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Extraction of brown rice extract and application in refined palm olein during accelerated storage

Supasit Chooklin

Department of Food Science and Technology, Faculty of Agro-Industry, Rajamangala University of Technology Srivijaya, Nakhon Sri Thamarat, 80240, Thailand

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Abstract

This research aimed to determine optimum condition of the phenolic compounds from brown rice extract from Sung Yod Phatthalung and to determine their applications as an antioxidant in refined palm olein. Brown rice was extracted with solvent concentration (40 - 80% ethanol, vv^{-1}), pH (2 - 8) and extraction time (5 - 60 min). Total phenolic content and antioxidant activities as determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) of brown rice extract were measured. The optimum conditions were as follows: ethanol concentration, 60% (vv^{-1}); pH 6 and extraction time, 25 min. Under the above-mentioned conditions, the experimental total phenolic content and DPPH of brown rice was 1.30 mg ferulic acid equivalents/g of dry sample and 86.55%, respectively. The oxidation properties (peroxide value, PV and thiobarbutitutic acid reactive substance, TBARS) of antioxidant in refined palm oil were compared with other synthetic antioxidants (butylated hydroxyanisole, BHA and butylated hydroxytoluene, BHT) at different concentration. It has been observed that the highest efficiency of BHT, followed by brown rice extract, BHA and control. The results revealed that the brown rice extract to be a potential antioxidant for stabilization of refined palm olein.

Introduction

Rice plays an important role as a main cereal food for people for a very long time since pre-historic time especially in Asia and other continents (Bhattacharee *et al*., 2002). Generally, people in the continent and in other regions consume it as a staple food for about more than 90% of world's population. In addition to acting as a main food, rice also is a plentiful source of a variety of biologically active compounds such as polyphenolic compounds (Zielinski and Kozlowska, 2000).

Phenolic compounds and other their related compounds have been a interested point of research and scientific arena as their significantly promising potential to reduce occurrence of important degenerative diseases e.g. coronary heart disease (CHD), tumor formation and cancer. Additionally, phenolic compounds also provide effective protection against other lipid oxidation associated damage as well. Previously, most artificially chemical food additives employed to retard oxidative reaction in foods and food products. Such additives particularly synthetic phenolic compounds are butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and propyl gallate (PG) and so forth. Currently, as a huge consumer's demand of natural or naturally like products leads to restricted or permitted dosage of artificially made food additives whereas nature based food additives is more accepted and preferred (Soong and Barlow, 2004).

Among many cereal grains, rice is unique in that it make up of special types of phenolic compounds like ferulic acid, *p*-coumaric acid and diferulate. As of the availably latest literature reviews, these constituents are not found in large amount in any fruits or vegetables (Adom and Liu, 2002). Furthermore, unpolished rice grains are composed of many other substantially health-promoting components e.g. dietary fibers, phytic acids, vitamin B complex, vitamin E and gamma aminobutyric acid (GABA) than commonly milled rice grains. These biologically functional constituents stayed at large in germ and bran layers which are taken out during polishing and milling (Champagne *et al*., 2004). Numerous literatures reported a finding out of ferulic acid and *p*-coumaric acid as a principal phenolic compound in unpolished rice as in free, conjugated and insoluble bound form. Up to the present time, there have no literally and distinctly tangible reports on successfully efficient extraction of phenolic components from brown rice. To achieve more eco-friendly procedures, less solvent utility, less time-consuming extraction as well as increment of the extracted output and enhanced quality of the products, a wide range of innovated extraction technique have been experimented for

*Corresponding author.
Email: *supasit_c@yahoo.co.th*
Tel: +66 8647 82887; Fax: +66 8647 82887

such purposes.

Among experimentally new promising and potential methods of choice for extraction of theses biologically active compounds, extraction by ultrasound radiation in the range of 20-100 KHz frequency is widely accepted and used due to its reproducibility, time efficiency, simpler maneuvering, less detrimental and toxic solvent usage, operation in lower temperature and lower energy consumption as well (Chemat *et al*., 2008). Possible mechanism of achieved extraction through sonication process is considered to be owing to its effects in cell wall rupture, consequently, it results in more efficient contact between solvent and as a result, attainably extractable compositions in attached cell wall (Vinatoru, 2001). Nonetheless, economical and commercial feasibility is limited because a multitude of scientific investigation still required until authentically high efficient method of extraction in practice is accomplished. A wide range of factors have been taken into account in the respect of extraction efficiency such as extraction procedure, type and quantity of solvent involved, solvent concentration, temperature and duration of extraction (Pinelo *et al*., 2005; Banik and Pandey, 2007; Silva *et al.*, 2007).

