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Effect of various heat treatments on rancidity and some bioactive compounds of rice bran

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ABSTRACT

Rice bran contains natural phytochemicals but unstable against hydrolytic and oxidative rancidity. Various heat treatments, such as dry-heating (DH), freeze-drying followed by dry-heating (FDDH), microwave heating (MH), autoclaving (AC), and ethanol vapor (EV) treatment were applied to rice bran and their effects on the storage stability at room temperature were evaluated. The free fatty acid (FFA) content of untreated rice bran gradually increased from 2.14 to 19.81% during 24 weeks, whereas those of the treated rice brans were not or marginally changed. The FFA content in DH samples was greater than that in FDDH samples, indicating the positive effect of freeze-drying prior to DH. Among the treatments, autoclaving was most effective in retarding the FFA formation. The tocol content in the rice brans varied with the treatments, ranging from 181.4 mg/kg (EV) to 310.6 mg/kg (AC), whereas that in untreated rice bran was 216.3 mg/kg. Amounts of other bio-functional components such as phytosterols and policosanols were either unchanged or increased by the treatments. Policosanol level was increased by all the heat treatments, but not affected by EV treatment, indicating that the thermal treatments induced the release of free policosanols.

1. Introduction

Rice bran (RB), a natural by-product obtained from the milling process of rice, is the outer layer of rice grain. It contains the layers of seed coat and germ with minor amounts of broken endosperm which comprises about 10% of the total rice grain (Poural et al., 2009). In Korea, approximately 6,200,000 tons of rice are produced annually, yielding about 500,000 tons of rice bran. Rice bran contains many unique bioactive compounds, such as gamma-oryzanol, phytosterols, tocopherols, tocotrienols and ferulic acids (Loypimai et al., 2009; Sharma et al., 2004). These bioactive compounds are known to have antioxidant activities and various health-beneficial effects on blood pressure, lipid profiles, glucose metabolism, and coronary management (Cicero and Derosa, 2005; Shirakawa et al., 2006).

However, rice bran is rarely utilized for the residual bioactive components because it is unstable to hydrolytic and oxidative rancidity (Tao et al., 1993). When bran layers are removed from the endosperm during the rice milling process, the lipids in rice bran come in contact with reactive enzymes such as lipases. The lipases, which are endogenously present, cause the hydrolysis of neutral bran oil to free fatty acids (FFA), leading to the development of hydrolytic rancidity (Ramezanzadeh et al., 1999). This hydrolytic rancidity contributes to the development of off-flavor and taste and the acidity increase. In addition, the FFA is highly susceptible to oxidation which usually occurs by the action of the inherent lipoxygenase. As a result, the FFA undergoes further oxidative decomposition with the generation of rancid off-flavors in rice bran (Ramezanzadeh et al., 1999). The formation of these compounds is quite rapid and their presence in rice bran makes it unsuitable for human consumption or extraction of edible oil and bioactive components (Loypimai et al., 2009). Therefore, rice bran is currently used as an additive to animal feeds, or as a fertilizer.

Various stabilization methods (inactivating lipases) of rice bran such as dry or moist heating (Sharma et al., 2004), microwave heating (Ramezanzadeh et al., 1999; Tao et al., 1993), ohmic heating (Loypimai et al., 2009), extrusion (Mujahid et al., 2005), γ-irradiation (Ramarathnam et al., 1989), refrigeration and acidification (Amarasinghe et al., 2009) have been conducted to improve its utilization in food. The inactivation of lipolytic

Abbreviations: AC, autoclaving; AOCS, American Oil Chemists' Society; DH, dry-heating; EV, ethanol vapor; FDDH, freeze-drying followed by dry-heating; FFA, free fatty acids; MH, microwave heating; RB, rice bran; RH, relative humidity.

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enzymes by chemicals such as hydrochloric acid, acetic acid, acrylonitrile and propanol has also been studied (Prakash, 1996). Depending on the stabilization treatment, lipolytic enzymes may be either reversibly inhibited or permanently denatured. For the effective stabilization of rice bran, the treatment should sufficiently inactivate lipolytic enzymes but minimize the decomposition of bioactive components. However, study on the effect of stabilization treatments on the bioactive constituents in rice bran has not been performed. In this study, various heat treatments including dry-heating, freeze-drying followed by dry-heating, microwave heating, autoclaving, and ethanol vapor treatment were applied to rice bran and their effects on the storage stability as well as some of the lipophilic bioactive compounds in rice bran were investigated.

2. Materials and methods

2.1. Materials

Rice bran (*Oryzae sativa* L., cultivar: Ilpum) was provided by CJ CheilJedang Co. (Seoul, Korea). The rice bran was screened through a 60-mesh sieve to have a uniform particle size, and then stored in a freezer at −20 °C until analysis. The initial moisture content of the raw rice bran was 9.1%. Tocopherol and tocotrienol isomers were purchased from Merck (Darmstadt, Germany). Phytosterols and polycosanols used as standards for the quantitative analysis were purchased from Sigma–Aldrich (St. Louis, MO, U.S.A.). Solvents and other chemicals used in this study were analytical grade.

