

## Evaluation of GABA, crude protein and amino acid composition from different varieties of Malaysian's brown rice

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### Abstract

The main objective of present study was to investigate the  $\gamma$ -aminobutyric acid (GABA), crude protein and total glutamic acid composition of Malaysian's brown rice varieties. Significant ( $p < 0.05$ ) difference was observed among the content of GABA, crude protein and total glutamic acid of thirty five Malaysian's brown rice varieties. As shown in results, GABA contents in Malaysian brown rice seeds ranged between 0.01 and 0.1 (mg/g). The quantity of glutamic acid and protein contents varied between 10.1-15.2 (mg/g) and 6.99-10.17 %, respectively. A significant ( $p < 0.05$ ) positive correlation ( $r^2 = 0.820$ ) exists between the concentration levels of protein and glutamic acid. On the other hand, a significant ( $p < 0.05$ ) positive correlation ( $r^2 = 0.507$ ) was also observed between the glutamic acid and GABA contents.

**Key words:** Brown rice;  $\gamma$ -Aminobutyric acid; Glutamic acid; Protein

**Abbreviations:** GABA\_  $\gamma$ -aminobutyric acid; GAD\_ glutamate decarboxylase; AABA\_ L- $\alpha$ -amino-n-butyric acid; PITC\_ phenylisothiocyanate

### Introduction

Due to increasing market demands on protein ingredients, several studies have been carried out on the novel proteins obtained from various sources (Lamsal et al., 2007; El Nasri and El Tinay, 2007). However, for a novel protein to be useful for food processing application, it should possess desirable functional and nutritional qualities (Mu et al., 2009). In many countries, particularly in tropical regions of the world, cereal grains, legumes or beans, roots and tubers are an important part of human diet (Emmy Hainida et al., 2008). Rice (*Oryza sativa* L.) is one of the most important crops for more than a third of the world's population in addition to wheat and corn.

World production of rice has risen steadily from about 200 million tons of paddy rice in 1960 to 645 million tons in 2007. Rice accounts for over 22% of global energy intake. More than 90% of the world's rice is grown and consumed in Asia. Rice is classified according to the degree of milling that makes a brown rice different from white rice. Brown rice or "hulled rice" is un-milled or partly milled rice produced by polishing process of rice. Brown rice grains are rich in more nutritional components, such as dietary fibers, phytic acids, vitamins B and E and gamma amino butyric acid (GABA) than the ordinary milled rice grains. These bio-functional components are

found in the germ and bran layers (Champagne et al., 2004).

GABA is a well known non-protein amino acid that is widely distributed throughout the biological world, and has been reported to accumulate in plants and mammalian tissues; also in bacteria and in yeast (Wang et al., 2006; Komatsuzaki et al., 2007). It is one of the major inhibitory neurotransmitters in the central nervous system. Many neurological disorders, such as seizures, Parkinson's disease, Stiff-man syndrome, and schizophrenia are related to alterations of the GABA levels in the brain (Bao et al., 1995). Medical studies have proven that GABA plays a remarkable role in treatment of various diseases. It has long been reported to reduce blood pressure by intravenous administration in experimental animals (Stanton, 1963, Lacerda et al., 2003) and in human subjects (Elliot and Hobbiger, 1959). Several studies have shown that plant extracts containing high levels of GABA are effective for treatment of alcohol-related symptoms (Cha and Oh, 2000), blood pressure regulation (Nakagawa and Onoto, 1996), cardiovascular diseases and diabetes management (Hagiwara et al., 2004). Moreover, Okada et al. (2000) stated that intake of GABA for 8 consecutive weeks' suppressed blood pressure and improved sleeplessness, and autonomic disorder observed during the menopausal or presenile period.

GABA is primarily synthesized by the decarboxylation of L-glutamic acid which is catalyzed by glutamate decarboxylase (GAD) enzyme and pyridoxal phosphate as a cofactor (Park et al., 1999). Glutamic acid also serves as a precursor for many other substances including glutathione (Wen et al., 2004) and arginine (Donald and Carl, 1950) in addition to GABA. Glutamic acid is an amino acid found in abundance in plant proteins. Recently, an increasing interest has been focused on the utilization of GABA as a bioactive component for food. Many vegetables, fruits and different plants and products can be counted as sources of GABA. Because of the importance of GABA as a bio-functional component in brown rice, the selection of the suitable Malaysian brown rice varieties containing high GABA content looks to be necessary to satisfy the consumer demands. The main objective of this research was to evaluate the variation of GABA, crude protein and total glutamic acid contents of thirty five Malaysian's brown rice varieties.

