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Superfine grinding improves functional properties and antioxidant capacities of bran dietary fibre from Qingke (hull-less barley) grown in Qinghai-Tibet Plateau, China

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Hydrochloric acid (PubChem CID: 24845468)
Sodium hydroxide (PubChem CID: 24845831)
Potassium chloride (PubChem CID: 4873)
Sodium chloride (PubChem CID: 5234)
Potassium dihydrogen phosphate (PubChem CID: 516951)
Disodium hydrogen phosphate (PubChem CID: 24203)
Gallic acid (PubChem CID: 370)
2,4,6-Tri (2-pyridyl)-s-triazine (PubChem CID: 113228)
1,1-Diphenyl-2-picrylhydrazyl (PubChem CID: 74358)

ABSTRACT

The effect of particle size of hull-less barley (HLB) bran DF on antioxidant and physicochemical properties was investigated. HLB bran and extracted DF was ground by regular and superfine grinding, their particle sizes were determined using laser diffraction method. The results showed that superfine grinding could significantly pulverize DF particles to micro-scale; the particle size distribution was close to a Gaussian distribution. The soluble DF in HLB bran was increased effectively with superfine grinding. Insoluble DF with submicron scale showed increased total phenolic content (TPC), DPPH radical scavenging activity and ferric reducing antioxidant power (FRAP). With particle size reduction, the water retention capacity (WRC), swelling capacity (SC), oil binding capacity (OBC), and nitrite ion absorption capacity (NIAC) were significantly ($p < 0.05$) increased and the water holding capacity (WHC) had no significant change. A kind of health beneficial DF with higher soluble DF content, WRC, SC, OBC, NIAC and antioxidant activity was obtained using superfine grinding.

Practical applications:

Superfine grinding could effectively pulverize insoluble dietary fibre particles to submicron scale. The technical advantage of superfine grinding is that water soluble dietary fiber is significantly increased. Such improvement could promote the bioavailability of dietary fiber, such as β-

Abbreviations: DF, Dietary fibre; FRAP, Ferric reducing antioxidant power; HLB, Hull-less barley; IDF, Insoluble dietary fibre; NIAC, Nitrite ion absorption capacity; OBC, Oil binding capacity; SDF, Soluble dietary fibre; SC, Swelling capacity; TDF, Total dietary fibre; TPC, Total phenolic content; WHC, Water holding capacity; WRC, Water retention capacity.

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glucan. It is clear that superfine grinding technology will play an important role in the milling industry to produce health beneficial products. Techniques and data presented in this manuscript would be of use to food manufacturers including millers who are seeking to obtain new dietary fibre sources. Current findings also could draw attentions of manufacturers in whole food production chain to turn food waste into value-added products which carry health promotion benefits.

1. Introduction

Hull-less barley (*Hordeum vulgare* L. *var. nudum* Hook. f) differs from regular hulled barley as it is a barley line with fewer hulls to cover the caryopsis (Thomason et al., 2009). Qingke, a cultivar of hull-less barley, is the main staple food crop in Qinghai-Tibet Plateau, China and is being used as an essential food crop for human consumption, as brewing material and as an important feed source (Yang et al., 2013). Growing attention has been paid to hull-less barley due to its specific attributes, such as high glucan content, high dietary fibre (DF) content, high feeding value, and high malt quality (Du et al., 2014a,b).

DF has attracted increasing interests in recent years as many studies have revealed that it might be involved in disease prevention and health promotion of consumers (Zhu et al., 2014). Functional properties of dietary fibre, such as water holding capacity, cation binding and sorption of bile acids, play significant roles in the prevention of diet-dependent diseases, e.g. obesity, atherosclerosis and colon cancer (Mehta, 2005; Dziedzic et al., 2012). In addition, nitrite scavenging capacity of dietary fibre may be a factor in the possible role for protecting against gastric cancer development (Moller et al., 1988). There is continuous interest in providing options of dietary sources to increase DF intake of human being (Niu et al., 2014). Increasing DF intake is likely to reduce the risks of cardiovascular diseases, type 2 diabetes, weight gain and obesity. However, the average worldwide ingestion of DF is still considerably lower than the recommended daily intake levels (Phillips, 2013). So, it is necessary to add DF from new sources through modern emerging food processing technologies to food products.

