

ผลของสารสกัดโปรตีนจากรำข้าวต่อการยับยั้งการเกิดสีน้ำตาลในผักและผลไม้บด

Effect of Rice Bran Protein Extract on Browning Inhibition in Vegetable and Fruit Puree

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บทคัดย่อ

การศึกษามลของสารสกัดโปรตีนจากรำข้าวต่อการยับยั้งการเกิดสีน้ำตาลเนื่องจากเอนไซม์ในผักและผลไม้บด โดยนำรำข้าวที่สกัดไขมันมาสกัดโปรตีนด้วยด่าง และตกตะกอนที่ pH นำมาบดผสมกับมันฝรั่ง กัวยหอม หรือแอปเปิล แล้ววัดค่าสี L*, a* และ b* พบว่าค่าการเกิดสีน้ำตาลของมันฝรั่ง กัวยหอม และแอปเปิล บดผสมกับสารสกัดโปรตีนจากรำข้าวที่ระยะเวลาการเก็บ 6 ชั่วโมง มีค่าเท่ากับ 27.16, 13.29 และ 18.22 ตามลำดับ ซึ่งต่ำกว่าตัวอย่างที่บดผสมกับน้ำกลั่น ($p \leq 0.05$) ค่า L* ของมันฝรั่ง กัวยหอม และแอปเปิล บดผสมกับสารสกัดโปรตีนจากรำข้าวที่ระยะเวลาการเก็บ 6 ชั่วโมง มีค่า 52.79, 52.68 และ 37.19 ตามลำดับ ซึ่งสูงกว่าตัวอย่างที่บดผสมกับน้ำกลั่น ($p \leq 0.05$) และพบว่าที่ระยะเวลาการเก็บ 6 ชั่วโมง สารสกัดโปรตีนจากรำข้าวมีค่าร้อยละการยับยั้งการเกิดสีน้ำตาลในมันฝรั่งบด สูงกว่ากัวยหอม และแอปเปิลบด ซึ่งมีค่าร้อยละ 46.68, 16.76 และ 10.24 ตามลำดับ ($p \leq 0.05$) ดังนั้นสารสกัดโปรตีนจากรำข้าวสามารถยับยั้งการเกิดสีน้ำตาลในผักและผลไม้บดโดยเฉพาะในมันฝรั่งบด

ABSTRACT

Effect of Rice bran protein extract (RBPE) on browning inhibition in vegetable and fruit puree was investigated. Protein from defatted rice bran was prepared by alkaline extraction and pH precipitation. The obtained RBPE was blended with potato, banana, or apple and their color values (L*, a* and b*) values were measured. It was found that browning values of potato, banana, and apple puree treated with RBPE at 6 hrs storage were 27.16, 13.29 and 18.22, respectively, which were lower than those treated with distilled water (DW) ($p \leq 0.05$). The L* values of potato, banana and apple puree treated with RBPE at 6 hrs storage were 52.79, 52.68 and 37.19, respectively, which were higher than those treated with DW ($p \leq 0.05$). At 6 hrs storage, RBPE showed higher % browning inhibition in potato puree than banana and apple puree with the values of 46.68, 16.76 and 10.24 %, respectively ($p \leq 0.05$). Therefore, RBPE could inhibit browning in vegetable and fruit puree, especially potato puree.

Key Words: browning, rice bran protein, potato, banana, apple

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INTRODUCTION

Browning has a major impact on many vegetables and fruits quality. It might also produce undesirable changes in flavor, texture and nutritional values of the product during handling, processing and storage, resulting in shorter shelf life and reduce market value (Fu *et al.*, 2005; Jang *et al.*, 2002). There are several methods to inhibit browning such as applying chelators, reducing agents, acidulants, PPO inhibitor or avoiding exposure to oxygen (Zhu *et al.*, 2007; Buta and Moline, 2001). Among the compounds that inhibit browning, sulfite is widely used for prevention of the browning reaction. However, they could create undesirable flavor and a problem for human health (Zhu *et al.*, 2007; McEvily *et al.*, 1992) and they have been banned by the Food and Drug Administration for most fresh food application (FDA, 1986). In addition, there is an increasing consumer demand for substituting synthetic compounds with natural substances (Jang *et al.*, 2002). Therefore, the natural antibrowning agents have been widely investigated.

