



# Effects of $\gamma$ -irradiation on phenolics content, antioxidant activity and physicochemical properties of whole grain rice

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## HIGHLIGHTS

- Effects of irradiation on physicochemical properties of three rice were investigated.
- The bound phenolics and its antioxidant capacity were increased with the dosage increase.
- The color parameters of flour were slightly affected by irradiation.
- The viscosity and gel hardness were decreased continuously with the dosage increase.

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## ABSTRACT

Three rice genotypes with different color were gamma irradiated at a dose of 2, 4, 6, 8 and 10 kGy. The aim of this study was to investigate the effect of gamma irradiation on the phenolics content and the antioxidant activity, as well as physicochemical properties of whole grain rice. The bound phenolics content in all the genotypes were significantly increased with the increase of dose of irradiation. Gamma irradiation at high dose significantly increased the free, bound and total antioxidant activities of three rice genotypes except for the free antioxidant activities of red rice. Though the color parameters were slightly changed, these changes could not be visibly identified. Rapid visco-analyzer (RVA) viscosities and gel hardness decreased continuously with the increase of the irradiation doses. It is suggested that gamma irradiation enhanced the antioxidant potential and eating quality of whole grain rice.

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## 1. Introduction

Rice is the staple food for more than half the population in the world. The commonly consumed rice is white rice, also called polished rice or milled rice, in which the bran layer and embryo of rice grain have been removed by polishing. Contrary to milled rice, brown rice grain is the rice with only the hull removed, and thus retains the embryo and bran. Whole grains have become popular in western countries, but now gradually accepted in developing countries with the improvement of living standards. Consumption of the whole grain brown rice has brought many health benefits including reduced risks of chronic diseases such as cardiovascular disease, type-2 diabetes, some cancers and all-cause mortality (Liu, 2007). Brown rice whole grains have more fiber, antioxidants such as vitamin E and trace minerals compared to milled rice, which are responsible for its health benefits (Bao, 2012).

It has been widely known that irradiation is one of the processing technologies available for killing some insects and microbes, and has been proven successfully in ensuring the safety and extending the shelf life of foods as well as selectively inactivating and removing antinutritional factors in food (Mahapatra et al., 2005; Siddhuraju et al., 2002). In 1981, the Food and Agriculture Organization of the United Nations (FAO)/International Atomic Energy Agency (IAEA)/World Health Organization (WHO) joint committee have declared that irradiation dose up to 10 kGy introduced no special nutritional problem, and for cereal products, doses  $\leq 1$  kGy can eliminate insects, and doses  $\geq 2$  kGy can eliminate bacteria and reduce cooking time (Lacroix and Ouattara, 2000). In 1999, FAO/IAEA/WHO Study Group and International Conference advocated that food irradiated to any dose appropriate to achieve technological objectives is both safe and nutritionally adequate (Kong et al., 2009).

Effects of the gamma irradiation on the physicochemical properties and nutritional quality of different foods have been reported in recent years (Liu et al., 2012; Mukisa et al., 2012; Chung and Liu, 2010; Singh and Datta, 2010; Kim et al., 2009a,

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2009b; Aziz et al., 2006). These studies have proved that gamma irradiation is effective to decontaminate cereals and other foods (Mukisa et al., 2012; Aziz et al., 2006; Aouidi et al., 2011), modify starch physicochemical properties (Liu et al., 2012; Mukisa et al., 2012; Chung and Liu, 2010; Wang and Yu, 2009), and alter nutritional and antioxidant activities (Singh and Datta, 2010; Kim et al., 2009a, 2009b; Aziz et al., 2006; Shawrang et al., 2008). Effects of gamma irradiation on the physical properties of milled rice have been well documented in the literatures (Bao et al., 2001a; Bao and Corke, 2002; Sung et al., 2008; Yu and Wang, 2007). However, as far as we are aware, no study has been on the effects of gamma irradiation on the whole grain rice.

Antioxidant activity is an important parameter to evaluate the functional property of foods. Many studies have been conducted on the effects of gamma irradiation on the phenolics and antioxidant capacity of fruits (Hussain et al., 2010), medicinal plants (Jeong et al., 2009; Brandstetter et al., 2009), mushroom (Jeong et al., 2009), and tea (Jo et al., 2003), etc. Some studies showed that gamma irradiation did not alter the radical scavenging activities and total polyphenols of extracts of some medicinal plants (Jeong et al., 2009; Brandstetter et al., 2009), dried spices (Nagy et al., 2011) and olive leaves (Aouidi et al., 2011). Others reported that low or mild gamma irradiation could increase or slightly increase the total phenolics content, and enhance the antioxidant activities of seeds of soybean and cumin (Dixit et al., 2010; Kim et al., 2009a) and peach fruit (Hussain et al., 2010).

Effects of gamma irradiation on the free, bound and total phenolics content and antioxidant capacity of whole grain rice, as well as physicochemical properties have not been reported. The objective of this study was to determine the effects of gamma irradiation on free, bound and total phenols, free, bound and total antioxidant capacity, color parameters, paste viscosity profiles and textural property of whole grain brown rice.