## **Materials and methods**

### ***Reagents and standards***

L- $\alpha$ -amino-n-butyric acid (AABA), triethylamine, phenylisothiocyanate (PITC), amino acid standard

AAS18 and GABA standard were purchased from Sigma-Aldrich (St. Louis, MO, USA). Acetonitrile, methanol (HPLC grade), sulphuric acid (nitrogen-free), sodium hydroxide, hydrochloric acid (37%) and ammonium acetate were provided by Merck (Darmstadt, Germany). Glacial acetic acid ( $\geq 99.7\%$ ) was supplied by Fisher Scientific, (Leics, UK).

### ***Malaysian brown rice varieties***

Thirty five Malaysian brown rice varieties cultivated by Malaysian Agriculture Research and Development Institute (MARDI) were used as our samples in this study. Samples were harvested in 2005 and stored in a refrigerator at 4 °C for 6 months. Cleaned paddy rice was de-husked by using a Satake rice machine (Satake Engineering Co. LTD, Tokyo, Japan), then the husked brown rice was ground into flour by a mortar and a pestle and subsequently sieved through a 355  $\mu\text{m}$  screen for analytical use.

### ***Crude protein analysis***

The crude protein content of 35 brown rice varieties were measured according to the AOAC (1995) official method and protein content was calculated by multiplying conversion factor of 5.95 (Ohtsubo et al., 2005).

### ***Quantitative analysis of GABA and total amino acids***

Total amino acid and GABA contents was determined by using a reversed-phase high performance liquid chromatography (RP-HPLC) (Rozan et al., 2000). Brown rice seeds were weighed (the weights of samples were based on the percentage of their protein divided by four) and hydrolyzed by adding 15 ml 6N HCl to the sample, mixing well in a stoppered test tube for 24 hour at 110 °C. Hydrolysates were derivatized with phenylisothiocyanate and subsequently, 20  $\mu\text{L}$  of the sample was injected into the HPLC system equipped with HPLC Photodiode Array Detector (model MD-2010; JASCO, Tokyo, Japan). The linear gradient system was used with buffer A (0.1 M ammonium acetate, pH 6.5) and buffer B (0.1 M ammonium acetate containing acetonitrile and methanol, 44:46:10, v/v, pH 6.5) at flow-rate of 1 ml/min, by using a C18 reversed phase column (Thermal C18 5U, 250  $\times$  4.6 mm) in an oven at 43 °C. The UV absorption detection at a wavelength of 254 nm was employed to measure the total content of amino acids. The results were analyzed by using the Borwin chromatography software (Version 1.5, Jasco Co. Ltd., Japan).

**Table 1.** Total amino acid profile and protein contents of brown rice varieties (mean  $\pm$  standard deviation) expressed as mg/g

