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Physicochemical and functional properties of dietary fiber from foxtail millet (*Setaria italic*) bran

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ABSTRACT

Agricultural byproduct-foxtail millet (*Setaria italic*) bran was explored to extract dietary fiber by enzymatic methods and the physicochemical and functional properties of foxtail millet bran dietary fiber (FMBDF) were investigated. Results revealed that foxtail millet bran was a good source for dietary fiber production. The studies of physicochemical and functional properties showed that the water holding capacity and swelling power of FMBDF were 3.24 g/g and 2.06 ml/g, respectively. Meanwhile, FMBDF exhibited good adsorption capacities to lipophilic substances such as lard (3.34 g/g), peanut oil (2.32 g/g) and cholesterol (5.19 mg/g). Its bile salts adsorption capacity (143.03 μmol/g for sodium cholate and 76.65 μmol/g for sodium taurocholate) indirectly reflected its cholesterol-lowering effect. The chemical composition and microstructure studies of FMBDF demonstrated that these had a lot to do with its functional properties. The results indicate that FMBDF has a great potential to be used as functional ingredient in food products.

1. Introduction

Millet, an annual grass species, belongs to the grain. It is an important food crop and a staple diet in many areas of Asia, especially in north China. Millet bran, a really inexpensive and easily obtained by-product, consisting of a small number of embryo, pericarp, testa, aleurone layer, and endosperm, contains nutritious elements such as protein, fat, minerals, vitamins, and dietary fiber. Unfortunately, lots of millet bran is not fully utilized and even discarded.

In recent years, many beneficial nutrients particularly dietary fiber were lost in our increasingly sophisticated modern diet. “Rich people's diseases” like obesity, high blood pressure, cardiovascular disease and diabetes have arisen along with the change of dietary habit. Therefore, a large number of high-value novel foods, known as functional foods or nutraceuticals appear in our daily life (Arvanitoyannis and Van Houwelingen-Koukaliaroglou, 2005), which suggests that the public awareness of nutrition and health is increasing. Simultaneously, health care research substantiated that the active substances of plant origin such as dietary fiber has beneficial properties on people's health (Devi et al., 2014).

Dietary fiber (DF), a carbohydrate-based polymer with beneficial physiological properties, is generally derived from certain cereals, legumes, vegetables and fruits (Chen et al., 2014). Defined as the edible part of plants or analogous carbohydrates, dietary fiber is undigested and unabsorbable in human's small intestine. While, it can be completely or partially fermented in large intestine, and includes polysaccharides, oligosaccharides, lignin and associated plant substances (Bangoura et al., 2013). Attributed to the features in reducing the transit time of chyme in the small intestine and increasing stool volume, decreasing blood total cholesterol and low-density lipoprotein cholesterol levels, and lowering blood sugar and insulin levels, its beneficial effects on the reduction of blood cholesterol (Anderson et al., 2009), cardiovascular (Güroy et al., 2013), and colon cancer (Rodríguez et al., 2006) have attracted the attention of researchers. General, dietary fibers can be divided into the water-soluble and the water-insoluble, recorded as SDF and IDF. IDF accounts for 2/3–3/4 of natural fibers, and high-viscosity SDF can serve as a therapeutic agent for hypercholesterolemia. A research showed that dietary fiber's composition, source

Abbreviations: ANOVA, one-way analysis of variance; BBC, bile salts binding capacity; CBC, cholesterol binding capacity; DF, dietary fiber; FMB, foxtail millet bran; FMBDF, foxtail millet bran dietary fibers; HPLC, high-performance liquid chromatography; IDF, insoluble dietary fiber; NIAC, nitrite ion absorption capacity; OBC, oil-binding capacity; SC, swelling capacity; SDF, soluble dietary fiber; WHC, water holding capacity.

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and preparation method could affect the effectiveness of its adsorption characteristics (Galisteo et al., 2008), like water holding capacity, swelling power, and binding capacities for oil and cholesterol to a large extent *in vitro*.

Different cereal sources have been used to prepare dietary fibers such as barley grains, oat bran and wheat bran (Caprez et al., 1986; Zhang et al., 2011a). However, millet bran, as an agricultural by-product rich in dietary fiber, was rarely studied and utilized. The objective of this work was to explore the potential of millet bran as a dietary fiber source. The physicochemical and functional properties of millet bran dietary fiber were also determined.

2. Materials and methods

2.1. Materials and chemicals

Foxtail millet (*Setaria italic*) bran was obtained from Qinshui Country (Shanxi Province, China). Peanut oil, lard and eggs were purchased from local market. Cholesterol (≥99%) and sodium taurocholate (≥97%) were supplied by Sigma-Aldrich. Heat-stable α-amylase (≥4000 μ g^{-1}), neutral protease (>50 μ mg^{-1}), amyloglucosidase (>100 μ mg^{-1}), pancreatin and sodium cholate (>98%) were purchased from Aladdin Chemistry Co., Ltd. The other chemicals used were of analytical grade.

