



# Phenolic acid composition, antioxidant activity and phenolic content of tarhana supplemented with oat flour



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## ABSTRACT

In this study, oat flour (OF) was used to replace wheat flour in tarhana formulation at the levels of 10, 20, 30 and 40% (w/w). Control sample did not contain OF. The results showed that addition of OF caused increases in levels of phenolic acids within tarhana samples. The most abundant phenolic acids were vanillic and ferulic acids, and they were followed by gallic acid. Tarhana samples with OF also showed higher antioxidant activities than control sample did. Compared with the control sample, the total phenolic content level increased with the increase in the amount of OF. The results of sensory analysis showed that OF addition neither caused any undesirable taste nor an odor and panelists emphasised a sweet taste as the OF amounts were increased. Therefore, tarhana supplemented with OF can be claimed to be a good source of minerals, phenolics and antioxidants as compared to tarhana without OF.

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## 1. Introduction

Tarhana is a traditional Turkish fermented cereal food made from cereal flours, yogurt and various vegetables. It is a good source of B vitamins, minerals, organic acids and free amino acids. Thanks to its rich content, it is known to be a healthy food for children, adults, and for patients (Daglioglu, 2000). After all ingredients are mixed and homogenised and dough is obtained, it is fermented at 30–35 °C for 1–5 days (Gocmen, Gurbuz, Roussef, Smoot, & Dagdelen, 2004; Temiz & Pirkul, 1991), then it is dried and ground by a mill (Tarakci, Dogan, & Koca, 2004). An important part of tarhana consumed in Turkey is homemade and therefore it is sun-dried. From a commercial point of view, tarhana is produced by using modern drying techniques on industrial scale (Tarakci et al., 2004). Tarhana is mainly consumed as soup in Turkey. Tarhana powder is first mixed with cold water (1:5) and then allowed to dissolve for about half an hour, and finally cooked for 20 min with occasional stirring. As soon as it is boiled, some butter is added to the soup and it is consumed at 70 °C. Cheese and roasted bread pieces can also be added upon request (Ozdemir, Gocmen, & Kumral, 2007).

Regional diversity of the amount and the type of ingredients, as well as the processing techniques in Turkey, affects chemical composition, nutritional content and sensory attributes of tarhana (Degirmencioglu, Gocmen, Dagdelen, & Dagdelen, 2005; Tarakci et al., 2004). Basically, 4 different types of tarhana have been defined by the Turkish Standardization Institute: (a) flour tarhana,

(b) goce tarhana, (c) semolina tarhana, and (d) mixed tarhana. Using wheat flour, cracked wheat and semolina separately or as combinations in the recipe causes some differences (Daglioglu, 2000). Cereal and legume flours other than wheat flour can also be used in the production of tarhana (Ozdemir et al., 2007). One of them is oat flour which is known as rich in bioactive compounds, dietary fiber and  $\beta$ -glucan.

The use of oats in human nutrition has been increasing owing to the fact that they contain beneficial nutritional properties (Webster, 2002). Oats are also useful for the control of diabetes, lipid profile (Butt, Tahir-Nadeem, Khan, & Shabir, 2008), total and low density lipoprotein (LDL) cholesterol levels (Kestin, Moss, Clifton, & Nestel, 1990).

In recent years, the interest in oat-based foods for human consumption has increased dramatically because oats are rich in  $\beta$ -glucan and phenolic (Gray et al., 2000; Liu, Zubik, Collins, Marko, & Meydari, 2004; Malkki, Myllymaki, Teinila, & Koponen, 2004; McMullen, 2000; Peterson, Emmons, & Hinns, 2001). The most important bioactive compounds of oats are phenolic compounds. Some oat phenolics have great potential as nutraceuticals while some others are powerful antioxidants. Before the development of the commercial potential of synthetic antioxidants, oat flour was used as antioxidant to extend the shelf life of milk powder, butter, ice cream and some cereal products for many years (Webster, 2002). The antioxidant potential of oats has been recognised for many years. The US FDA Authorization of the Heart Health's remarks connected with the consumption of oats is especially significant. In the early 1960s, the first researchers who investigate oat antioxidants drew attention to their potential as a sanitary material (Webster, 2002). In addition to its nutritional

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and antioxidant properties, phenolic compounds also influence multiple sensorial food properties, such as flavour, astringency, and colour. Phenolic compounds contribute to aroma and taste of numerous food products of plant origin (Rodriguez et al., 2009).