Superfine grinding technology is a new type of food processing that is employed to produce powders with outstanding properties such as high solubility, dispersion, adsorption, chemical reactivity and fluidity (Wu et al., 2012). This technology plays an important role in application of DF (Zhao et al., 2009). Compared with other samples ground with traditional mechanical methods, superfine powder bears good physical properties like lowering the interfacial tension and effect of steric hindrance (Zhu et al., 2012; Ting et al., 2014). To date, the superfine grinding technology has also been applied in biotechnology (Wu et al., 2012) and foodstuffs, such as ginger powder (Zhao et al., 2009), *Astragalus membranaceus* powder (Zhao et al., 2010), wheat bran DF (Zhu et al., 2010) and wine grape pomace DF (Zhu et al., 2014). Since the decrease of particle sizes might increase the particle surface area and cause the release of some antioxidant compounds (Rosa et al., 2013), it is valuable to find out functional properties and antioxidant properties of DF as affected by superfine pulverization. Our research group previously investigated that the physicochemical and antioxidant properties of Qingke flour DF as affected by superfine grinding (Du et al., 2014b). In this work, it is the first time to evaluate the effect of superfine pulverization technology on producing hull-less barley (HLB) bran DF in the submicron range, and to investigate the effects of superfine grinding on the antioxidant and functional properties of insoluble DF from hull-less barley. This study can be used as a basis for further studies for production of high DF products.

2. Materials and methods

2.1. Materials and chemicals

Bran from Qingke (hull-less barley, *Hordeum vulgare* L.) grown in Qinghai-Tibet Plateau was purchased from Qinghai Xinlvkang Food Co., Ltd. (Qinghai, China) during the 2012 crop year. Phosphate-buffered saline (PBS, pH 7.0), protease and α-amylase were purchased from Beijing Aoboxing Biotechnology Co., Ltd. (Beijing, China). Gallic acid, 1,1-diphenyl-2-picrylhydrazyl (DPPH), 2,4,6-tri (2-pyridyl)-s-triazine (TPTZ) were purchased from National Standard Samples Center (Beijing, China). All other chemicals were of analytical grade.

2.2. Preparation of insoluble dietary fibre (IDF) with regular grinding

IDF was prepared from hull-less barley bran with regular grinding using the procedure described by Deng et al. (2011) with minor modifications. Briefly, HLB bran (100 g, 0.42 mm) was sonicated in pH 6.0 water (2000 mL) at 20 °C for 20 min α-Amylase (1.0 g) was added the mixture, and then the mixture was incubated at 60 °C for 3.5 h. Subsequently, protease (0.5 g) was added into the mixture, and then the mixture was incubated at 55 °C for 3 h. Finally, the residue was washed with cold water and dried in a vacuum oven at 50 °C overnight to yield HLB bran IDF. This IDF was ground by a regular laboratory mill (FZ102, Tianjin Taisite Co. Ltd., Tianjin, China). This regular-ground IDF was for further study.

2.3. Preparation of HLB bran and IDF with superfine grinding

HLB bran and the regular-ground IDF were further pulverized by mini-type airflow pulverization instrument (QLM-80K, Shangyu City Heli Powder Engineering Co., Ltd., Zhejiang, China), respectively. The working pressure was set at 70 Mpa and the working frequency was set at 40 Hz. Superfine-ground HLB bran and superfine-ground IDF were sealed with aluminum foil and kept in a desiccator for further study.

2.4. Determination of DF content in regular and superfine-ground HLB bran

Total dietary fibre (TDF), IDF and soluble dietary fibre (SDF) contents in ground bran powder were determined as AOAC methods (AOAC, 1996; AOAC, 2005).

2.5. Particle size determination of HLB IDF

A laser diffraction particle size analyzer (LA-920, Horiba Limited, Japan) was applied to determine the particle size distribution of superfine-ground IDF. The ground powder was suspended in ethanol directly in the measuring cell (small volume unit sample module) and the suspensions were analyzed when the obscuration was between 70 and 80%.

2.6. Extraction of antioxidant components from IDF

An amount of 3.0 g of HLB bran IDF yielded with regular grinding and superfine grinding was defatted by stirring for 1 h in 15 mL *n*-hexane with a magnetic stirrer at room temperature and filtered. The residues obtained after filtration were dried and then extracted in triplicates with 80% methanol (30 mL) by stirring with a magnetic stirrer for 2 h. The mixture obtained was subsequently filtered and filled up to 100 mL with 80% methanol, and then stored at 4 °C until further study.

