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Mild high hydrostatic pressure pretreatments applied before soaking process to modulate wholegrain brown rice germination: An examination on embryo growth and physicochemical properties

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ABSTRACT

This investigation aimed to examine the effects of a novel processing pattern, combining high hydrostatic pressure (HHP) with germination, on the embryo growth and physicochemical characteristics of wholegrain brown rice (WBR). WBR grains were firstly subjected to mild HHP stress (30–90 MPa/5 min) and then incubated at 37 °C for 52 h for obtaining germinated samples (GBR). The results showed that HHP shock resulted in a delayed embryo growth of WBR grains, maintaining acceptable sprouting rates ranging from 65% to 76% when germination was finished. The contents of gama-aminobutyric acid in GBR were greatly increased responding to HHP stress, showing pressure intensities dependent. Total digestible and resistant starch contents in samples stressed at 60/90 MPa were decreased, mainly associated with high pressure-induced amorphization as revealed by SEM imaging and FTIR, which promoted starch hydrolysis during germination. Besides, the levels of zinc and iron were influenced by HHP pretreatments due to the high pressure-mediated degradation behavior for phytic acids. The storability of HHP-stressed GBR grains was significantly enhanced through reducing free fatty acids formation and maintaining color stability during a storage testing. These results obtained from the current work demonstrated that mild HHP stress pretreatment prior to germination process could be used as a promising strategy to modulate certain physicochemical characteristics of WBR products.

1. Introduction

The ever growing demand for safe, nutritious and high-quality foods has encouraged persistent research and development of emerging non-thermal processing techniques (e.g., pulsed electric field, high pressure, plasma, etc.), among which high hydrostatic pressure (HHP) becomes the most successfully commercialized approach (Huang, Wu, Lu, Shyu, & Wang, 2017; Misra et al., 2017), showing the capability to provide organoleptically satisfactory, microbiologically safe and nutritionally intact foods. Regarding plant-derived food matrix, HHP treatments have been reported to improve the bioaccessibility of bioactive components, especially substantial phytochemicals with beneficial effects such as phenolic acids, vitamin E and β-carotene (Knockaert et al., 2011; Nuñez-Mancilla, Pérez-Won, Uribe, Vega-Gálvez, & Di Scala, 2013). In addition, biochemical reactions can be accelerated to accumulate functional components following a prolonged storage after HHP processing (Barba, Poojary, Wang, Olsen, & Orlien, 2017; Ueno et al., 2010). In these cases, the increase in bioactive constituents is principally related to pressure induced decompartmentalization owing to physical damage of cell wall, thus facilitating the release of intracellular micronutrients (increased extractability) as well as the accessibility of enzymes to the related substrates.

On the other hand, it should be noted that metabolic response to high pressure shock may occur during germination process as a result of the pressure-modified gene expression and related metabolic pathways (Liu, Zhang, Duan, & Wu, 2008), thus exerting an influence on the quality of germinated products. Interestingly, although the related research and application of HHP technique in food sector, mainly in food preservation, have been reported for over decades, there is very scarce information concerning the modulation of food functionality and quality via HHP-induced biological effects.

Wholegrain brown rice (WBR) is an important functional food and its health benefits have been well-recognized, but it remains a less popular consumer choice globally due to rice bran-derived intrinsic shortcomings compared with polished/white rice (Mohan et al., 2017). Several studies have recently validated the great potential effectiveness

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of HHP technique in promoting the acceptability of WBR consumption by shortening cooking time, softening texture and palatability, and improving micronutrients bioaccessibility (Xia, Wang, Xu, Mei, & Li, 2017; Yu et al., 2017; Yu, Ge, Zhu, Zhan, & Zhang, 2015). Simultaneously, it was reported that germination process in combination with high pressure technique was employed to enhance the formation of various bioactive substance within WBR grains (Kim, Lee, Jang, Li, et al., 2015; Kim, Lee, Jang, Park, et al., 2015; Kim et al., 2017), in which rough rice was firstly germinated and then subjected to HHP treatments for promoting biochemical reactions. However, according to these researches, the increased bioactivities for WBR were achieved mainly depending on the modification of food matrix microstructure due to accelerated mass transfer from high pressure-derived turbulence and shear force, and no literature has reported the application of HHP pretreatments before WBR germination as well as their related physicochemical changes and biomolecular effects to our best knowledge so far.

Therefore, in the present investigation, the effects of HHP stress prior to steeping process on functionality and quality characteristics of germinated WBR grains (GBR) were investigated, and accordingly the examined physicochemical parameters included gama-aminobutyric acid (GABA), starch digestibility in vitro and minerals as well as storage stability. Our pilot work previously suggested that HHP shock before germination at the pressures higher than 100 MPa largely compromised seed viability (Xia, Wang, & Li, 2018), so this study adopted mild HHP stress (30, 60 and 90 MPa/5 min) pretreatments to induce WBR germination in order to further reveal the relationship between the applied pressure and changes in physicochemical properties.

