

Development of healthy colored rice snack containing concentrated pineapple juice

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Abstract

Rice with pigment or colored rice is a good source of antioxidant such as anthocyanin and phenolic compound, which can reduce risk of cancer and coronary heart disease. The objective of this study was to develop healthy colored rice snack containing concentrated pineapple juice. Concentrated pineapple juice (50°Brix) was prepared by vacuum evaporator at 50°C with the rate of agitation at 85.0 SP and 645 mmHg for 13.5 h. Puffed colored rice was prepared by a microwave oven at two different temperatures (180 and 200°C) and times (6 and 7 min) using a 2×2 factorial in randomized complete block design. Five formulations of the healthy colored rice snack using mixture design technique were obtained. Then, the physical (texture and color), chemical (total phenolic compound, TPC; anthocyanin; DPPH assay and proximate analysis), and sensory properties of all samples were determined. The results showed that the best condition of puffed rice preparation was at 180°C for 6 min. Formulation 2, which consisted of 37% puffed rice, 34% concentrated pineapple juice, 4% gelatin, 5% sugar, 5% honey and 15% dried fruit, has the highest value in anthocyanin, DPPH, ash, protein, fat and crude fiber when compared to the others ($p<0.05$). The sensory using 30 panelists showed that formulation 2 had the highest value in overall appearance (6.4), color (6.5), odor (6.7), hardness (6.3), crispiness (6.5) and overall liking (7.0). Therefore, this formulation could be used for further development of healthy colored rice snack to obtain more consumer acceptability.

Keywords: colored rice, rice snack, total phenolic compound, anthocyanin, concentrated pineapple juice.

1. Introduction

Rice (*Oryza sativa* L.) is the principle cereal consumed in Asia and the primary staple food being consumed by nearly half of the world's population (Zhai *et al.*, 2001). It is also the main export product of Thailand. There are different varieties of rice that contain color pigments. The cultivars of pigmented rice have long history for people consumption, especially in Southeastern Asia (Hu *et al.*, 2003). In addition, pigmented rice composed of high content of

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phenolic compounds and it contains anthocyanin pigments with notable antioxidant (Hu *et al.*, 2003; Oki *et al.*, 2002). There is an evidence that phenolic substances act as antioxidants by preventing the oxidation of LDL-lipoprotein, platelet aggregation and damage of red blood cells (Cheynier, 2005). Anthocyanin pigments are flavonoids, a group of phenolic compounds that play an important biological role in reducing the risk of oxidative damage, cancer, and cardiovascular disease (Osawa *et al.*, 1992; Harborne, 1997; Harborne and Williams, 2000; Lazze *et al.*, 2003; Acquaviva *et al.*, 2003).

Pineapple (*Ananus comosus*) is the most popular fruit in Chiang Rai province. It is a wonderful tropical fruit having exceptional juiciness, vibrant tropical flavour and immense health benefits. There are many cultivars with various in size, shape, odor, and flavor. It contains important health-promoting compounds that have protective or disease preventive properties such as bromelain and vitamin C (Kongsuwan *et al.* (2009). Bromelain act as anti-inflammatory, immune system modulator, and it helps to enhance of absorption of drugs and inhibit of platelet aggregation (Taussig and Batkin, 1988). According to Joy (2001), vitamin C is the body's primary water soluble antioxidant, against free radicals that attack and damage normal cells.

Snack foods are commonly foods that are eaten between main meals. According to Fellows and Hilmi (2011), snack foods have a tendency to have a lower nutritional value because most of them provide significantly amounts of carbohydrate and fat. Colored rice and pineapple contain high amount of bioactive compounds, which promote human health by reducing the concentration of radioactive oxygen species and free radicals. Therefore, objective of this study was to develop colored rice snack containing concentrated pineapple juice. Then physical, chemical and sensory qualities of the products were determined.

