# Development of Ready-to-Eat Color Rice Product Enriched With Natural Amino Acids

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# Abstract

A meat-free diet and the aging society cause some of the health problems. One of that was the deficiency of some essential amino acid in the diets. Riceberry has recently been developed in Thailand. This new variety of colored rice has a dark purple seed coat containing anthocyanin which acts as an antioxidant. Similar to normal rice, riceberry lacks lysine which plays an important role in maintaining body systems through hormone release and muscle mass maintenance. To enrich ready-to-eat rice products, red bean was added as a natural amino acid source. This formulated product had higher protein and ash content, while energy, fat and carbohydrate content were not different from the control. The essential amino acid profile of the product was complete and product shelf life was 6 months with an unchanged texture. Anthocyanin and antioxidant capacity of the product decreased with storage time. This product suited a wide range of consumers and was convenient for modern lifestyles that demand healthy, ready-to-eat foodstuffs.

Keywords: riceberry, colored rice, red bean, lysine

# 1. Introduction

Demand for meat-free diets is increasing and this has resulted in emerging health problems through lack of proteins. Hypoalbuminemia results from replacement of cow milk by rice milk (Massa et al., 2001) as a symptom caused by protein malnutrition (Steele, Yokum, & Armstrong, 1998) which and is related to heart failure (Filippatos et al., 2011). Apart of the lifestyle changing that causes the malnutrition, the aging is also the cause of undernutrition. Lamy, Mojon, Kalykakis, Legrand, and Butx-Jorgensen (1999) reported that oral problems resulted in eating difficulties which affected health conditions in the elderly (Pirlich & Lochs, 2001).

Rice is the staple carbohydrate source of Asian cuisine but unfortunately, it lacks essential amino acids, particularly lysine (Sotelo, Hernandez, Montalvo, & Sousa, 1994; Moonggnarm & Saetung, 2010). It is well known that lack of lysine leads to kwashiorkor in infants (Albanese, Higgons, Hyde, & Orto, 1956) and retards mental and physical development in children (Galili & Amir, 2013). Furthermore, in adults and the elderly, lysine also plays an important role in maintaining body systems such as growth hormone release (Isidori, Lo, Monaco, & Cappa, 1981) and muscle mass increase in the elderly (Fuller et al., 2011). To solve lysine deficiency in rice, new genetically engineered rice cultivars (Wang & Galili, 2016) were introduced but some consumers had doubts about their safety. Another option for improving rice amino acid profiles is by adding other sources of lysine plant protein. Bressani and Valiente (1962) suggested that black bean could supplement polished rice with lysine at low cost (Bressani, 2010).

Following the health and wellness trend, unpolished rice is now preferred by consumers over polished rice due to the health benefits of fiber, while colored rice has gained increasing interest. Riceberry has become the popularly consumed rice in Thailand as this variety contains micronutrients, particularly anthocyanins, which are known for their antioxidant capacity. Unpolished rice takes longer to cook because the bran layer retards water

penetration into the grain (Mohapatra & Bal, 2006). This phenomenon also occurs in red bean cooking (Zamindar, Baghekhandan, Nasirpour, & Sheikhzeinoddin, 2013). To solve this problem, a longer soaking time before cooking resulted in a softer texture (Tian et al., 2014; Meng et al., 2018). Pretreatment of rice and red bean also affects product eating quality. Rewthong, Soponronnarit, Taechapairoj, Tungtrakul, and Prachayawarakorn (2011) and Syafutri, Pratama, Syaiful, and Faizal (2016) reported that blanching or boiling resulted in a harder texture of cooked rice than steaming or using a rice cooker. Moreover, heat treatment was also efficient in beany flavor removal (Yuan, Chang, Liu, & Xu, 2008). Here, ready-to-eat brown rice products were developed using the riceberry variety enriched with red kidney bean to complement the amino acid profile. Moreover, product shelf life was evaluated based on textural and antioxidant properties.