Rice is one of the important staple food for world's population. Rice are produced annually in 2011 about 690 million metric tons (IRRI, 2012). This huge amount of production results in commensurate amount of rice by-products (Sereewatthanawut *et al.*, 2008). Rice bran is the major by-product generated about 3-8% of rice grain during milling process (Amissah *et al.*, 2003). It is a source of protein, mineral, unsaturated fat, vitamin and dietary fiber (Abdul-Hamid and Luan, 2000). Due to its high content of hypoallergenic protein (Helm and Burks, 1996), rice bran has a high potential to be used as food ingredients in food applications.

There were several studies about inhibition of browning in vegetable and fruit by proteins, peptides and amino acids such as proteins, protein hydrolysates and amino acids from milk (Kahn, 1995), peptide from honey (Oszmianski and Lee, 1990; Ates, 2001), hen egg white lysozyme (Li *et al.*, 2006), wheat bran albumin (Campas-Ríos *et al.*, 2012), wheat bran globulin (Ortiz-Estrada *et al.*, 2012), sericin (Thongsook and Tiyaboonchai, 2011), peptides from onion (Gnangui *et al.*, 2010); however, protein from rice bran has not been investigated. Therefore, the objective of this research was to study the effect of rice bran protein extract on browning inhibition in vegetable and fruit puree.

MATERIALS AND METHODS

Materials

Potato (*Solanum tuberosum* L.), banana (*Musa* (AAA Group) 'Gros Michel'), apple (*Malus pumila* cv. Fuji) were purchased from local market in Bangkok, Thailand. Full-fatted rice bran from the aromatic rice (*Oryza sativa* L. cv. Khao Dawk Mali 105) was obtained from Patum Rice Mill and Granary Public Co. Ltd., Thailand. All chemical reagents were analytical grade.

Preparation of Defatted Rice Bran

Full-fatted rice bran was sieved through a 50 mesh screen and extracted twice with hexane at a ratio of rice bran to hexane at 1:3 (w/v) for 30 min, centrifuged at 4,000xg at 25 °C for 30 min. The defatted rice bran was air-dried overnight under hood, ground and sieved through a 50 mesh screen and stored in aluminium foil bag at -20 °C until used (Wang *et al.*, 1999).

Preparation of Rice Bran Protein Extract

Rice bran protein was extracted by dispersing defatted rice bran in distilled water (DW) at a ratio of rice bran to DW at 1:4 (w/v). The pH was adjusted and maintained at 9.5 with 1.0 N NaOH during stirred with an overhead stirrer at 500 rpm at room temperature for 45 min, then centrifuged at 15,200xg at 25 °C for 30 min. The supernatant was adjusted to pH 4.5 with 1.0 N HCl and centrifuged at the same conditions. The obtained precipitate was rice bran protein extract (RBPE) (Gnanasambandam and Hettiarachchy, 1995).

Browning Inhibition in Potato, Banana and Apple Puree by RBPE

RBPE was dispersed in DW at 5% (w/v) and blended with potato, banana or apple at a ratio of potato, banana or apple to RBPE at 2:1 (w/v) for 20 seconds. DW was used as control. The color (L^* , a^* , b^*) values of the samples were measured by a Spectrophotometer (CM-3500D, Minolta) at 0, 1, 2, 3, 4, 5 and 6 hr(s) during storage at room temperature. Browning value was calculated from $(\Delta L^*/L_0^*) \times 100$; when ΔL^* was the change of L^* at storage time and L_0^* was the initial L^* measurement (Labuza *et al.*, 1990) and % browning inhibition was calculated from $((\Delta E^*_{\text{control}} - \Delta E^*_{\text{inhibitor}}) \times 100 / \Delta E^*_{\text{control}})$; when ΔE^* was calculated from $((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)^{1/2}$ (Girelli *et al.*, 2004).