2. Materials and Methods

2.1 Materials

Pineapple (100% yellow color of fruit peel) was purchased from Nang Lae district, Chiang Rai, Thailand. Colored rice (Smile Rice Brand) was purchased from Chaiyaphum province. Gelatin (Telephone Brand), dried fruit (Confetth Brand), brown sugar (Mitr Phon Brand), honey (Yellow Farm Brand) and 100% apple juice (Malee Brand) were purchased from supermarket in Chiang Rai.

All chemicals were analytical grade. Sodium hydroxide, potassium chloride, sodium carbonate, ethanol, methanol, sulfuric acid, concentrated hydrochloric acid, petroleum ether, boric acid, copper sulfate, potassium sulfate were obtained from Quality Reagent Chemical

(QReC[®], Thailand). The 2,2-Diphenyl-1-picryl-hydrazyl radical, and gallic acid were purchased from SIGMA-ALDRICH[®] INC., USA. Folin-ciocalteu's phenol reagent was purchased from Merck Company, Germany. Peptone water, plate count agar and potato dextrose agar were purchased from Criterion Company, USA.

2.2 Methods

2.2.1 Preparation of concentrated pineapple juice

Pineapple was manually peeled and the juice was delivered by hydrolic pressure. Concentrated pineapple juice (50°Brix) was prepared by vacuum evaporator (EVAP-50) at 50°C with the rate of agitation at 85.0 SP and 645 mmHg for 13.5 h (Nindo *et al.*, 2007). Samples were kept in LDPE plastic bag at -40°C.

2.2.2 Optimal condition of puffed rice preparation

According to method of Maisont and Narkrugsa (2010), rice was soaked in distilled water and 2% salt solution (NaCl) for 24 h. at room temperature. Then rice was drained and cooked by a steamer for 1 h. After that, cooked rice was dried at 50°C for 11 h using a tray-dryer, until the moisture content was 11%. Puffed rice was prepared by a microwave oven (SHARP/R-7A56) at two different temperatures (180 and 200°C) and times (6 and 7 min) using a 2×2 factorial in randomized complete block design. Puffed rice were determined for physical and chemical quality. The best condition of puffed rice was selected to make healthy colored rice snack containing concentrated pineapple juice.

2.2.3 Optimal formulation of colored rice snack containing concentrated pineapple juice

Formulation of rice snack was performed using mixture design technique. The factors were percentage of puffed rice, concentrated pineapple juice and gelatin were 40–50%, 45–55% and 5–10%, respectively. Five formulations of healthy colored rice snack containing concentrated pineapple juice were then summarized as shown in Table 1. In order to improve flavor of the product, 100% apple juice was mixed with concentrated pineapple juice at the ratio 1 : 4 before mix with other ingredients.

Brown sugar was dissolved with honey and concentrated pineapple juice. Then, gelatin was gradually added and stirred until it completely dissolved. Next, puffed rice and dried fruit were added. The mixture was pressed in a molding block and baked at 130°C for 15 min. Colored rice snacks were left at room temperature until it was cool down and removed from the molding block. The snacks were cut into pieces (1.5 cm × 3.5 cm × 0.7 cm) and dried at 70°C for 45 min in a desk oven. After leave at room temperature until cool, the samples were packed in laminated plastic bag for the determination of physical, chemical and sensory characteristics.

Table 1 Composition of colored rice snack containing concentrated pineapple juice

Formula	Percent (%)					
	Puffed rice	Conc. pineapple juice	Gelatin	Brown sugar	Honey	Dried fruit
1	30	41	4	5	5	15
2	37	34	4	5	5	15
3	34	34	7	5	5	15
4	30	38	7	5	5	15
5	33	36	6	5	5	15

2.2.4 Physical analysis

Determination of color

According to Sun-Waterhouse *et al.* (2010), color of healthy colored rice snack containing concentrated pineapple juice was measured in triplicate using a colorimeter (Color Quest XE) and expressed as CIE L*, a*, b* values (L* value defines the lightness, a* value the red-greenness and b* value the blue-yellowness, respectively).

