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Food Chemistry

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# From rice bag to table: Fate of phenolic chemical compositions and antioxidant activities in waxy and non-waxy black rice during home cooking

Yayuan Tang, Weixi Cai, Baojun Xu\*

Food Science and Technology Program, Beijing Normal University-Hong Kong Baptist University United International College, Zhuhai, Guangdong 519085, China

## ARTICLE INFO

## Article history:

Received 13 September 2014  
 Received in revised form 23 January 2015  
 Accepted 1 February 2015  
 Available online xxxx

## Chemical compounds studied in this article:

Condensed tannin/proanthocyanidin  
 (PubChem CID: 108065)  
 Anthocyanin (PubChem CID: 145858)  
 Anthocyanin 3'-O-beta-D-glucoside  
 (PubChem CID: 56928084)  
 Cyanidin-3-glucoside (PubChem CID:  
 197081)  
 Peonidin-3-glucoside (PubChem CID:  
 443654)  
 DPPH (PubChem CID: 2735032)  
 (+)-Catechin (PubChem CID: 9064)  
 Gallic acid (PubChem CID: 370)  
 Trolox (PubChem CID: 40634)

## Keywords:

Black rice  
 Thermal processing  
 Phenolic compounds  
 Anthocyanin  
 Cyanidin-3-glucoside  
 Antioxidants

## ABSTRACT

The objectives of this study were to systematically analyze degradation rate of functional substances, such as total phenolic content (TPC), total flavonoid content (TFC), condensed tannin content (CTC), monomeric anthocyanin content (MAC), cyanidin-3-glucoside (Cy3glc), and peonidin-3-glucoside (Pn3glc), as well as antioxidant activities in cooked waxy and non-waxy black rice through different home cooking manners. Results showed that greater phenolics and antioxidant capacities were detected in non-waxy rice rather than waxy one. All processed black rice exhibited significantly ( $p < 0.05$ ) lower TPC, TFC, CTC, MAC, Cy3glc, Pn3glc, and antioxidants as compared to the raw rice. Different processing methods significantly degraded the content and activities of antioxidants of both waxy and non-waxy black rice. Under the same cooking time, black rice porridge retained more active substances than that of cooked rice by rice cooker. Therefore, to maintain bioavailability of active components, black rice porridge may gain more health promoting effects.

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## 1. Introduction

Nowadays, colored rice (*Oryza stiva* L.) consumption is increasing rapidly. Both waxy and non-waxy black rice are particularly important colored rice species, and derive their names from their naturally purple or black pigments that have been confirmed as anthocyanins (Kong & Lee, 2010). Rice is cultivated primarily in Asian countries, such as China, Japan and Korea, and is generally consumed as an ingredient in refreshments by people who are

living in those countries (Tananuwong & Tewaruth, 2010). It has been shown that black rice has a beneficial contribution to nutritional and therapeutic values in comparison to white rice, and these extra values make the pigmented rice important sources of amino acids (especially essential amino acids), vitamins and some trace minerals (such as Fe, Zn and Cu), as well as rich natural colorants (Jiang, Liu, Long, & Sheng, 1999).

It has been reported that black rice might provide health benefits to reducing risk of chronic diseases, such as cardiovascular problems, cancers (Xia et al., 2006), diabetes and its complications (Walter & Marchesan, 2011), as well as iron-deficiency anemia (Wang & Guo, 2007), because of the existences of phytochemicals in black rice, such as phenolic compounds (Shen, Jin, Xiao, Lu, & Bao, 2009).

\* Corresponding author at: 28, Jinfeng Road, Tangjiawan, Zhuhai, Guangdong 519085, China. Tel.: +86 756 3620636; fax: +86 756 3620882.

E-mail address: [baojunxu@uic.edu.hk](mailto:baojunxu@uic.edu.hk) (B. Xu).

Phenolics, defined as molecules with at least one aromatic ring bearing one or more hydroxyl groups, are one of the major bioactive substances in black rice (Shen et al., 2009). Among phenolic substances, flavonoids are a large subclass, containing two or more aromatic rings and each ring bearing at least one aromatic hydroxyl and connected with a carbon bridge. Anthocyanins (one group of water-soluble flavonoids) are the predominant pigments and functional phenolics in black rice. They have been proved to be essentially substantial foundations for anti-oxidation effect, because of their notably strong free-radical scavenging effects in black rice (Chiang et al., 2006; Shen et al., 2009; Zhang, Guo et al., 2006; Zhang, Zhang et al., 2006). Five anthocyanins have been separated and identified. These anthocyanins are malvidin, pelargonidin-3,5-diglucoside, cyanidin-3-glucoside, cyanidin-3,5-diglucoside and peonidin-3-glucoside (Hou, Qin, Zhang, Cui, & Ren, 2013; Zhang, Guo et al., 2006; Zhang, Zhang et al., 2006).

Rice is usually cooked before its consumption, and there are two ordinarily home-cooked products in Asian countries: rice porridge and cooked rice. A negative correlation between thermal processing and the concentration of some functional ingredients, such as phenolic compounds, has been proved (Walter et al., 2013). Hiemori, Koh, and Mitchell (2009) reported that all cooking processes could generate a dramatic decrease in anthocyanin content of black rice.