Statistical analysis

The experiment was performed in three replications. Statistically significant difference was assessed by one-way analysis of variance. Significant difference ($p \leq 0.05$) among treatments were detected by using Duncan's multiple range tests.

RESULTS AND DISCUSSION

Browning Inhibition in Potato, Banana and Apple Puree by RBPE

Browning values of potato, banana and apple puree treated with RBPE after storage at room temperature for 6 hrs compared to DW are shown in Figure 1. Browning values of potato and apple puree

treated with RBPE were lower than those treated with DW through the storage time ($p \leq 0.05$) and browning value of banana treated with RBPE were lower than those treated with DW at 2-6 hrs storage ($p \leq 0.05$). It indicated that potato, banana and apple puree treated with RBPE had lower browning color than those treated with DW.

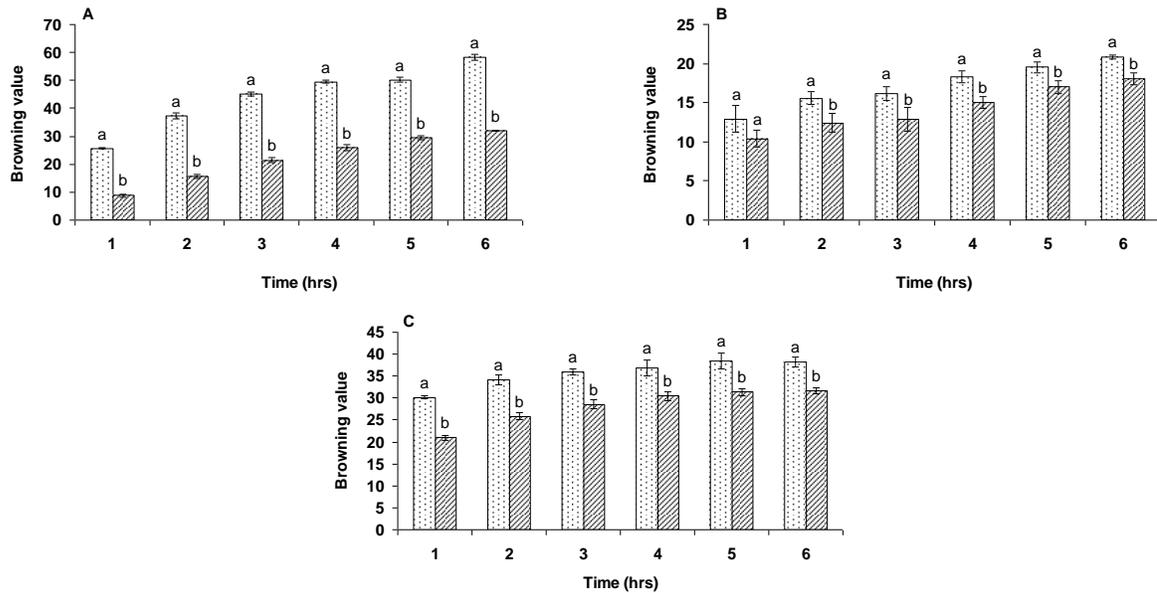


Figure 1 Browning values of vegetable and fruit puree treated with DW () and RBPE () storage at room temperature for 6 hrs; A = potato, B = banana, C = apple

^{a, b} Means with different letters are significantly different ($p \leq 0.05$) at the same storage time.