Determination of texture

The hardness of the sample was measured in triplicate using a texture analyzer (TA.XT. Plus texture analyzer, UK). The force (N) required for a knife probe to cut 10 mm into the sample was measured (Sun-Waterhouse *et al.*, 2010).

2.2.5 Sample extraction

Two grams of ground sample was extracted with 80% methanol for 6 h at room temperature (27°C) with shaking. The total volume of extract was adjusted to 50 ml with methanol 80%. Then samples extract were centrifuged with ultrasonic centrifuge (BECKMEN/AVANTI J-301) at 2500 g for 10 min. Sample extract were stored at -40°C until use for measurement of total phenolic compound, anthocyanin and DPPH assay (Chen *et al.*, 2012).

2.2.6 Chemical analysis

Determination of total phenolic compound (TPC)

TPC was determined using the Folin-Ciocalteu method (Yao *et al.*, 2004). One thousand µg/ml gallic acid stock solution was prepared by dissolve 0.0100 g gallic acid with distilled water in 10 ml volumetric flask 0, 200, 400, 600, 800 and 1000 µl stock solution was then pipetted into 10 ml volumetric flask to adjust the concentration of standard to 0, 20, 40, 60, 80, and 100 µg/ml, respectively with distilled water. 0.5 ml of extracted sample or

standard was added into the test tube (triplicate). 2.5 ml of 10% v/v folin-ciocalteu's reagent and 2.0 ml of 7.5% sodium carbonate was added. Then, the mixture was left at room temperature for 1 h. The absorbance at 765 nm was detected by spectrophotometer and using water as blank.

Determination of antioxidant activity (DPPH assay)

Antioxidant activity was determined by free radical scavenging activity using the DPPH assay (Molyneux, 2004). Briefly, 50 μ l of sample extract or standard (Trolox concentration 0–1000 μ l) was mixed with 1950 μ l DPPH solution and left at dark place for 30 min. The absorbance at 517 nm was measured by spectrophotometer and methanol as blank. The percentage inhibition activity was calculated from the following equation:

$$\% \text{ inhibition} = ((A_0 - A_1)/A_0) \times 100$$

Where A_0 = Absorbance of the control; A_1 = Absorbance of the extract/standard.

Determination of total anthocyanin content

Total anthocyanin content was determined using the pH difference method (AOAC, 2005). Briefly, buffer (pH 1.0) was prepared by 1.86 g KCl and adjust pH to 1.0 (± 0.05) with 37% HCl. Buffer solution (pH 4.5) was prepared by 54.43 g $\text{CH}_3\text{CO}_2\text{Na} \cdot 3\text{H}_2\text{O}$ and adjust pH to 4.5 (± 0.05) with 37% HCl. 1 ml of extract sample was pipette into test tube (triplicate). Then, buffer solution pH 1.0 and pH 4.5 was added. The absorbance was measured at both 520 and 700 nm (absorbance need to be measured within 20–50 min of preparation). Total anthocyanin content was calculated following equation;

$$\text{Anthocyanin content (cyaniding-3-glucoside equivalents, mg/l)} = (A \times \text{MW} \times \text{DF} \times 10^3) / (\epsilon \times 1)$$

Where $A = (A_{510} - A_{700}) \text{ pH } 1.0 - (A_{510} - A_{700}) \text{ pH } 4.5$;

MW (molecular weight) = 449.2 g/mol for cyaniding-3-glucoside (cyd-3-glu);

DF = dilution factor established in D;

1 = path length in cm;

ϵ = 26900 molar extinction coefficient, in $\text{L} \times \text{mol}^{-1} \times \text{cm}^{-1}$, for cyd-3-glu;

and 10^3 = factor conversion from g to mg.

Determination of moisture content

The moisture content was determined using hot air oven according to AOAC method No. 925.10 (AOAC, 2000).