Variety	Asp	Glu	Ser	Gly	His <sup>a</sup>	Arg <sup>a</sup>	Thr <sup>a</sup>	Ala	Pro	Tyr
Malinja	6.9 $\pm$ 0.2	12.4 $\pm$ 0.4	5.8 $\pm$ 0.4	3.2 $\pm$ 0.2	1.2 $\pm$ 0.3	3.9 $\pm$ 0.3	1.7 $\pm$ 0.06	4.2 $\pm$ 0.4	3.2 $\pm$ 0.3	1.4 $\pm$ 0.4
Mahsuri	8.2 $\pm$ 0.2	14.5 $\pm$ 0.4	4.6 $\pm$ 0.06	3.8 $\pm$ 0.1	1.5 $\pm$ 0.2	4.7 $\pm$ 0.1	2.4 $\pm$ 0.2	4.8 $\pm$ 0.6	3.7 $\pm$ 0.3	0.7 $\pm$ 0.2
Ria	7.1 $\pm$ 0.1	11.8 $\pm$ 0.6	4.7 $\pm$ 0.1	3.6 $\pm$ 0.4	1.5 $\pm$ 0.1	3.8 $\pm$ 0.2	2.0 $\pm$ 0.1	4.3 $\pm$ 0.2	3.2 $\pm$ 0.1	1.5 $\pm$ 0.1
Bahagia	7.3 $\pm$ 0.2	13.3 $\pm$ 0.2	3.5 $\pm$ 0.2	3.4 $\pm$ 0.2	1.4 $\pm$ 0.4	4.1 $\pm$ 0.1	2.1 $\pm$ 0.1	4.3 $\pm$ 0.2	3.4 $\pm$ 0.1	1 $\pm$ 0.1
Murni	8 $\pm$ 0.4	13.6 $\pm$ 0.4	3.6 $\pm$ 0.3	3.8 $\pm$ 0.5	1.3 $\pm$ 0.2	4.3 $\pm$ 0.2	2.3 $\pm$ 0.2	4.7 $\pm$ 0.09	3.8 $\pm$ 0.2	0.9 $\pm$ 0.1
Masria (P)	7.9 $\pm$ 0.2	14 $\pm$ 0.4	3.7 $\pm$ 0.2	4 $\pm$ 0.1	1.5 $\pm$ 0.3	4.7 $\pm$ 0.5	2.6 $\pm$ 0.2	4.8 $\pm$ 0.1	3.7 $\pm$ 0.1	1 $\pm$ 0.1
Jaya	7.6 $\pm$ 0.5	14 $\pm$ 0.7	4.3 $\pm$ 0.5	3.9 $\pm$ 0.2	1.5 $\pm$ 0.2	4.4 $\pm$ 0.3	2.5 $\pm$ 0.0	4.8 $\pm$ 0.2	3.8 $\pm$ 0.3	0.9 $\pm$ 0.1
S.Malaysia. I	6.4 $\pm$ 0.3	12.6 $\pm$ 0.4	3.6 $\pm$ 0.2	3.4 $\pm$ 0.3	1.1 $\pm$ 0.1	4.1 $\pm$ 0.1	1.9 $\pm$ 0.5	4.5 $\pm$ 0.1	3.3 $\pm$ 0.2	1 $\pm$ 0.1
S.Malaysia. II	6.9 $\pm$ 0.3	12.5 $\pm$ 0.2	3.7 $\pm$ 0.6	3.2 $\pm$ 0.2	1 $\pm$ 0.1	4.3 $\pm$ 0.2	1.7 $\pm$ 0.2	4.2 $\pm$ 0.4	3.1 $\pm$ 0.1	0.8 $\pm$ 0.0
P.Malaysia. I	6.7 $\pm$ 0.2	12.6 $\pm$ 0.3	3.6 $\pm$ 0.2	3.4 $\pm$ 0.1	1.2 $\pm$ 0.2	4.2 $\pm$ 0.1	1.8 $\pm$ 0.4	4.2 $\pm$ 0.2	3.2 $\pm$ 0.1	0.9 $\pm$ 0.2
Setanjung	6.4 $\pm$ 0.2	12.8 $\pm$ 0.6	3.6 $\pm$ 0.1	3.4 $\pm$ 0.2	1.4 $\pm$ 0.3	3.9 $\pm$ 0.6	1.9 $\pm$ 0.4	4.2 $\pm$ 0.0	3.1 $\pm$ 0.3	1 $\pm$ 0.1
Sekencang	8.1 $\pm$ 0.2	14.1 $\pm$ 0.4	3.6 $\pm$ 0.4	3.9 $\pm$ 0.1	1.3 $\pm$ 0.1	4.4 $\pm$ 0.1	2.3 $\pm$ 0.1	4.9 $\pm$ 0.5	3.6 $\pm$ 0.2	1.3 $\pm$ 0.1
Sekembang	6 $\pm$ 0.4	11.1 $\pm$ 0.6	2.7 $\pm$ 0.2	2.8 $\pm$ 0.6	0.8 $\pm$ 0.04	3.4 $\pm$ 0.3	1.4 $\pm$ 0.0	3.5 $\pm$ 0.1	2.8 $\pm$ 0.2	1.1 $\pm$ 0.1
Kadaria	8.3 $\pm$ 0.3	14.3 $\pm$ 0.2	4.7 $\pm$ 0.1	4.3 $\pm$ 0.1	1.6 $\pm$ 0.1	4.6 $\pm$ 0.2	2.1 $\pm$ 0.1	4.6 $\pm$ 0.3	3.8 $\pm$ 0.3	1.5 $\pm$ 0.1
Pulut Siding	6.6 $\pm$ 0.2	13.2 $\pm$ 0.1	3.9 $\pm$ 0.2	3.5 $\pm$ 0.2	1.4 $\pm$ 0.2	4.2 $\pm$ 0.1	2.3 $\pm$ 0.2	4.2 $\pm$ 0.3	3.2 $\pm$ 0.1	1.4 $\pm$ 0.2
Manik	6.6 $\pm$ 0.5	11.9 $\pm$ 0.6	3.1 $\pm$ 0.07	3.2 $\pm$ 0.1	1.1 $\pm$ 0.1	3.8 $\pm$ 0.1	2.1 $\pm$ 0.08	3.8 $\pm$ 0.2	3.1 $\pm$ 0.3	1 $\pm$ 0.1
Muda	7.7 $\pm$ 0.5	13.9 $\pm$ 0.8	3.9 $\pm$ 0.1	3.7 $\pm$ 0.2	1.8 $\pm$ 0.5	4.9 $\pm$ 0.2	2.2 $\pm$ 0.1	4.5 $\pm$ 0.1	3.8 $\pm$ 0.2	2.2 $\pm$ 0.1
Seberang	7.6 $\pm$ 0.2	13.3 $\pm$ 0.5	4.4 $\pm$ 0.2	3.8 $\pm$ 0.5	1.5 $\pm$ 0.3	5.9 $\pm$ 0.3	2.8 $\pm$ 0.3	4.6 $\pm$ 0.3	3.5 $\pm$ 0.1	2.3 $\pm$ 0.1