2.2. Heat treatments

The rice bran was subjected to different heat treatments as described below. For dry-heating (DH), a portion of rice bran (500 g) was transferred into shallow pans and spread uniformly in a layer of about 0.5 cm thickness. The pans were then placed in a preheated oven at 80 or 100 °C for 1 h (Sharma et al., 2004). The treated rice bran was immediately cooled to room temperature. For freeze-drying followed by dry-heating (FDDH), rice bran was freeze-dried using a freeze-dryer (SFDSM06, Samwon Freezing Engineering Co., Busan, Korea) until the moisture content reached below 3%, and then dry-heating was carried out at 80 or 100 °C as described.

For microwave heating (MH), the moisture content of the raw rice bran was first adjusted to 20% by adding deionized water (Tao et al., 1993). Before heating the rice bran sample in a domestic microwave oven (1200 W; Samsung Electronics Corp. Korea), the microwave chamber was preheated at 100% power for 3 min to establish a constant initial temperature and effective heating. The rice bran sample in a sealed polyethylene bag (spread out 1.5 cm layer) was placed on a microwave-resistant glass tray and subjected to microwave-heating at 100% power for 2 or 5 min (Ramezanzadeh et al., 1999).

Rice bran was also autoclaved at 121 °C for 20 min (AC), or treated with ethanol vapor (EV) following the method described by Champagne et al. (1992). For the EV treatment, an aluminum container (11 cm height × 8.5 cm diameter) equipped with a wire mesh screen at the bottom position was used. Ethanol (95%, v/v) was added to the container, and rice bran was placed on the screen above ethanol. The container was then heated at 100 °C and rice bran was exposed to ethanol vapor for 5 or 20 min. After the treatment, the sample was transferred to a stainless steel pan and allowed to cool to room temperature. For the storage test, all heat-treated samples were stored in sealed polyethylene bags for 24 weeks at room temperature (20–25 °C).

2.3. Color measurement

Color values were measured using a colorimeter (CR-10, Konica Minolta Sensing Inc., Japan). The CIE color values were recorded as L^* (lightness), a^* (redness) and b^* (yellowness). The total color differences (ΔE) between the control and treated samples were calculated using the equation (Yueh et al., 2002): $\Delta E = (\Delta L^2 + \Delta a^2 + \Delta b^2)^{1/2}$

2.4. Moisture content

The moisture content was determined by drying the samples (1 g) in a hot air oven at 105 °C for 3 h and measuring the weight changes. All the data from quantitative analysis are given on a dry basis.

2.5. Oil extraction

Twenty grams of rice bran were dispersed in *n*-hexane (120 mL), and the dispersion was magnetic-stirred at room temperature (20–25 °C) for 6 h. The extract was filtered through a glass fiber filter paper, and the extraction was repeated with the residue using 120 mL *n*-hexane. Two extracts were combined, and then evaporated until *n*-hexane was removed using a rotary evaporator under reduced pressure at 40 °C. The dried extracts were then analyzed for the amounts of free fatty acid (FFA), tocols, phytosterols and policosanols.

2.6. Free fatty acids

Rice bran samples were taken at 0, 2, 4, 6, 8 and 24 weeks of storage, and the FFA content was determined using a standard titration method (AOCS, 1989). The rice bran oils (1 g) extracted as described, were dissolved in a mixture (100 mL) of ethanol and diethyl ether (1:1, v/v) and titrated with a KOH/ethanol solution (0.01 N). The FFA content was calculated as oleic acid equivalent, expressed as percentage of total lipids.

2.7. Analysis of tocols

The tocopherol and tocotrienol contents in the rice bran samples treated in different procedures were determined after storage for 4 weeks according to the method reported by Ha et al. (2006) with some modification. For analysis of tocopherols and tocotrienols, the rice bran oils extracted were saponified. In brief, bran oil sample (1 g) and 5% pyrogallol solution in ethanol (4 mL) were placed in a round bottomed flask fitted with a reflux condenser and heated on a hot plate. When the mixture started boiling, the condenser was removed and 50% aqueous potassium hydroxide solution (1 mL) was added for saponification. The 20 mL of water and 30 mL of diethyl ether were added to the samples and transferred to a separating funnel. Extraction with diethyl ether (30 mL × 2) was repeated twice. The pooled diethyl ether layer was washed three times with 20 mL of distilled water, filtered through anhydrous sodium sulfate, and then evaporated at 40 °C. The remaining residue was diluted with 10 mL *n*-hexane, filtered through a Millipore 0.45 μm membrane, and then the filtrate was injected into an HPLC (PU-1580; JASCO) with a fluorescence detector (FP-1520; JASCO). A Lichrospher Si-60 column (250 × 4.6 mm, Merck Co.) was used and the mobile phase was *n*-hexane/2-propanol (99:1) at 1.0 mL/min. External standards of each tocopherols and tocotrienols were used for measuring the residual amounts of the tocols in the linear measuring range of 0.5–40 μg/mL.