Determination of ash content

Two grams of ground sample was placed in crucible and weighed. Then, the crucibles were placed in a muffle furnace at 525°C for 4 h. Next, the crucible was removed to cool, and stored in desiccators until samples were weighed (Jame, 1995).

Determination of protein content

Protein content was determined by using Kjeldahl method (Nielsen, 2003c). One gram of ground sample was exactly weighed and placed in digestion tube. Catalyst (5 g) and concentrated sulfuric acid (12 ml) were added. Then, digestion tubes were placed on digestion block and covered digestion block with exhaust system turned on. Samples were digested until digestion completed (about 45 min, at 420°C) and the samples should be clear. Samples were then taken off the digestion block and allowed cool with the exhaust system still turn on. Twenty-five ml of boric acid solution was prepared into an Erlenmeyer flask. Sample tube was put in distillation system (Foss Tecator, Denmark). In this process, a set volume of NaOH solution will delivered to the tube and steam generator will distill the sample for set period of time (4 min). The color of boric acid was changed from red to green. Four drops of methyl red indicator were added and titrated each sample and blank (standardized HCl solution) to red color. Recorded and calculated protein content.

Determination of fat content

Two grams of ground sample was exactly weighed in a paper and placed in the thimble. The thimbles were placed in a Soxhlet extractor (Foss Tecator, Denmark). Pre-dried extraction cups were weighed and then 70 ml petroleum ether was added by dispenser. Set program, following instruction. After finished, the cup was heated in the oven at 105°C for 2 h and removed it to cool to room temperature in a desiccators (Nielsen, 2003a).

Determination of crude fiber content

Briefly, 1 g of ground sample was exactly weighed in a pre-dried crucible. Crucibles were placed in the Fibertec hot extraction (Foss Tecator, Denmark), added 150 ml of hot 1.25% w/v sulfuric acid, diatomaceous earth and filter aid. 2–4 drops of *n*-octanol (prevent foaming) were added and continue boiling for 30 min. Filter the aid, washed with hot distilled water and added 150 ml 1.25%w/v hot sodium hydroxide. Crucibles were placed in the fibertec cold extraction unit (Foss Tecator, Denmark) and filled crucible with 25 ml of acetone, leaved for 10 min and filtered. Dried the crucibles and contents in the furnace at 130°C for 2 h then cooled, and weighed. Ignited the crucible and contents in the furnace at 525±25°C for 4 h, cooled, and weighed (AOAC, 2000).

Sensory evaluation

Thirty untrained panelists were determined the sensory evaluation of healthy colored rice snack containing concentrated pineapple juice; color, crispness, odor, hardness, sweetness, overall appearance, and overall liking using a 9-Point hedonic scale (from 1 = extremely dislike to 9 = extremely like) (Altan *et al.*, 2008).

3. Results and Discussion

3.1 Optimal condition of puffed rice

Table 2 shows that temperature and time affect to physical and chemical properties of puffed rice. The lowest temperature and time (180°C, 6 min) gave significantly highest in antioxidant activity (632.21 μmol trolox/100 dry basis) and anthocyanin content (1.95 mg Cyd-3-glu/100g) ($p < 0.05$). This was due to the loss of anthocyanin content at high temperature and long time. The results showed that the loss of anthocyanin is being reflected in the decrease of antioxidant capacity. According to Patras *et al.* (2010) and Rupasinghe *et al.* (2008), anthocyanin is easily oxidized by heat treatment and susceptible to oxidative degradation during various steps of processing and storage. However, total phenolic compounds of each condition were not significantly differences ($p \geq 0.05$). In addition, moisture content of the sample at 180°C, 6 min was significantly highest (6.89%) compared to the others ($p < 0.05$). Therefore, this treatment gave the sample lowest in hardness texture (40.68 N) due to high amount of moisture ($p < 0.05$).