Makmur	5.6 $\pm$ 0.2	10.1 $\pm$ 0.1	3 $\pm$ 0.1	3 $\pm$ 0.1	0.9 $\pm$ 0.05	3.5 $\pm$ 0.1	1.7 $\pm$ 0.2	3.5 $\pm$ 0.1	2.7 $\pm$ 0.3	1.1 $\pm$ 0.2
MR 84	7.2 $\pm$ 0.1	13.6 $\pm$ 0.5	3.7 $\pm$ 0.1	4.1 $\pm$ 0.1	0.9 $\pm$ 0.04	4.4 $\pm$ 0.3	2.8 $\pm$ 0.2	4.7 $\pm$ 0.2	3.6 $\pm$ 0.1	1.4 $\pm$ 0.1
MR 81	6.4 $\pm$ 0.3	11.5 $\pm$ 0.4	3.5 $\pm$ 0.2	3.3 $\pm$ 0.1	1.1 $\pm$ 0.1	3.8 $\pm$ 0.1	1.5 $\pm$ 0.1	3.8 $\pm$ 0.1	2.9 $\pm$ 0.4	1 $\pm$ 0.1
MR 103	6.5 $\pm$ 0.2	11.07 $\pm$ 0.0	3.1 $\pm$ 0.1	3.1 $\pm$ 0.2	1.1 $\pm$ 0.1	3.7 $\pm$ 0.2	1.8 $\pm$ 0.1	3.8 $\pm$ 0.4	3 $\pm$ 0.1	1.1 $\pm$ 0.1
MR 106	6.1 $\pm$ 0.3	12.2 $\pm$ 0.6	4 $\pm$ 0.1	3.1 $\pm$ 0.1	1.6 $\pm$ 0.3	4 $\pm$ 0.1	2.1 $\pm$ 0.1	4.1 $\pm$ 0.1	3.1 $\pm$ 0.1	1.7 $\pm$ 0.2
Pulut Hitam 9	6.8 $\pm$ 0.2	13.1 $\pm$ 0.1	4.4 $\pm$ 0.2	3.8 $\pm$ 0.5	1.6 $\pm$ 0.1	4.8 $\pm$ 0.6	2.5 $\pm$ 0.3	4.4 $\pm$ 0.2	3.5 $\pm$ 0.2	2 $\pm$ 0.1
MR 123	7.7 $\pm$ 0.4	14.7 $\pm$ 0.3	4.7 $\pm$ 0.2	4.4 $\pm$ 0.3	1.6 $\pm$ 0.1	5.2 $\pm$ 0.1	2.8 $\pm$ 0.3	4.8 $\pm$ 0.1	3.9 $\pm$ 0.3	2.2 $\pm$ 0.0
MR 127	6.6 $\pm$ 0.2	13.5 $\pm$ 0.4	4.4 $\pm$ 0.4	3.1 $\pm$ 0.1	1 $\pm$ 0.1	4.8 $\pm$ 0.2	2.6 $\pm$ 0.2	4.4 $\pm$ 0.2	3.6 $\pm$ 0.3	2.2 $\pm$ 0.1
MR 159	5.8 $\pm$ 0.6	11.8 $\pm$ 0.4	3.5 $\pm$ 0.2	3.4 $\pm$ 0.2	1.2 $\pm$ 0.1	3.9 $\pm$ 0.4	2.4 $\pm$ 0.2	3.8 $\pm$ 0.1	3.2 $\pm$ 0.1	1.8 $\pm$ 0.1
MR 167	7.7 $\pm$ 0.2	13.9 $\pm$ 0.1	4.6 $\pm$ 0.1	3.5 $\pm$ 0.2	1.9 $\pm$ 0.09	4.6 $\pm$ 0.1	1.9 $\pm$ 0.0	4.5 $\pm$ 0.3	3.3 $\pm$ 0.2	2.6 $\pm$ 0.1
MR 185	6.2 $\pm$ 0.4	11.2 $\pm$ 0.3	3.2 $\pm$ 0.3	3.4 $\pm$ 0.4	1.3 $\pm$ 0.1	3.7 $\pm$ 0.5	1.8 $\pm$ 0.4	3.8 $\pm$ 0.2	2.5 $\pm$ 0.2	1.6 $\pm$ 0.2
MR 211	7.2 $\pm$ 0.2	13.8 $\pm$ 0.7	4.3 $\pm$ 0.3	3.2 $\pm$ 0.1	1.5 $\pm$ 0.2	4.7 $\pm$ 0.1	2.1 $\pm$ 0.1	4.5 $\pm$ 0.1	3.5 $\pm$ 0.2	2 $\pm$ 0.1
MRQ50	7 $\pm$ 0.2	13.9 $\pm$ 0.6	4.1 $\pm$ 0.1	3.8 $\pm$ 0.1	1.3 $\pm$ 0.2	4.3 $\pm$ 0.0	2.2 $\pm$ 0.2	4.4 $\pm$ 0.0	3.3 $\pm$ 0.1	2.1 $\pm$ 0.1
MR 219	7.6 $\pm$ 0.2	13 $\pm$ 0.9	3.6 $\pm$ 0.4	3.7 $\pm$ 0.2	1.5 $\pm$ 0.2	4.7 $\pm$ 0.3	2.1 $\pm$ 0.09	4.8 $\pm$ 0.4	3.5 $\pm$ 0.3	2.3 $\pm$ 0.1
MR 220	7.9 $\pm$ 0.5	14.8 $\pm$ 0.7	4.4 $\pm$ 0.2	4.1 $\pm$ 0.1	1 $\pm$ 0.1	4.6 $\pm$ 0.1	2.0 $\pm$ 0.1	4.8 $\pm$ 0.1	3.6 $\pm$ 0.3	2.1 $\pm$ 0.1
MRQ74	9 $\pm$ 0.2	15.2 $\pm$ 0.6	3.3 $\pm$ 0.2	4.2 $\pm$ 0.3	0.8 $\pm$ 0.03	4.6 $\pm$ 0.2	2.5 $\pm$ 0.4	5.8 $\pm$ 0.3	3.8 $\pm$ 0.2	1.7 $\pm$ 0.1
MR 232	8 $\pm$ 0.3	13.6 $\pm$ 0.5	3.4 $\pm$ 0.3	3.2 $\pm$ 0.1	1.6 $\pm$ 0.1	3.9 $\pm$ 0.2	2.1 $\pm$ 0.1	5.1 $\pm$ 0.1	3.3 $\pm$ 0.1	0.8 $\pm$ 0.1
Mean	7.13	13	3.89	3.56	1.3	4.3	2.14	4.40	3.36	1.47
S.D.	0.28	0.42	0.21	0.21	0.16	0.21	0.2	0.2	0.2	0.12
CV%	3.92	3.2	5.39	5.89	12.5	4.98	9.4	4.73	5.95	8.16

