

# กิจกรรมต้านอนุมูลอิสระ สารประกอบฟีนอลิกทั้งหมด และปริมาณกรดแกมมาอะมิโนบิวทีริกในสารสกัดจากข้าวกล้องงอกและข้าวฮางอกของไทย

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## บทคัดย่อ

วิธีดั้งเดิมของไทยในการแปรรูปข้าวกล้องให้เป็นข้าวกล้องงอกและแปรรูปข้าวเปลือกให้เป็นข้าวฮางอก เป็นวิธีที่ช่วยในการเพิ่มคุณค่าทางโภชนาการ, ปรับปรุงเนื้อสัมผัสและเพิ่มปริมาณสารออกฤทธิ์ทางชีวภาพของข้าว ในงานวิจัยนี้ ข้าวไทย 4 พันธุ์ ได้แก่ ข้าวเหนียว, ข้าวหอมมะลิ, ข้าวเหนียวดำและข้าวหอมแดง ได้ถูกนำมาแปรรูปให้เป็นข้าวกล้องงอกและข้าวฮางอก จากนั้นจึงนำไปสกัดด้วยน้ำและนำไปวัดระดับของกิจกรรมต้านอนุมูลอิสระ สารประกอบฟีนอลิกทั้งหมดและปริมาณกรดแกมมาอะมิโนบิวทีริก (สารกาบา) ซึ่งผลการทดลองแสดงให้เห็นว่าข้าวกล้องงอกและข้าวฮางอกของทุกพันธุ์ข้าว ยกเว้นข้าวหอมแดง มีเปอร์เซ็นต์ผลได้ของการสกัดสูงกว่าข้าวที่ไม่ได้รับการกระตุ้นให้เกิดการงอก โดยเฉพาะอย่างยิ่ง สารสกัดจากข้าวฮางอกของข้าวเหนียวดำถูกพบว่ามีผลได้ของการสกัดคือ 9.39% นอกจากนี้ สารสกัดจากข้าวฮางอกของข้าวเหนียวและข้าวหอมมะลิมีกิจกรรมต้านอนุมูลอิสระสูงกว่าข้าวกล้องงอกและข้าวกล้องของพันธุ์ข้าวดังกล่าวอย่างมีนัยสำคัญทางสถิติ ซึ่งการทดสอบกิจกรรมในการดักจับอนุมูลอิสระ DPPH แสดงให้เห็นว่า ข้าวฮางอกของข้าวหอมมะลิมีกิจกรรมต้านอนุมูลอิสระสูงสุด คือ  $1.66 \pm 0.30$  มิลลิกรัมเทียบเท่า BHA ต่อกรัมน้ำหนักแห้งของข้าว นอกจากนี้ ข้าวฮางอกของข้าวเหนียวดำถูกพบว่ามีสารประกอบฟีนอลิกทั้งหมดสูงสุด คือ  $1.42 \pm 1.10$  มิลลิกรัมเทียบเท่า gallic acid ต่อกรัมน้ำหนักแห้งของข้าว ถึงแม้ว่าค่าดังกล่าวจะไม่แตกต่างอย่างมีนัยสำคัญทางสถิติกับค่าที่พบในข้าวอื่นๆ อาจเนื่องมาจากผลที่ได้จากการทดลองมีความแปรปรวนสูง การแปรรูปข้าวเหนียว, ข้าวหอมมะลิและข้าวเหนียวดำให้อยู่ในรูปข้าวฮางอกนั้นถูกพบว่าเป็นการเพิ่มปริมาณสารกาบาในข้าวได้อย่างมีนัยสำคัญทางสถิติ โดยเฉพาะอย่างยิ่ง ข้าวฮางอกของข้าวเหนียวดำและข้าวหอมมะลินั้นถูกพบว่าการเพิ่มขึ้นของสารกาบาสูงขึ้น 4 ถึง 7 เท่า