2.2. Preparation of foxtail millet bran dietary fiber

2.2.1. Pretreatment of FMB

FMB was defatted with *n*-hexane. After filtration, the sediment was dried in the fume hood. The dry defatted FMB was milled to pass through 40 mesh sieve and stored at −20 °C in a sealed container.

2.2.2. Preparation of foxtail millet bran dietary fiber

Defatted FMB was treated enzymatically using the method of standard AOAC (2000) with a slight change. defatted FMB (10 g) was suspended in water with a material-liquid ratio 1:10 (W/V) and heated in water bath at 100 °C for 15 min. Then the mixture was incubated at 50 °C for 30 min. After adjusting the pH to 5.5, the mixture was treated with heat stable α-amylase (200 μ g^{-1}) at 95 °C for 1 h, then digested with amyloglucosidase (pH 4.2) at 60 °C for 1 h. Finally, neutral protease (250 μ g^{-1}) was added for further reaction at 55 °C and pH 7.0 for 2 h. Reaction mixture was transferred to boiling water for 15 min to inactive all the enzymes, and then precipitated with 4 vol of 95% ethanol overnight. The residue was oven-dried and marked as foxtail millet bran dietary fibers (FMBDF).

2.3. Proximate analysis of FMB and FMBDF

On the basis of dry weight, Moisture, ash, protein, fat and starch were determined in duplicated according to the standard methods of GB/T5009.1 (2003) and GB/T5009.9 (2008). For FMB, DF content was calculated by subtracting the total percent values of other measurements from 100. For FMBDF, soluble, insoluble and total dietary fibers contents were determined by AACCI method 32-07(2000). Cellulose, hemicellulose and lignin of FMBDF were determined following the method of Xiong et al. (2005).

2.4. Monosaccharide and uronic acid determination

1 mg SDF or IDF was hydrolyzed with 300 μl 4 mol l^{-1} trifluoroacetic acid at 110 °C for 2 h and cooled at room temperature. The reaction solution was dried at 40 °C for 4 h and washed three times with 2 ml methanol each time to remove the excess trifluoroacetic acid. Then, the hydrolytes of SDF and IDF were derivatized with 2-aminobenzoic acid and analyzed on a HPLC system (SHIMADZU LCMS2020) equipped with a fluorometric detector and a ODS HyperClone C18 column (250 mm × 4 mm, 5 μm). The excitation and emission was 360 nm and 425 nm, respectively. The standards of galacturonic, mannose, glucose, galacturonic acid, arabinose, xylose, glucose acid and rhamnose were treated accordingly and applied on HPLC analysis.

2.5. Molecular weight assay

The molecular weight of SDF was determined by gel-permeation chromatography, in combination with a HPLC (Agilent 1100, USA). Sample dissolved in distilled water was passed through a 0.45 μm filter and applied to a TSK G4000PWX gel column (7.8 × 300 mm), eluted with distilled water at a flow rate of 0.98 ml min^{-1}, and detected by a differential refractive index detector (RID, Alltech 3300). Standard Dextrans (M_W ranging from 12600 to 289000) were injected, and then the elution time was plotted against the logarithm of molecular weight with gel-permeation chromatography software. The molecular weight of SDF was calculated by comparing their retention time with those of standards under the same HPLC conditions by constructing a calibration curve. The Dextran calibration curve of was Lg(Mw) = 5.431–0.019RT (RT: retention time of Dextran). $R^2 = 0.9996$.

2.6. Scanning electron microscopy

The scanning electron images of FMB and FMBDF were gathered using a scanning electron microscope (XL-30ESEM, Philips, Holland). The samples were prepared according to the method described by Zhang et al. (2011b) with slight modification. Briefly, crushing samples were mounted on the observation platform after dried at 105 °C to constant weight, and sputter-coated with gold for 5 min at 2 mbar. Then the samples were observed with 1200 × and 5000 × fold magnification at an acceleration voltage of 15 kV and high vacuum condition.

2.7. Physicochemical and functional properties

2.7.1. Physicochemical properties

Water holding capacity (WHC) and oil-binding capacity (OBC) of FMBDF were estimated according to the method reported by Sangnark and Noomhorm (2003). Swelling capacity (SC) was determined by the method reported by Zhang et al. (2011a).

2.7.2. Functional properties

2.7.2.1. Cholesterol binding capacity (CBC). On account of the bad aqueous solubility of commercial cholesterol, egg yolk was used to replace cholesterol according to the method of Zhang et al. (2011b) with slight modification. The yolk was separated from the egg and diluted with 9 vol of distilled water. FMBDF (1.0 g) were mixed with 25 ml diluted yolk. The mixture was adjusted to pH 7.0 or 2.0 and incubated in a shaking water bath at 120 rpm and 37 °C for certain time (simulated the conditions in small intestine and stomach, successively). Diluted yolk was set as negative group. Reaction mixture was centrifuged at 4000 rpm for 20 min. 0.02 ml supernatant was used to determine the cholesterol content.

CBC was calculated as follows:

$$\mathrm{CBC} = \frac{(C_{blank} - C_d) - \left(C_{yolk} - C_{blank}\right)}{W_1} \times 25 \quad (1)$$

where C_{yolk}, C_{blank}, and C_d were the concentrations of cholesterol in