TAA: Total amino acid

TEAA: Total essential amino acid

<sup>a</sup> Essential amino acid

### **Statistical analysis**

Data were expressed as the mean  $\pm$  SD in triplicate. Pearson's correlation was applied to investigate the possible correlations between GABA, glutamic acid and protein contents. The data analysis was performed using the Minitab v. 14 statistical package (Minitab Inc., PA, USA).

### **Result and discussion**

The amino acid composition of the thirty five Malaysian's brown rice varieties is shown in Table 1. There was a wide variation for most of the amino acids among the different varieties. The values of different amino acids were ranged respectively: aspartic acid (5.60-9 mg/g brown rice powder), glutamic acid (10.10-15.20 mg/g), serine (2.70-5.80 mg/g), glycine (2.80-4.40 mg/g), histidine (0.8-1.90 mg/g), arginine (3.40-5.90 mg/g), threonine (1.40-2.80 mg/g), alanine (3.50-5.80 mg/g), proline (2.50-3.90 mg/g), tyrosine (0.7-2.60 mg/g), valine (1.60-2.60 mg/g), methionine (1-1.80 mg/g), isoleucine (0.9-4 mg/g), leucine (3.13-12.30 mg/g), phenylalanine (0.8-4.50 mg/g), lysine (1.60-2.80 mg/g).

A relatively low variation of the amino acid content in milled rice was reported by previous researchers (Houston et al., 1969). Sekhar and Reddy (1982) found an extensive variation range for most of the amino acids in rice varieties. In present study we measured highly different amino acid levels in various varieties, as it is demonstrated by the coefficient of variation percent (CV%). GABA, Met and His had the highest CV%, 20, 12.5, and 14.6, respectively. However, the values obtained from the other amino acid were not as varied and had low CV%, such as Leu (1.6), Ile (3.73), Glu (3.2) and Asp (3.92) (Table 1).

