10% moisture content before de-husking. 150 g of conditioned barley was de-husked with the rice polisher (Khera, Delhi). The de-husked grains were then debranned and further ground.

Flours Analysis

Proximate Composition Analysis

The flour samples from different barley cultivars, non-germinated and germinated, were tested using standard analysis methods for their ash, fat, protein, and fiber content (AOAC 1990). The carbohydrate content was calculated by difference. All the measurements were replicated thrice and reported on a dry weight basis (dwb).

Color Characteristics of Flours

A Hunter Colorimeter with an optical sensor (Hunter Associates Laboratory Inc., Reston, VA, USA) was used to measure the color of flours using the L*, a*, and b* color system. The color difference (ΔE) was calculated as:

$$\Delta E = \left\{ \left(\mathrm{dL}^*\right) + \left(\mathrm{da}^*\right) + \left(\mathrm{db}^*\right) \right\}^{1/2}$$

The lightness is indicated by the L* value, which ranges from 0 to 100. The degree of red-green color is determined by the a* value, with a more positive a* value suggesting more red. The degree of yellow-blue color is indicated by the b* value, with a more positive b* value suggesting more yellow. Redness intensity (RI) was calculated for each sample as: $RI = \mathrm{a}^*/\mathrm{b}^*$.

Total Phenolic Content (TPC) of Flours

TPC of flours from different barley cultivars non-germinated (published by Punia Bangar et al. 2022; Bangar et al.

Table 1 Physicochemical composition of flours from non-germinated and germinated barley cultivars

Barley cultivars	Ash (%)		Fat (%)		Protein (%)		Crude fiber (%)		Carbohydrate (%)		Amylose (%)	
	Non-germinated	Germinated	Non-germinated	Germinated	Non-germinated	Germinated	Non-germinated	Germinated	Non-germinated	Germinated	Non-germinated	Germinated
BH-393	1.57^{b}	$1.63^{b}_{\uparrow 3.8}$	4.1^{bc}	$3.2^{c}_{\downarrow 21.9}$	13.3^{cd}	$14.3^{c}_{\uparrow 7.5}$	1.4^{ab}	$1.8^{b}_{\uparrow 28.5}$	74.5^{a}	$72.4^{b}_{\downarrow 2.8}$	18.9^{b}	$17.2^{d}_{\downarrow 8.9}$
BH-932	1.38^{a}	$1.48^{a}_{\uparrow 7.2}$	4.2^{c}	$2.9^{c}_{\downarrow 30.9}$	12.9^{b}	$13.3^{b}_{\uparrow 3.1}$	1.3^{ab}	$1.4^{a}_{\uparrow 7.6}$	74.7^{a}	$72.1^{b}_{\downarrow 3.4}$	16.8^{a}	$14.6^{b}_{\downarrow 13}$
BH-902	1.68^{c}	$1.71^{c}_{\uparrow 1.7}$	3.2^{a}	$2.1^{ab}_{\downarrow 34.3}$	13.6^{c}	$14.1^{c}_{\uparrow 3.6}$	1.4^{ab}	$1.6^{ab}_{\uparrow 14.2}$	76.8^{b}	$73.5^{c}_{\downarrow 4.2}$	16.3^{a}	$13.1^{a}_{\downarrow 19.6}$
BH-885	1.77^{d}	$1.95^{e}_{\uparrow 10.1}$	4.0^{b}	$2.7^{b}_{\downarrow 32.5}$	13.1^{cd}	$13.7^{bc}_{\uparrow 4.5}$	1.4^{ab}	$1.5^{ab}_{\uparrow 7.1}$	74.4^{a}	$71.3^{a}_{\downarrow 4.1}$	19.4^{c}	$19.3^{e}_{\downarrow 0.5}$
DWR-52	1.58^{b}	$1.69^{bc}_{\uparrow 6.9}$	3.4^{a}	$2.1^{ab}_{\downarrow 38.2}$	12.8^{b}	$13.2^{b}_{\uparrow 3.1}$	1.5^{b}	$1.7^{ab}_{\uparrow 13.3}$	77.1^{b}	$72.6^{b}_{\downarrow 5.8}$	17.5^{ab}	$14.5^{b}_{\downarrow 17.1}$
PL-172	1.61^{bc}	$1.76^{d}_{\uparrow 9.3}$	3.3^{a}	$1.9^{a}_{\downarrow 42.4}$	11.6^{a}	$12.5^{a}_{\uparrow 7.7}$	1.1^{a}	$1.3^{a}_{\uparrow 18.1}$	79.5^{c}	$76.3^{d}_{\downarrow 4}$	18.2^{b}	$15.3^{c}_{\downarrow 15.9}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) and decrease (↓) from control samples for corresponding properties.