2.7. Assays for antioxidant properties of IDF

2.7.1. Total phenolic content (TPC)

The TPC of samples was determined using the Folin-Ciocalteu reagent-based colorimetric assay as described by Singleton et al. (1999). Phenolic content was calculated as gallic acid equivalents (GAE) and reported as mg/g dry matter. Briefly, 0.5 mL appropriately diluted extract (or gallic acid standard at 0, 50, 100, 150 or 200 ppm) was mixed with 0.5 mL of 2 N Folin-Ciocalteu reagent and 7.5 mL deionized water and allowed to stand for 10 min at room temperature; then 3 mL of 20% (w/v) Na_2CO_3 was added to the reaction mixture, and then it was placed in a 40 °C water bath for 20 min. After the 20 min reaction period, the samples were cooled to room temperature and the absorbance measured at 760 nm (Dong et al., 2013).

2.7.2. DPPH free radical scavenging capacity

DPPH radical scavenging capacity of HLB bran IDF was evaluated according to the method of Xu and Chang (2007) with slight modifications. DPPH radicals have a maximum absorption at 515 nm, which discolors with addition of antioxidants. The DPPH$^{\cdot}$ solution in methanol (6×10^{-2} mM) was prepared freshly, and 3 mL of this solution was mixed with 100 μL sample solution. The mixture was incubated for 20 min at 37 °C in a water bath, and then the decrease in absorbance at 515 nm was measured (A_S). A blank absorbance (A_B) was measured with 100 μL of methanol to replace sample. Free radical scavenging activity was calculated using the following formula:

$$\%\ \text{Scavenging rate} = [(A_S - A_B)/A_B] \times 100$$

Where A_B = Absorbance of the blank, and A_S = Absorbance of sample.

2.7.3. Ferric reducing antioxidant power (FRAP)

The FRAP was performed as described previously (Xu and Chang, 2007; Du and Xu, 2014). The absorbance of all samples, blank, and standard solutions were measured against the blank at 593 nm by the spectrophotometer under condition of 37 °C. FRAP values were expressed as mmol Fe^{2+}/100 g of sample. All measurements were done based on the triplicate extracts.

2.8. Functional properties of IDF

2.8.1. Water-holding capacity (WHC) of HLB bran IDF

Accurately weighed dried HLB bran IDF (1.0 g) was loaded into a graduated test tube, 30 mL of water was added and it was hydrated for 18 h. The supernatant was removed by passing through a sintered glass crucible (G4) under vacuum. The hydrated weight of IDF was recorded, and it was dried at 105 °C for 2 h to obtain the residual dry weight (Raghavendra et al., 2004).

$$\text{WHC (g/g)} = (\text{Hydrated weight} - \text{Dry weight})/\text{Dry weight}$$

2.8.2. Water retention capacity (WRC) of HLB bran IDF

Accurately weighed dried HLB bran IDF (1.0 g) was loaded into a graduated centrifuged tube, 30 mL of water was added and it was hydrated for 18 h, followed by centrifugation at 3000 × g for 20 min, and the supernatant solution was removed by passing through a sintered glass crucible (G4) under vacuum. The hydrated weight was recorded and then sample was dried at 105 °C for 2 h to obtain its dry weight (Raghavendra et al., 2004).

$$\text{WRC (g/g)} = (\text{Hydrated weight after centrifugation} - \text{Dry weight})/\text{Dry weight}$$

2.8.3. Swelling capacity (SC) of HLB bran IDF

Accurately weighed dried HLB bran IDF (0.2 g) was placed in a graduated test tube, 10 mL of water was added, and then IDF was hydrated for 18 h. The final volume attained by sample was measured after 18 h (Raghavendra et al., 2004).

$$\text{SC (mL/g)} = \text{Volume occupied by sample}/\text{Original sample weight}$$

2.8.4. Oil-binding capacity (OBC) of HLB bran IDF

OBC was determined by the method of Sangnark and Noomhorm (2003) with slight modifications. A portion of dried IDF (5.0 g) was mixed with peanut oil in a centrifugal tube and left for 1 h at room temperature (25 °C). The mixture was then centrifuged at 1500 × g for 10 min, the supernatant was decanted and the pellet was recovered by filtration through a nylon mesh. OBC was expressed as follows:

$$\text{OBC (g/g)} = (\text{Pellet weight} - \text{Original dry weight})/\text{Original dry weight}$$

2.8.5. Nitrite ion absorption capacity (NIAC) of HLB bran IDF

A portion of dried IDF (1.0 g) was mixed with 100 mL 1 mol/L $NaNO_2$ solution in a 250-mL conical flask. The pH was adjusted to 2.0. The mixture was incubated at 37 °C for 75 min with continuous mild agitation. The residual concentration of nitrite ion was measured. NIAC was expressed as follows:

$$\text{NIAC } (\mu g/g) = (\text{Nitrite ion before absorption} - \text{Nitrite ion after absorption})/\text{Dry weight}$$

2.9. Statistical analysis

All results in this work were expressed as mean ± standard deviation of three replicates. Data in triplicate were analyzed by one-way analysis of variance using SPSS 11.5 software package for Windows (SPSS Inc, U.S.A.).