The results indicated that low concentration of histidine was observed in all varieties studied and cysteine was not detected as it was destroyed during acid hydrolysis and its values are therefore not reported (Barbeau and Hilu, 1993). Among the thirty five varieties studied (see Table 1), MRQ74 had the highest level of aspartic acid (9 mg/g), glutamic acid (15.20 mg/g) and alanine (5.8 mg/g) contents; while Makmur showed the least content of aspartic acid (5.60 mg/g), glutamic acid (10.10 mg/g), valine (1.60 mg/g), methionine (1 mg/g) and lysine (1.60 mg/g). Furthermore, Sekembang had the least content of serine (2.7 mg/g), glycine (2.8 mg/g), histidine (0.8 mg/g), arginine (3.4 mg/g), threonine (1.4 mg/g), alanine (3.5 mg/g) and proline (2.8 mg/g). Mahsuri, Seberang, Malinja and Bahagia also showed the least content of tyrosine (0.7 mg/g), isoleucine (0.9 mg/g),

leucine (3.13 mg/g) and phenylalanine (0.80 mg/g), respectively. The total amino acids (TAA) ranged between 47.9 mg/g (Makmur) and 78.5 mg/g (MR123) with a CV% of 4.8. The composition of TAA in different varieties was compared with that of recommended standards of Food and Agriculture Organization (FAO). The results showed that Malaysian brown rice varieties had lower TAA values compared with FAO standard. The total essential amino acids (TEAA) were ranged from 18.9 (Makmur) to 36.1 mg/g (MR123) with a CV% of 5.18. When the composition of TEAA including lysine, methionine, threonine, leucine, isoleucine, valine, histidine, arginine and phenylalanine in different varieties were compared with (FAO) it was revealed that these values had comparable values than that of the FAO. The present results are also in agreement with the results reported in oilseed where Asp and Glu are the major abundant amino acids (Olaofe et al., 1994). Rice was found to be rich in glutamic acid, arginine, leucine, threonine and methionine, while lysine had the lowest concentrations of these amino acids. Even though lysine content of rice is the highest among the other cereals, it is the limiting amino acid for rice itself, followed by threonine and tryptophan (Sotelo et al., 1994; Sekhar and Reddy 1982). Protein content of different varieties was varied from 6.99 to 10.17 %. The highest protein content was observed in MRQ74 (10.17%), Sekencang (9.88%) and Kadaria (9.82%), respectively. Conversely, Makmur, Sekembang and MR232 had the least protein content of 6.99, 7.38 and 7.62 %, respectively (Table 1).

The present work indicated that there was a considerable difference between the GABA content of thirty five Malaysian brown rice varieties. As shown in Table 1, the difference between the highest and the lowest GABA contents was around 10 times. MRQ74 had the highest GABA contents (0.10 mg/g) among all the varieties, whereas MR 232 showed the least GABA content (0.01 mg/g). Kihara et al. (2007) reported the significant differences in GABA contents of 43 barley varieties. Saikusa et al. (1994) also observed the considerable difference in GABA concentrations of brown rice varieties. The authors explained that the observed differences could be due to genetic variations of different brown rice varieties. MRQ74 is considered to be a promising source for GABA among thirty five Malaysian brown rice varieties as it had higher GABA contents; while, negligible GABA concentration was shown in MR232 compared to the other varieties (Table 1). The results also showed that there was a considerable variation in the total glutamic acid content among thirty five varieties. MRQ74 had the highest glutamic

