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ANTIOXIDANT PROPERTIES OF RICE BRAN OIL FROM DIFFERENT VARIETIES EXTRACTED BY SOLVENT EXTRACTION METHODS

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Abstract

Antioxidant properties of rice bran oil from different rice bran varieties; Rice Bran-Bario (RB-Bario), Rice bran-Lowland (RB-Low) and Rice Bran- Upland rice (RB-Up), collected from different cultural plots, were assessed. Measurement of antioxidant properties was evaluated by TPC (total phenolic content), DPPH scavenging activities and reducing power of extracts.. The study shows that antioxidant efficacy of rice bran was found the highest in RB-Up, followed by RB- Low and RB-Bario. The antioxidant properties were related to the rice bran origin and water irrigation demand by particular variety. RB –Up has a unique plantation condition which takes least amount of water retention which contribute to the highest antioxidant activity. Extraction solvents used shows that Upland (16.15%) and Lowland (16.16%) yielded the highest amount in conserving the crude fat oil in rice bran extract compared to Bario.

Keywords: Antioxidant, extraction, rice bran oil

1.0 INTRODUCTION

Rice bran is a by-product from rice milling process. It is mostly used as animal feeds because of the quick hydrolysis of oil into free fatty acids. Chemical and nutritional properties of bran produced have been assessed for their potential in both industrial and domestic application. Biochemical instability of rice bran occurs immediately after milling. Free fatty acid formation has been reported as the time of storage extended without stabilization process done on the rice bran [1]. Consequently, this formation of unnecessary acid affects the composition of the rice bran. Rice bran is rich in natural antioxidants such as tocopherols, tocotrienols, oryzanol and phenolic compound. These bioavailable materials have shown their potential as antioxidant and anti-cancer agent which protect body from free radical damage [2]. Antioxidant and other composition in the rice bran differ according to their variety and species [3]. Recently, rice bran oil had been produced as cooking oil. It has been reported that rice bran oil could lower aortic cholesterol ester and lower the cholesterols level similarly as oat bran [4]. There were also high value products potentially produced from rice bran for food and pharmaceutical industries [5]. Solvent is one of the factors that affected the extraction condition of rice bran oil. The increasing of vitamin E composition of rice bran oil from extraction using hexane has been reported with the temperature range 40-60 °C compared to Isopropanol [6]. While, when ethanol was used, the amounts of extracts obtained were from 4-12 times higher than extracts achieved with hexane [7]. Thus,

the foundation idea for this work was to investigate the antioxidant properties of the rice bran oil decanted from different type of solvents from different rice bran varieties.

2.0 EXPERIMENTAL

2.1 Material

2,2-diphenyl-1-picrylhydrazyl (DPPH), Follin Ciocalteu reagent, sodium carbonate from Sigma-Aldrich, ethanol, methanol, hexane, potassium ferricyanide and tricholoroacetic acid from Merck. Gallic acid from Fluka.

2.2 Methods

2.2.1 Rice Bran Preparation

Different varieties of rice bran were selected from local suppliers; Bario (RB-Bario), Lowland (RB-Low) and Upland (RB-Up) rice bran. Fresh rice bran was screened to pass through a 710 µm aperture sieve to remove broken grains, hull fragments, paddy kernels and foreign materials. Rice bran will be stored in sealed polyethylene bags in refrigerator at ±4 °C to control the growth of rice bran free fatty acid (FFA).

2.2.2 Stabilization of Rice Bran

Stabilization of rice bran was carried out according to method of Malekian *et al*. [8] with some modification. The stabilization of rice bran was performed using a microwave oven with 550W output power. 100 grams of each sample was heated in preheated oven for 3 minutes at 120°C or ±550W and it was cooled down at room temperature overnight. This procedure was repeated three times to make sure the stabilization of the rice bran. All samples were placed in chiller at 4°C until analysis.

2.2.3 Soxtherm Lipid Extraction

Rice bran was extracted by weighing samples (10 g) into cellulose thimbles (22 mm x 80 mm, What-man). The timbers were covered with cotton wool, placed in the pre-dried Gerhardt's extraction flask (250 ml) with additional of Gerhardt's boiling stones as catalyzer. The flask and sample then connected to the Soxtherm system for extraction. 140 ml of N-hexane were used to extract oil by refluxing on an electric mantle at solvent boiling point for 4 h. The extracts were dried in the oven for an hour and proceed another one hour in desiccator under vacuum condition. Total crude fat content was determined gravimetrically and samples were stored.

2.3 Analysis of the Rice Bran Oil

2.3.1 Total Phenolic Content of Rice Bran Oil

The concentration of phenolic compound in the extracts was determined using spectrophotometric methods [9] with some modification. Solvents of extraction were used in the analysis as a diluent. The mixture was prepared by mixing 1.0ml of diluted extract, 5.0ml of Follin Ciocalteu's reagent and 15ml. 7.5% $CacO_3$. The samples were incubates for 40 to 45 minutes before the absorbance taken. The absorbance used was 700nm. All the result then was read as (mg/ml) from the calibration curve (ppm of GA/g of extract).

2.3.2 DPPH Scavenging Activity of Rice Bran Oil

The antioxidant activity of sample extracts determined following a reported method [9] with modification. Solution of DPPH was prepared with methanol as a diluent. 0.1mL of extract was added and concentration of the DPPH was settled. The mixture was shaken and being incubated under dark condition for almost 45 minutes. The decrease in absorbance was measured at 517 nm against blank by using spectrophotometer. The antioxidant activity then calculated as equations below to express antioxidant activity.

= (A 517nm, control - A 517 nm,sample) / A 517 nm,control x 100

2.3.3 Reducing Power of Rice Bran Oil

About (2.5, 5, 10, 20, 40 mg/ml) of extract were mixed with 2.5ml, 2.0 M Phosphate buffer which concentration adjusted (pH 6.6). Mixture of sample then mixed with 1 % potassium ferricyanide (2.5ml) and was incubated for 30 minutes at room temperature. Then 10% tricholoacetic acids were added and centrifuged the mixture for 10 minutes. The upper layer was collected and mixed with distilled water as well as 1% ferric chloride (0.5ml). The mixture was measured by UV-Visible at 700nm

3.0 RESULTS AND DISCUSSION

3.1 Crude Fat Analysis

The extractability of rice bran oil using soxhlet methods has been reported in which ranging between 18-20% [10]. Figure 1, shows the crude fat of rice bran oil from different variety (Bario, Lowland and Upland) being extracted using three type of solvents (Ethanol, Hexane and Methanol). RB-Bario exhibits higher yields of oil decanted in 3 hours of extraction time with ±12.72% followed by RB-Up and RB-Low types with using hexane as solvent. The total oil decanted shows slightly low than previous study done by [10] which report that 14% of lipid was

extracted from 25g of rice bran. The upland and lowland variety exhibits higher crude oil decanted compared to Bario in 4 hours times of extraction; upland (16.15%) and lowland (16.16%). This amount of extract agreed the previous study by [13]. Based on the ANOVA analysis of data there are significant result obtained between the time of extraction with $p<0.05$.

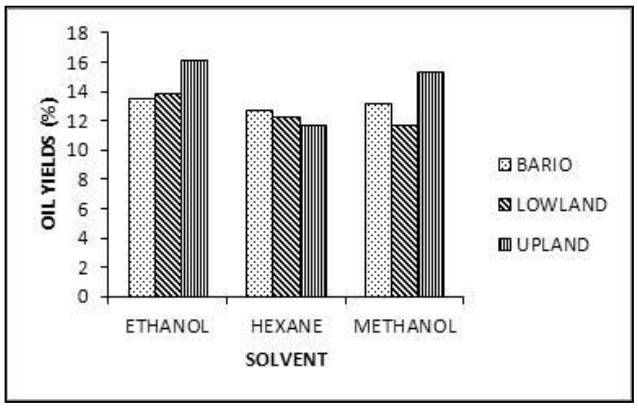

Figure 1 Oil yield obtained from different type of rice bran and solvents using Soxtherm extraction method

3.2 Total Phenolic Content of Rice Bran Oil

The total phenolic content (TPC) was determined with Follin-Ciocalteu method with modification and result expressed as gallic acid. As shown in Table 1, the phenolic content was different with three various solvent, the higher phenolic content was determined in RB-Low with Ethanol as solvent of extraction (±509.0 ppm), while RB-Bario type give the higher measurement with hexane as solvent for ex-traction (±447.3 ppm). The phenolic compound may contribute directly to antioxidant activity [11]. Then, their different content of phenol content in this type of rice bran oil may affect by their variety and ecological planted. The water retention and highland rice to be known as factors of differences

Table 1 Total phenolic content of three varieties of rice bran oil extracted using different solvent

Type of Rice Bran	Total Phenol content (Gallic acid eq.) (ppm/g of bran)		
	Ethanol	Hexane	Methanol
RB-Bario	136.9±0.56	447.3±10.4	61.4±3.90
RB-Low	509.0±0.10	173.1±0.73	49.4±1.10
RB-Up	225.7±1.73	189.9±0.12	62.1±0.31

3.3 Radical scavenging activity of rice bran oil

Figure 2 shows the DPPH activities of different type of rice bran oil extracted using different solvent, the. Lowland type exhibit high antioxidant activities in almost all solvent extract with ethanol (92.96%), followed by Bario type with methanol solvent (84.25%) and upland rice bran with ethanol solvent extraction (80.13%). General tendency of extract Methanol > Ethanol > Hexane found for DPPH scavenging activities at ($p<0.05$).

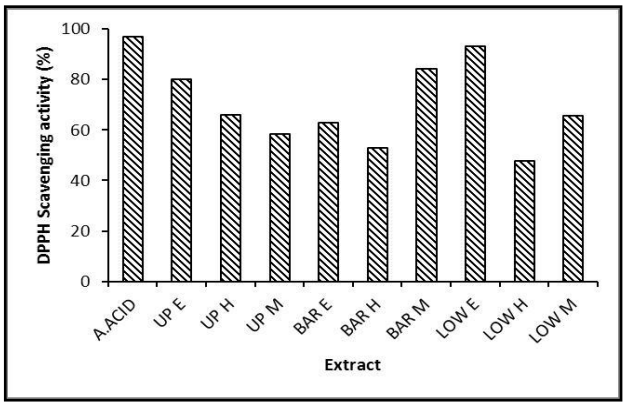

Figure 2 DPPH activities of three varieties of rice bran oil extracted using different solvent

Based on previous research, methanol extract had superior comparatively amount of antioxidant compositions because of the possibility of more polar phenolic compounds and lipids contains in the extract [10]. The different of activities showed the ionic transfer-ring within extraction process in different solvents. Rice bran extracts have a high tendency to donate hydrogen atoms, the results of DPPH free radical-scavenging might be due to hydrogen donation ability [12].

3.4 Reducing Power Determination

Figure 1, 2, 3, 4 and 5 shows the reducing power of rice bran oil extracted by different type of solvents. As shown, the reducing power of rice bran oil increased with increasing concentration. Upland rice bran oil exhibit the higher reducing power with methanol solvent at 700 nm were 3.558 compared to Bario (3.263) and Lowland (3.504). Unstable relation between phenolic content and scavenging activities might due to prolonged time taken, this is because rice bran were known to be hydrolyzed into free fatty acids in storage. The highest values of absorption for reducing power in methanolic the rice bran oil extracts were observed in Upland rice variety ($p<0.05$). Result obtained had demonstrated that com-pounds of rice bran oil extract were electron donor and can terminate radical chain reaction [2].

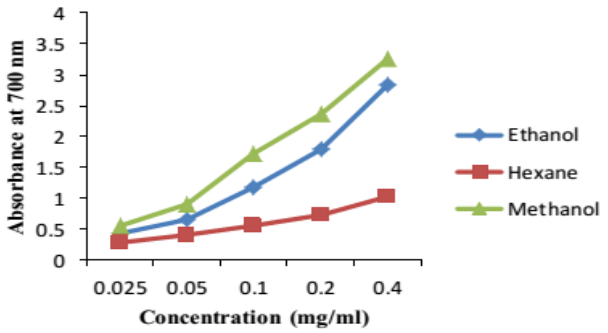

Figure 3 Reducing capacity of Bario bran oil extracts

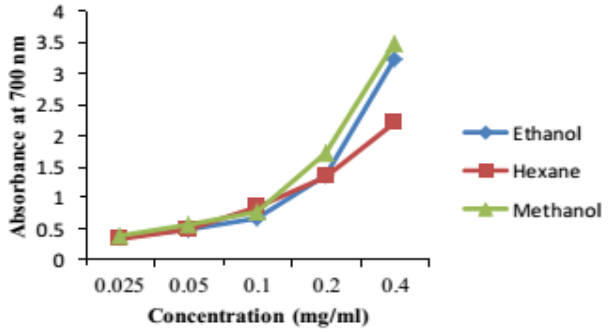

Figure 4 Reducing capacity Lowland bran oil extracts

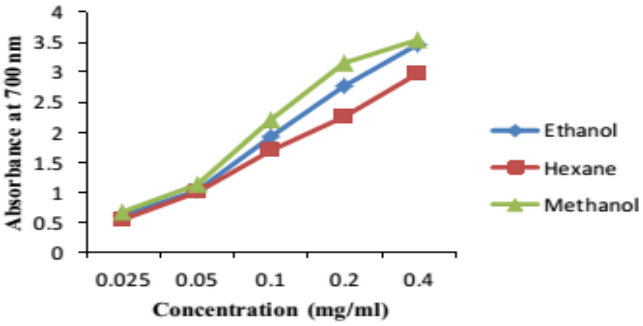

Figure 5 Reducing capacity Upland bran oil extracts

4.0 CONCLUSION

Extraction of antioxidant and phenolic content were varied with type of rice bran (Bario, Lowland and Upland) and solvent (Ethanol, Hexane and Methanol) extraction. Upland rice bran oil with Methanol produced a significantly greater yield and reducing power than other two varieties. The strong antioxidative activity of rice bran oil extracts might due to the existence of tocopherols and tocotrienols compound in the extracts

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References

[1] Lakkakula, N. R., Lima, M. and Walker, T. 2004. Rice Bran Stabilization And Rice Bran Oil Extraction Using Ohmic Heating. *Journal of Bioresource Technology*. 92: 157-161.

[2] Nam, S. H., Choi, S. P., Kang, M. Y., Kozukue, N. and Friedman, M. 2005. Antioxidative, Antimutagenic And Anti-Carcinogenic Activities Of Rice Bran Extracts In Chemical And Cell Assay. *Journal of Agricultural and Food Chemistry*. 53: 816-822.

[3] Lloyd, B. J., Sienbenmorgen, T. J. and Beers, K. W. 2000. Effects of Commercial Processing on Antioxidants in Rice Bran. *Journal of Cereal Chemistry*. 77 (5): 551-555.

[4] Hegsted, M. and Windhauser, M. M. 1993. Reducing Human Heart Disease Sick With Rice Bran. *Louisiana Agriculture*. 36(3): 22-24.

[5] Hangrove, K. L. 1994. Processing And Utilization Of Rice Bran In The United States. In Rice Science and Technology: Marshall, W.E., Wadsworth, J. I. Eds. Dekker: New York.

[6] Hu, W., Wells, J. H., Shin, T. S., and Godber, J. S. 1996. Comparison Of Isopropanol And Hexane For Extraction Of Vitamin E And Oryzanols From Stabilized Rice Bran. *Journal of the American Oil Chemists Society*. 73: 1653-1656.

[7] Ferriera-Dias, S., Valente, D. G., and Abreu, J. M. F. 2003. Comparison Between Ethanol And Hexane For Oil Ex-Traction From Quercus Suber L. Fruits. Institute Superior de Agronomia,Lisbon. 54: 378-383.

[8] Malekian, F. Rao, R. M., Prinyawiwatkul, W., Marshall, W. E., Windhauser, M., and Ahmedna, M. 2000. Lipase And Lipoxygenase Activity, Functionality, And Nutrient Losses In Rice Bran During Storage. Bulletin No. 870. Baton Rouge: LSU Agricultural center, Louisiana Agricultural Experiment Station. 1-68.

[9] Singleton. V. L., and Rossi, J. A. 1965. Colorimetry Of Total Phenolic With Phophomolybdic-Phosphotungestic Acid Reagent. *American Journal of Enology and Viticulture*. 16: 144-158.

[10] Iqbal, S., bhanger, M. I. and Anwar, F. 2005. Antioxidant Properties And Components Of Some Commercially Available Varieties Of Rice Bran In Pakistan. *Journal of Food Chemistry*. 93: 265-272.

[11] Roger, E. J., Rice, S. M., Nicolosi, R. J., Carpenter, D. R., McClelland, C. A. and Romanczyk Jr., L. J. 1993. Identification And Quantitation Of Gamma Oryzanol Components And Simultaneous Assessment Of Tocopherols In Rice Bran Oil. *Journal of The American oil Chemists' Society*. 70(3): 301-307.

[12] Shimada, K., Fujikawa, K., Yahara, K., and Nagamura, T. 1992. Antioxidative Properties Of Xanthan On The Autoxidation Of Soybean Oil In Cyclodextrin Emulsion. *Journal of Agricultural and Food Chemistry*. 40: 945-948.

[13] Xu, Z. and Godber, J. S. 1999. Purification And Identification Of Component Of Gamma Oryzanol In Rice Bran Oil. *Journal of Agriculture Food Chemistry*. 47: 2724-27.