In this study, tarhana was supplemented with oat flour so as to improve its functional and nutritional qualities. The aim of this research was to determine the effects of oat flour addition on the phenolic acid composition, antioxidant activity, phenolic and mineral contents of tarhana.

## 2. Materials and methods

### 2.1. Materials

Wheat flour (*Triticum aestivum*) (Type 650), containing 13.3% protein (db), 0.64% ash (db) and 13.4% moisture, was used and supplied from the Toru Flour Milling Co., Ltd. (Bandirma/Turkey). Salt (NaCl), yoghurt, dried onion, tomato and paprika pastes were purchased from the local markets in Bursa, Turkey. Stabilised oat meal (inactivated fat hydrolysing enzymes) was purchased from Eti Food Co., Ltd. (Bozuyuk, Bilecik, Turkey). Oat meal was ground by using a hammer mill (Falling Number-3100 Laboratory Mill, Perten Instruments AB, Huddinge, Sweden) and sieved through 212  $\mu\text{m}$  sieve. It was kept in refrigerator until analysed.

### 2.2. Production of tarhana

To prepare tarhana samples, flour 100%, yogurt 50%, dried onion 2.5%, tomato paste 2.5%, paprika paste 7.5% and salt (NaCl) 7.5% were used (w/w, flour base). Oat flour (OF) were used to replace wheat flour at the levels of 10, 20, 30 and 40% (w/w) whereas control sample did not contain OF. The ingredients were mixed in a mixer (Electrolux Ditomix 5, GA, USA) for 5 min and the resulting dough samples were fermented at 30 °C in a fermentation cabinet (Efe Co., Ltd., İzmir/Turkey) for 3 days. The fermented dough samples were dried at  $50 \pm 2$  °C in an air-convection oven (Efe Co., Ltd., İzmir/Turkey) to 9–10% moisture content. After drying process, tarhana samples were ground into a rough powder by a hammer mill equipped with a 1-mm opening screen. The resulting powders were stored in a glass jar (0.5 L) and kept in a refrigerator until analysed.

### 2.3. Determination of minerals

All solutions were prepared with analytical reagent grade chemicals and ultra-pure water (18 M $\Omega$  cm resistivity) generated by purifying distilled water with the TKA Ultra Pacific and Genpura water purification system (Germany). Suprapur HNO<sub>3</sub> (67% v/v) was purchased from Merck (Darmstadt, Germany). Standard stock solutions containing 1000 mg L<sup>-1</sup> of each element (K, Ca, P, Mg, Fe, Cu, Zn and Mn) were purchased from Merck (Darmstadt, Germany) and used to prepare calibration standards. Working standards were prepared in 0.3% (v/v) HNO<sub>3</sub> on a daily basis and used without further purification. 1000 mg L<sup>-1</sup> standard stock solutions (Merck, Darmstadt, Germany) were prepared in 0.3% HNO<sub>3</sub> for internal standard solution. Argon (99.9995% pure, Linde, Turkey) was used as carrier gas.

Sample digestion was carried out using the Milestone MLS 1200 (Italy) microwave digestion system. The samples were homogenised and then approximately 0.5 g of them was weighed directly on PTFE flasks after adding 6 mL of HNO<sub>3</sub> and subjected to following digestion program: 250 W (2 min), 0 W (2 min), 250 W (6 min), 400 W (5 min) and 600 W (5 min). After cooling at room temperature, sample solutions were transferred into 50 mL polyethylene flasks. 100  $\mu\text{L}$  of internal standard solution

(1 mg L<sup>-1</sup>) was added and then the digested samples were diluted to 25 mL prior to analysis by ICP-OES.