L^* value is the lightness. The decrease of L^* value means darker color that related with a higher browning formation (Sapers and Douglas, 1987). From Figure 2, it was found that L^* values of potato puree treated with RBPE were higher than that treated with DW through 1-6 hr ($p \leq 0.05$). The L^* values of banana puree treated with RBPE were higher than that treated with DW through 2-6 hr ($p \leq 0.05$) and L^* values of apple puree treated with RBPE were higher than that treated with DW through the storage time ($p \leq 0.05$). It indicated that potato, banana and apple puree treated with RBPE had lower browning color than those treated with DW, which were in accordance with the results of browning value.

According to the results of browning values and L^* values, it showed that RBPE was more effective in browning inhibition in potato, banana and apple puree than DW. Proteins, peptides and amino acids can affect PPO activity in at least two ways: by reacting with the *o*-quinone and by chelating on copper at the active site of PPO (Kahn, 1995). Moreover, there were several researchers reported that rice bran contains antioxidative peptides and sulfur amino acids (Parrado *et al.*, 2006; Sereewatthanawut *et al.*, 2008; Adebiji

et al., 2009). Thus, antioxidative activity of peptides in RBPE might play the role in browning inhibition in potato, banana and apple puree. Browning of potato and apple were also inhibited by whey protein (Tien *et al.*, 2001), dipeptide (Girelli *et al.*, 2004) and honey (Oszmianski and Lee, 1990).

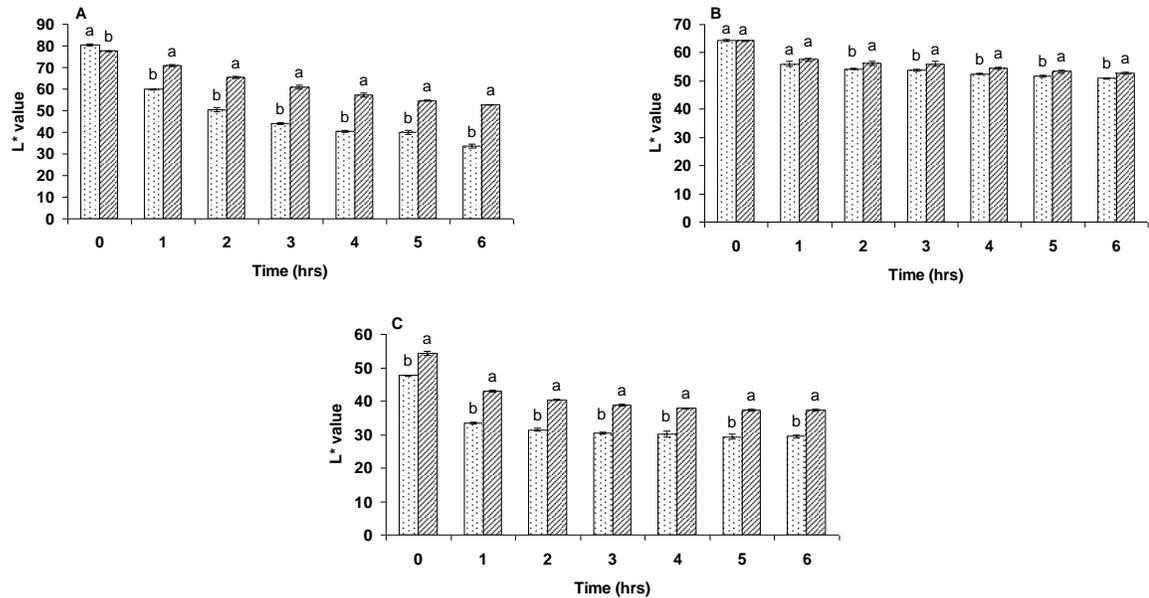


Figure 2 L* values of vegetable and fruit puree treated with DW (▨) and RBPE (▩) storage at room temperature for 6 hrs; A = potato, B = banana, C = apple

^{a, b} Means with different letters are significantly different ($p \leq 0.05$) at the same storage time.