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เมื่อเทียบกับข้าวกล้องของพันธุ์ข้าวดังกล่าว ถึงแม้ว่าข้าวกล้องของข้าวหอมแดงจะถูกพบว่ามีปริมาณสารกาบาสูงที่สุด คือ  $116.07 \pm 0.91$  ไมโครกรัมต่อกรัมน้ำหนักแห้งของข้าว แต่ข้าวกล้องอกและข้าวฮางอกของข้าวชนิดนี้ถูกพบว่ามีปริมาณสารกาบาต่ำกว่ามาก การวิเคราะห์ด้วย Two-way ANOVA พบว่าพันธุ์ข้าวและวิธีการแปรรูปมีความสัมพันธ์กันและส่งผลต่อกิจกรรมต้านอนุมูลอิสระ ( $P = 0.002$ ) และปริมาณสารกาบา ( $P < 0.001$ ) อย่างมีนัยสำคัญทางสถิติ ดังนั้น ชนิดของพันธุ์ข้าวและวิธีการแปรรูปอาจจะมีบทบาทสำคัญต่อปริมาณสารออกฤทธิ์ทางชีวภาพบางชนิดในข้าว ข้อมูลที่ได้จากการวิจัยนี้อาจจะเป็นประโยชน์ต่อการพัฒนาผลิตภัณฑ์จากข้าวกล้องอกและข้าวฮางอกเพื่อนำไปใช้ในเชิงโภชนาการ การบำบัดรักษาและเครื่องสำอางในอนาคต

**คำสำคัญ:** ข้าวกล้องอก ข้าวฮางอก ผลได้จากการสกัด กิจกรรมต้านอนุมูลอิสระ สารประกอบฟีนอลิก ทั้งหมด กรดแกมมาอะมิโนบิวทิริก

# Antioxidant Activities, Total Phenolic Compounds and $\gamma$ -Aminobutyric Acid Contents of Extracts Derived from Thai Pre-Germinated Brown Rice and Pre-Germinated Rough Rice

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## ABSTRACT

Thai traditional methods for processing brown rice (BR) into pre-germinated brown rice (GBR) and rough rice into pre-germinated rough rice (GRR) have been used to increase nutritional values, improve textures and increase bioactive compounds of rice. In this study, four Thai rice cultivars, including waxy rice, jasmine rice, purple waxy rice and red hawn rice, were processed into GBR and GRR, extracted using distilled water and determined for levels of antioxidant activities, total phenolic compounds and  $\gamma$  aminobutyric acid (GABA) contents. The results showed that GBR and GRR of all rice cultivars, except red hawn rice, had higher percent yield of extraction than those without pre-germination. Especially, the extract of GRR of purple waxy rice was found to have the highest yield at 9.39%. Moreover, the extracts of waxy rice and jasmine rice in the form of GRR were found to have levels of antioxidant activities significantly higher than their GBR and BR. Indeed, DPPH scavenging assay showed that GRR of jasmine rice had the maximum antioxidant activities at  $1.66 \pm 0.30$  mg BHA equivalents/g dry weight of rice. In addition, GRR of purple waxy rice was found to have the highest total phenolic compounds at  $1.42 \pm 1.10$  mg gallic acid equivalents/g dry weight of rice, although not significantly different from the others perhaps due to high variation of data. Moreover, GRR processing method was also shown to significantly increase the levels of GABA in waxy rice, jasmine rice and purple waxy rice. In particular, GRR of purple waxy rice and jasmine rice were found to have 4-7 fold increase of GABA when compared to their

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BR. Although BR of red hawn rice was found to have the highest level of GABA at  $116.07 \pm 0.91$   $\mu\text{g/g}$  dry weight of rice, its GBR and GRR were found to have much lower GABA contents. Two-way ANOVA analysis suggested that there were significant interactions between rice cultivars and processing methods on antioxidant activities ( $P = 0.002$ ) and GABA contents ( $P < 0.001$ ). This may suggest that types of rice cultivars and processing methods may play important roles on certain rice's bioactive compounds. The data from this study could be beneficial for the future development of pre-germinated brown rice and pre-germinated rough rice products for nutritional, therapeutic and cosmetic purposes.

**Keywords:** pre-germinated brown rice, pre-germinated rough rice, yield, antioxidant activity, total phenolic compounds,  $\gamma$ -aminobutyric acid

## Introduction

Rice (*Oryza sativa* L.) is one of the world's most important staple foods. It is estimated that global milled rice production in 2012 is 488.2 million tones, of which 468.4 million tones are for consumption [1]. In many Asian countries, there are several techniques that have been used to process rice grains to have better taste, quality and shelf-life. For example, parboiling is the hydrothermal treatment that rough rice (paddy) grains with intact husks are partially boiled before being dried and stored. The grains of rough rice that are treated with heat are tougher and thus have better milling quality. Parboiling can also improve the quality of rice grains to have better texture, color, aroma, fiber, shelf-life and preservation of nutrition and bioactive compounds, particularly  $\gamma$ -oryzanol [2-4].  $\gamma$ -oryzanol in rice has been found to decrease plasma lipid and lipoprotein cholesterol concentrations as well as aortic cholesterol ester accumulation in hypercholesterolemic hamsters [5, 6].