Table 2 Physical and chemical analysis of puffed rice

Treatment	Texture (Hardness, N)	TPC (mgGAE/100g)	DPPH (μmol trolox/100g)	Anthocyanin (mg C-3-G/100g)	Moisture (%)
180°C, 6 min	40.68±4.03 ^b	0.19±0.01	632.21±21.86 ^a	1.95±0.09 ^a	6.89±0.20 ^a
180°C, 7 min	47.77±2.69 ^a	0.19±0.01	542.79±10.23 ^c	1.88±0.11 ^a	5.67±0.40 ^b
200°C, 6 min	48.80±1.36 ^a	0.19±0.00	591.17±8.32 ^b	1.78±0.18 ^a	5.77±0.13 ^b
200°C, 7 min	46.28±2.59 ^a	0.18±0.00	568.68±12.07 ^{bc}	1.33±0.10 ^b	5.32±0.35 ^b

Note: Value are expressed as means±SD (n=3). Values in the same column sharing with different letters are expressed as significantly different ($p < 0.05$).

3.2 Optimal formulation of healthy colored rice snack containing concentrated pineapple juice

3.2.1 Determination of physical characteristics

Color value of healthy colored rice snack containing concentrated pineapple juice was determined as shown in Table 3. The L* values of formulation 4 (24.39) were appeared to be significantly lightness when compared with other formulations ($p < 0.05$). Formulation 1 showed the lowest of redness and yellowness (4.80 and 9.29, respectively). However, the texture of all samples were not significantly differences among these formulations ($p > 0.05$).

Table 3 Physical analysis of healthy colored rice snack containing concentrated pineapple juice

Formula	Color			Texture (Hardness, N)
	L*	a*	b*	
1	22.48±0.42 ^c	4.80±1.24 ^b	9.29±0.17 ^c	50.01±6.10
2	22.77±0.52 ^c	6.93±1.29 ^a	12.60±0.56 ^b	52.43±6.26
3	23.88±0.46 ^{ab}	6.19±0.47 ^a	14.74±0.91 ^a	55.37±2.79
4	24.39±0.64 ^a	7.00±0.48 ^{ab}	15.90±1.94 ^a	53.52±2.36
5	23.35±0.20 ^{bc}	7.39±0.11 ^a	16.49±0.83 ^a	53.95±9.11

Note: Value are expressed as means±SD (n=3). Values in the same column sharing with different letters are expressed as significantly different ($p<0.05$).

3.2.2 Determination of chemical characteristics

The chemical analysis of the colored rice snack containing concentrated pineapple juice is shown in Table 4. There was no significant different in TPC among five formulations however DPPH (474.49 μmol trolox/100 dry basis) and anthocyanin (2.28 mg C-3-G/100g) of formulation 2 was significantly highest compared to the others ($p<0.05$). Because of formulation 2 contained highest content of puffed colored rice (37%) which is high in anthocyanin and antioxidant activity and lead to increasing in anthocyanin and DPPH (Hu *et al.*, 2003). The moisture content of formulation 2, 3 and 4 (9.05, 9.05, 9.08%, respectively) was significantly lower than formulation 1 and 5 (13.41 and 11.81%, respectively) ($p<0.05$) because these formulations contained lower amount of concentrated pineapple juice (34–38%). Moisture content of products from formulation 2, 3 and 4 were in range of moisture content of commercial rice snack products (8.68–9.48%). In addition, formulation 2 contained highest value in protein (4.83%), ash (1.29%), fat (0.98%) and crude fiber (1.40%) content when compared with the others ($p<0.05$). It might be due to the highest content of puffed colored rice in formulation 2.