3. Results and discussion

3.1. Particle size of superfine-ground HLB bran IDF

The particle size of HLB bran IDF with superfine grinding was distributing in a range from 2.98 μm to 67.52 μm with a mean particle size of 14.06 μm, which belongs to submicron (Fig. 1). However, the mean particle size of hull-less barley IDF with regular grinding is 74.31 μm. The results indicated that superfine pulverization using mini-type airflow pulverization instrument could cut down the sizes of the IDF particles to a submicron. Zhu et al. (2010) obtained the smaller particle size of superfine wheat bran DF using multidimensional swing high-energy nano-ball-milling. The mean

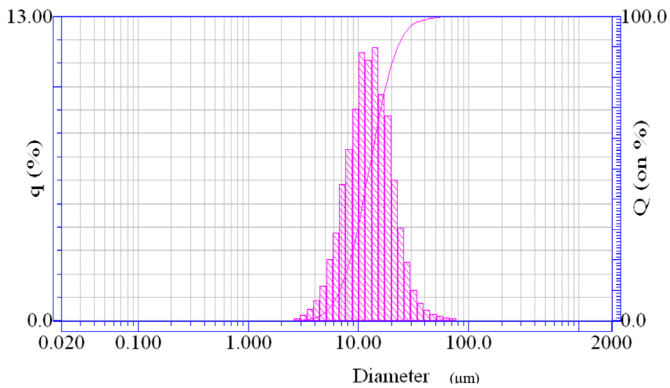

Fig. 1. Particle size distribution of hull-less barley IDF with superfine grinding (The abscissa is the diameter (μm) of the particle size; the ordinates q% (quantity %) is the particle size distribution, and Q% is under size %).

Table 1
Effect of superfine grinding on dietary fibre content in hull-less barley bran*.

Treatments	TDF (%)	IDF (%)	SDF (%)	IDF:SDF
Regular grinding	76.5 ± 0.51^a	68.2 ± 0.30^b	6.46 ± 0.40^a	10.5
Superfine grinding	72.0 ± 0.26^a	16.1 ± 0.53^a	54.4 ± 0.07^b	0.30

*Results were expressed as means ± standard deviation (n = 3). Values in the same column with different letters are significantly ($p < 0.05$) different.

particle size of the superfine DF was 343.5 nm. It was then speculated that the different phenomena were depending on different treatments and experimental instruments. However, these results were almost similar with our previous study in which the mean particle size of wine grape pomace DF was 7.06 μm using the same instrument (Zhu et al., 2014).

3.2. Dietary fibre profiles of regular and superfine-ground HLB bran

The TDF content of HLB bran was 76.5% (shown in Table 1), and SDF content of regular-ground HLB bran was increased from 6.5% to 54.4%, while IDF reduced from 68.2% to 16.1% with superfine grinding treatment, suggesting that superfine grinding causes a redistribution of fibre components among soluble and insoluble. The decrease of IDF content after superfine grinding is caused by the degradation of hemicellulose, cellulose and lignin, which are turned into some small molecular compounds (Zhu et al., 2010). The key to balancing SDF and IDF intake is consuming different types of foods that incorporates the different sources of each fiber type. SDF can help to lower blood cholesterol and control blood glucose. The reasons are as follows: SDF absorb water and produce a gel, which reduces digestion. It delays the emptying of your stomach, which assists control weight. Slower stomach emptying may also influence blood sugar levels and have a beneficial effect on insulin sensitivity, which may help control diabetes. Most foods have soluble fiber and insoluble fiber, but the amount of each type can vary depending on the food. There are also different kinds of soluble and insoluble fiber. Many studies indicate that SDF is more important than IDF in many health aspects (Galisteo et al., 2008), such as preventing obesity and controlling diabetes. It is also an interesting challenge to turn IDF into SDF. In this research, it is good to note that the SDF content of HLB bran with superfine grinding was higher than regular-ground HLB bran. It provides one useful modification method to produce high quality DF in food industry.