Table 1. Continued

Variety	Val <sup>a</sup>	Met <sup>a</sup>	Ile <sup>a</sup>	Leu <sup>a</sup>	Phe <sup>a</sup>	Lys <sup>a</sup>	GABA	Protein <sup>a</sup>	TAA	TEAA
Malinja	2 ± 0.5	1.3 ± 0.2	3 ± 0.5	3.1 ± 0.3	1.2 ± 0.2	2.1 ± 0.1	0.02 ± 0.01	8.02 ± 0.4	56.62	19.5
Mahsuri	2.2 ± 0.1	1.2 ± 0.1	3 ± 0.2	6.7 ± 0.1	1 ± 0.1	2.5 ± 0.1	0.05 ± 0.01	9.15 ± 0.1	65.5	25.2
Ria	2 ± 0.1	1.4 ± 0.3	3.3 ± 0.1	5.4 ± 0.1	1.4 ± 0.1	2.2 ± 0.1	0.05 ± 0.02	8.25 ± 0.2	59.2	23
Bahagia	2.2 ± 0.1	1.3 ± 0.1	2.9 ± 0.1	3.2 ± 0.2	0.8 ± 0.1	2.3 ± 0.2	0.02 ± 0.01	7.87 ± 0.3	56.5	20.3
Murni	2.2 ± 0.1	1.4 ± 0.2	3.3 ± 0.1	4.9 ± 0.1	1 ± 0.1	2.4 ± 0.1	0.03 ± 0.02	8.88 ± 0.1	61.5	23.1
Masria (P)	2.5 ± 0.1	1.5 ± 0.3	3.2 ± 0.1	6.7 ± 0.1	1.2 ± 0.1	2.8 ± 0.1	0.04 ± 0.01	9.7 ± 0.0	65.8	26.7
Jaya	2.1 ± 0.1	1.3 ± 0.2	3.1 ± 0.1	6.1 ± 0.1	1.2 ± 0.1	2.4 ± 0.1	0.04 ± 0.01	9.61 ± 0.3	63.9	24.6
S.Malaysia I	2.2 ± 0.2	1.3 ± 0.1	3.1 ± 0.1	5.2 ± 0.1	1.1 ± 0.0	2 ± 0.1	0.03 ± 0.01	8.21 ± 0.2	56.8	22
S.Malaysia II	1.9 ± 0.1	1.1 ± 0.1	2.8 ± 0.1	5.5 ± 0.1	1 ± 0.1	2 ± 0.1	0.02 ± 0.01	8.59 ± 0.0	55.7	21.3
P.Malaysia I	2.1 ± 0.1	1 ± 0.2	3 ± 0.1	5.7 ± 0.1	1.2 ± 0.1	2.1 ± 0.0	0.03 ± 0.01	8.46 ± 0.2	56.9	22.3
Setanjung	2.1 ± 0.0	1.2 ± 0.3	3.2 ± 0.1	5.9 ± 0.1	1.1 ± 0.1	2 ± 0.1	0.05 ± 0.03	8.2 ± 0.1	57.2	22.7
Sekencang	2.3 ± 0.1	1.5 ± 0.3	3.8 ± 0.1	7.4 ± 0.2	1.4 ± 0.1	2.5 ± 0.1	0.08 ± 0.01	9.89 ± 0.1	66.4	26.9
Sekembang	1.9 ± 0.1	1.2 ± 0.0	3.1 ± 0.1	7.6 ± 0.1	1.1 ± 0.2	1.8 ± 0.2	0.06 ± 0.02	7.38 ± 0.0	52.3	22.3
Kadaria	2.2 ± 0.2	1.1 ± 0.1	3.9 ± 0.1	6.5 ± 0.1	1.4 ± 0.1	2.4 ± 0.2	0.05 ± 0.01	9.82 ± 0.2	67.3	25.8
Pulut Siding	2.3 ± 0.2	1.3 ± 0.2	3.6 ± 0.1	4.7 ± 0.1	1.4 ± 0.1	2 ± 0.1	0.03 ± 0.01	8.06 ± 0.2	59.2	23.2
Manik	1.9 ± 0.1	1.1 ± 0.2	2.9 ± 0.1	5.8 ± 0.2	1 ± 0.1	2 ± 0.2	0.05 ± 0.02	7.65 ± 0.0	54.4	21.7
Muda	2.3 ± 0.1	1.1 ± 0.2	4 ± 0.2	4.2 ± 0.1	2.9 ± 0.2	2.3 ± 0.1	0.09 ± 0.01	8.8 ± 0.1	65.4	25.7
Seberang	2.4 ± 0.1	1.2 ± 0.1	0.9 ± 0.1	3.7 ± 0.1	3.4 ± 0.1	2.4 ± 0.1	0.08 ± 0.02	9.02 ± 0.2	63.7	24.2
Makmur	1.6 ± 0.1	1 ± 0.1	2.7 ± 0.1	4.5 ± 0.1	1.4 ± 0.1	1.6 ± 0.1	0.03 ± 0.01	6.99 ± 0.0	47.9	18.9
MR 84	2.1 ± 0.1	1.1 ± 0.1	3.7 ± 0.1	8.1 ± 0.1	1.8 ± 0.1	2.4 ± 0.1	0.05 ± 0.01	8.56 ± 0.0	65.6	27.3
MR 81	1.8 ± 0.1	1.2 ± 0.3	3 ± 0.1	5.7 ± 0.1	1.3 ± 0.1	1.9 ± 0.1	0.03 ± 0.02	8.2 ± 0.2	53.7	21.3
MR 103	1.7 ± 0.1	1.1 ± 0.2	2.9 ± 0.1	4.9 ± 0.1	2.1 ± 0.1	2 ± 0.1	0.02 ± 0.01	7.85 ± 0.1	52.9	19.6
MR 106	1.9 ± 0.1	1.1 ± 0.1	3.2 ± 0.1	8.6 ± 0.1	3.4 ± 0.1	1.9 ± 0.2	0.07 ± 0.02	8.24 ± 0.1	62.1	27.8
Pulut Hitam 9	2.3 ± 0.1	1.4 ± 0.2	3.5 ± 0.1	12.2 ± 0.1	4 ± 0.2	2.4 ± 0.1	0.06 ± 0.02	8.67 ± 0.3	72.7	34.7
MR 123	2.3 ± 0.2	1.7 ± 0.3	3.6 ± 0.1	12.3 ± 0.1	4 ± 0.1	2.6 ± 0.1	0.09 ± 0.01	9.8 ± 0.3	78.5	36.1
MR 127	2.3 ± 0.1	1.3 ± 0.2	3.6 ± 0.1	11.1 ± 0.1	4.4 ± 0.1	2.2 ± 0.1	0.09 ± 0.01	8.5 ± 0.2	71.1	31
MR 159	1.9 ± 0.2	1.2 ± 0.1	3 ± 0.2	7.8 ± 0.1	3.2 ± 0.1	1.9 ± 0.1	0.04 ± 0.02	7.63 ± 0.1	59.8	26.5
MR 167	1.9 ± 0.1	1.4 ± 0.2	3.2 ± 0.1	10.5 ± 0.1	3.9 ± 0.1	2.1 ± 0.1	0.09 ± 0.03	8.55 ± 0.1	71.5	31.4
MR 185	1.7 ± 0.1	1.3 ± 0.1	2.8 ± 0.1	10.9 ± 0.1	3.6 ± 0.2	2 ± 0.2	0.04 ± 0.02	7.69 ± 0.1	61	29.1
MR 211	2.4 ± 0.1	1.6 ± 0.2	3.7 ± 0.1	11 ± 0.2	3.4 ± 0.1	2.3 ± 0.1	0.07 ± 0.01	9.3 ± 0.1	71.2	32.7
MRQ50	2 ± 0.1	1.4 ± 0.3	3.5 ± 0.1	11.2 ± 0.1	3.4 ± 0.1	2.1 ± 0.1	0.09 ± 0.01	8.9 ± 0.3	70	31.4
MR 219	2.4 ± 0.2	1.4 ± 0.1	3.3 ± 0.1	8 ± 0.1	4.5 ± 0.1	2.5 ± 0.1	0.08 ± 0.01	8.55 ± 0.3	68.9	30.4
MR 220	2.6 ± 0.1	1.8 ± 0.4	3.5 ± 0.1	10 ± 0.2	1.9 ± 0.1	2.6 ± 0.1	0.1 ± 0.03	8.72 ± 0.2	71.7	30
MRQ74	2.5 ± 0.2	1.7 ± 0.3	3.7 ± 0.1	11.4 ± 0.1	1.3 ± 0.1	2.6 ± 0.1	0.12 ± 0.04	10.17 ± 0.2	74.1	31.1
MR 232	2.3 ± 0.1	1.2 ± 0.2	3.4 ± 0.1	4.9 ± 0.1	1.2 ± 0.2	2.3 ± 0.1	0.01 ± 0.00	7.62 ± 0.2	60.3	22.9
Mean	2.12	1.29	3.21	7.18	2.1	2.2	0.05	8.55	62.78	25.79
S.D.	0.12	0.18	0.12	0.12	0.11	0.11	0.01	0.15	3	1.33
CV%	6	14.6	3.73	1.6	5.4	5.1	20	1.8	4.8	5.18

