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Molecular characterization of water-extractable arabinoxylan from wheat bran and its effect on the heat-induced polymerization of gluten and steamed bread quality

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ABSTRACT

Enhanced bread quality by water-extractable arabinoxylan (WEAX) depends on its inherent structural features. To clarify the underlying mechanism, the current study prepared WEAX with varied structures via graded ethanol precipitation from wheat bran, and their effects on the steamed bread quality in relation to the heat-induced physicochemical changes of dough components were evaluated. The results showed that WEAX with a lower molecular weight (Mw), higher branched degree and ferulic acid content possessed a superior improved effect on the steamed bread quality. The gelatinization of starch was partially inhibited by WEAX, with a more distinct effect by the lower Mw and higher branched WEAX. However, WEAX suppressed the short-term retrogradation of starch, especially for the higher Mw and lower branched WEAX. Both of the heat-induced polymerization degree and rate of gluten were reduced by WEAX, and these reductions were more evident by the lower Mw and higher branched WEAX. The partial inhibition of gluten polymerization by WEAX contributed substantially to the enlarged loaf volume and softer textural property of steamed bread. The current study can provide a theoretical basis for the exploitation of WEAX as a nutritious and technofunctional dough improver.

1. Introduction

Steamed bread is a traditional Chinese staple food and is widely consumed in Asian countries. Due to developed milling techniques, flour has been extensively refined and losses of nutrients such as minerals and dietary fiber have become an issue in wheat-based products. Thereby, the fortification of bioactive compounds is gaining a wide range of prevalence due to their functional and nutraceutical importance. Vitamins, minerals, and dietary fiber are often enriched to some extent by the incorporation of finely grinded wheat bran due to its enriched nutrients and significant important roles in human vitality (Rosa-Sibakov, Poutanen, & Micard, 2015). Accounting for ~25% of the wheat kernel weight, wheat bran is the main byproduct of the wheat milling industry, which is mainly comprised of fiber and protein. The relatively high protein content makes bran suitable for animal feed. However, the presence of arabinoxylan (AX) reduces the rates of digestion and the absorption of nutrients in monogastric animals. Moreover, as a food additive, the involvement of wheat bran can hinder the development of gluten network by competing for water and acting as physical barriers. This hindrance significantly degrades the organoleptic quality of the final product (Hemdane et al., 2018).

As the major hemicelluloses of wheat bran, AX occupies ~70% of non-starch polysaccharides. AX is composed of β-1,4 linked D-xylopyranosyl residues as a backbone substituted with monomeric α-L-arabinofuranose units at the second and/or third carbon positions. Ferulic acid can be coupled to the fifth carbon position of arabinose through an ester linkage (Kiszonas, Fuerst, & Morris, 2013). AX can be categorized into water-extractable AX (WEAX) and water-unextractable AX (WUAX) fractions. As a soluble dietary fiber, WEAX is considered to perform more significant physiological functions due to its hydrophilic characteristics and more available access to the microbial fermentation in the large intestine. The produced xylooligo-saccharides by the hydrolysis of WEAX are prebiotics, which can selectively stimulate the growth and/or activity of probiotics (Mendis & Simsek, 2014). Moreover, WEAX is generally regarded as a dough improver, which could enhance the organoleptic quality of bread regardless of its nutritional value. Therefore, WEAX holds great potential to be exploited as both a technofunctional and nutritional supplement for enhancing the wheat-

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based food quality. Meanwhile, as a staple food, steamed bread provides an ideal matrix for the dietary fiber delivery to the consumers, and thus improves the national nutrition. Therefore, the contribution of wheat bran to the health benefits and the organoleptic quality of bread can be maximized through the selective fractionation of the AX fractions (Koegelenberg & Chimphango, 2017).

Improved bread quality by WEAX is attributed to the interactions between the dough components and WEAX. Much emphasis has been placed on the WEAX-gluten interactions during the development of dough in the mixing stage, since the rheological property of dough conferred by gluten is suggested to be a pivotal indicator for the end-use quality of dough. However, the results of the relevant studies are not always in accordance with each other. Wang, Vliet, and Hamer (2004) suggested that WEAX enhanced the gluten strength and elasticity by enlarging the particle size of glutenin macropolymers and competing for water with gluten moderately, whereas Turner, Soh, Ganguli, and Sissons (2008) found that WEAX hindered the formation of gluten network and diminished the dough strength and elasticity. This discrepancy was probably related to the different interactions between WEAX and gluten, which mainly depended on the structural composition of WEAX including the molecular weight, branched degree and the bounded ferulic acid groups (Saeed, Pasha, Anjum, & Sultan, 2011).

Heating is the last but most important step in the bread making procedure. A series of physicochemical variations, such as the evaporation of water, volume expansion, protein transition and starch gelatinization, take place during heating. Among them, the wheat protein transition is of major importance in establishing the bread structure. The transition of gluten is accompanied by decreased solubility and proceeds to a point where the gas vesicle walls are fixed and expansion ends (Stathopoulos, Tsiami, Dobraszczyk, & Schofield, 2006). With the increased heating temperature, the disulfide-mediated polymerization of gluten is initiated and the heat-induced formation of permanent gluten structures is vital in establishing the loaf volume and texture of final products (Lagrain, 2007). Meanwhile, starch gelatinization also contributed non-negligibly to the textural profiles of baked bread (Lagrain, Wilderjans, Glorieux, & Delcour, 2012; Tao, Xiao, Wu, & Xu, 2018). Therefore, modifications in the heat-induced polymerization of gluten and the gelatinization of starch directly affect the quality of the final products. However, the influence of WEAX with different structures on the heat-induced structural changes of dough components remains unclear.