In addition, pre-germination is also a cost-effective and simple process that uses lightly milled brown rice (BR) to be pre-germinated, steamed to stop enzymatic activities and finally dried for long term storage. The resulting product is pre-germinated brown rice (GBR). Pre-germination can activate amylolytic and proteolytic enzymes in rice grains which breakdown large substrates into small molecules and hence increase nutrition, particularly crude lipid, crude protein and glucose [7-9]. This process also improves texture and enhances several important bioactive compounds, especially  $\gamma$ -aminobutyric acid (GABA),  $\gamma$ -oryzanol, antioxidants, tocopherols, tocotrienols, phenolic compounds and ascorbic acid in a variety of rice cultivars [7, 10-12]. Consumption of GBR products that contain high levels of GABA has also been found to associate positively with recovery of psychological disorders, such as depression and memory deficit, in mouse models [13, 14].

In Thailand, particularly in the Northeast region, there is a process that uses rough rice grains with intact husks to be pre-germinated, steamed, dried and finally milled to obtain the product so-called pre-germinated rough rice (GRR) (Ministry of Science and Technology, Thailand, <http://siweb.dss.go.th>). This technique for producing GRR is easier when compared to GBR because GRR uses rough rice grains which are more convenient for pre-germination and they are also better at resisting oxidation and bacterial and fungal contaminations. Furthermore, GRR grains have very good milling quality because they are treated with heat before milling. Most importantly, GRR has been found to have the levels of GABA,  $\gamma$ -oryzanol, crude protein, total free amino acids,  $\alpha$ -tocopherol, thiamine, niacin, pyridoxine, lysine and leucine significantly higher than GBR [8, 11, 15-18].

In this study, four rice cultivars, including waxy rice, jasmine rice, purple waxy rice and red hawn rice, were used for processing into GBR and GRR which were then extracted

using distilled water and determined for levels of antioxidant activities (mg BHA equivalents), total phenolic compounds (mg gallic acid equivalents) and GABA contents ( $\mu\text{g}$ ) per gram of dry weight of rice. These results of GBR and GRR were also compared with BR (control). The knowledge obtained from this study could benefit the future development of GBR and GRR as nutritional, therapeutic and cosmetic products.

## Materials and Methods

### Rice samples

Four cultivars of *Oryza sativa* L., including waxy rice (RD6), jasmine rice (Khao Dawk Mali 105 or KDML105), purple waxy rice and red hawn rice, were purchased as rough rice from the local farmers in Udonthani which is located in the northeast region of Thailand. Laboratory de-husker was used for milling rough rice to obtain brown rice (BR) which was used as control group.

### Pre-germinated brown rice (GBR) and pre-germinated rough rice (GRR) preparations

Preparations of GBR and GRR were performed using the method previously described by Moongngarm and Saetung (2010) [11] with some modifications. GBR was prepared by steeping BR in distilled water for 12 h at room temperature (28-30°C), whilst the soaking water was changed every 4 h. Then, steeped BR was spread onto double layers of cheese clothes and then covered by another set of double layers of cheese clothes and placed into plastic container. Sterile distilled water was sprayed regularly to maintain 90-95% relative humidity for induction of pre-germination for 24 h. Then, pre-germinated brown grains were steamed for 5-10 minutes to stop enzymatic reactions and dried at 45°C to lower moisture content to be approximately 10%.

GRR were prepared by steeping rough rice in distilled water at room temperature (28-30°C) for 24 h and soaking water was changed every 6-8 h. Then, steeped rough rice was placed onto cheese clothes and put into plastic container to induce pre-germination for 24 h in the same conditions as GBR preparation. Pre-germinated rough grains were steamed for 10-15 minutes and dried at 45°C to reduce moisture content to be approximately 10%. Husks of these dried grains were removed by laboratory de-husker to obtain GRR.

