Effect of Germination Time of GABA of Karen Landrace Rice in Banka, Ratchaburi

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Abstract

Karen landrace rice, which is upland rice, has abundant amount in Banka, Ratchaburi because this landscape is optimum for cultivating. Generally, rice has many benefits, such as reduced risk of cardiovascular diseases, diabetes, obesity, and cancer due to its antioxidant activity and bioactive phytochemical compounds. Therefore, this study focused on determination of antioxidant activity, total phenolic, and GABA contents of three varieties of Karen landrace rice (Chupoeng, Nangpania, and Kaodaeng). The antioxidant activity of the brown rice (BR) and germinated brown rice (GBR) extracts was investigated using DPPH free radical scavenging assay. The total phenolic and GABA contents were also determined by colorimetric assay. GBR extracts showed significantly higher antioxidant activity and total phenolic content than BR extracts ($p<0.05$). The GBR of Chupoeng (89.92 ± 0.38 %) and Kaodaeng (109.91± 6.21 mg Gallic acid/g of extract) revealed the highest antioxidant activity and total phenolic content respectively. Moreover, an influence of germinated time was studied by varying to 0, 12, 24, and 36h. The optimum germinated time that showed the highest GABA content of rice extracts is 12 h. The 12 h showed a significantly difference and Kaodaeng (22.38 ± 0.21 mg GABA/g of dry weight) exhibited the highest GABA contents compared with other varieties.

Keywords : Karen, landrace rice, germination, GABA

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Introduction

Rice is an important cereal and is a staple food of over half the world's population (Ebuehi and Oyewole, 2008). Moreover, rice is a well-known product that is associated with reduced risk of chronic diseases such as cardiovascular disease, diabetes, obesity, and cancer (Okarter and Liu, 2010). The beneficial effects associated with rice consumption are partly due to the unique phytochemical content of rice. However, phytochemicals in Karen landrace rice have not been evaluated as much as those other whole grain.

Rice (*Oryza sativa* L.) is an important source of the human diet and contains greater levels of particular phenolic acids, such as ferulic, coumaric and caffeic acids than other grains (Ti et al. 2014; Bunzel et al. 2001; Shahidi and Naczk, 1995). These compounds are present in different fraction from rice grain. Rice grain has a hard husk protecting the kernel. When the husk is removed, brown rice remains. After removing the bran and embryo, polished rice remain that is endosperm. Normally, rice is consumed as polished rice. However, the rice bran has many amount of fiber and bioactive phytochemicals, which can improve human health (Ti et al. 2014).

In addition, an important phytochemical of rice that can be found after germinating is γ-amino-n-butyricacid or GABA. GABA plays an important role in the central nervous system and human brain. It can lower hypertension, promote the sleepiness and has thebenefit for human health. Moreover, GABA from rice germ has been used to anticancer (Kawabata et al., 1999; Mori et al., 1999). The GABA content in rice depends on rice varieties and germinated time (Varanyanond et al., 2005).

Karen landrace rice has many amounts in Banka, Ratchaburi. It is not available in the market and its information on the phytochemical and antioxidant characteristics is lacking in literature. Therefore, this study could enhance the utilization of these local rice varieties in the development of value added products. The objective of this study is to evaluate the antioxidant capacity, total phenolic content, and GABA content ofthree varieties of Karen rice; Chupoeng, Nangpania, and Kaodaeng.

Materials and Methods

Three varieties of Karen landrace rice (Chupoeng, Nangpania, and Kaodaeng) were obtained from Banka, Ratchaburi province. Each sample was separated in to two parts. First part of each rice sample was milled to separate the husks from the brown rice (BR). Another part, germinated brown rice (GBR), was germinated by varying time. The condition for germination was i) soaking for 24 h, ii) germinating for 12, 24, and 36 h at room temperature, iii) steaming rice sample at 80°C for 20 min, and iv) drying the germinated rice grains in an oven at 50°C, 48 h. The samples were packed in air tight containers.

All of brown and germinated brown rice were extracted by methanol (Arab et al., 2011). Three grams of each sample were shaken at 120 rpm for 24 h. The supernatant was removed and used as crude extract to determine the antioxidant activity, total phenolic content, and GABA content.

Antioxidant activity was determined by DPPH radical scavenging assay. This assay was modified from Madhujith and Shahidi (2009). Briefly, 200 mg/L of crude extract was added to 2 mL of 0.3 mM DPPH radical solution, which was freshly made. After 15 min of incubation at room temperature, the mixture solution was measured by UV/VIS spectroscopy (Biochrom, Cambridge, UK) at 517 nm.

Total phenolic content was analyzed by the Folinciocalteu (FC) colorimetric method Ti et al. (2014). Briefly, 125 μL of above extract was reacted with 125 μL FC reagent and 0.5 mL distilled water for 6 min. 1.25 mL of 7% sodium carbonate solution was added with water to make up the total volume to 3 mL. The mixture was incubated for 1 h at room temperature. The absorbance was measured at 765 nm using Biochromlibra s11 spectrometer (Biochrom, Cambridge, UK). Gallic acid was used as the standard. Total phenolic content was expressed as mg gallic acid equivalents (GAE) per 1 g dry weight (DW) of sample.