Consequently, to investigate in detail and to provide deeper knowledge related to this above-mentioned topic, we put an endeavor to examine possibly optimal conditions of brown rice extraction. Application of the crude extract in refined palm olein was also determined.

Materials and Methods

Materials

Rice samples and preparation

Sung Yod Phatthalung rice, which is one of the recommended rice cultivating in Southern Thailand, was obtained from Phatthalung Rice Research Center. These grains were milled to separate husk from brown rice. Then the brown rice was ground and pass through 0.25 mm sieve screen. The samples were analyzed proximate compositions according to the Association of Official Agricultural Chemists (AOAC, 2000).

Chemicals and reagents

BHA and BHT were obtained from Sigma Chemical Co. Ethanol and Folin-Ciocalteu's phenol reagent was obtained from Merk. Ferulic acid, 2,2-Diphenyl-1- picrylhydrazyl (DPPH) and 2-thiobarbituric acid were purchased from Fluka Analyticals. All chemicals and reagents were of A.R. grade. The refined edible palm olein used in this study was purchased from a local market in 2.5 L packing; the oil is free of any synthetic antioxidant.

Methods

Extraction of phenolic compounds

The dried ground of brown rice (5 g) was subjected to ultrasound – assisted using 100 ml of pH (2 – 8), ethanol concentration (40 - 80% vv^{-1}) and extraction time (5 – 60) min. The extracts were filtered through filter paper (whatman no.1). Then solvents were removed using a rotary evaporator at 60^{o}C under reduced pressure to remove solvent and then was lyophilized to dryness. The extracts obtained were used for the determination of total phenolic content and antioxidant activities. Total phenolic content and antioxidant activities of brown rice extracts were determined using the Folin-Ciocalteu reagent (Singleton *et al*., 1999) and DPPH radical scavenging activity (Brand-Williams *et al*., 1995).

Accelerated oxidation study

The brown rice extracts (0.02%, 0.1% and 0.5% ww^{-1}), BHT and BHA at a concentration of 0.02% ww^{-1} was added in to the refined palm olein. The oils samples were stirred to ensure that it was completely dissolved. All samples were kept in an oven at 60^{o}C and were removed from the oven after 0, 3, 6, 9 and 12 days for oxidative stability determination. A 25 ml of each sample was stored separately under the same conditions in a small, open glass container for performing peroxide value (PV) and thiobarbituric acid reactive substances (TBARS) determination. A sample of each treatment was removed after 0, 3, 6, 9 and 12 days, flushed with nitrogen, covered with aluminium foil and stored at -20^{o}C until further analysis was carried out.

Results and Discussion

Proximate composition

The proximate composition of Sung Yod brown rice variety from Phatthalung Province, which is one of the recommended rice cultivating in Southern Thailand, is evaluated. The results found that moisture, lipid, dietary fiber ash and protein was 10.54 ± 0.10%, 2.62 ± 0.04%, 4.25 ± 0.01%, 1.43 ± 0.21% and 10.12 ± 0.16%, respectively, which agreed with the results from Sawaddiwong *et al.* (2008) and Sompong *et al.* (2011). Protein influences the nutritional quality of rice. In this study the protein content was appreciably high (>7%) and thus are interesting for food products. The total carbohydrate content was higher than 70%, and thus it is considered as good source of energy.