## 2. Experimental part

### 2.1. Materials and $\gamma$ -irradiation treatment

Three rice genotypes whose colors were distinctly different, including one white rice (BP605), one red rice (BP144) and one black rice (BP602), were grown and harvested in the farm of Zhejiang University in 2009. Fresh rice samples were air-dried until the moisture content was reduced to about 13%, and stored in air-tight plastic bags at room temperature for three months. About 10 g rice grain samples in the air-tight plastic bags were irradiated with  $\gamma$ -rays at room temperature. The treatment was performed in a  $^{60}\text{Co}$  irradiator with a dose rate of 1.0 kGy/h at the Institute of Nuclear Agricultural Sciences, Zhejiang University, China. The five doses used were 2, 4, 6, 8, and 10 kGy. The dose field was measured by a Fricke dosimeter. After irradiation, the rice grains were dehulled on a Satake Rice Machine (Satake Co., Japan), and then milled and passed through a 100 mesh sieve on a cyclone sample mill (UDY Corporation, Fort Collins, CO).

### 2.2. Chemicals and reagents

Methanol (MeOH), ethanol (EtOH), hexanes, ethyl acetate, and hydrochloric acid (HCl) were of analytical grade and were purchased from Sinopharm (Sinopharm chemistry reagent CO., Ltd, China). Sodium carbonate ( $\text{Na}_2\text{CO}_3$ ), potassium peroxydisulfate, sodium hydroxide (NaOH) and gallic acid were of analytical grade and were purchased from BBI (Toronto, Ontario, Canada). Folin–Ciocalteu reagent, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) and 2,2-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) were purchased from Sigma (St. Louis, MO).

### 2.3. Extraction of free phenolics

Free phenolic compounds of rice grain samples were extracted using the method previously reported by Zhang et al. (2010) with minor modification. Briefly, 0.5 g rice grain flour was blended with 10 mL of chilled acidified methanol (99% methanol and 1 M HCl 85:15, v/v). The mixture was then centrifuged at  $2500 \times g$  for 10 min. The supernatant was collected, and the remaining pellet was again extracted with 10 mL of chilled acidified methanol for another two times. All supernatants were pooled and made up to a final volume of 30 mL with chilled acidified methanol. The extracts were stored at 4 °C in dark until use. The remaining pellet was used to extract the bound phenolics.

### 2.4. Extraction of bound phenolics

Bound phenolics of rice grain prepared as forward described were extracted using the method previously reported by Zhang et al. (2010) with minor revises. Briefly, after 0.5 g of rice grain powder was extracted for free phenol, the residues were then digested with 2 M sodium hydroxide at room temperature for 2 h on a mechanical shaker. The mixture was neutralized with an appropriate amount of hydrochloric acid and extracted with hexane to remove lipids. The final solution was extracted with ethyl acetate. The ethyl acetate fraction was evaporated to dryness under nitrogen gas. Phenolics compounds were reconstituted in 10 mL of chilled acidified methanol (99% methanol and 1 M HCl 85:15, v/v) and stored at 4 °C in dark until use.

### 2.5. Free and bound phenolics content

The phenols content was assayed by the Folin–Ciocalteu colorimetric methods with a slight modification as before (Cai et al., 2004). Briefly, aliquots (0.15 mL) of appropriately diluted extracts or standard solutions were mixed with 1.0 mL 0.5 N Folin–Ciocalteu reagent and then the reaction was neutralized with 7.5% sodium carbonate. Then measure the absorbance at 760 nm using a spectrophotometer (UNICAM UV300, Thermo Scientific, USA) after incubating for 2 h at room temperature. The phenolics contents were expressed as mg of gallic acid equivalent (GAE) per 100 g of dry weight. The total phenolics content calculated as the sum of free and bound phenols.

### 2.6. Antioxidant capacity

The antioxidant capacity was expressed as radical cation  $\text{ABTS}^+$  scavenging activity. It was conducted by using a spectrophotometer method by the improved 2,2-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) radical cation (Cai et al., 2004; Re et al., 1999). The  $\text{ABTS}^+$  solution was prepared by  $\text{ABTS}$  (7 mM): potassium (2.45 mM) peroxydisulfate (V/V=2:1) agitated via an agitator in dark for 14 h. Then 80% ethanol was used to adjust the absorbance to  $0.700 \pm 0.020$  at 734 nm using the spectrophotometer (UNICAM UV300, Thermo scientific, USA). After mixture 3.9 mL  $\text{ABTS}^+$  solution and 0.1 mL standard solution or appropriately diluted extracts together thoroughly for about 6 min, the absorbance at 734 nm was measured. Results were expressed as mM of Trolox equivalent antioxidant capacity (TEAC) per 100 g of dry weight.

### 2.7. Color parameters of rice flour

The color parameters of wholemeal flour were measured by Minolta Chroma Meter CR-301 (Minolta Co., Osaka, Japan), standardized with calibration plate sets CR-A47 and a white plate. Color measurements were expressed as tristimulus parameters,

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