# 2. Method

# 2.1 Material

Riceberry, purple-black color Thai brown rice, was purchased from the Rice Science Center Kasetsart University, while red bean was purchased from a local market.

# 2.2 Rice and Red Bean Preparation

Rice and red bean were separately soaked in 50 °C water for 4 hr to obtain moisture contents at approximately 30 and 50%, respectively. Then, the soaked rice was steamed for 15 min while the red bean was pressure cooked at 8 psi for 15 min. The precooked rice and red bean were kept in closed containers until required for further experiments.

# 2.3 Effect of Phosphate Salt

Starch products undergo retrogradation during storage, resulting in increased hardness or stiffness of the products. To retard this phenomenon, phosphate salts were used to increase water absorption in the products and slow down starch retrogradation. Phosphate salt concentration in the sample was 0.15% by wt. Textures of riceberry and red bean were analyzed to compare with the control sample which was not added with phosphate salt.

# 2.4 Preparation of Ready to Eat Rice Mixed With Red Bean

Precooked rice 100 g was mixed with 50 g precooked red bean in a retort pouch, then 20 ml of 0.3% salt water was added before sealing and sterilizing using a water spray retort (Model: ARS-100/20; HISAKA, Japan). The sterilized temperature was 118 °C with processing time of 40 min. Proximate analysis of the samples was determined according to AOAC (2006).

# 2.5 Texture Analysis

Changes of textural properties of riceberry and red bean during 6 months of storage at ambient temperature were determined separately using a Texture Analyzer (TA-Tx2i, Stable Micro, UK) equipped with a 25 mm diameter cylinder probe (P/25). Test speed was 1 mm/sec and pressed to 60% strain. Hardness of the samples was analyzed.

# 2.6 Anthocyanin and Antioxidant Capacities

The 5 g of sample was treated with 50 ml of 1 mM citric acid in 80% ethanol for 3 hr. Clear supernatant was then collected and kept at -20 °C until analyzed for its anthocyanin content (Pedro, Granato, & Rosso, 2016). Moreover, antioxidant capacities were determined for total phenolic content (Kaur & Kapoor, 2002), DPPH (Brand-Williams, Cuvelier, & Berset, 1995) and FRAP (Benzie & Strain, 1999).

# 2.7 Amino Acid Profile of the Product

The amino acid profile of the enriched product was analyzed using HPLC according to the method of Herbert, Barros, Ratola, and Alves (2000).

# 2.8 Data Analysis

All experiments were carried out in triplicate. Results were reported as average values with standard deviations. Analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) at p = 0.05 were used to determine differences between treatments using SPSS version 12.0.

# 3. Results and Discussion

# 3.1 Effect of Phosphate Salt

The rice and red bean were precooked by steeping in warm water at 50 °C and pressure cooked. This process not only precooked the seeds but also improved cooking and eating quality of the rice (Park, Chae, & Yoon, 2009).

Cooked or gelatinized rice products undergo natural starch reconstruction or retrogradation. This results in changes of physical properties, especially texture which becomes harder and firmer due to loss of water in the gel matrix.

The most common method to retard the retrogradation process is by adding food additives. Among these, phosphate salt is commonly selected as it is cheap, efficient and recognized as safe among food manufacturers. Additionally, phosphate salt increases water binding capacity in many products including rice (Chang & Regenstein, 1997; Singh, Kaur, Singh, & Sekhon, 1999). Lee, Han, and Rhee (2002) found that sodium phosphate salt retarded the retrogradation process. Here, riceberry and red bean with phosphate salt added had a softer texture (Table 1). This result concurred with Wang, Hou, Hsu, and Zhou (2011) who reported that adding phosphate salt yielded softer texture in non-fried instant noodle.

Table 1. Texture of control and phosphate salt added riceberry and red bean kept in ambient temperature for 24 hr

Sample		Hardness (g)	
Riceberry	Control	3,367.54±483.86b	
	Phosphate added	2,650.79±563.08a	
Red bean	Control	3,762.71±152.89a	
	Phosphate added	3,512.40±87.57a	

*Note.* Different letters in the same column of each raw material indicate statistical differences (p < 0.05).