2022) and germinated (at 24 h and 48 h) was determined by following the Folin-Ciocalteau method described by Gao et al. (2002). Briefly, 200 mg of ground barley samples were extracted with acidified methanol (HCl/methanol/water, 1:80:10, v/v) (4 mL) at room temperature for 2 h. The extracts obtained were oxidized with a Folin-Ciocalteau reagent, and the reaction mixture was neutralized with sodium carbonate. The mixture was incubated at room temperature for 90 min, and its absorbance using a spectrophotometer (Systronics-106, Ahmedabad) was measured at 725 nm. Acidified methanol was used as a blank. Gallic acid was used as the standard, and the results are expressed as µg gallic acid equivalents (GAE)/g of flour.

Total Flavonoids Content (TFC) of Flours

TFC of flours from different barley cultivars non-germinated (published by Punia Bangar et al. 2022; Bangar et al. 2022) and germinated (at 24 h and 48 h) was determined by following the method described by Jia et al. (1998). 1.25 mL distilled water was used to dilute the barley extract (250 µL). The sodium nitrite (75 µL of a 5% solution) was added, and the mixture was allowed to stand for 6 min. In addition, 150 µL of a 10% aluminum chloride solution was added, and the combination was allowed to react for 5 min. After that, the solution was then stirred well with 0.5 mL of 1 M sodium hydroxide. A spectrophotometer was used to detect the absorbance at 510 nm. The standard was catechin, and the results were expressed as g of catechin equivalents (CE)/g of flour.

Antioxidant Activity (AOA) of Flours

Antioxidant Potential Radical 1,1-Diphenyl-2-Picrylhydrazyl (DPPH) Radical Scavenging Activity The activity of DPPH radical scavenging was determined using a modified version of the Brand-Williams et al.'s method described (1995). Ground non-germinated (published by Punia Bangar et al. 2022; Bangar et al. 2022) and germinated (at 24 h and 48 h) barley cultivars samples were extracted with 100% methanol for 2 h at ambient temperature. The extracts were mixed with DPPH solution (6×10^{-5} mol/L of methanol). The absorbance (A) of the mixture at 515 nm was determined at 0 and 30 min. Methanol was used as a blank, and AOA was calculated as percent discoloration.

$$\%AOA = \left(1 - \left(A\ of\ sample_{t=0}/A\ of\ control_{t=30}\right) \times 100\right.$$

Metal Chelating Activity (MCA) MCA of non-germinated (published by Punia Bangar et al. 2022; Bangar et al. 2022) and germinated (at 24 h and 48 h) barley cultivars extract was measured by the method described by Dinis et al. (1994). The extract (0.5 mL) was mixed with 50 µL

Table 2 Hunter color characteristics of flours from germinated and non-germinated[§] barley cultivars

Barley cultivars	L*		a*		b*	
	Non-germinated[§]	Germinated	Non-germinated[§]	Germinated	Non-germinated[§]	Germinated
BH-393	91.2^{c}	$80.3^{d}_{\downarrow 11.9}$	0.59^{a}	$1.23^{a}_{\uparrow 108}$	9.25^{b}	$10.27^{ab}_{\uparrow 11}$
BH-932	89.5^{a}	$79.2^{c}_{\downarrow 11.5}$	1.38^{d}	$1.99^{c}_{\uparrow 44.2}$	11.35^{e}	$13.5^{d}_{\uparrow 18.9}$
BH-902	92.7^{d}	$80.6^{d}_{\downarrow 13}$	0.63^{a}	$1.53^{b}_{\uparrow 142}$	7.75^{a}	$9.95^{a}_{\uparrow 28.3}$
BH-885	90.4^{b}	$79.3^{c}_{\downarrow 12.2}$	1.13^{c}	$2.33^{e}_{\uparrow 106}$	11.52^{f}	$13.6^{d}_{\uparrow 18}$
DWR-52	90.6^{b}	$78.4^{b}_{\downarrow 13.4}$	1.03^{b}	$2.15^{d}_{\uparrow 108}$	10.72^{d}	$12.2^{c}_{\uparrow 13.8}$
PL-172	89.2^{a}	$77.6^{a}_{\downarrow 13}$	1.08^{b}	$2.28^{de}_{\uparrow 111}$	9.81^{c}	$10.88^{b}_{\uparrow 10.9}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) and decrease (↓) from control samples for corresponding properties. L* = darkness (0) or lightness (100); a* = redness (+); b* = yellowness (+);
[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