Total phenolic content

The total phenolic content of brown rice extracts obtained by ultrasound-assisted extraction was determined by the Folin-Ciocalteu assay. The results of this colorimetric method, expressed as mg of ferulic acid/g dry sample. The crude phenolic compounds present in brown rice were extracted by ultrasound-assisted extraction method was 1.28 ± 0.05 mg FAE/g DW. The enhancement in extraction obtained by using an ultrasound is mainly attributed to the effects of acoustic cavitations produced in the solvent by the passage of an ultrasound wave. Ultrasound also exerts a mechanical effect, allowing greater penetration of solvent into the sample matrix, increasing the contact surface area between solid and liquid phase; as a result the solute quickly diffuses from solid phase to the solvent (Rostagno *et al*., 2003).

Effect of ethanol concentration on total phenolic compounds of brown rice

The ethanol-water mixture (40 - 80% vv^{-1}) was used to extract the total phenolic compounds at ultrasonic extraction time 25 min and ultrasonic extraction pH 6. The results are shown in Figure 1. When ethanol concentration increased from 40% to 60% vv^{-1}, the total phenolic content of the extracts increased from 1.12 to 1.31 mg FAE/g DW. When ethanol concentration reached 70% vv^{-1}, the total phenolic content in brown extracts decreased, and at a concentration of 80% vv^{-1}, the total phenolic content was 1.03 mg FAE/g DW. When 80% vv^{-1} ethanol was used to extract brown rice, some lipid components were also extracted, which may limit the extraction of phenolics in brown rice. Thus, the increasing ethanol concentration allowed higher yield of phenolic compounds in the extract, according to a better solubility of phenolic compounds in ethanol. However, at ethanol concentration higher than 70%, the phenolic content in the extracts were decreased due to the co-extraction of lipid to the extract. A similar effect was reported in the extraction of phenolic antioxidants from peanut skins (Neptoe *et al.*, 2005), wheat bran (Wang *et al*., 2008) and brown rice (Sawaddiwong *et al.*, 2008), heat-stabilized defatted rice bran (Onofre and Hettiarachchy, 2007) where phenolics extracted increased with an increase in the ethanol concentration to 60% vv^{-1}, and then decreased with the further increase in the ethanol percentage.

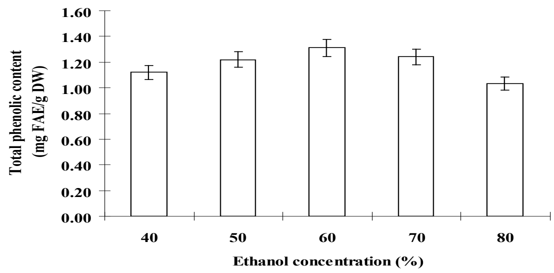

Figure 1. Effect of ethanol concentration on the total phenolics content from brown rice. The vertical bars represent the standard deviation (n = 3). Extraction conditions under sonication: pH, 6.0; extraction time, 25 min; extraction temperature, 30°C

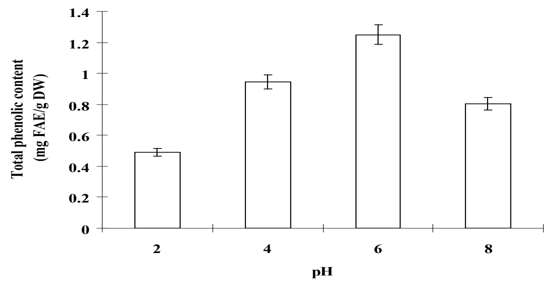

Figure 2. Effect of pH on the total phenolics content from brown rice. The vertical bars represent the standard deviation (n = 3). Extraction conditions under sonication: ethanol concentration, 60% vv^{-1}; extraction time, 25 min; extraction temperature, 30°C

Effect of pH on extraction of total phenolic compounds

The influence of pH (2 - 8) on total phenolic compound of brown rice extracts were studied at the conditions of 60% vv^{-1} ethanol as extraction solvent and extraction time 25 min. The effects of pH on the content of phenolics in brown rice extracts are shown in Figure 2. When extraction pH increased from 2 to 6, the total phenolic content of extracts increased from 0.49 to 1.25 mg FAE/g DW. When pH reached 8, the total phenolic content in brown rice decreased. Therefore, extraction of phenolic compounds appeared to be pH dependent. This could be explained by considering that the highest stability of phenolic compound in weak acid solvent (pH 6) but unstability of phenolic compound is found other pH. A similar effect of using solvent at various pH in the extraction of phenolic compounds from black glutinous rice (Tananuwong and Tewaruth, 2010) and various yam cultivars (Chen *et al*., 2008) had been reported. They found that total phenolic contents for black glutinous rice and all yams were the highest at pH 6.8 and 5, respectively. Thus, pH 6 was selected for further study to extract phenolic from brown rice.

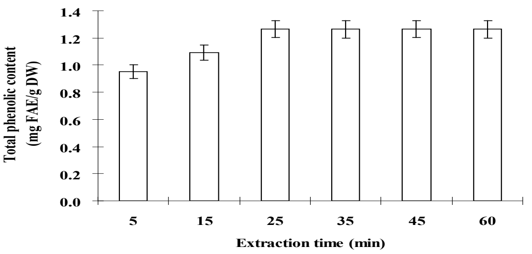

Figure 3. Effect of extraction time on the total phenolics content from brown rice. The vertical bars represent the standard deviation (n = 3). Extraction conditions under sonication: ethanol concentration, 60% vv^{-1}; pH, 6.0; extraction temperature, 30^{o}C.

Effect of extraction time on extraction of total phenolic compounds

The contents of total phenolic compounds extracted from brown rice at different times of sonication are presented in Figure 3. A significant increase of the total phenolic content was observed over the extraction time (5 – 60 min), and the total phenolic content reached a maximum of around 1.26 mg FAE/g DW. Mass transfer controls solvent extraction of any component from a plant matrix. When the solvent saturates with the extracted compound the concentration gradient becomes null and the phenomena stops (Rodrigues *et al*., 2008). In an ultrasound extraction of phenolic acid from brown rice mass transfer stops after 25 min and the process can be interrupted.

The results obtained from these experiments should that the optimum extraction conditions for maximum of total phenolic content from brown rice extracts obtained by the ultrasonic-assisted extraction conditions were 60% vv^{-1} ethanol concentration, pH 6.0 and 25 min extraction time for maximum total phenolic content (1.30 mg FAE/DW) and antioxidant activity (86.55%).

Accelerated oxidation studies

Peroxide value is often used as a measurement of this oxidative deterioration of oil, fat and fatty acids, hence, efficacy of an antioxidant extract to prevent the onset of oxidation could be predicated by the rate of formation of these primary oxidation products. The effect of the addition of antioxidants on PV is shown in Figure 4. From the results obtained, PV of refined palm olein samples with additives was in the range 9.58 ± 14.54 meq/kg for stabilized samples after storage upto 12 days, while maximum of PV for control (without antioxidant added) sample was 16.16 meq/kg. The addition of antioxidant decreased PV of refined palm olein. At all stages, highest PV was observed for control sample followed by BHA, brown rice extract and BHT, respectively (Figure 4a). Brown rice, at all the concentrations, controlled peroxide

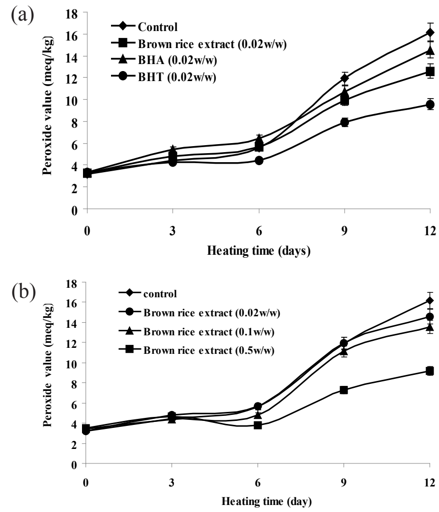

Figure 4. Effects on peroxide value of refined palm olein at 60^{o}C of addition of antioxidants-butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and brown rice extract-at different concentrations: (a) 0.02% ww^{-1}; and (b) 0.02–0.5% ww^{-1}.