The mineral content of the tarhana samples (K, Ca, P, Mg, Fe, Cu, Zn and Mn) were measured using the microwave (Milestone MLS 1200, Italy) nitric acid digestion procedure according to the method described by Sahan, Basoglu, and Gucer (2007) and it was followed by induction coupled plasma optical emission spectrometry (Perkin Elmer 2100 ICP-OES). The emission intensities were obtained for the most sensitive lines free of spectral interference. The analyses were performed at the following flow rates: (a) plasma gas of 15 L min<sup>-1</sup>, (b) auxiliary gas of 1 L min<sup>-1</sup>, and (c) sample of 0.8 mL min<sup>-1</sup>. The mineral eluates were monitored at different wavelengths: 317.9 nm-Ca, 214.9 nm-P, 285.2 nm-Mg, 766.5 nm-K, 206.2 nm-Zn, 238.2 nm-Fe, 327.4 nm-Cu and 257.6 nm-Mn. All chemical analysis were carried out in duplicate on each sample.

### 2.4. Extraction of free phenolic compounds

Free phenolic compounds were extracted according to Vitali, Vedin, Dragojevic, and Sebecic (2009) with slight modifications. Samples (10.0 g dry weight-dw) were defatted twice with 20 mL of hexane at 30 °C by an ultrasonic homogeniser (Scientz-IID, Ningbo Scientz Biotechnology Co., Ltd., Zhejiang, China). The defatted samples (2.0 g dry weight-dw) were mixed with 20 mL of HClconc/methanol/water (1:80:10, v/v) mixture and shaken with a laboratory rotary shaker (JB50-D; Shanghai Shengke Instruments, Shanghai, China) at 250 rpm for 2 h at 20 °C, and then the mixtures were centrifuged at 3500 rpm for 10 min at 4 °C in a centrifuge (Eppendorf 5417R, Germany). The supernatants (*free/extractable phenolic compounds*) obtained after centrifugation were stored at -20 °C until used.

### 2.5. Extraction of bound phenolic compounds

Bound phenolic compounds were extracted according to Vitali et al. (2009) with slight modifications. After free phenolic extraction, the residues were combined with 20 mL of methanol/H<sub>2</sub>SO<sub>4</sub> conc (10:1) mixtures. The mixtures were placed in a water bath at 85 °C for 20 h, and then cooled at room temperature. They were centrifuged at 3500 rpm for 10 min at 4 °C in a centrifuge (Eppendorf 5417R, Germany). The supernatants (*bound/hydrolyzable phenolic compounds*) were stored at -20 °C until used.

### 2.6. Determination of phenolic content

Phenolic contents (*free, bound and total phenolics*) were determined based on the Folin-Ciocalteu colorimetric method as described by Xu et al. (2009) with slight modifications. To summarise briefly, an aliquot (0.5 mL) of appropriately diluted extracts, 2.5 mL of deionised water and 0.5 mL of 1.0 M Folin-Ciocalteu reagent were mixed within 10 mL volumetric flasks and vortexed for 10 min at room temperature. After 30 min, 1.5 mL of 7.5% sodium carbonate solution was added and mixed thoroughly. The absorbance of the reaction mixtures was measured using a spectrophotometer (UV-Mecasys, Optizen 3220) at 750 nm wavelength after incubation for 30 min at room temperature. Methanol was used as the blank, and gallic acid (GA) was used for calibration of the standard curve (0–500 mg/L). Phenolic content was expressed as gallic acid equivalents (milligrams of GAE per gram DW).

### 2.7. Determination of phenolic acids

Phenolic acids were analysed according to validated methods with slight modifications in HPLC elution conditions (Mattila, Pihlava, & Hellström, 2005; Verardo, Serea, Segal, & Caboni, 2011;

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