of ferrous chloride (2 mM/L) and added 1.6 mL of 80% methanol. The reaction was initiated after 5 min by adding 5 mM/L ferrozine (100 µL), and the mixture was shaken on a vortex. The mixture was incubated at room temperature (25 °C) for 10 min. The absorbance of the solution was recorded at 562 nm on a spectrophotometer (Systronics, Ahmedabad). The chelating activity of the extract for $Fe^{2+}$was calculated as follows:

$$Iron\left(Fe^{2+}\right)\ chelating\ activity\ (\%) = \{1 - (Absorbance\ of\ sample/Absorbance\ of\ control)\} \times 100$$

ABTS^{+} Scavenging Capacity ABTS^{+} scavenging activity was measured following the method described by Re et al. (1999). This involved a diammonium salt of free radical ABTS, 2, 2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid). ABTS^{+} was generated by the oxidation of ABTS with potassium persulfate. 3 mL of ABTS cation solution was mixed with 30 µL extract in a disposable microcuvette, and the decrease of absorption was measured after 1 min of incubation. A standard curve was prepared using different vitamin C concentrations similar to the DPPH assay. ABTS^{+} scavenging property was expressed as vitamin C in µmol/g of non-germinated (published by Punia Bangar et al. 2022; Bangar et al. 2022) and germinated (at 24 h and 48 h) barley.

Results and Discussion

Physicochemical Composition

The chemical composition of flours from germinated barley cultivars varied significantly ($P < 0.05$) (Table 1). Germination increased ash, protein, and fiber content with values ranging from 1.48 to 1.95%, 12.5 to 14.3%, and 1.3 to 1.8%, respectively. Germinated flour has a higher ash concentration than non-germinated flour, which can be attributed to lower total soluble solids and likely dry matter loss (Chinma et al. 2015). Researchers reported that the increase of protein content by germination could be attributed to the net synthesis of enzyme protein which might have resulted in the production of some amino acids during protein synthesis (Ali and Elozeiri 2017; Nkhata et al. 2018). The higher soluble dietary fiber of germinated flour than non-germinated counterparts could be attributed to new primary cell walls (Benincasa et al. 2019; Budhwar et al. 2020). However, fat and carbohydrate content among cultivars decreased after germination and ranged from 1.9 to 3.2% and 71.3 to 76.3%, respectively. Amylose content of barley flour decreased after germination and ranged between 13.1 and 19.3%, respectively, among cultivars. Chinma et al. (2015) reported that decreased amylose contents of flours from germinated grains could be due to amylase activity in respiration metabolism during germination.

Hunter Color Characteristics of Flours

Color parameters of flour from different non-germinated (published by Punia Bangar et al. 2022; Bangar et al. 2022) and germinated barley cultivars were evaluated using a hunter color lab (Table 2). The L* value of germinated flours decreased, and the values ranged from 77.6 to 80.6; the highest for cv.BH-902 and the lowest for cv.PL-172 were observed. The higher L* value of flours from cv.BH-902 and cv.BH-393 indicates their light color. The a* value increased after germination and ranged from 1.23 to 2.33; the highest and the lowest values were observed for cv.BH-885 and cv.BH-393, respectively. The b* value also increased during germination, ranging from 9.95 to 13.6 among cultivars. ΔE ranged from 78.3 to 82.4 among germinated cultivars. A decrease in L* and an increase in a* and b* values of oats after germination have been reported by other studies as well (Tian et al. 2010; Li et al. 2020). An increase in a* and b* values could be due to more starch and protein hydrolysates formed, and the Maillard reaction occurs during drying

