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# Total phenolic, flavonoid content, and antioxidant activity of flour, noodles, and steamed bread made from different colored wheat grains by three milling methods

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ABSTRACT

The objective of this study was to evaluate the effects of wheat variety, food processing, and milling method on antioxidant properties. Black wheat variety Heibaoshi 1 had the highest total phenolic content (659.8 μg gallic acid equivalents $g^{-1}$), total flavonoid content (319.3 μg rutin equivalents $g^{-1}$), and antioxidant activity, whereas light purple wheat variety Shandongzimai 1 had the lowest total flavonoid content (236.2 μg rutin equivalents $g^{-1}$) and antioxidant activity. Whole wheat flour and partially debranned grain flour had significantly higher total phenolic contents, total flavonoid contents, and antioxidant activity than refined flour ($P < 0.05$). Compared with flour, total phenolic contents, total flavonoid contents and antioxidant activity decreased in noodles and steamed bread, whereas noodles had slightly higher total phenolic and flavonoid content than steamed bread. Antioxidant activities (by ferric reducing ability of plasma assay) of steamed bread made from whole wheat flour, partially debranned grain flour, and refined flour were 23.5%, 21.1%, and 31.6% lower, respectively, than the corresponding values of flour. These results suggested that black whole wheat flour and partially debranned grain flour are beneficial to human health.



## 1. Introduction

Free radicals contribute to cancer, atherosclerosis, malaria, and rheumatoid arthritis and neurodegenerative diseases [1]. In the scientific and medical communities, antioxidants are considered to have the ability to scavenge free radicals and reduce oxidative damage [2,3]. Accordingly, increased consumption of fruits and vegetables containing high levels of antioxidants has been recommended. However, wheat, one of the most important grains in the world, is not only a source of basic nutrients, such as carbohydrates, proteins, and vitamins, but also a source of antioxidants, such as flavonoids

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and phenolic acids [4]. Wu et al. [5] reported that the antioxidant activity of whole grain including whole wheat bread ranged from 1303 to 2479 μmol trolox equivalent (TE) per 100 g, whereas the average values of 24 types of fruit and 22 types of vegetables were 2200 and 1200 μmol TE per 100 g, respectively. These results indicated that whole grains have pronounced antioxidant activities that should not be overlooked. Regular consumption of these bioactive compounds could reduce the risk of cardiovascular diseases and cancer [6].

These bioactive compounds are mainly located in the outer membranes of the grain [7]. The bran fraction contains markedly higher concentrations of phenolic acids than those in white flour [8]. Conventional modern milling methods, which remove most of the bran and germ, reduce the amounts of total phenolics and flavonoids in wheat products [4]. Wang et al. [9] found that the grain phenolic acid concentrations ranged from 54 μg $g^{-1}$ in flour produced at 60% extraction rate to 695 μg $g^{-1}$ in flour produced at 100% extraction rate. Given that the regular consumption of whole wheat products reduces the risk of heart disease and cancer [4,6], gradual or whole-wheat milling has been developed to retain nutrients and bioactive compounds [10,11]. However, food processing also affects the antioxidant properties of foods to different extents. Wu et al. [5] found that processing methods (including cooking and hulling) affected oxygen radical absorbance capacity. Chlopicka et al. [12] reported that total flavonoid content (TFC) of flour was approximately 2–4-fold higher than that in breads. Noodles and steamed bread are the most widely consumed wheat products in China. Researchers have focused on flour noodle and steamed bread sensory qualities [13,14], and there is little information on the antioxidant properties of these products.

Recently, much attention has been focused on colored wheat varieties. Black-grained wheat has been reported to have high free radical scavenging ability and phenolic content [15]. Purple and black wheat varieties have high protein content and antioxidant activity (AOA), owing to the presence of phenolic acid and vitamin C [16,17]. Similarly, green wheat bran has high AOA, which is positively correlated with pigmentation [18]. The relationships between grain color and nutrient qualities have been reported by Zong et al. [19]. Colored wheat, which has high levels of anthocyanins, suppresses oxidation and nitric oxide formation in vitro [20]. However, there has been some disagreement about the relationship between total phenolic content and antioxidant activity and grain color. Mpofu et al. [21] found that grain color does not appear to be a factor in the expression of antioxidant-related parameters. Colored wheat with lower antioxidant activity was also reported by Liu et al. [17]. In our previous report, some white wheat varieties showed a higher total phenolic content (TPC) and antioxidant activities than black wheat varieties [22]. Similar results have been found in rice [23]. To clarify the effect of grain color, milling methods and food types on antioxidant content and its activities, three colored wheat varieties (deep purple, light purple, and black) and one white wheat variety were milled to different degrees (yielding whole wheat flour, partially debranned grain flour, and refined flour). TPC, TFC, and AOA of the different flours, Chinese fresh noodles, and steamed bread were analyzed.

## 2. Materials and methods

### 2.1. Materials

Four different colored wheat (*Triticum aestivum* L.) varieties were collected from the 2013 harvest at Henan Agricultural University Experimental Station: white Yumai 49-198 (YU), deep purple Jizi 439 (JZ), light purple Shandongzimai 1 (SDZM), and black wheat Heibaoshi 1 (HBS). Seeds were cleaned and stored at room temperature and damaged seeds were removed. Three milling methods were used, yielding whole wheat flour (WWF), partially debranned grain flour (PGF), and refined wheat flour (RF). To prepare WWF, whole wheat kernels were milled in a Cyclotec 1093 mill (Foss Tecator, Höganäs, Sweden) without removal of bran or germ. To prepare PGF, whole wheat kernels were first stripped of the bran layer using a grain polisher (TYT200, Tianyang Machinery Co. Ltd., Shandong, China) and then milled. Hulling degree was calculated as the difference between the initial sample weight and the hulled grain weight, relative to the initial sample weight, and was approximately 5.0%. To prepare RF, wheat kernels were milled into refined flour in a Brabender Junior laboratory mill (method 26-21A, AACC, 1995). All sample flours were passed through 80-mesh sieves (sieve size: 0.180 mm).

### 2.2. Noodle and steamed bread making

Chinese fresh noodles were prepared as described by Zhang et al. [24]. Fresh noodles (50 g) were cooked in 1000 mL distilled water, rinsed with cold water, and allowed to cool to room temperature. The noodles were then dried and milled into noodle powder with a small grinder (FW100, Taisite Co. Ltd., Tianjin, China).

Chinese steamed breads were prepared as described by Chen et al. [25]. After cooking, steamed breads were allowed to cool at room temperature, sliced 1.5 cm thick, and oven-dried at 40 °C. The steamed bread slices were milled into bread powder with the FW100 grinder. For each sample, two replicates were prepared for noodles and steamed breads.

### 2.3. Extract preparation

Total phenolics were extracted from the samples by the method of Moore [26], with slight modifications. Briefly, flour samples (2.0 g) were mixed with 16 mL of methanol containing 1% HCl for 24 h at 24 °C. The procedure was repeated twice. The methanol extracts were centrifuged at 4000 × 9.81 (m $s^{-2}$) for 15 min and the resulting supernatants were pooled and stored at 4 °C.

### 2.4. TPC

TPC was determined by the method of Singleton [27], with slight modifications. Extracts (0.5 mL) were mixed with 5 mL of Folin–Ciocalteu reagent (1 mol), neutralized with 4 mL saturated sodium carbonate (75 g $L^{-1}$), and kept at room temperature for 2 h. Absorbance at 765 nm was measured with a spectrophotometer. TPC was expressed as gallic acid equivalents (mg GAE $g^{-1}$ dry weight).