# 3.2 Nutrition Value of Products

Nutrient contents of control and the enriched samples were determined. Overall energy and energy from fat were similar (Table 2). Fat and carbohydrate contents were slightly lower than control, while ash was slightly higher in enriched samples. Protein content of enriched samples was significantly higher than control due to added red bean. Results concurred with Rodríguez-Bürger, Mason, and Nielsen (1998) who reported that addition of black bean increased protein and ash content compared to rice alone. Legumes contain protein at around 24-26% (Bressani, 2010), while protein content in rice is about 7% (Shih, 2003). Therefore, the combination of rice and legume altered the protein content in rice products.

	Table 2. Nutrition	values c	of control	and	enriched	samples
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Detail	Control	Enriched sample
Total energy (kcal/100 g)	210.87±7.57a	207.53±13.00a
Energy from fat (kcal/100 g)	20.25±0.13a	19.49±1.46a
Moisture (%)	49.19±1.89a	49.79±2.94a
Carbohydrate (%)	42.73±1.66a	41.23±2.88a
Protein (%)	4.93±0.20a	5.79±0.01b
Fat (%)	2.25±0.01a	2.17±0.16a
Ash (%)	0.91±0.01a	1.04±0.11a

*Note.* Different letters in the same row indicate statistical differences (p < 0.05).

# 3.3 Essential Amino Acid Content

Development of food blends vegetable protein to obtain the essential amino acid pattern of animal protein. A mixture of cereal grains and legume is a commonly used example. The legume provides lysine amino acid which is lacking in cereal grains (Bressani, 2010). Essential amino acid contents in the enriched products are shown in Table 3. Rice is a cereal crop that normally lacks lysine. Saikusa, Horino, and Mori (1994) reported that lysine content in brown rice kernels was 0.7 mg/100 g, while lysine in bean was 1.96 g/100 g dry matter and lysine content increased to 2.12 g/100 g when cooked (Rodríguez-Bürger et al., 1998). Red bean was reported as high in lysine content, and the cooking method did not affect the amount of lysine in cooked beans (Audu & Aremu, 2011). Our results suggested that adding red bean increased amino acid content in ready to eat rice products.

Amino acid	Content (mg/100 g as eaten)	Content (mg/100 g dry basis)
Alanine	254	505.88
Arginine	345	687.11
Aspartic acid	350	697.07
Cystine	126	250.95
Glutamic acid	702	1,398.13
Glycine	305	607.45
Histidine	142	282.81
Isoleucine	268	533.76
Leucine	400	796.65
Lysine	141	280.82
Methionine	99	197.17
Phenylalanine	324	645.29
Proline	65	129.46
Serine	294	585.54
Threonine	203	404.30
Tryptophan	493	981.88
Tyrosine	163	324.64
Valine	265	527.78

Table 3. Essential amino acid content in enriched samples

#### 3.4 Shelf life of the Enriched Product

#### 3.4.1 Texture Analysis

Product texture change during storage for 6 months at room temperature was monitored by a texture analyzer. Riceberry and red bean hardness slightly increased with longer storage time (Table 4). Grain hardness increased as the result of starch retrogradation (Perdon, Siebenmorgen, Buescher, & Gbur, 1999; Kingcam, Devahastin, & Chiewchan, 2008). C. E. Park, Y. S. Kim, K. J. Park, and B. K. Kim (2012) stated that increasing grain hardness related to loss of moisture from starch gel during storage. Moreover, change in hardness of rice and red bean was not greatly affected by storage time due to the contribution of the outer layer of the grain (Mariotti, Sinelli, Catenacci, Pagani, & Lucisano, 2009) that could prevent the moisture loss which would retard the texture changes.