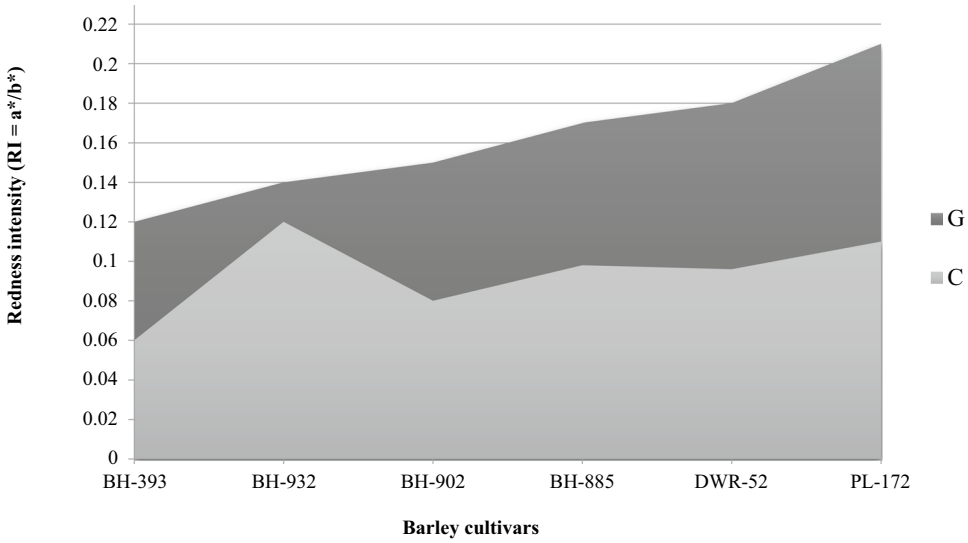

Fig. 1 Graphic representation of redness intensity (RI = a*/b*) between germinated (G) and non-germinated (C)[§] barley cultivars

treatment (Kleekayai et al. 2022). Overall, RI increased from non-germinated (range: 0.06–0.12) to germinated (range: 0.12–0.21) (Table 2 and Fig. 1).

TPC of Flour

TPC significantly ($P < 0.05$) increased (up to 35.3%) among non-germinated barley cultivars upon germination for 24 h. The highest TPC of 4548 µg GAE/g was observed for cv.BH-885, while cv.BH-393 showed the lowest value (3814 µg GAE/g) (Table 3). Sharma and Gujral (2010) reported the increase in TPC of barley flours when germinated for 24 h with values ranging between 2477 and 2723 µg GAE/g. Further, TPC increased among all cultivars when germination was allowed for 48 h, with values ranging between 3898 and 4604 µg GAE/g. The % increase was from 13.6 to 42.3%.

Table 3 Total phenolic content (TPC) of flours from non-germinated[§] and germinated (at 24 and 48 h) barley cultivars

Barley cultivar	TPC (µg GAE/g) non-germinated[§]	TPC (µg GAE/g) germinated at 24 h	TPC (µg GAE/g) germinated at 48 h
BH-393	3256^{d}	$3814^{a}{}_{\uparrow 17.1}$	$3898^{a}{}_{\uparrow 19.7}$
BH-932	3056^{c}	$3906^{b}{}_{\uparrow 27.8}$	$4034^{b}{}_{\uparrow 32}$
BH-902	3761^{e}	$4121^{c}{}_{\uparrow 9.5}$	$4275^{d}{}_{\uparrow 13.6}$
BH-885	3922^{f}	$4548^{d}{}_{\uparrow 15.9}$	$4604^{e}{}_{\uparrow 17.3}$
DWR-52	2922^{b}	$3938^{b}{}_{\uparrow 34.7}$	$4075^{b}{}_{\uparrow 39.4}$
PL-172	2890^{a}	$3911^{b}{}_{\uparrow 35.3}$	$4113^{c}{}_{\uparrow 42.3}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) from control samples for corresponding properties.

[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

The rise in phenolic compounds in germinated seeds could be attributed to the breakdown of the cell wall during germination, which leads to an increase in free phenolic forms (Van Hung 2016; Šimić et al. 2017).

TFC of Flour

The changes in TFC during barley germination are shown in Table 4. Upon germination for 24 and 48 h, a significant ($P < 0.05$) increase (up to 61.2% and 68.8%) in TFC was observed among flours from different cultivars. TFC significantly ($P < 0.05$) varied among cultivars, and the values ranged from 2778 to 3545 µg CE/g and 2937 to 3711 µg CE/g when germinated for 24 h and 48 h, respectively. The highest TFC was observed for cv.BH-885. Pradeep and Guha (2011) reported that the flavonoids content of the germinated millet extract showed an increase of up to 4.6% compared to native millet. This may be attributed to the biochemical changes of millet during germination, which leads to the production of these secondary plant metabolites (Sharma et al. 2018; Gowda et al. 2022).