### Extraction

For extraction, BR, GBR and GRR were finely ground using food processor. The rice powders were sieved (40 mesh) and extracted using sterile distilled water as solvent at the ratio of 1: 5 w/v. The mixtures were shaken at 150 rpm at room temperature (28-30°C) for 12 h and

then centrifuged at 4000 rpm for 10 min to separate supernatants from debris. The supernatants were evaporated at 45°C using vacuum rotary evaporator to obtain concentrated extracts. Percent yield of extraction was calculated as follows:

$$\% \text{ yield of extraction} = [\text{weight of extract (g)}/\text{dry weight of rice (g)}] \times 100$$

### **DPPH scavenging assay**

DPPH scavenging assay was used for evaluation of antioxidant activities of the rice extracts as described previously [20]. Extract (100 µl) was mixed with freshly prepared 0.1 mM DPPH<sup>o</sup> solution (1.9 ml). The mixture was incubated for 30 min at room temperature in the dark and used for measurement of absorbance at 517 nm. BHA (butylated hydroxyanisole) was used as standard reagent. Levels of antioxidant activities of the extracts were calculated and expressed as BHA equivalents (mg BHA equivalents/g of dry weight of rice). The experiments were performed in three replicates.

### **Total phenolic compound measurement**

Level of total phenolic compounds was measured by Folin-Ciocalteu method as previously described by Butsat and Siriamornpun (2010) [20]. Extract (200 µl) was mixed with Folin-Ciocalteu reagent (diluted 1:10 with sterile distilled water) (1 ml). The mixture was shaken for 1 min and topped up with 10% Na<sub>2</sub>CO<sub>3</sub> (800 µl). Then, sterile distilled water was used to top up the mixture to have the total volume of 5 ml. Finally, the mixture was incubated at room temperature in the dark for 2 h and used for measurement of absorbance at 760 nm. Gallic acid was used as standard reagent. Levels of total phenolic compounds of the extracts were calculated and expressed as gallic acid equivalents (mg gallic acid equivalents/g of dry weight of rice). The experiments were carried out in three replicates.

### **γ-aminobutyric acid (GABA) measurement**

Level of GABA content was measured by the Food Research and Testing Laboratory (FRTL), Faculty of Science, Chulalongkorn University, Thailand, using HPLC technique as described previously [21].

### **Statistical analysis**

Data were subjected to Two-way ANOVA analysis (SPSS version 16.0). Significant differences between the treatments were determined using Duncan's multiple range test at  $P < 0.05$ .

## Results

### Percent yield of extraction

Most of the rice cultivars were shown to have increase in % yield of extraction after pre-germination (Table 1). GRR of waxy rice, jasmine rice and purple waxy rice were shown to have higher % yield than BR and GBR of the same cultivars. The highest % yield was found in the extract of GRR of purple waxy rice (9.39%), followed by GRR of jasmine rice (8.19%) and waxy rice (5.73%). Surprisingly, the % yield of red hawn rice was found to be reduced after pre-germination.

**Table 1** Percent yield of BR, GBR and GRR extraction.

Rice cultivar	% yield of extraction		
	BR	GBR	GRR
Waxy rice	0.66	2.58	5.73
Jasmine rice	1.48	2.36	8.19
Purple waxy rice	1.31	2.28	9.39
Red hawn rice	3.84	0.42	0.78

### Antioxidant activities

In this study, DPPH scavenging assay was used to indicate the antioxidant activities of the rice extracts (Table 2). The maximum antioxidant activity was found in the extract of GRR of jasmine rice ( $1.66 \pm 0.30$  mg BHA equivalents/g dry weight of rice), followed by GRR of purple waxy rice and waxy rice ( $1.30 \pm 0.67$  and  $1.16 \pm 0.21$  mg BHA equivalents/g dry weight of rice, respectively); nonetheless, all of which were not significantly different. Furthermore, GRR of waxy rice and jasmine rice were shown to have levels of antioxidant activities significantly higher than their GBR and BR ( $P < 0.05$ ). Even though most of the rice cultivars were shown to boost certain levels of antioxidant activities after pre-germination, red hawn rice was found to have sharp decline of antioxidant activities after pre-germination. Two-way ANOVA analysis suggested that there was significant interaction between rice cultivars and processing methods on antioxidant activities ( $P = 0.002$ ).