Table 4. Texture of riceberry and red bean in product stored for 6 months

Storage time (month)	Riceberry Hardness (g)	Red bean Hardness (g)
	3,677.88±717.78ab	5,763.71±301.63a
2	3,347.83±513.80a	6,558.64±755.53b
1	3,595.26±530.67ab	5,934.86±215.11a
)	3,789.64±231.56b	6,339.05±317.47ab

*Note.* Different letters in the same column indicate statistical differences (p < 0.05).

# 3.4.2 Anthocyanin and Antioxidant Capacities

Anthocyanin content in the products significantly decreased after processing and then remained constant during storage time (Table 5). Because of their highly reactive nature, anthocyanins readily degrade or react with other constituents in the media, while water addition resulted in the formation of colorless chalcone (Htwe et al., 2010) or brown colored compounds. Moreover, anthocyanin decomposed during thermal processing (Walter *et al.*, 2013), resulting in a color change. Min, McClung, and Chen. (2014) reported that anthocyanin decreased by 78% after wet cooking due to heat decomposition and leaching of water-soluble anthocyanin. In addition, anthocyanin degraded with storage time (Patras, Brunton, O'Donnell, & Tiwari, 2010) due to oxidation (Rhim, 2002) and phenolic complexation (Reed, Krueger, & Vestling, 2005).

Phenolic content in the product also decreased with storage time (Table 6) and 70% of phenolic compounds were lost when the product was kept for 6 months. Wallace and Guisti (2008) reported that phenolic compounds stored in berry added yogurt reduced. Reduction of phenolic compounds was influenced by oxidation (Abramovic, Butinar, & Nikoliê, 2007). Phenolic compounds were affected by heating processes less than anthocyanins since they were more thermally stable and less water-soluble (Min et al., 2014).

Storage time (month)	Anthocyanin content (mg/g db)
Raw material	69.36±6.31b
0	26.37±2.32a
3	23.84±1.10a
6	23.67±0.31a

Table 5. Anthocyanin content in product stored for 6 months

*Note.* Different letters in the same column indicate statistical differences (p < 0.05).

Antioxidant capacity of the stored product was determined for DPPH and FRAP with results shown in Table 6. Storage decreased antioxidant capacity of the product. Our results concurred with Gujral, Angurala, Sharma, and Singh (2011) who suggested that antioxidant activity of pressure cooked pulse was lower than raw pulse. Antioxidant activity was reduced by antioxidants leaching to water with thermal decomposition (Xu & Chang, 2007). At longer storage time, antioxidant capacity decreased, related to the phenolic content in the sample (Kalt, McDonald, & Donner, 2000; Kim, Jeong, & Lee, 2003; Piljac-Žegarac, Valek, Martinez, & Belščak, 2009). Brownmiller, Howard, and Prior (2008) indicated that this was the result of anthocyanin decomposition.

Table 6. Phenolic content and antioxidant capacity in product stored for 6 months

Storage time (month)	Phenolic content (µg/g db)	DPPH (µg gallic eq/g db)	FRAP (µg trolox eq/g db)
Raw material	260.57±6.03c	440.30±4.24d	520.00±10.94c
0	258.02±3.85c	158.89±1.10b	160.69±17.12b
3	251.12±1.80b	163.38±2.31c	145.86±1.19ab
6	184.53±2.15a	144.76±1.55a	133.26±2.83a

*Note.* Different letters in the same column indicate statistical differences (p < 0.05).

# 4. Conclusions

An upsurge in the popularity of meat-free diets and an aging society have resulted in many health problems because of essential amino acid deficiency. Rice is the main source of carbohydrate in Asian cuisine and a popular new variety of colored rice has entered the Thai market, namely riceberry that contains anthocyanin which acts as an antioxidant. However, similar to normal rice, riceberry lacks lysine. To enrich the rice product, red bean which is rich in minerals was added to complete the amino acid profile. The formulated product of mixing riceberry and red bean was high in protein and ash. The essential amino acid profile of the product was completed. Shelf life was 6 months with unchanged textural properties but anthocyanin and antioxidant capacities of the product decreased with storage time. This product is popular with a wide group of consumers and convenient for fast-paced lifestyles that demand healthy products with minimal preparation time.

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