The results of % Browning inhibition of potato, banana and apple calculated from total color changes between sample treated with DW and RBPE are shown in Table 1. It indicated that RBPE could inhibit browning in potato puree more effective than banana and apple puree, respectively ($p \leq 0.05$). This might be due to the specific inhibition of RBPE on potato PPO isozymes more than PPO from the others. The PPO isozymes in plants were reported to be different (Flurkey, 1986).

Table 1 % Browning inhibition of RBPE in vegetable and fruit puree storage at room temperature for 6 hrs

Source	% Browning inhibition (at storage time; hrs)					
	1	2	3	4	5	6
Potato	67.02 ± 2.45a	58.13 ± 2.99a	52.88 ± 1.60a	49.27 ± 1.17a	43.66 ± 1.55a	46.68 ± 0.81a
Banana	21.89 ± 4.43b	23.31 ± 2.54b	23.23 ± 1.62b	19.82 ± 3.31b	16.89 ± 0.58b	16.76 ± 1.89b
Apple	22.74 ± 2.98b	17.97 ± 4.05b	14.96 ± 2.21c	10.45 ± 6.65c	11.76 ± 2.16c	10.24 ± 0.97c

^{a, b} Means with different letters are significantly different ($p \leq 0.05$) at the same storage time.

CONCLUSION

RBPE could inhibit browning in potato, banana and apple puree. RBPE showed % browning inhibition in potato puree higher than banana and apple puree. Therefore, RBPE had a potential to be used as a browning inhibitor in vegetable and fruit puree, especially potato puree.

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REFERENCES

- Abdul-Hamid, A. and Y.S. Luan. 2000. Functional properties of dietary fibre prepared from defatted rice bran. *Food Chem.* 68: 15-19.
- Adebiyi, A.P., A.O. Adebiyi, J. Yamashita, T. Ogawa and K. Muramoto. 2009. Purification and characterization of antioxidative peptides derived from rice bran protein hydrolysates. *Eur. Food Res. Technol.* 228: 553-563.
- Amissah, J.G.N., W.O. Ellis, I. Oduro and J.T. Manful. 2003. Nutrient composition of bran from new rice varieties under study in Ghana. *Food Control.* 14: 21-24.
- Ates, S., S. Pekyardimci and C. Cokmus. 2001. Partial characterization of a peptide from honey that inhibits mushroom polyphenol oxidase. *J. Food Biochem.* 25: 127-137.
- Buta, J.G. and H.E. Moline. 2001. Prevention of browning of potato slices using polyphenoloxidase inhibitors and organic acids. *J. Food Quality.* 24: 271-282.
- Campas-Ríos, M.D.J., J.N. Mercado-Ruiz, M.A. Valdéz-Covarrubias, A.R. Islas-Rubio, A.M. Mendoza-Wilson and R.R. Baladrán-Quintana. 2012. Hydrolysates from wheat bran albumin as color-adding agents and inhibitors of apple polyphenol oxidase. *J. Food Biochem.* 36: 470-478.
- Flurkey, W.H. 1986. Polyphenoloxidase in higher plants. *Plant Physiol.* 81: 614-618.
- Food and Drug Administration. 1986. Sulfiting agents; revocation of GRAS status for use on fruits and vegetables intended to be served or sold raw to consumers. *Fed. Regist.* 51: 25021-25026.
- Fu, B., H. Li, X. Wang, F.S.C. Lee and S. Cui. 2005. Isolation and identification of flavonoids in licorice and a study of their inhibitory effects on tyrosinase. *J. Agric. Food Chem.* 53: 7408-7414.