TAA: Total amino acid; TEAA: Total essential amino acid

<sup>a</sup> Essential amino acid; \* Protein (g/100g)

acid content (15.2 mg/g), whereas Makmur showed the lowest content (10.1 mg/g) (Table 1).

The results showed that GABA content had a significant positive correlation with the glutamic acid ( $r^2 = 0.507$ ) and protein ( $r^2 = 0.501$ ) contents. As stated by Oh and Choi (2001), glutamic acid as the precursor of GABA is one of the most abundant amino acids in brown rice. Results also demonstrated that glutamic acid and protein contents also had a significant positive correlation ( $r^2 = 0.820$ ).

## Conclusions

The results of current study revealed that the amino acids variation (CV%) is different among Malaysian brown rice varieties probably due to genetic and environmental factors. Some of the rice varieties were observed to have superior concentration level of total amino acids compared to USDA. These values can be exploited in the breeding of amino acid superior rice varieties in Malaysia. Aromatic varieties such as MRQ74 possessed better amino acid profile among other varieties. The results also showed a relatively high variation between GABA contents of Malaysian brown rice varieties depending on the variety. GABA contents of the brown rice varieties having high GABA concentration (e.g. MRQ74, MRQ50 and MR 220) were 4 to 10 times higher than that of the brown rice varieties (e.g. MR 232, Malinja and Bahagia) with low GABA contents. Among Malaysian brown rice varieties some aromatic species such as MRQ74 and MRQ50 are considered to be a promising source for GABA. Also it was demonstrated that two popular Malaysian brown rice varieties (MR 219 and MR 220) had the high GABA content. In contrast, MR232 which is the last released variety (released in 2006) was found to have negligible GABA content compared to other varieties. Protein and glutamic acid contents varied in different brown rice varieties. The results revealed a positive correlation between protein, glutamic acid and GABA contents. The present study could be useful when brown rice varieties are selected based on their GABA contents for functional food consumption. Taking into consideration the novel nutraceutical applications of brown rice, difficulty of measuring GABA and glutamic acid by using HPLC, the protein content could be considered as an indicator for selection of the brown rice varieties with high GABA contents. This observation could be utilized in breeding varieties with improved amino acid composition.

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