Against this background, the purpose of this study is to investigate the structural features of WEAX prepared by graded ethanol precipitation from wheat bran. The impact of different WEAX fractions on the heat-induced changes of dough components in relation to their steamed bread characteristics was evaluated to clarify the underlying mechanism of the improved bread qualities.

2. Materials and methods

2.1. Materials

Commercial steamed bread flour (10% protein, 74% carbohydrate, 13% moisture) and active dry yeast (Angel brand, Hubei, China) were purchased from a local supermarket. Wheat bran was obtained from Yuchen Co., Ltd. (Taixing, Jiangsu, China), and alpha-amylase (Termamyl 120 L) and amyloglucosidase (AMG 300 L) were purchased from Novozymes (Bagsvaerd, Denmark). Lichenase (EC 3.2.1.73) was obtained from Megazyme International Ireland Ltd. (Bray, Ireland). Deionized water was used throughout the experiment. All used reagents were of analytical grade unless otherwise specified.

2.2. Extraction of water-extractable arabinoxylan (WEAX) from wheat bran

The extraction of WEAX was performed according to Mansberger et al. (2014) with modifications. The wheat bran was heated at 130 °C for 150 min in an oven to inactivate the enzymes. The wheat bran (50 g) was added to 500 mL deionized water with 0.01 mL α-amylase and stirred at 65 °C for 90 min. Afterwards, the solids were removed by centrifugation at 5000*g* for 10 min using an Avanti J-25 Beckman centrifuge (Beckman Coulter, Inc., New York, NY, USA). The heat-stable α-amylase was added to eliminate the starch contaminants from the aqueous extract (95 °C, 1 h). The pH value was lowered to 6.5 and 0.3 mL amyloglucosidase was added (55 °C, 24 h). Then, the enzymes were inactivated by heat treatment for 30 min at 120 °C. The resulting solution was filtered (type 595, Whatman, Maidstone, England). To bind the protein, 10% of a bentonit solution (0.2% mass) was added. After stirring at room temperature for 30 min, the solution was cleared again by centrifugation. The WEAX were fractionated by a graded ethanol precipitation, and increased volumes of ethanol were added to a solution of WEAX to obtain ethanol concentrations ranging from 20% to 70%. Solutions were allowed to stand overnight at 4 °C for each ethanol concentration. After centrifugation, the residues were redissolved in water (0.5%) and incubated with lichenase (40 °C, 1 h) to remove the remaining β-glucans. The enzymes were inactivated by heat (100 °C, 10 min), and the solutions were centrifuged (13,000 g, 20 min). The supernatant was dialyzed against deionized water at 4 °C for 72 h using membrane tubing with a 3500 Da molecular weight cut off (Spectrum Laboratories, Inc., CA, USA), and then the solutions were freeze-dried and stored at −80 °C. The fractionated WEAX were designated as F20, F30, F40, F50, F60 and F70 (the numbers refer to the saturation level of ethanol at which the material was collected). In the current study, the yield and AX content of the F20 and F70 fractions were quite low on the basis of the total amount of material recovered after fractionation, and thus they were not used for the further study.

2.3. Molecular characterization of WEAX

2.3.1. Molecular weight

The molecular weight of WEAX was conducted on an Agilent 1200 Series high performance liquid chromatography (HPLC) system (Agilent Technologies, Santa Clara, CA, US). The aliquots of WEAX samples (1.0 mg) were solubilized in 0.3% NaCl and centrifuged (10000 g, 10 min). The solutions (20 μL) obtained were separated on a Shodex KW-804 column by elution with 0.3% NaCl and monitored with a refractive index detector. The flow rate was 0.7 mL/min and the temperature was set at 30 °C. The molecular weight markers were Shodex standard P-82 pullulans (1.0 mg/mL) with molecular weights of ranging from 0.59×10^4 to 78.8×10^4.

2.3.2. Monosaccharide composition

Samples were initially hydrolyzed in 1 M H_2SO_4 at 97 °C for 3 h. The hydrolysate was diluted (1:20) and derivatized with 1-phenyl-3-methyl-5-pyrazolone (PMP) at the anomeric carbon atoms. An Agilent XDB-C18 column (4.6 mm × 250 mm, 5 μm) at 30 °C and a flow rate of 0.8 mL/min were used to perform the chromatography. The separation of PMP-monosaccharide derivatives was separated with eluent containing 40 mM ammonium acetate in 10% acetonitrile (pH 6.8) and a gradient of 8% (v/v) to 100% (v/v) acetonitrile over the 20 min. Total WEAX content was then calculated as 0.88 times the sum of monosaccharide xylose and corrected arabinose (Marcotuli et al., 2015).

2.3.3. Ferulic acid (FA) determination

FA quantification was done following the method described by Hartmann, Piber, and Koehler (2005). About 50 mg WEAX was saponified with 3 mL of 4 M NaOH under nitrogen atmosphere for 18 h at room temperature with intermittent mixing. Then samples were centrifuged (3000 g, 10 min) and acidified to pH 2 using 2 M HCl. The FA was extracted three times from the samples with 3 mL ethyl acetate. The combined samples were dried under nitrogen atmosphere followed by dissolution in 2 mL of H_2O/MeOH mixture on 1:1 ratio and