

Effects of Rice Bran Extract on Browning and Polyphenol Oxidase Activity in Vegetable and Fruit

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ABSTRACT

The effects of rice bran extract (RBE) on browning in potato, banana and apple slices compared with distilled water (DW) were investigated by measuring the changes of color values. Potato slice treated with RBE had lower browning value than that treated with DW during storage 9 to 12 hrs ($p \leq 0.05$). The browning values of potato treated with RBE and DW stored for 12 hrs were 4.72 and 7.64, respectively. Banana slice treated with RBE had lower browning value than that treated with DW during storage 2 to 12 hrs ($p \leq 0.05$). The browning values of banana treated with RBE and DW stored for 12 hrs were 19.76 and 27.17, respectively. Whereas, apple slice treated with RBE was slightly different from that treated with DW during storage 0 to 9 hrs. The browning values of apple treated with RBE and DW stored for 9 hrs were 5.21 and 7.84, respectively. Potato, banana and apple slices treated with RBE had higher L^* value but lower a^* value than that treated with DW. Regarding to the effect of RBE on polyphenol oxidase (PPO) activity. It was found that RBE had more inhibitory effect on PPO activity of potato than that of banana and apple with the %PPO inhibition of 96.11, 51.43 and 12.3, respectively. In conclusion, RBE could reduce browning and inhibit PPO activity in potato more than that of banana and apple.

Key words: browning, polyphenol oxidase, rice bran

INTRODUCTION

Fresh vegetables and fruits have a problem from the enzymatic browning by polyphenol oxidase (PPO) during harvesting, processing and storage. Enzymatic browning causes to reduce quality, change color, off-flavor and loss in nutritional quality of fruits and vegetables (Severini *et al.*, 2003). To prevent browning, sulfites have been used extensively because of its highly effectiveness in controlling browning and inexpensive price. However, sulfites were subjected to regulatory restrictions because

of adverse effects on health (Son *et al.*, 2000). Thus, the extract from natural sources; for example, honey (Oszmainski and Lee, 1990), pineapple juice (Lozzano-de Gonzalez *et al.*, 1993), grapefruit seed extract (Park *et al.*, 1999), enokitake extract (Jang *et al.*, 2002) and onion extract (Kim *et al.*, 2005) were searched for replacing the chemical additives in prevention of enzymatic browning reaction. Rice bran is one of major by product from rice milling industry and is also an interesting source of phenolic compounds and antioxidant activity. Kim and Chun (1996) reported that rice bran contained

ferulic acid, sinapic acid, syringic acid, p-coumaric acid and vanillic acid. Phenolic compounds in rice bran may involve in inhibition of enzymatic browning in vegetable and fruit. Therefore, the main objective of this research was to investigate the effect of rice bran extract on browning and polyphenol oxidase activity in vegetable and fruit.

MATERIALS AND METHODS

Preparation of rice bran extract (RBE)

Full-fat rice bran was obtained from Patum Rice Mill and Granary Public Co. Ltd. The full-fat rice bran was milled and screened pass 50 mesh sieve, extracted with distilled water at 1:3 (w/v) by using overhead stirrer at 700 rpm for 30 min and then centrifuged at 8,000xg at 20°C for 30 min. The supernatant, rice bran extract (RBE) was used immediately for further study.

Fresh vegetable and fruit slice preparation and treatment

Potatoes (*Solanum tuberosum*), bananas (*Musa* (AAA group) Gros Michel') and apples (*Malus pumila* cv. Fuji) from local market were peeled. Potato and apple were cut at a size of 1.5×3 cm and banana was sliced across at the thickness of 0.5 cm. The slice samples were dipped in 200 ml RBE or distilled water (control) for 5 min at room temperature (25°C). The color values (L*, a* and b*) of the samples were measured by spectrophotometer (CM-3500D, Minolta) at 0, 1, 2, 3, 6, 9 and 12 hr(s) during storage at room temperature and the browning values (($\Delta L^*/L_0^*$) × 100) were calculated; when ΔL^* is equal to $L_0^* - L^*$; L* is the L* value at any time and L_0^* is the initial L* measurement (Labuza *et al.*, 1990).