GABA content was analyzed by method that modified by Watchararparpaiboon et al. (2010). Briefly, 0.3 mL of extracted sample was mixed with borate buffer (pH 9) 6 mL, 1 mL of 6% phenol, and 0.4 mL of 7.5% sodium hypochlorite reagent. The mixture was reacted under cool-heat-cool condition. The absorbance was measured at 630 nm using Biochromlibra s11 spectrometer (Biochrom, Cambridge, UK).

Data were reported as mean ± SD for triplicate determinations of each sample. Different samples were analyzed with ANOVA followed by the LSD at p = 0.05.

Results and Discussion

The DPPH free radical scavenging activity method has been used to evaluate the reducing substances. The efficiency of reducing substances based on the reduction of methanolic DPPH solution in the presence of a proton-donating substance. (Soares et al., 1997).The scavenging effect of all extracts is shown in Table 1. The highest DPPH•-scavenging activity of BR extracts was found in Kaodaeng (88.40%) and the lowest activity was in Chupoeng (81.85%). For GBR extracts, the highest DPPH•-scavenging activity was in Chupoeng(89.92%) and the lowest activity was in Kaodaeng (88.63%).However, DPPH free radical-scavenging of all rice extracts was less than that of BHT (92.97%) at the same concentration.The results of DPPH free radical-scavenging of GBR show slightly higher than that of BR (Chiemela et al., 2015).

The total phenolic content was determined following modified FC colorimetric method, and results were expressed as gallic acid equivalents, shown in Table 2. Higher the total phenolic content was observed for GBR, than BR. For extracts compositions, the total phenolic content was significant greater for GBR than BR (p<0.05). For the effect of varieties, the total phenolic content shows no significant at p<0.05. The highest of total phenolic content of BR and GBR are Kaodaeng (91.88 and 109.91 mg Gallic acid/g sample, respectively). The increasing of total phenolic content of GBR compared to BR could be due to biosynthesis of phenolic compounds caused by enzyme hydrolysis during germination (He et al., 2011). Phenolic compounds are the most important contributors to the antioxidant capacity of cereal grains, and play an important role in the prevention of diseases (Zielínski et al., 2001).

The GABA contents of rice samples revealthe change depending on germinated time. The germinated time at 12 h, that is the optimum time, has the highest GABA contents of all extracts. Kaodaeng shows the highest GABA contents compared with other varieties (Table 3).For the effect of germinated time, the GABA content shows no significant between 24 and 36 h and 0 and 36 h but the GABA content of 12 and 24 h shows significant at p<0.05.

Table 1 DPPH radical-scavenging activity (%) of mathanolic BR and GBR extracts (conc. 200 mg/L) from Karen landrace rice: Chupoeng, Nangpania, and Kaodaeng

Species of rice	BR	GBR (at 12 h)
Chupoeng	81.85 ± 0.55 [a]	89.92 ± 0.38 [b]
Nangpania	85.00 ± 1.72 [a]	87.22 ± 1.27 [b]
Kaodaeng	88.40 ± 0.34 [a]	88.63 ± 0.17 [b]

Result represent mean ± standard deviation (n=3). In each line, different letters mean significant differences (p=0.05).

Table 2 Effect of germination on the total phenolic contents of three Karen landrace rice: Chupoeng, Nangpania, and Kaodaeng

Species of rice	Total Phenolic Content (mg Gallic acid/g of BR or GBR)	
	BR	GBR (at 12 h)
Chupoeng	88.05 ± 1.82 [a]	104.45± 5.68 [b]
Nangpania	69.92± 1.21 [a]	101.71± 5.76 [b]
Kaodaeng	91.88± 1.15 [a]	109.91± 6.21 [b]

Result represent mean ± standard deviation (n=3). In each line, different letters mean significant differences (p=0.05).

Table 3 GABA contents of three Karen landrace rice (Chupoeng, Nangpania, and Kaodaeng) varying germinated time.

Species of rice	GABA Content (mg GABA/g of dry weight)			
	0 h	12 h	24 h	36 h
Chupoeng	6.21± 0.14 [a]	21.28 ± 0.18	16.25 ± 0.01	13.81± 0.03 [b]
Nangpania	7.26± 0.09 [a]	22.04 ± 0.07	11.57 ± 0.05	11.50± 0.12 [b]
Kaodaeng	5.42± 0.07 [a]	22.38 ± 0.21	8.35 ± 0.04	0.42± 0.00 [b]

Result represent mean ± standard deviation (n=3). In each line, different letters mean significant differences (p=0.05).

Conclusion

Extractions of GBR from three Karen landrace rice (Chupoeng, Nangpania, and Kaodaeng) reveal the higherantioxidant capacity and total phenolic contents than extraction of BR. The GBR of Chupoeng and Kaodaeng variety show the highest antioxidant capacity and total phenolic content respectively. For germinated time, 12 h is an optimum time and has the highest GABA contents. When compared with other varieties, Kaodaeng has the highest GABA contents.

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