2. Materials and methods

2.1. Raw materials and chemicals

WBR samples Sanqiuding SZ (*Oryza sativa* L.) recently harvested were obtained from a local market, which were sealed and stored at −20 °C until use. Standard solutions of sodium hydroxide (0.5 M) and the enzymes including pepsin (P110927), α-amylase (A109181) and amyloglucosidase (A107823) were bought from Aladdin Chemistry Co., Ltd. (Shanghai, China), while the other chemicals, such as nitric acid, sodium bicarbonate, hydrochloric acid, were purchased from Sinopharm Chemical Reagent Co., Ltd., China. All chemicals were of analytical grade, unless otherwise stated.

2.2. Sample preparation

Three replicates of WBR grains were mixed with de-ionized water (2:1; w/v), vacuum-packed using high-density polyethylene bags and then subjected to high pressure stress using a hydrostatic pressurization unit (HHP-750; BaoTou KeFa Co., Ltd., China) equipped with a 5-L cylinder. Water was used as the medium to transmit pressure. The samples were pressurized under 30, 60 and 90 MPa with a holding time of 5 min at room temperature (21 ± 0.5 °C), respectively. The time to reach the target pressure varied between 0.2 and 0.5 min when the pressure started to increase, whereas the pressure-relief process was immediate (< 4 s). The grains without HHP treatments were set as control group. After HHP pretreatments were finished, all WBR grains were immediately subjected to germination process.

The subsequent germination treatment was conducted according to previously used procedures (Xia, Tao, et al., 2017). WBR grains were immersed into the de-ionized water (3:10, w/v), followed by the incubation at 37 °C in darkness in a climatic cabinet (RGX-260B, Hualian Med., China). This process continued for 52 h, during which germination performance was recorded. The germinated grains pretreated at 30, 60 and 90 MPa were marked as H30GBR, H60GBR and H90GBR, respectively.

2.3. Analysis of germination performance

To test germination percentage, the ratio of germinated grains in fifty kernels of WBR grains was countered at 0, 16, 24, 36 and 52 h. After 16-h soaking, obvious germination phenomenon could be distinguished, and there were no great changes in germination rates when the samples were soaked for 52 h. For evaluating the sprout length of GBR grains, sprouts were cut off close to the pericarp and then measured using a Verniercaliper. The grains for each treatment were determined in three replicates.

2.4. Analysis of free amino acids (FAA)

Individual FAA components, including protein amino acids and GABA, were extracted and quantified. The extraction procedures followed the description of Barba et al. (2017). The ground GBR samples which had been sieved by an 80-mesh screen were mixed thoroughly with HCl (0.1 M) and then allowed for an ultrasonic bath (20 min/25 °C). The mixture was centrifuged (3000*g*) for 20 min, and the collected supernatants were filtered with 0.45 μm filters prior to injecting into an L-8900 automated amino acid analyzer (Hitachi, Tokyo, Japan).

2.5. Measurements of mineral elements

The contents of mineral elements, including zinc (Zn), iron (Fe) and phosphorus (P) were measured using an inductively coupled plasma-optical emission spectrophotometer (ICP-OES) (Thermo Scientific iCAP 6300, Cambridge, UK), as previously described (Xia, Wang, Yu, & Li, 2017). The limits of detection (LOD) for zinc and iron were within a range of 0.03–0.05 mg/L and the LOD for phosphorus was 0.08 mg/L, which were sensitive enough for the quantitative analysis of selected minerals in this work.

2.6. In vitro starch digestibility

According to the determination of starch in food from the Standardization Administration of China (GB/T 5009.9-2008), 250 mg of samples were weighed and rinsed with 10 mL of alcohol (80%, v/v) in order to remove soluble carbohydrates. Then, the concentrations of digestible starch (DS) and resistant starch (RS) were simultaneously determined according to the approach modified by Morales, Escarpa, and González (1997), with slight modifications. The glucose released within digestion process was used to define the contents of DS and RS fractions, with a correction factor of 0.9 (Xia, Wang, Xu, et al., 2017).

2.7. Lipid hydrolysis

Lipid hydrolysis of GBR grains was expressed by the formation of free fatty acids (FFA), which was quantified based on the method described by Jaisut, Prachayawarakorn, Varanyanond, Tungtrakul, and Soponronnarit (2009). FFA in ground samples was firstly extracted by ethanol (95%, v/v) and titrated with standard sodium hydroxide solutions using phenolphthalein as indicator until a faint pink color persisted for 0.5 min. FFA concentrations were indicated as meq FFA per 100 g sample, using oleic acid as equivalent.

2.8. Instrumental analysis of color parameters

To accomplish sample homogeneity, all rice flour samples were prepared by grinding GBR grains with a laboratory mill (GX-02, Gaoxiang, China), which were then screened through a 178-μm mess. The color coordinates were obtained in triplicate using a colorimeter (LabScan XE, HunterLab, Reston, VA) in the CIE-LAB system, and the results were indicated as lightness (L^*), redness (a^*) and yellowness (b^*). The overall color difference (ΔE) was determined with the following formula: