

Ladda Wattanasiritham 2015: Isolation and Characterization of Antioxidative Peptides from Khao Dawk Mali (KDML) 105 Rice Bran Protein Hydrolysates. Doctor of Philosophy (Food Science), Major Field: Food Science, Department of Food Science and Technology. Thesis Advisor: Associate Professor Chockchai Theerakulkait, Ph.D. 130 pages.

Antioxidant is very important in inhibiting oxidation process in food and biological systems. Antioxidative peptide is considered as antioxidant and can be produced through protein hydrolysis. This study was to prepare rice bran protein hydrolysates with antioxidant activity by enzymatic hydrolysis and the antioxidative peptides in the hydrolysate were isolated and identified.

Rice bran protein extracted from defatted rice bran using alkali extraction and isoelectric precipitation (AE-RBP) was prepared. AE-RBP was hydrolyzed with Alcalase 2.4L or papain and antioxidant activity of the hydrolysates were determined by 2,2-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity and Ferric Reducing Ability Power (FRAP) assay at hydrolysis time of 0, 30, 60, 90 and 120 min. The DPPH free radicals scavenging activity and FRAP value of the alcalase AE-RBP hydrolysates (AE-RBPHs) were 32.1 - 35.5 % and 951 - 1,018 $\mu\text{mol FeSO}_4/\text{ml}$ of hydrolysate, respectively and were not significantly different ($p > 0.05$) at different hydrolysis times. AE-RBP was freeze-dried (FD-AE-RBP) and it was found that freeze-drying did not effect on antioxidant activity of AE-RBP. DPPH radical scavenging activity and FRAP of the FD-AE-RBP was 41.9% and 92.6 $\mu\text{mol FeSO}_4/\text{g}$ protein, respectively. FD-AE-RBP was also combined with butyrate hydroxylanisol (BHA) or α -tocopherol and their antioxidant activities were evaluated.

Albumin, globulin, glutelin and prolamin were extracted from defatted KDML 105 rice bran based on the difference in their solubility. These protein fractions (native form and that denatured by dithiothreitol) were hydrolyzed with papain and trypsin at 37 °C for 3 h. The antioxidant activity of them and their hydrolysates were evaluated by Oxygen Radical Absorbance Capacity (ORAC). Among these protein fractions and their hydrolysates, trypsin-hydrolyzed denatured albumin hydrolysate exhibited the highest antioxidant activity and its ORAC value was 4.07 $\mu\text{mol Trolox eq./mg}$ protein. The trypsin-hydrolyzed denatured albumin hydrolysate was separated by RP-HPLC. The peptide fractions that showed high antioxidant activity were identified by UPLC MS/MS. The main MW of peptides were in the range of 800-1,500 Da and consisted of 6 to 20 amino acid residues. The peptide fractions that observed to be the highest antioxidant activity demonstrated typical characteristics of well-known antioxidative peptides with hydrophobic and aromatic amino acid residues. The amino acid sequence of copper ion (Cu^{2+})-chelating peptides were also demonstrated. The peptides with molecular weight of approximately 800-1,500 Da had high ability to donate an electron to free radical.

Student's signature

Thesis Advisor's signature

____ / ____ / ____