Table 4 Chemical analysis of healthy colored rice snack containing concentrated pineapple juice

Formula	TPC (mgGAE/100g)	DPPH (μmol trolox/100g)	Anthocyanin (mg C-3-G/ 100g)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Crude fiber (%)
1	0.15±0.01	422.02±10.97 ^{de}	1.64±0.08 ^b	13.41±0.32 ^a	3.72±0.09 ^d	0.50±0.03 ^e	1.16±0.02 ^b	1.23±0.04 ^b
2	0.16±0.00	474.49±7.12 ^a	2.28±0.21 ^a	9.05±0.04 ^c	4.83±0.02 ^a	0.98±0.01 ^a	1.29±0.02 ^a	1.40±0.05 ^a
3	0.16±0.00	432.59±2.36 ^{cd}	2.22±0.42 ^a	9.05±0.01 ^c	4.78±0.06 ^a	0.71±0.01 ^c	1.30±0.03 ^a	1.12±0.04 ^b
4	0.15±0.01	415.90±11.88 ^e	1.87±0.27 ^{ab}	9.08±0.02 ^c	4.07±0.05 ^b	0.62±0.02 ^d	1.29±0.03 ^a	1.22±0.14 ^b
5	0.16±0.00	454.07±4.03 ^b	2.24±0.05 ^a	11.81±0.31 ^b	3.91±0.09 ^c	0.80±0.01 ^b	1.27±0.03 ^a	1.24±0.01 ^b

Note: Value are expressed as means \pm SD (n=3). Values in the same column sharing with different letters are expressed as significantly different ($p<0.05$).

Table 5 Sensory evaluation of healthy colored rice snack containing concentrated pineapple juice

Formulation	Overall appearance	Color	Odor	Hardness	Crispness	Sweetness	Overall liking
1	5.90 \pm 1.30 ^b	6.33 \pm 1.12 ^{ab}	6.50 \pm 1.14 ^{ab}	5.67 \pm 1.02 ^b	5.57 \pm 1.28 ^b	6.57 \pm 1.07	5.97 \pm 1.25 ^b
2	6.43 \pm 1.10 ^a	6.50 \pm 1.04 ^a	6.67 \pm 0.88 ^a	6.30 \pm 0.99 ^a	6.53 \pm 1.17 ^a	6.70 \pm 0.84	6.97 \pm 0.76 ^a
3	6.50 \pm 1.25 ^a	6.50 \pm 1.07 ^a	6.40 \pm 0.93 ^{ab}	5.80 \pm 1.49 ^{ab}	6.17 \pm 1.37 ^a	6.37 \pm 1.27	6.33 \pm 1.06 ^b
4	6.43 \pm 1.19 ^a	6.40 \pm 1.25 ^{ab}	6.07 \pm 1.26 ^c	5.97 \pm 1.45 ^{ab}	6.07 \pm 1.26 ^{ab}	6.40 \pm 1.19	6.27 \pm 1.26 ^b
5	6.40 \pm 0.97 ^a	5.97 \pm 1.10 ^b	6.23 \pm 1.04 ^{bc}	6.07 \pm 1.23 ^{ab}	6.30 \pm 1.26 ^a	6.27 \pm 1.08	6.00 \pm 1.05 ^b

Note: Value are expressed as means \pm SD (n=3). Values in the same column sharing with different letters are expressed as significantly different ($p<0.05$).

3.2.3 Sensory evaluation

The sensory evaluation of healthy colored rice snack containing concentrated pineapple juice from 30 untrained panelists is shown in Table 6. Formulation 2 had significantly most acceptability of overall appearance (6.4), color (6.5), odor (6.7), hardness (6.3), crispiness (6.5), and overall liking (7.0) compared to the others ($p<0.05$).

4. Conclusion

This study found that the best condition for puffed rice preparation by a microwave oven was 180°C for 6 min. Higher of colored rice content in a formulation resulting in higher anthocyanin content and antioxidant activity of the product. When the concentrated pineapple juice increased then the moisture content of the colored rice snack was increased. The optimal formulation of colored rice snack containing concentrated pineapple juice consists of 37% puffed rice, 34% concentrated pineapple juice, 4% gelatin, 5% sugar, 5% honey and 15% dried fruit. This formulation was significant highest value of antioxidant activity, anthocyanin and nutrient compared to the others ($p<0.05$). Therefore, this formulation could be used for further development of healthy colored rice snack mixed with concentrated pineapple juice.

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