AOA of Flours

Antioxidant Potential Radical DPPH Radical Scavenging Activity

DPPH radical scavenging activity of flours from germinated (up to 24 h and 48 h) barley differed significantly ($P < 0.05$) among cultivars, and values ranged from 27.5 to 34.6% (Table 5). The highest DPPH was observed for cv.BH-885, and the lowest was for cv.PL-172. An increase in AOA by 21.2 to 50.2% was observed. An increase in the duration of germination to 48 h increased the DPPH by 47.4

Table 4 Total flavonoids content (TFC) of flours from non-germinated[§] and germinated (at 24 and 48 h) barley cultivars

Barley cultivars	TFC (μg CE/g) non-germinated[§]	TFC (μg CE/g) germinated at 24 h	TFC (μg CE/g) germinated at 48 h
BH-393	2011^{cd}	$3016^{c}{}_{\uparrow 49.9}$	$3242^{c}{}_{\uparrow 61.2}$
BH-932	2024^{d}	$3244^{e}{}_{\uparrow 60.2}$	$3321^{d}{}_{\uparrow 64}$
BH-902	1988^{b}	$2778^{a}{}_{\uparrow 39.7}$	$3076^{b}{}_{\uparrow 54.7}$
BH-885	2198^{e}	$3545^{f}{}_{\uparrow 61.2}$	$3711^{e}{}_{\uparrow 68.8}$
DWR-52	2002^{c}	$3109^{d}{}_{\uparrow 55.2}$	$3345^{d}{}_{\uparrow 67}$
PL-172	1968^{a}	$2856^{b}{}_{\uparrow 45.1}$	$2937^{a}{}_{\uparrow 49.2}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) from control samples for corresponding properties.

[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

Table 6 Metal chelating activity (MCA) of flours from non-germinated[§] and germinated (at 24 and 48 h) barley cultivars

Barley cultivars	MCA (%) non-germinated[§]	MCA (%) germinated at 24 h	MCA (%) germinated at 48 h
BH-393	44^{c}	$56^{de}{}_{\uparrow 27.2}$	$59.6^{e}{}_{\uparrow 35.4}$
BH-932	38^{b}	$46^{b}{}_{\uparrow 21}$	$48^{b}{}_{\uparrow 26.3}$
BH-902	38^{b}	$47.3^{c}{}_{\uparrow 24.4}$	$50.2^{c}{}_{\uparrow 32.1}$
BH-885	51^{d}	$57^{e}{}_{\uparrow 11.7}$	$59.7^{e}{}_{\uparrow 17}$
DWR-52	42^{bc}	$55^{d}{}_{\uparrow 30.9}$	$56.6^{d}{}_{\uparrow 34.7}$
PL-172	31^{a}	$39^{a}{}_{\uparrow 25.8}$	$42^{a}{}_{\uparrow 35.4}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) from control samples for corresponding properties.

[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

to77.2%, and the values ranged from 31.1 to 39.4%. Sharma and Gujral (2010) reported an increase in the AOA of barley cultivars after germination, with values ranging from 26.84 to 35.90%. Yang et al. (2001) studied the increase in DPPH for sprouted barleys and reported that the antioxidant compounds such as vitamin C and tocopherols also increased with the length of germination.

Metal Chelating Activity

MCA of flours from germinated barley differed significantly ($P < 0.05$) among cultivars (Table 6). Germination for 24 h resulted in an increase of 11.7 to 30.9%, with values ranging between 39 and 57% compared to non-germinated. Upon germination for 48 h, the MCA of flours increased (by 17 to 35.4%), and the values varied significantly ($P < 0.05$) among cultivars. Tian et al. (2004) reported that an increase in MCA might be due to increased alanine and amino butyrate and synthesis of vitamins C and E.

$ABTS^{+}$ Scavenging Activity

The antioxidant activity of flours from germinated barley (using $ABTS^{+}$) differed significantly ($P < 0.05$) among germinated cultivars (Table 7) compared to non-germinated. The germination of barley for both 24 and 48 h increased the $ABTS^{+}$ scavenging activity among all cultivars. Duenas et al. (2009) reported that germination is attributed to the biochemical metabolism of seeds during germination resulting in increased scavenging activity. Hydrolytic enzymes change the endosperm during germination and may free some of the bound components that have a role in antioxidant activity (Singh et al. 2019; Tarasevičienė et al. 2019, Doblado et al. 2007).