3.3. Functional properties of HLB bran IDF

The results of WHC, WRC, SC, OBC and NIAC were obtained in Table 2. As indicated in Table 2, the WHC, SC, OBC and NIAC of HLB bran IDF increased with superfine grinding. There were some other reports presenting the similar phenomena apart from this study. Zhao et al. (2009) suggested that the smaller particle size of ginger powder contributed a higher WHC even when their particle sizes decreased to 7.23–8.32 μm, because superfine grinding would lower the interfacial tension (Ting et al., 2014) and expose more polar groups, surface area and water-binding sites to the surrounding water. Additionally, WHC is associated with the porous matrix structure formed by polysaccharide chains which can hold large amounts of water through hydrogen bonds (Kethireddipalli et al., 2002). Moreover, it is in agreement with previously published results which indicated that the SC of coconut DFs was increased when its particle size decreased from 1127 to 550 μm (Raghavendra et al., 2004). Swelling property depends on many factors such as network density, solvent nature, polymer solvent interaction parameter. However, Sangnark and Noomhorm (2003) investigated that the smaller particle size of sugarcane bagasse DF was related with a lower OBC. It is not surprising because the reverse phenomena were depending on experimental parameters such as stirring grinding and all of these could alter the physical structure of DF (Sangnark and Noomhorm, 2003). The WRC of HLB bran DF had no significant ($p > 0.05$) change in this work. Furthermore, it is very useful to evaluate the NIAC of DF from different foods. There is a similar research, the NIAC of Qingke flour DF was increased using superfine grinding (Du et al., 2014b).

Table 2
Effect of superfine grinding on functional properties of hull-less barley bran IDF*.

Treatments	WHC (g/g)	WRC (g/g)	SC (mL/g)	OBC (g/g)	NIAC (μg/g)
Regular grinding	2.25 ± 0.07^a	4.76 ± 0.45^a	5.02 ± 0.08^a	1.74 ± 0.03^a	478 ± 17^a
Superfine grinding	19.53 ± 0.03^b	4.93 ± 0.51^a	6.79 ± 0.13^b	2.03 ± 0.09^b	674 ± 14^b

*Results were expressed as means ± standard deviation (n = 3). Values in the same column with different letters are significantly ($p < 0.05$) different. WHC, water holding capacity; WRC, water retention capacity; OBC, oil binding capacity.

Table 3
Effect of superfine grinding on TPC, DPPH scavenging activity and FRAP of hull-less barley bran IDF*.

Treatments	TPC (μg GAE/g)	DPPH free radical scavenging activity (%)	FRAP (mmol Fe^{2+}/100 g)
Regular grinding	306.3 ± 3.98^a	65.64 ± 0.51^a	306.2 ± 3.98^a
Superfine grinding	346.3 ± 1.50^b	90.14 ± 0.73^b	346.3 ± 1.50^b

*Results were expressed as means ± standard deviation (n = 3). Values in the same column with different letters are significantly ($p < 0.05$) different. TPC, Total phenolic content; FRAP, ferric reducing antioxidant power.

3.4. Antioxidant characteristics of HLB bran IDF

In this work, we measured the TPC, DPPH and FRAP values in HLB bran IDF with regular grinding and superfine grinding. As shown in Table 3, the TPC, DPPH and FRAP increased ($p < 0.05$) after superfine grinding. Most likely, the reason for this finding is the superfine grinding which altered or destroyed the DF matrix, thus causing some phenolic compounds released or exposed. The result was equivalent to the study performed by Zhu et al. (2010). They developed that the TPC and FRAP of wheat bran DF increased with superfine grinding. A completely different experimental result was reported in the study of Zhu et al. (2014). The results revealed that wine grape pomace DF with superfine grinding had decreased DPPH radical scavenging activity.

4. Conclusions

In the current work, the mini-type airflow pulverization instrument could mainly reduce the particle size of hull-less barley bran insoluble dietary fibre to micro-scale, redistribute fibre components from insoluble to soluble fractions and yield a kind of health beneficial dietary fibre with higher soluble dietary fibre content. Hull-less barley bran insoluble dietary fibre after superfine grinding had many remarkable characteristics: good water holding capacity, swelling capacity, oil binding capacity and nitrite ion absorption capacity capacity. Total phenolic content, ferric reducing antioxidant power and DPPH increased after superfine grinding. Strong correlation was observed among total phenolic content, ferric reducing antioxidant power and DPPH scavenging capacity. This study could become the useful basis for later research according to the superfine grinding of hull-less barley bran dietary fibre in food and pharmaceutical industries.

Conflicts of interest

None.

Compliance with ethics requirements

This article does not contain any studies with human or animal subjects.

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