**Table 2** Antioxidant activities of the BR, GBR and GRR.

Rice cultivar	Antioxidant activity* (mg BHA equivalents/g dry weight of rice)		
	BR	GBR	GRR
Waxy rice	0.11 ± 0.01 <sup>bcB</sup>	0.23 ± 0.03 <sup>abB</sup>	1.16 ± 0.21 <sup>abA</sup>
Jasmine rice	0.20 ± 0.04 <sup>bb</sup>	0.18 ± 0.13 <sup>abB</sup>	1.66 ± 0.30 <sup>aa</sup>
Purple waxy rice	0.65 ± 0.03 <sup>ca</sup>	0.36 ± 0.05 <sup>aa</sup>	1.30 ± 0.67 <sup>abA</sup>
Red hawn rice	0.85 ± 0.04 <sup>aa</sup>	0.05 ± 0.01 <sup>bb</sup>	0.15 ± 0.08 <sup>bb</sup>

**\*Note:**

1. Levels of antioxidant activities in the same column with the same lower-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .
2. Levels of antioxidant activities in the same row with the same upper-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .

**Levels of total phenolic compounds**

GRR of waxy rice, jasmine rice and purple waxy rice were found to have high levels of total phenolic compounds, despite no significant differences from GBR and BR (Table 3). GRR of purple waxy rice was shown to have the highest level of total phenolic compounds ( $1.42 \pm 1.10$  mg gallic acid equivalents/g dry weight of rice), followed by GRR of jasmine rice ( $0.75 \pm 0.53$ ); however both were not significantly different. Similar to the results of antioxidant activity measurement, most of the rice cultivars were shown to enhance certain levels of total phenolic compounds after pre-germination with the exception of red hawn rice that a large amount of total phenolic compounds was greatly reduced. There was no significant interaction between rice cultivars and processing methods on the levels of total phenolic compounds as indicated by Two-way ANOVA analysis ( $P = 0.284$ ).

**Table 3** Levels of total phenolic compounds of BR, GBR and GRR.

Rice cultivar	Total phenolic compounds* (mg gallic acid equivalents/g dry weight of rice)		
	BR	GBR	GRR
Waxy rice	0.02 ± 0.001 <sup>cA</sup>	0.03 ± 0.002 <sup>bA</sup>	0.06 ± 0.03 <sup>aA</sup>
Jasmine rice	0.08 ± 0.005 <sup>bA</sup>	0.05 ± 0.003 <sup>abA</sup>	0.75 ± 0.53 <sup>aA</sup>
Purple waxy rice	0.02 ± 0.003 <sup>cA</sup>	0.12 ± 0.05 <sup>aA</sup>	1.42 ± 1.10 <sup>aA</sup>
Red hawn rice	0.44 ± 0.03 <sup>aA</sup>	0.01 ± 0.006 <sup>bB</sup>	0.10 ± 0.06 <sup>ab</sup>

**\*Note:**

1. Levels of total phenolic compounds in the same column with the same lower-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .
2. Levels of total phenolic compounds in the same row with the same upper-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .

**GABA contents**

Waxy rice, jasmine rice and purple waxy rice were shown to have significant increase in their levels of GABA after processing into GRR, especially in GRR of jasmine rice and purple waxy rice with approximately 7-fold and 4-fold increases, respectively, when compared to BR (Table 4). GRR processing method was significantly superior to GBR in boosting the levels of GABA in waxy rice, jasmine rice and purple waxy rice. Even though BR of red hawn rice was found to have the highest GABA contents at  $116.07 \pm 0.91 \mu\text{g/g}$  dry weight of rice, its GBR and GRR were found to have much lower GABA contents. There was a significant interaction between rice cultivars and processing methods on GABA contents, as revealed by Two-way ANOVA ( $P < 0.001$ ).

