STUDY OF MILLING PROCESS AND ITS EFFECT ON VITAMIN B1 AND FOLIC ACID CONTENTS ON LOWLAND RICE PROMISING LINES

Pengaruh Penyosohan terhadap Kandungan Vitamin B1 dan Asam Folat pada Galur-galur Harapan Padi Sawah

Siti Dewi Indrasari, Shinta Dewi Ardhiyanti and Buang Abdullah

Indonesian Center for Rice Research, Jalan Raya No. 9, Sukamandi, Subang 41256, West Java
Phone +62 260 520157, Fax. +62 260 520158, E-mail: bbpadi@litbang.deptan.go.id
Corresponding author: dewindrasari@yahoo.com

Submitted 3 January 2013; Revised 25 February 2014; Accepted 24 April 2014

ABSTRACT

Rice is an important source of vitamin B1 and folic acid, but cultivated rice cultivars contain low of these nutrient. Breeding program had been conducted and several advanced lines with a high vitamin B and folic acid contents were found. This research aimed to study the contents of vitamin B1 and folic acid in the brown rice and milled rice of fifteen promising lowland rice lines. Dried paddy (14% moisture content) were husked in the mini husker (Satake THU 35A) to obtain brown rice and milled rice by using abrasive-type mini polisher (Satake TM-05, 1,450 RPM for 2 minutes). Vitamin B1 and folic acid in the brown rice and milled rice were determined by using HPLC equipped with fluorescence detector for vitamin B1 and UV detector for folic acid. Four out of fifteen rice lines studied showed a unique characteristic of vitamin B1 content in the brown rice and milled rice. Rice lines which have highest vitamin B1 in the brown rice were B10267-4-PN-6-2-3-2-2-2-3-3-2 (3.03 mg 100 g^{-1}), B10876H-MR-2 (2.13 mg 100 g^{-1}), B10531E-KN-1-2-PN-1-4-2 (1.33 mg 100 g^{-1}) and B12411E-RS*-1-2-1 (1.21 mg 100 g^{-1}). However after milling, 43-92% of their vitamin B1 were lost; the least loss was observed in B12411E-RS*-1-2-1 (1.21 mg 100 g^{-1}) which was only 4%. Three out of fifteen rice lines studied has highest folic acid in brown rice, i.e. BP400G-PN-12-3-6 (158.5 µg 100 g^{-1}), B10876H-MR-2 (152.0 µg 100 g^{-1}), and B11742-RS*-2-3-MR-5-5-1-Si-1-3 (100.0 µg 100 g^{-1}). After milling the line B10876H-MR-2 had the highest folic acid content (52.0 µg 100 g^{-1}), much higher than that of Ciherang variety as a control. This study indicates the importance of having a suitable milling instrument to minimize loss of vitamin B1 and folic acid. The study also suggests the importance of creating new rice cultivars that have high nutrient and resistant to milling process.

[***Keywords*:** Rice, promising lines, milling, vitamin B1, folic acid]