PPO extraction

Twenty five grams of potato, banana and apple each were homogenized for 30s with 50 ml of 0.2 M phosphate buffer, pH 6.5 and then centrifuged at 25,560xg at 4°C for 20 min. The

obtained supernatant was crude PPO extract used for study the effect of RBE on PPO activity (adapted from Galeazzi *et al.*, 1981).

PPO activity assay

The mixture contained 3 ml of 0.2 M catechol in phosphate buffer (pH 6.5) and 0.2 ml of the rice bran extract or distilled water was hold for 30s, then 0.02 ml of PPO extract was added and the increasing of absorbance at 420 nm was measured for 5 min. (adapted from Lozano-de-Gonzalez *et al.*, 1993). One unit of enzyme activity is defined as the amount of enzyme causing an increasing in absorbance of 0.001 at 420 nm per min under the assay condition (25°C, pH 6.5).

Statistical analysis

Three replications of each experiment were performed. All data were analyzed by Statistical Package for Social Science (SPSS) and tested by one-way analysis of variance. Significant difference ($p \leq 0.05$) among various treatments was detected by using Duncan's multiple range tests.

RESULTS AND DISCUSSION

Effect of rice bran extract on browning in fresh vegetable and fruit slices

Browning values of potato, banana and apple slices; that were dipped in rice bran extract (RBE) and distilled water (DW) for 5 min and stored at 25°C for 12 hrs were shown in Figure 1. Browning value of potato slice treated with RBE was slightly lower than that treated with DW during storage for 6 hrs; however, they were not significantly different ($p > 0.05$). After storage for 9 hrs, potato sliced treated with RBE had significantly lower browning value than that treated with DW ($p \leq 0.05$). Browning values of potato treated with RBE and DW stored for 12 hrs were 4.72 and 7.64, respectively. Banana slice treated with RBE had significantly lower browning value than that treated with DW during storage