**Table 4** Levels of GABA in BR, GBR and GRR.

Rice cultivar	GABA ( $\mu\text{g/g}$ dry weight of rice)*		
	BR	GBR	GRR
Waxy rice	$48.31 \pm 0.19^{\text{bB}}$	$32.78 \pm 0.58^{\text{aC}}$	$58.04 \pm 0.21^{\text{aA}}$
Jasmine rice	$4.38 \pm 0.11^{\text{dC}}$	$7.93 \pm 0.03^{\text{bB}}$	$28.79 \pm 0.65^{\text{bA}}$
Purple waxy rice	$7.19 \pm 0.01^{\text{cB}}$	$4.09 \pm 0.09^{\text{dC}}$	$26.81 \pm 0.21^{\text{cA}}$
Red hawn rice	$116.07 \pm 0.91^{\text{aA}}$	$5.33 \pm 0.07^{\text{cB}}$	$2.68 \pm 0.01^{\text{dC}}$

**\*Note:**

1. Levels of GABA in the same column with the same lower-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .
2. Levels of GABA in the same row with the same upper-case letter(s) are not significant different as determined by Duncan's multiple range test at  $P < 0.05$ .

**Discussion**

Pre-germination of rice grains is a practical and inexpensive method for making rice to have better quality and more nutrition and bioactive compounds. The common practice for pre-germination generally uses de-husked brown rice for pre-germination, which has been previously found to increase a number of bioactive compounds, especially GABA [22, 23]. In this study, Thai traditional method for pre-germination of rough rice with intact husks was employed and compared to the common practice. [24]. Here, the rice of colorless cultivars, including waxy rice and jasmine rice, and pigmented cultivars, including purple waxy rice and red hawn rice, were pre-germinated and processed into GBR and GRR and their levels of bioactive compounds were compared with their non-germinated BR.

Only waxy rice and jasmine rice that were processed into GRR were shown to have levels of antioxidant activities and GABA contents significantly higher than their GBR and BR. These results were similar to the previous reports that pre-germination of rough rice could increase levels of antioxidant activities and GABA contents better than pre-germination of brown rice [11, 15, 16-18]. This may be due to that GRR processing method involves pre-germination of grains while they are still in their husks and therefore all the bioactive compounds are efficiently maintained within their grains and protected from oxidation. However, GRR processing did not seem to significantly increase the levels of total phenolic compounds of all rice cultivars perhaps due to high variation (standard errors) of the data. Two-way ANOVA

analysis was conducted to examine the effect of rice cultivars and processing methods on the levels of total phenolic compounds, antioxidant activities and GABA contents. The results of analysis indicated that there were significant interactions between rice cultivars and processing methods only on the levels of antioxidant activities ( $P = 0.002$ ) and GABA contents ( $P < 0.001$ ). This may suggest that GRR processing may be beneficial for enhancing antioxidant activities and GABA contents of only rice cultivars used in this study.

However, GBR processing of all rice cultivars did not significantly enhance their levels of antioxidant activities, total phenolic compounds and GABA contents, when compared to their BR. In particular, GABA in GBR of waxy rice, purple waxy rice and red hawn rice were significantly lower than their BR. This may be due to the fact that GBR processing induces pre-germination of grains when their husks are already removed and therefore their bioactive compounds can be easily lost.

For red hawn rice, its GBR and GRR did not have higher levels of antioxidant activities, total phenolic compounds and GABA than its BR. This may be due to the fact that GBR and GRR of red hawn rice has much lower % yield of extraction than its BR. Therefore, when compared gram-by-gram of dry weight of rice, GBR and GRR were found to have levels of antioxidant activities, total phenolic compounds and GABA significantly lower than BR. Methods to increase % yield of extraction of red hawn rice, perhaps by changing the solvents for extraction, remain to be determined in the future experiments.

## Conclusion

In this study, only GRR processing seems to be beneficial for enhancing antioxidant activities (in waxy rice and jasmine rice) and GABA contents (in waxy rice, jasmine rice and purple waxy rice) and therefore GRR should be applied for future enhancement of bioactive compounds in rice. The results from this study may also suggest that processing methods for obtaining GBR and GRR may have different effects on different rice cultivars. Some bioactive compounds may be boosted after pre-germination, but some may not. Hence, to enhance the value of processed rice, the types of processing methods and rice cultivars should be carefully considered as they may play important roles on certain rice's bioactive compounds.

## Acknowledgement

We would like to thank to the Department of Biotechnology, Faculty of Technology, Mahasarakham University for providing laboratory facility. Thanks to Miss Rungkarn Changkul and Miss Manashanok Khamshompu for laboratory assistance. This work is financially supported by Mahasarakham University Research Funding and New Researcher Scholarship of CSTS (MOST) from Coordinating Center for Thai Government Science and Technology Scholarship Students (CSTS), National Science and Technology Development Agency (NSTDA).

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