value appreciably; revealing good antioxidant efficacy in stabilization of oil. A regular increase in PV as a function of storage time was observed for all the samples at all intervals. In Figure 4b, increasing brown rice extract concentration significantly reduced oil oxidation. Moreover, Maximum PV content of refined palm olein with 0.5% ww^{-1} brown rice extract was 9.19 meq/kg, which less than the other extract. This experiment indicated that 0.5% ww^{-1} brown rice extract was the most effective antioxidant, which were similar results to reported by Iqbal and Bhanger (2007) studied the stabilization of sunflower oil (SFO) by garlic extract during accelerated storage. Methanolic extract of garlic at three different concentrations, i.e. 250 (SFO-250), 500 (SFO-500) and 1000 ppm (SFO-1000) were added to preheated refined sunflower oil. It was reported that PV of SFO containing antioxidants was lower than that of SFO without antioxidants. The highest PV was observed for control sample followed by SFO-250, SFO-500, SFO-BHA, SFO-1000 and SFO-BHT, respectively.

TBARS measures the formation of secondary oxidation products i.e. aldehydes or carbonyls, which may contribute to off-flavour of oxidized oils. TBARS for all the samples were determined upto 12 days of storage under accelerated conditions (Figure 5). From the results obtained, TBARS of refined palm olein samples with additives was in the range 2.16 ± 2.65 mg MDA/kg oil for stabilized samples after storage upto 12 days, while maximum of TBARS for control (without antioxidant added) sample was 2.84 mg MDA/kg oil. The addition of antioxidant decreased TBARS of refined palm olein. At all stages, highest TBARS was observed for control sample followed

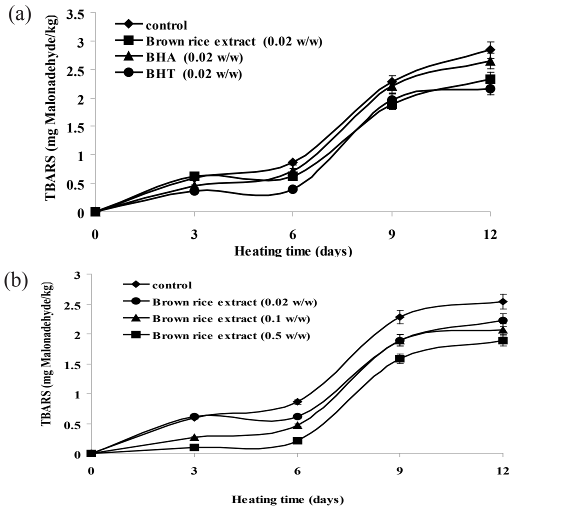

Figure 5. Effects on thiobarbituric acid reactive substances (TBARS) of refined palm olein at 60°C of addition of antioxidants at different concentrations: (a) 0.02% ww^{-1} and (b) 0.02–0.5% ww^{-1}.

by BHA, brown rice extract and BHT, respectively (Figure 5a). In Figure 5b, TBARS of refined palm olein samples with additives were in the range of 1.89 – 2.23 mg MDA/kg oil for stabilized samples and 2.54 mg MDA/kg oil for control sample after 12 days of storage. Brown rice extract inhibited the formation of TBARS at all concentrations. Tananuwong and Tewaruth (2010) reported that the addition of dried black glutinous rice crude extract at 500 and 1000 mg kg^{-1} (oil weight basis) could retard an increase in TBARS. Higher concentration of the extract showed greater antioxidant activities.

Conclusion

In order to achieve the maximum extraction performance by ultrasound-assisted extraction, a 60% (vv^{-1}) ethanol concentration extraction, pH 6 and 25 min of extraction time as optimal operating conditions. The addition of the antioxidants brown rice extract, BHA and BHT to refined palm olein resulted in the retardation of oxidative deterioration. The antioxidant activities of the brown rice extract was higher there those of control, BHA but lower than BHT. Therefore, the brown rice from Sung Yod Phatthalung can be condisered as a potential antioxidant source of natural original.

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References

Adom, K.K. and Liu, R.H. 2002. Antioxidant activity of grains. Journal of Agricultural and Food Chemistry 50: 6182-6187.

Association of Official Agricultural Chemists (AOAC). 2000. Association of Official Analytical Chemists: Official Methods of Analysis. 16th ed. AOAC. Washington, DC, USA.

Banik, R.M. and Pandey, D.K. 2007. Optimizing conditions for oleanolic acid extraction from Lantana camara roots using response surface methodology. Industrial Crops and Products 27: 241-248.