- Girelli, A.M., E. Mattei, A. Messina and A.M. Tarola. 2004. Inhibition of polyphenol oxidases activity by various dipeptides. *J. Agric. Food Chem.* 52: 2741-2745.
- Gnanasambandam, R. and N.S. Hettiarachchy. 1995. Protein concentrates from unstabilized and stabilized rice bran: preparation and properties. *J. Food Sci.* 60: 1066-1069, 1074.
- Gnangui, S.N., S.L. Niamke and L.P. Kouame. 2010. Partial characterization of a new peptide from ivorian red onion that inhibits polyphenol oxidase and enzymatic browning of edible yam (*Dioscorea cayenensis-rotundata* cv *Longb*). *Chiang Mai J. Sci.* 37: 464-475.
- Helm, R.M. and A.W. Burks. 1996. Hypoallergenicity of rice protein. *Cereal Food. World.* 41: 839-843.
- International Rice Research Institute. 2012. *World Rice Statistics*. Available Source: www.irri.org/science/ricestat/index.asp, September 24, 2012.
- Jang, M.S., A. Sanada, H. Ushio, M. Tanaka and T. Ohshima. 2002. Inhibitory effects of 'enokitake' mushroom extracts on polyphenol oxidase and prevention of apple browning. *Lebensm.-Wiss. u.-Technol.* 35: 697-702.
- Kahn, V. 1995. Effect of proteins, protein hydrolyzates, and amino acids on *o*-dihydroxyphenolase activity of polyphenol oxidase of mushroom, avocado and banana. *J. Food Sci.* 50: 111-115.
- Labuza, T.P., J.H. Lillemo and P.S. Taoukis. 1990. **Inhibition of polyphenol oxidase by proteolytic enzymes "Killer Enzymes"**. Symposium on The 20th International Symposium International Federation of Fruit Juice Producers. Paris.
- Li, B., Y. Huang and S.M. Paskewitz. 2006. Hen egg white lysozyme as an inhibitor of mushroom tyrosinase. *FEBS Letters.* 580: 1877-1882.
- McEvily, J.A., R. Iyengar and W.S. Otwell. 1992. Inhibition of enzymatic browning in foods and beverages. *Crit. Rev. Food Sci. Nutr.* 32: 253-273.
- Ortíz-Estrada, A.M., J.N. Mercado-Ruiz, J.M. García-Robles, A.R. Islas-Rubio, A.M. Mendoza-Wilson and R.R. Balandrán-Quintana. 2012. Wheat bran globulins: competitive inhibitors of mushroom tyrosinase. *Food Sci. Biotechnol.* 21: 633-635.
- Oszmianski, J. and C.Y. Lee. 1990. Inhibition of polyphenol oxidase activity and browning by honey. *J. Agric. Food Chem.* 38: 1892-1895.
- Parrado, J., E. Miramontes, M. Jover, J.F. Gutierrez, L.C. de Terán and J. Bautista. 2006. *J. Food Chem.* 98: 742-748.
- Sapers, G.M. and F.W. Douglas. 1987. Measurement of enzymatic browning at cut surfaces and in juice of raw apple and pear fruits. *J. Food Sci.* 52: 1258-1285.

- Sereewatthanawut, I., S. Prapintip, K. Watchiraruji, M. Goto, M. Sasaki and A. Shotipruk. 2008. Extraction of protein and amino acids from deoiled rice bran by subcritical water hydrolysis. **Bioresource Technol.** 99: 555-561.
- Tien, C.L., C. Vachon, M.A. Mateescu and M. Lacroix. 2001. Milk protein coatings prevent oxidative browning of apples and potatoes. **J. Food Sci.** 66: 512-516.
- Thongsook, T. and W. Tiyaboonchai. 2011. Inhibitory effect of sericin on polyphenol oxidase and its application as edible coating. **Int. J. Food Sci. Tech.** 46: 2052-2061.
- Wang, M., N.S. Hettiarachchy, M. Qi, W. Burks and T. Siebenmorgen. 1999. Preparation and functional properties of rice bran protein isolate. **J. Agric. Food Chem.** 47: 411-416.
- Zhu, Y., Z. Pan and T.H. Mchugh. 2007. Effect of dipping treatments on color stabilization and texture of apple cubes for infrared dry-blanching process. **J. Food Process. Pres.** 31: 632-648.