ABSTRAK

Beras merupakan sumber penting vitamin B1 dan asam folat, namun kandungan kedua nutrisi tersebut pada varietas-varietas padi yang dibudidayakan masih rendah. Program pemuliaan tanaman telah mengembangkan galur harapan padi yang mengandung vitamin B1 dan asam folat tinggi. Penelitian ini bertujuan mempelajari kandungan vitamin B1 dan asam folat pada beras pecah kulit dan beras giling dari 15 galur padi sawah. Gabah kering giling diproses menjadi BPK menggunakan Satake Mini Husker *dan BG disosoh menggunakan* Satake Mini Polisher *selama 2 menit. Vitamin B1 pada beras pecah kulit dan beras giling diukur menggunakan HPLC dengan detektor fluoresen, sedangkan asam folat menggunakan detektor UV. Empat dari 15 galur harapan padi mempunyai karakteristik yang unik pada kandungan vitamin B1 pada beras pecah kulit dan beras giling. Galur padi yang mengandung vitamin B1 tinggi pada beras pecah kulit yaitu B10267-4-PN-6-2-3-2-2-2-3-3-2 (3,03 mg 100 g^{-1}), B10876H-MR-2 (2,13 mg 100 g^{-1}), B10531E-KN-1-2-PN-1-4-2 (1,33 mg 100 g^{-1}), dan B12411E-RS*-1-2-1 (1,21 mg 100 g^{-1}). Namun setelah disosoh, 43-93% kandungan vitamin B1 hilang ,yang paling sedikit pada galur B12411E-RS*-1-2-1 (1,21 mg 100g^{-1}) yaitu hanya 4%. Tiga dari 15 galur padi mengandung asam folat tinggi, yaitu BP400G-PN-12-3-6 (158,5 µg 100 g^{-1}), B10876H-MR-2 (152,0 µg 100 g^{-1}), dan B11742-RS*-2-3-MR-5-5-1-Si-1-3 (100,0 µg 100 g^{-1}). Setelah proses penyosohan, galur B10876H-MR-2 mempunyai kandungan asam folat tertinggi (52,0 µg 100 g^{-1}), jauh lebih tinggi dibanding Ciherang sebagai kontrol. Studi ini mengindikasikan pentingnya alat penyosoh yang dapat meminimalkan kehilangan kandungan vitamin B1 dan asam folat. Studi ini juga menyarankan pentingnya mengembangkan galur harapan padi yang mempunyai kandungan gizi tinggi dan tahan terhadap proses penyosohan.*

[***Kata kunci*:** Padi, galur harapan, penyosohan, vitamin B1, asam folat]

INTRODUCTION

Rice is the staple food of more than three billion people, mainly in Asia (Deepa *et al.* 2008). It feeds nearly half of the world's population and accounts for more than 50% of their daily calorie intake (Maclean *et al*. 2002).

Rice is an important source of vitamin B1 (Lebiedzin'ska and Szefer 2006) and folic acid needed by humans. Vitamin B1 (thiamine), the precursor of the cofactor thiamine diphosphate, is required in the processes of carbohydrates and amino acids (Jordan

2003; Settembre *et al*. 2003; Nosaka 2006). Thiamine diphosphate is synthesized in the human body from thiamine, but the body is unable to synthesize thiamine *denovo* (new thiamine). Severe deficiency of vitamin B1 causes *beri-beri* disease in human (Lonsdale 2006). Folic acid is categorized as a water soluble vitamin B. Deficiency of folic acid is linked to birth defects, cardiovascular disease, cancer, and other health disorders (Lucock 2000; Scott *et al*. 2000).

Rice accounted for nearly 50% of total energy consumption of Indonesian people. An average daily energy consumption per capita in 2010 was 927.05 kcal (Kementerian Pertanian 2011). Approximately 94.8% of the total rice production in 2008 was from irrigated/lowland rice field. Therefore, to meet the national consumption as the primary goal of food self-sufficiency program, the Indonesian Center for Rice Research (ICRR) has conducted breeding of new rice varieties having high yield. In addition to high yield, rice quality, especially nutritional quality should be taken into consideration in breeding program. Through conventional breeding method, ICRR has developed several promising rice lines that have a high vitamin B and or folic acid content. Fifteen promising lines have low to high amylose and are resistant to bacterial leaf blight (BLB) and brown planthopper (BPH) (Table 1).

Generally, Indonesian consumed cooked milled rice with 100% milling degree; meaning rice aleuron layers rich in vitamins and minerals, including vitamin B1 and folic acid have been completely removed leading to nutritional deficiency. Based on the type of milling equipment, methods of milling from brown to milled rice can be divided into two, i.e. friction and abrasive methods (Balittan Sukamandi 1989). The friction method produces translucent milled rice because the process does not erode the endosperm of rice grains, but only in the form of inter-particle friction due to the rotation of steel. While the abrasive method produces whiter milled rice but less translucent because the rough surface of grinding stone erodes aleurone layer (Balittan Sukamandi 1989). The use of the abrasive type (Satake) produces better physical quality characteristics of rice than the Grainman friction one (Indrasari *et al*. 2006). Rice milling units in Indonesia generally use abrasive methods.