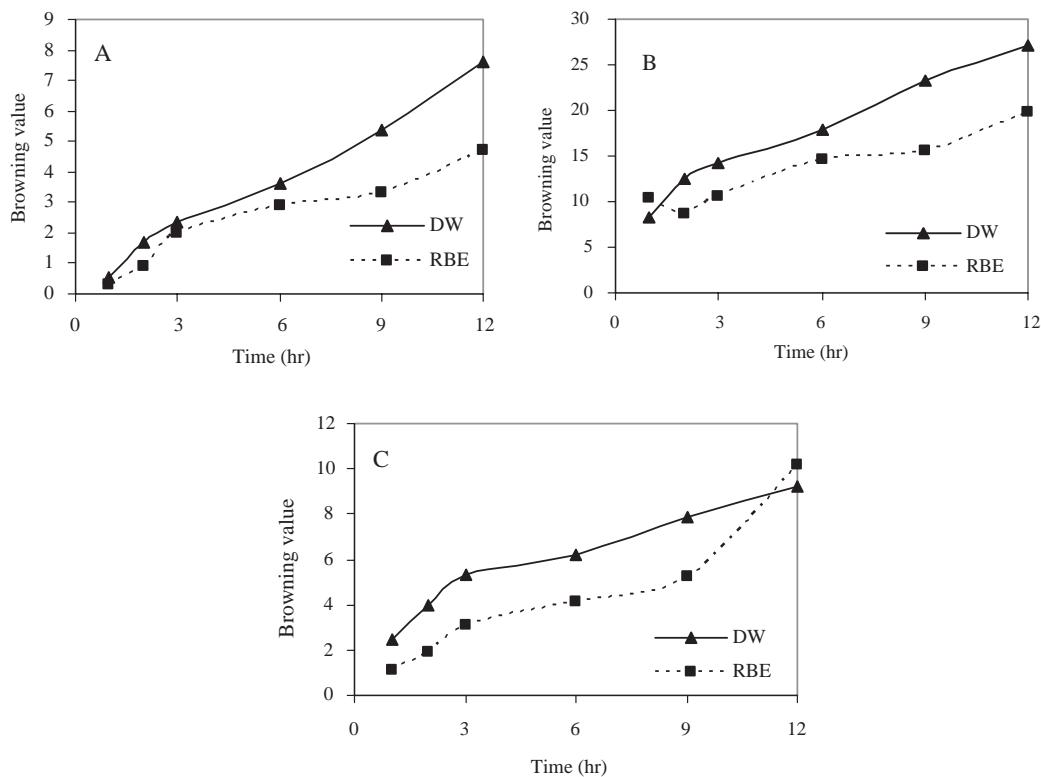


Figure 1 Browning values $((DL^*/L_0^*) \times 100)$ of vegetable and fruit slices treated with distilled water (DW) and rice bran extract (RBE) stored at 25°C for 12 hrs; (A) potato, (B) banana and (C) apple slices.

for 2 to 12 hrs ($p \leq 0.05$). Browning values of banana treated with RBE and DW stored for 12 hrs were 19.76 and 27.17, respectively. Browning values of banana treated with RBE and DW stored for 12 hrs were 19.76 and 27.17, respectively. Browning value of apple slice treated with RBE was also slightly lower but was not significantly different from that treated with DW during storage for 9 hrs. Their browning values of apple were 5.21 and 7.84, respectively. The results indicated that the immersion of potato, banana and apple slices in RBE decreased the development of browning occurring during storage. RBE could reduce the browning of potato and banana slices better than that of apple slice. This might be due to different chemical compositions of these fruit

and vegetables, especially the different type and activity of PPO isozymes (Labuza *et al.*, 1990); therefore, different PPO isozymes could be inhibited by RBE at different extent.

L^* value of potato slice treated with RBE and DW were not significantly different during storing for 9 hrs ($p > 0.05$). After storage for 12 hrs, potato slice treated with RBE had significantly lower L^* value than that treated with DW ($p \leq 0.05$). The a^* value of potato slice treated with RBE tended to be lower, but was not significantly different from that treated with DW. However, the b^* value of potato slice treated with RBE decreased less than that treated with DW after storage for 12 hrs (Figure 2).

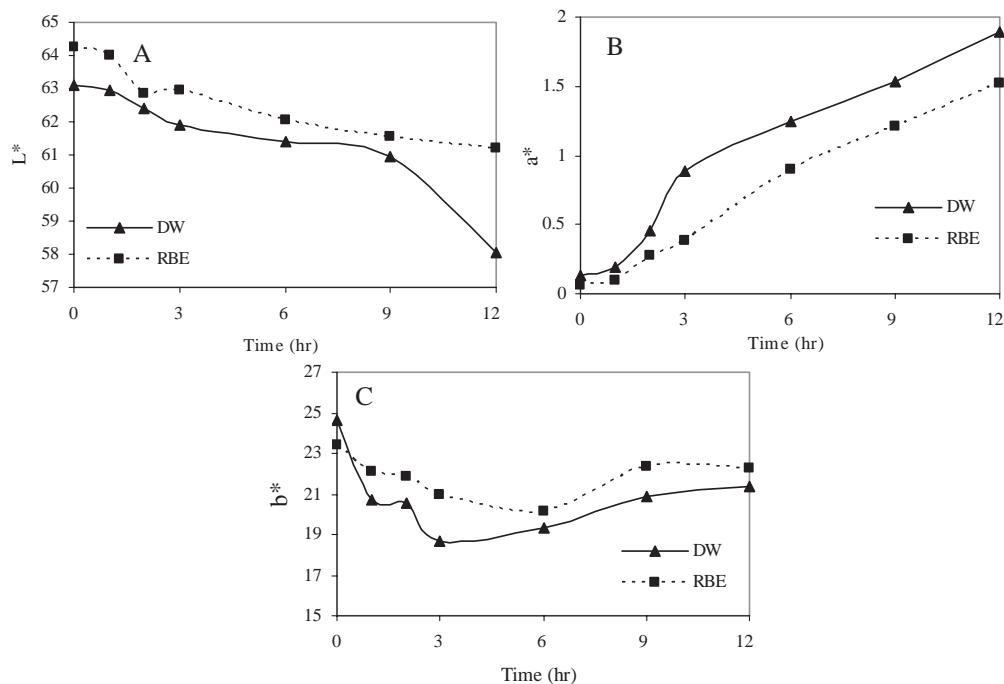


Figure 2 L*, a* and b* values of potato slice treated with distilled water (DW) and rice bran extract (RBE) stored at 25°C for 12 hrs; (A) L*, (B) a* and (C) b*.

L* value of banana slice treated with RBE was higher than that treated with DW during storage for 9 to 12 hrs ($p \leq 0.05$). The a* value of banana slice treated with DW increased more rapidly than that treated with RBE during storage for 2 hrs. However, after storage for 3 hrs, a* value of banana slice treated with DW increased slowly, while the b* value of banana slice treated with DW decreased more rapidly than that treated with RBE after storage for 9 hrs (Figure 3).

Apple slice treated with RBE showed higher L* value than that treated with DW during storage for 3 to 12 hrs ($p \leq 0.05$). The a* value of apple slice treated with DW increased more sharply than that treated with DW during the first 3 hrs of storage. After storage for 6 hrs, the a* value of apple slice treated with RBE increased as the similar extent as that treated with DW. The b* value of apple slice treated with DW increased more than that treated with RBE during storage for 12 hrs (Figure 4).

Moline *et al.* (1999) reported that the decrease of L* value means darker color and the increase of a* value means higher red color. Thus, the decrease of L* value and the increase of a* value are related with a high browning color. The results showed that the change of L* values in potato, banana and apple slices treated with RBE was less than that treated with DW, but the change of a* values was more than that treated with DW. Therefore, the results of the change of L* and a* values indicated that browning in potato increased less than that of banana and apple during storage for 12 hrs.

According to the results of browning values, the changes of L*, a* and b* values, it showed that RBE was more effective in browning inhibition of potato slice than those of banana and apple slices. Kim and Chun (1996) reported that rice bran had phenolic acid such as ferulic acid, sinapic acid, syringic acid, p-coumaric acid and vanillic acid. Miyazawa *et al.* (2003) found that

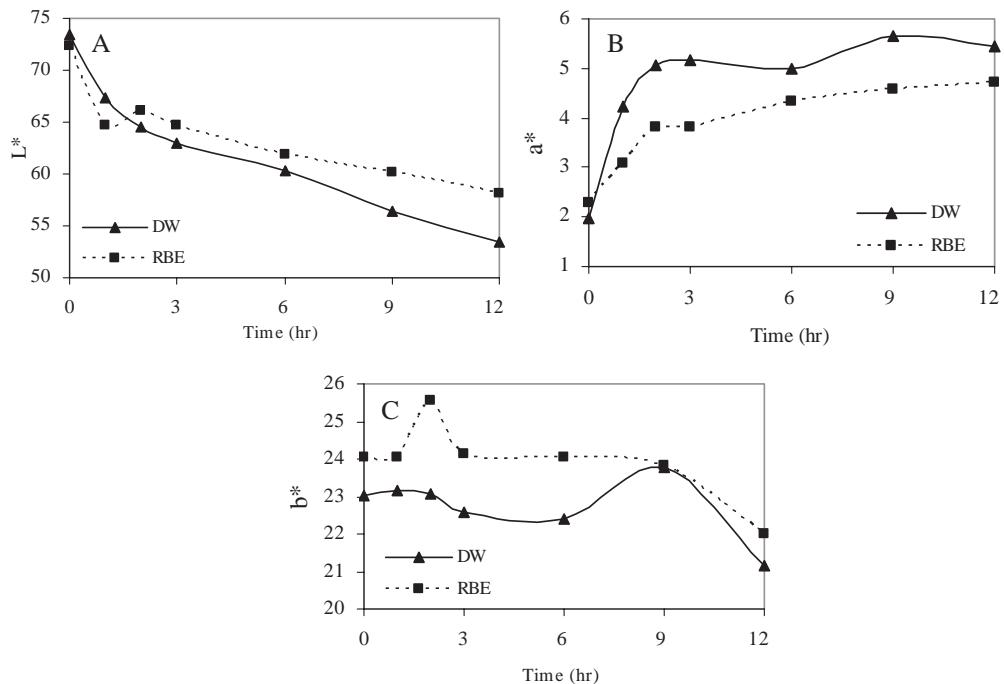


Figure 3 L^* , a^* and b^* values of banana slice treated with distilled water (DW) and rice bran extract (RBE) stored at 25°C for 12 hrs; (A) L^* , (B) a^* and (C) b^* .

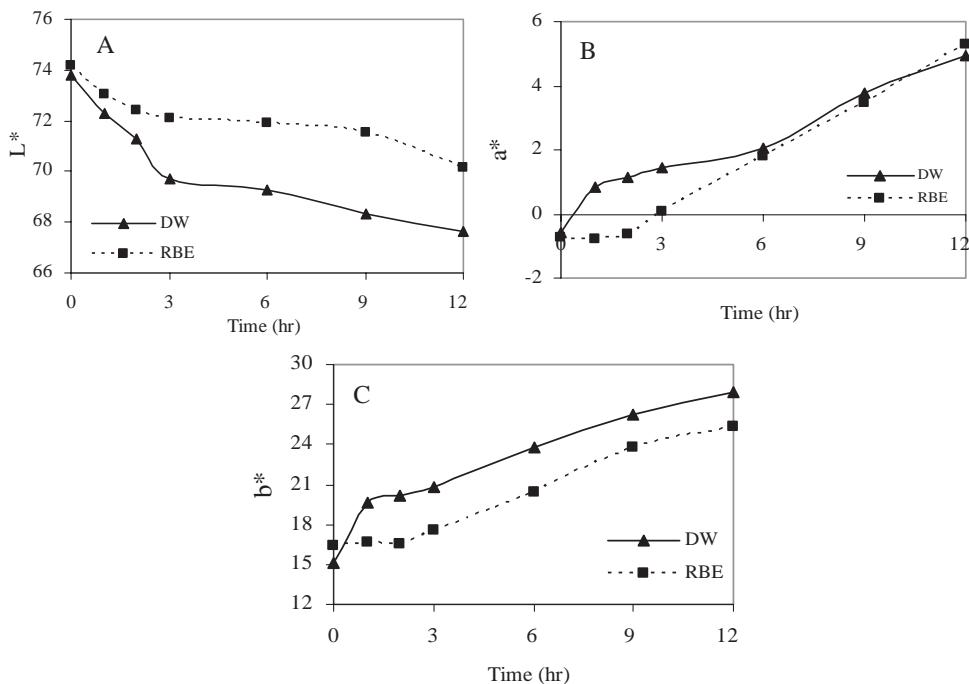


Figure 4 L^* , a^* and b^* values of apple slice treated with distilled water (DW) and rice bran extract (RBE) stored at 25°C for 12 hrs; (A) L^* , (B) a^* and (C) b^* .

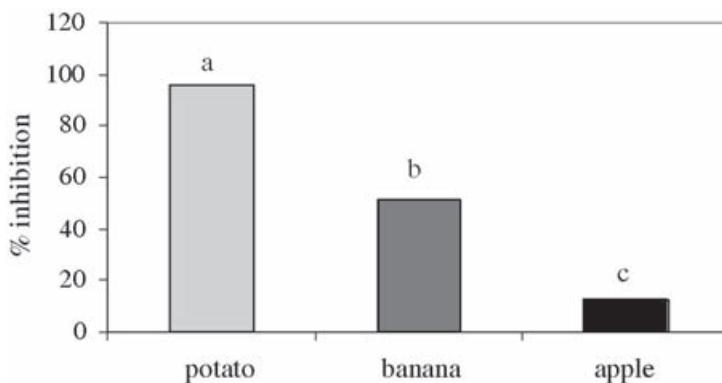


Figure 5 % Inhibition of potato, banana and apple PPO activity by rice bran extract.