Bhattacharee, P., Singhal, R.S. and Kulkarni, P.R. 2002. Basmati rice: A review. International Journal of Food Science and Technology 37(1): 1-12.

Brand-Williams, W., Cuvelier, M.E. and Berset, C. 1995. Use of a free radical method to evaluate antioxidant activity. LWT-Food Science and Technology 28(1): 25-30.

Champagne, E.T., Wood, D.F., Juliano, B.O. and Bechtel, D.B. 2004. The rice grain and its gross composition. In: Champagne, E.T. (Ed.), Rice. American Association of Cereal Chemistry, pp.77-107. USA: St. Paul.

Chemat, F., Tomao, V. and Virot. M. 2008. Ultrasound-assisted extraction in food analysis (edited by Semith Otles), Handbook of food analysis instruments, pp.85-103. USA: CRC press.

Chen, Y.T., Kao, W.T. and Lin, K.W. 2008. Effects of pH on the total phenolic compound, antioxidative ability and the stability of dioscorin of various yam cultivars. Food Chemistry 107: 250-257.

Iqbal, S. and Bhanger, M.I. 2007. Stabilization of sunflower oil by garluc extract during accerelated storage. Food Chemistry 100: 246-254.

Neptoe, V., Grosso, N.R. and Guzman, C.A. 2005. Optimization of extraction of phenolic antioxidants from peanut skins. Journal of the Science of Food and Agriculture 85: 33-38.

Onofre, F.O. and Hettiarachchy. 2007. Extraction, quantification, and characterization of phenolics extracted with the aid of sonication from rice bran. Cereal Chemistry 84(4): 337 – 342.

Pinelo, M., Rubilar, M., Jerez, M., Sineiro, J. and Nunez, M.J. 2005. Effect of solvent, temperature, and solvent-to-solid ratio on the total phenolic content and antiradical activity of extracts from different components of grape pomace. Journal of Agricultural and Food Chemistry 53: 2111-2117.

Rodrigues, S., Pinto, G.A.S. and Fernandes, F.A.N. 2008. Optimization of ultrasound extraction of phenolic compounds from coconut (*Cocos nucifera*) shell powder by response surface methodology. Ultrasonics Sonochemistry 15: 95 – 100.

Rostagno, M.A., Palma, M. and Barroso, C.G. 2003. Ultrasound-assisted extraction of soy isoflavones. Journal of Chromatography A 1012: 119-128.

Sawaddiwong, R., Jongjareonrak, A. and Benjakul, S. 2008. Phenolic content and antioxidant activity of germinated brown rice as affected by germination temperature and extraction solvent. KMITL Science Journal 8(2): 54-58.

Sompong, R., Siebenhandl-Ehn, S., Linsgerger-Martin, G. and Berghofer, E. 2011. Physicochemical and antioxidative properties of red and black rice varieties from Thailand, China and Sri Lanka. Food Chemistry 124: 132-140.

Silva, E.M., Rogez, H. and Larondelle, Y. 2007. Optimization of extraction of phenolics from Ingaedulis leaves using response surface methodology. Separation and Purification Technology 55: 381-387.

Singleton, V.L., Orthofer, R. and Lamuela-Raventos, R.M. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Method in Enzymology 299: 152-178.

Soong, Y.Y. and Barlow, P.J. 2004. Antioxidant activity and phenolic content of selected fruit seeds. Food Chemistry 88(3): 411-417.

Tananuwong, K. and Tewaruth, W. 2010. Extraction and application of antioxidants from black glutinous rice. Food Science and Technology 43: 476–481.

Vinatoru, M. 2001. An overview of the ultrasonically assisted extraction of bioactive principles from herbs. Ultrasonics Sonochemistry 8(3): 303 – 313.

Wang, J., Sun, B. Cao, Y. Tian, Y. and Li, X. 2008. Optimisation of ultrasound-assiated extraction of phenolic compounds from wheat bran. Food Chemistry 106: 804-810.

Zielinski, H. and Kozlowska. H. 2000. Antioxidant activity and total phenolics in selected cereal grains and their different morphological fractions. Journal of Agricultural and Food Chemistry 48: 2008-2016.