Brown rice is unpolished whole rice grain produced by removing only the hull or husk using a mortar and pestle or rubber rolls (Babu *et al*. 2009). Nutrient contents of the brown rice or whole-grains are hypothesized to contribute positively to human health due to their fiber, minerals, vitamins (e.g. vitamins B, D and E), phenolic compounds, phytoes-trogens (lignans), and other phytochemicals (Slavin *et al*. 1999). There is a significant research describing components in brown rice which have potential for nutritional impact (Rukmini and Raghuram 1991; Eggum 1979; Amissah *et al*. 2003; Goffman *et al.* 2003; Storck *et al.* 2005; Shen *et al.* 2009). Brown rice is not commonly used by Indonesian though its nutrient content is better than that of polished rice usually consumed. This lower nutrition is due to milling process that removes the outer bran (aleurone layer) of rice grain. The bran layer contains very important nutrients such as thiamine.

Villareal and Juliano (1989) studied the variety of thiamine contents of 30 developed rice varieties at the International Rice Research Institute (IRRI) and five local pigmented varieties from the Philippines. The thiamine contents of IRRI varieties ranged from 0.29 to 0.52 mg 100 g^{-1} (wet basis) and 0.33 to 0.46 mg 100 g^{-1} (wet basis) for local pigmented rice varieties. Compared to varieties from IRRI, the Philippines local varieties just have the slightest different thiamine content. Tapol variety, purple-black rice from the Philippines, had the highest content of thiamine (0.46 mg 100 g^{-1}) (Villareal and Juliano 1989; Sotelo *et al*. 1990; Wang *et al*. 1997).

Njavara is a wild variety of rice exclusively grown in Kerala, South India since ancient time, and is used mainly for Ayurvedic treatment. According to the Indian indigenous system of medicine or Ayurveda, Njavara is regarded as a special rice variety with beneficial properties for the circulatory, respiratory and digestive system. The thiamine content of Njavara (0.52 mg 100 g^{-1}) is 27-32% higher than that of Jyothi (0.35 mg 100 g^{-1}) and IR64 (0.40 mg 100 g^{-1}), the two were non-medicinal rice varieties (Deepa *et al*. 2008). Swain *et al.* (1978) reported that 0.45 mg 100g^{-1} for thiamine in wild rice is comparable to that of Njavara. The high thiamine content in Njavara rice could be useful in treating muscle weakness, neuritis and other symptoms related to deficiency of vitamin B1 (Menon 2004).

Information about rice nutrient content, especially vitamin B1 and folic acid, is important and still limited. This study will be valuable not only for consumers, but also for rice breeders in developing specific rice varieties suitable for better nutrition and production. Therefore, this research aimed to study the content of vitamin B1 and folic acid in brown rice and milled rice of 15 promising lowland rice lines.

MATERIALS AND METHODS

Experimental Design

The research was conducted in 2011 at the Grain Quality Laboratory of the Indonesian Center for Rice Research (ICRR) in Sukamandi, Subang, West Java and at the Chemistry Laboratory of the Center for Nutrition and Food at Bogor, West Java. The research was arranged in a completely randomized design with 15 promising lines and Ciherang variety (control) as the treatment. The experiment was replicated two times.

Source and Preparation of Rice Milling

Samples of rice grains of 15 lines were obtained from the Muara Experimental Station, Bogor, and Ciherang variety obtained from Sukamandi Experimental Station, Subang was used as a control (Table 1). Three lines were derived from Ciherang and the other lines were from various varieties. Almost all of the promising lines had medium amylose content, only three lines that had low amylose content, and one line had very low amylose content.

Milling Process

Dried rice grains (350 g; 14% moisture content) were husked in the mini husker (Satake THU 35A) to obtain brown rice. The brown rice was then polished in the abrasive-type mini polisher (Satake TM-05) at 1,450 RPM for 2 minutes to obtain milled rice.

Analysis of Vitamin B1 and Folic Acid

Vitamin B1 was determined according to Roche (1976). A total of 50 g samples was weighed, then added to 150 ml H_2SO_4 and then boiled (reflux) for 15 minutes.

Tabel 1. Sources and characters of 15 promising rice lines and the control Ciherang used in the study.