Table 5 DPPH radical scavenging activity of flours from non-germinated[§] and germinated (at 24 and 48 h) barley cultivars

Barley cultivars	DPPH (%) non-germinated[§]	DPPH (%) germinated at 24 h	DPPH (%) germinated at 48 h
BH-393	21.3^{c}	$28.4^{b}{}_{\uparrow 33.3}$	$31.4^{a}{}_{\uparrow 47.4}$
BH-932	20.5^{bc}	$29.3^{c}{}_{\uparrow 42.9}$	$32.1^{b}{}_{\uparrow 56.5}$
BH-902	24.9^{d}	$30.2^{d}{}_{\uparrow 21.2}$	$39.4^{e}{}_{\uparrow 58.2}$
BH-885	25.8^{e}	$34.6^{e}{}_{\uparrow 34.1}$	$38.5^{d}{}_{\uparrow 49.2}$
DWR-52	19.8^{b}	$28.2^{b}{}_{\uparrow 42.4}$	$35.1^{c}{}_{\uparrow 77.2}$
PL-172	18.3^{a}	$27.5^{a}{}_{\uparrow 50.2}$	$31.1^{a}{}_{\uparrow 69.9}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) from control samples for corresponding properties.

[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

Conclusion

The present study has been shown that germination of different barley cultivars increased ash, protein, and fiber content, antioxidant properties (total flavonoids content, total phenolic content, antioxidant activity, metal chelating activity, and $ABTS^{+}$ scavenging activity), and a*, b*, and redness intensity values related to color parameters, whereas the L* value of germinated flours decreased. Several studies have shown that when grains are processed, soaked, and sprouted, digestibility and nutrient absorption improve, as well as the levels of vitamins, minerals, proteins, and antioxidants, and the findings of this study highlight the benefit of germination in helping to improve barley valorization and other application perspectives. Germination seems to represent

Table 7 $ABTS^+$ scavenging activity of flours from non-germinated[§] and germinated (at 24 and 48 h) barley cultivars

Barley cultivars	$ABTS^+$ scavenging activity (μmol/g) non-germinated	$ABTS^+$ scavenging activity (μmol/g) germinated at 24 h	$ABTS^+$ scavenging activity (μmol/g) germinated at 48 h
BH-393	15.4^{b}	$17.6^{c}{}_{\uparrow 14.2}$	$21.2^{b}{}_{\uparrow 37.6}$
BH-932	17.8^{d}	$19.6^{e}{}_{\uparrow 10.1}$	$22.2^{c}{}_{\uparrow 24.7}$
BH-902	16.8^{c}	$18.3^{d}{}_{\uparrow 8.9}$	$20.2^{a}{}_{\uparrow 20.2}$
BH-885	15.1^{b}	$16.7^{b}{}_{\uparrow 10.5}$	$21.7^{b}{}_{\uparrow 43.7}$
DWR-52	13.2^{a}	$15.5^{a}{}_{\uparrow 17.4}$	$20.6^{a}{}_{\uparrow 56}$
PL-172	15.3^{b}	$16.9^{b}{}_{\uparrow 10.4}$	$23.6^{d}{}_{\uparrow 54.2}$

Means followed by similar superscript within the column do not differ significantly ($P < 0.05$). Subscripts denote the percentage increase (↑) from control samples for corresponding properties.

[§]Results from previous studies (Punia Bangar et al. 2022; Bangar et al. 2022)

an excellent green food developing process for improving the nutritional content of grains.

Acknowledgements Thanks to GAIN (Axencia Galega de Innovación) for supporting this review (grant number IN607A2019/01).

Author Contribution SPB: Methodology, data curation, formal analysis, writing-original draft preparation; KSS: project administration, supervision, writing—review and editing; MT: writing—original draft preparation, writing—review and editing; VM: writing—original draft preparation; JML: resources, writing—review and editing.

Data Availability All the data used in the manuscript are available in the tables and figures.

Code Availability Not applicable.

Declarations

Ethics Approval Not applicable.

Consent to Participate All authors have given their full consent to participate.

Consent for Publication All authors have given their full consent for publication.

Conflict of Interest The authors declare no competing interests.

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