Although a plenty of literature support the potential beneficial effects of black rice on health, little is known about the influence of hydrothermal cooking approaches on functional components and antioxidant activities in cooked black rice. The present study was intent to evaluate functional compositions and antioxidant activities in waxy and non-waxy black rice as affected by home cooking. The changes in total phenolic content (TPC), total flavonoid content (TFC), condensed tannin content (CTC), monomeric anthocyanin content (MAC), cyanidin-3-glucoside and peonidin-3-glucoside, as well as antioxidant capacities in waxy and non-waxy black rice with two ordinarily home-cooking methods were investigated. The changes in these functional substances under different cooking time (20, 25, 30, and 35 min) were also investigated. The consequences of this research can assist the public to understand in details the loss of these bioactive substances in black rice after home cooking.

## 2. Materials and methods

### 2.1. Chemicals and reagents

Folin–Ciocalteu reagent, 2-diohenyl-1-picrylhydrazyl (DPPH), (+)-catechin, and 2,4,6-tri (2-pyridyl)-s-triazine (TPTZ) were purchased from Shanghai Yuanye Biological Technology Co., Ltd (Shanghai, China). Anthocyanin standards of cyanidin-3-glucoside and peonidin-3-glucoside were supplied by ChemFaces Biochemical Co., Ltd. (Wuhan, China). 2,6-Di-tert-butyl-p-cresol was purchased from Lingfeng Chemical Reagent Co., Ltd. (Shanghai, China). 6-Hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) and high-performance liquid chromatography (HPLC)-grade acetonitrile was obtained from Sigma–Aldrich Co. (Shanghai, China). Vanillin was offered by Wenzhou Dongsheng Chemical Reagent Factory (Zhejiang, China). Absolute ethanol was from Tianjin Fuyu Fine Chemical Co., Ltd. (Tianjin, China). Other chemical reagents were supplied by Tianjin Damao Chemical Reagent Co., Ltd (Tianjin, China). All chemicals were of analytic grade unless specially mentioned.

### 2.2. Black rice samples

The rice species used in this research was *Oryza stiva* L. *indica* and included its waxy and non-waxy variants. Both of them were cultivated in Guilin, Guangxi Province, China.

### 2.3. Soaking time and determination of hydration rate

According to the method of Xu and Chang (2008a, 2008b) with slight modifications, black rice (20 g) were rinsed under running tap water, and soaked in 100 mL of tap water at room temperature for 24 h. Within the initial 6 h, the weight gain of sample was measured hourly as water absorption (moisture content), followed by each 2 h for determination over the next 10 h. The last measurement was done at 24 h. After each weighing, samples were placed back into the soaking water. The curve about water absorption was created by plotting the kinetic moisture content (%) with soaking time.

### 2.4. Cooking approaches and cooking time

Two thermal processes were performed for black rice. Processing method 1 (black rice porridge making): about 40 g of waxy and non-waxy black rice soaked for 2.5 h was separately added with 240 mL water (containing soaking water), and cooked on an electric hot plate cooker. After boiling, black rice was cooked for 20 min, 25 min, 30 min, and 35 min in each different sample. Processing methods 2 (cooked black rice making): presoaked (2.5 h) waxy and non-waxy (approximately 40 g) was respectively placed into 120 mL water (containing soaking water), and cooked by an automatic house-hold rice cooker (Luby Electronic Co., Ltd, Guangdong, China) for 25 min. All these cooked samples were freeze-dried by freeze-drier (Labconco Corporation, Kansas City, MO, U.S.A.), and then stored in 4 °C refrigerator for further studies.

### 2.5. Color measurement

Color attributes of black rice samples were measured by Colorimeter (CR-410, Konica Minolta, Japan). The color was expressed in  $L^*a^*b^*$ , where the  $L^*$  represents lightness ( $L^* = 0$  yields black and  $L^* = 100$  denotes white), the  $a^*$  expresses red (+) or green (–), and the  $b^*$  indicates yellow (+) or blue (–).  $L^*$ ,  $a^*$  and  $b^*$  parameters were measured against a white background plate which were directly obtained from the apparatus.

### 2.6. Extraction of phenolics from rice samples

Accurately, 0.5 g of each ground dry sample was weighed, and extracted with 5 mL of acidic 70% acetone (acetone/water/acetic acid = 70:29.5:0.5, v/v/v) in a set of capped centrifuge tubes by shaking on an orbital shaker at ambient temperature for 3 h and setting in the dark for 12 h. Then the extracts were centrifuged at 4000 rpm for 10 min, and the supernatants were removed. Residues were mixed with 5 mL of extracting solvent for the second time and the third time. All extracts were combined and stored at 4 °C in the dark. The extraction of every sample was conducted in triplicate.

### 2.7. Determination of total phenolic content (TPC)

Total phenolic content (TPC) was evaluated according to the method of Xu and Chang (2007) with slight modifications. TPC was demonstrated as milligrams of gallic acid equivalents (mg GAE/g black rice).

### 2.8. Determination of total flavonoid content (TFC)

The total flavonoid content (TFC) was determined by a colorimetric method using (+)-catechin as the standard (Xu & Chang, 2007). TFC was expressed as (+)-catechin equivalents (mg CAE/g black rice).

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