^{abc}Means with different letters are significantly different ($p \leq 0.05$)

phenolic compounds in black rice bran such as protocatechuic acid and protocatechuic acid methyl ester exhibited an inhibitory effect on tyrosinase activity. Some researchers also reported that p-coumaric acid, ferulic acid and sinapic acid were effective in inhibition of enzymatic browning in potato (Macrae and Duggleby, 1968) and apple (Pifferi *et al.* 1974; Walker and Wilson, 1975). These phenolic compounds in RBE might be the compounds that inhibit the browning of the vegetable and fruit. The browning inhibitory effect of RBE may be due to the inhibition of the PPO isozyme activities in these vegetable and fruit.

Inhibitory effect of rice bran extract on vegetable and fruit PPO activity

The inhibitory effects of RBE on potato, banana and apple PPO activity were shown in Figure 5. RBE inhibited PPO activity of potato was more than that of banana and apple with % inhibition of 96.11, 51.43 and 12.3, respectively ($p \leq 0.05$). The results confirmed that RBE could inhibit enzymatic browning in potato slice more effective than those in banana and apple slices.

CONCLUSION

Rice bran extract could reduce browning in potato more effective than that in banana and

apple during storage. RBE also had more inhibitory effect on PPO activity of potato than that of banana and apple. The rice bran extract showed a high potential to be used as a natural antibrowning agent for fresh potato.

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LITERATURE CITED

Galeazzi, M.A.M., V.C. Sgarbieri and S.M. Constantinides. 1981. Isolation, purification, and physicochemical characterization of polyphenol oxidase (PPO) from a dwarf variety of banana (*Musa cavendishii* L.). *J. Food Sci.* 46: 150-155.

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.

Kim, I.H. and H.S. Chun. 1996. Composition of

fatty acid and phenolic acid in rice with the different milling fractions. **J. Korean Society of Food Sci. Nutri.** 25: 721-726.

Kim M. J., C. Y. Kim and I. Park. 2005. Prevention of enzymatic browning of pear by onion extract. **Food Chem.** 89: 181-184.

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, France.

Lozzano-de-Gonzalez, P.G., D.M. Barrett, R.E. Wrolstad and R.W. Durst. 1993. Enzymatic browning inhibited in fresh and dried apple rings by pineapples juice. **J. Food Sci.** 58: 399-404.

Macrae, A.R. and R.G. Duggleby. 1968. Substrate and inhibitors of potato tuber phenolase. **Phytochemistry.** 7: 855-861.

Miyazawa, M., T. Oshima, K. Koshio, Y. Itsuzaki and J. Anazai. 2003. Tyrosinase inhibitor from black rice bran. **J. Agric. Food Chem.** 51: 6953-6956.

Moline, H.E., J. G. Buta and I.M. Newman. 1999. Prevention of browning of banana slices using natural products and their derivatives. **J. Food Quality.** 22: 499-511.

Oszmianski, J. and C.Y. Lee. 1990. Inhibition of polyphenol oxidase activity and browning by honey. **J. Agric. Food Chem.** 38: 1892-1895.

Park, W., D. Lee and S. Cho. 1999. Effect of grapefruit seed extract and antibrowning agents on the keeping quality of minimally processed vegetables. **Acta Hort.** 483: 325-330.

Pifferi, P. G., L. Baldassari and R. Culteria. 1974. Inhibition by carboxylic acids of and o-diphenol oxidase for *Prunus avium* Fruit. **J. Sci. Food Agric.** 25: 263-270.

Severini C., A. Baiano, T. De Pilli, R. Romaniello and A. Derossi. 2003. Prevention of enzymatic browning in sliced potato by blanching in boiling saline solution. **Lebensm.-Wiss.u-Technol.** 36: 657-665.

Son, S.M., K.D. Moon and C.Y. Lee. 2000. Kinetic study of oxalic acid inhibition on enzymatic browning. **J. Agric. Food Chem.** 48: 2071-2074.

Walker, J.R.L. and E.L. Wilson. 1975. Studies on the enzymatic browning of apples. Inhibition of apple o-diphenol oxidase by phenolic acids. **J. Sci. Food Agric.** 26: 1825-183.