Promising line	Character	Crossing
B12411E-MR-9-1-3	High yield potential, resistant to bacterial beaf blight (BLB), medium amylose, resistant to brown plant hopper (BPH)	Ciherang/IR71218-39-3-2-MR-11//Ciherang
B12411E-RS*1-2-1	High yield potential, medium amylose, resistant to BLB, resistant to BPH	Ciherang/IR71218-39-3-2-MR-11//Ciherang
B10531E-KN-1-2-PN-1-4-2	High yield potential, medium amylose, resistant to BLB, resistant to BPH	Bio9-MR-V3-11-PN-5//IR64*3/IRBB 21
B10544E-KN-73-3-PN-2-2-3	High yield potential, medium amylose, resistant to BLB	Memberamo//IR51672
B10876H-MR-2	High yield potential, resistant to BLB, aromatic rice, very low amylose	HSPR-45-9/Sintanur
B10267-4-PN-6-2-3-2-2-2-3-3-2	High yield potential, medium amylose, resistant to BLB	Cina VI-2/Suban
B11822-MR-3-22-1-1-3-SI-3-2	High yield potential, medium amylose, resistant to BPH	Widas/BP342E-MR-1-3//BP342E-MR-1-3
B12411E-MR-9-5-1	High yield potential, low amylose, resistant to BLB	Ciherang/IR71218-39-3-2-MR-11//Ciherang
B11823-MR-1-27-2-3-1-SI-1-1-MR-2	High yield potential, low amylose, good quality, resistant to BPH	BP140F-MR-1-KN-1/Code//BPI40F-MR-1-KN-1
B11824E-MR-1-54-1-2-2-SI-2-3	High yield potential, low amylose, good quality, resistant to BPH	Gilirang/Code//Gilirang
B11742-RS*2-3-MR-5-5-1-Si-1-3	High yield potential, medium amylose, resistant to BPH	BP360E-MR-79-PN-2/IR71218//BP360E
B11143D-MR-1-PN-3-MR-3-SI-2-3-PN-2	High yield potential, medium amylose	Beronaja/BP364B-MR-33-3-PN-5-1//Tukad Balian
B11143D-MR-1-PN-3-MR-3-SI-2-3-PN-3	High yield potential, medium amylose, resistant to BPH	Beronaja/BP364B-MR-33-3-PN-5-1//Tukad Balian
BP400G-PN-12-3-6	High yield potential, medium amylose	B10369B/Maros//Memberamo/IR66738
B10533F-KN-11-1	High yield potential, medium amylose, resistant to BPH	IR64*3/IRBB 21//Bio9-MR-V4-5-KN-4
Ciherang	High yield potential, medium amylose	IR18349-53-1-3-1-3/3*IR19661-131-3-1-3//4*IR64

The solution was sampled and cooled. The pH was adjusted to 4.5 by adding 30 ml of 0.3 M Na acetate. Then, Taka diastase plus 0.2 g enzyme was dissolved in 10 ml of distilled water and added into the solution. The solution was left overnight at 37°C, cooled and added with 1 ml Bromocresol green indicator. The pH was set to 5 by adding 0.2 N NaOH. Distilled water was added until the volume reached 350 ml.

Subsequently, 200 ml of the filtrate was put into stoppered Erlenmeyer, then added to 100 ml alcohol, phenol, phenol red and four drops of 0.2 N NaOH until red color appeared. Then, 1.6 ml $NaNO_2$, 1.6 ml amino acetopheum (stored in refrigerator), 20 ml H_2O, 50 ml NaOH-$NaHCO_3$ and 8 ml xylol were added. The solution was allowed to settle for 1 hour, and shaken in every 15 minutes.

The solution was filtered using a vacuum separator; the solid remnant was removed and the liquid was filtered over anhydrous sodium sulphate. Further, the refined solution was analysed using HPLC (Waters 1525) according to Arella *et al*. (1996) with slight modification. One ml of sample extract was mixed with an alkaline solution of potassium ferric cyanide (1 ml of 1% potassium ferric cyanide solution and 24 ml of 3.75 M sodium hydroxide) to form fluorescent thyochrome. The content was shaken vigorously by hand, filtered through a 0.45 mm membrane filter into HPLC Chamber vials, and was injected immediately. A mixture of methanol and 0.05 M sodium acetate (30:70, v/v) were used as mobile phase and the separation was carried out with a C18 column (250 mm, 4.6 mm and 5 mm particle size). An isocratic elution was used with a flow rate of 1 ml minute^{-1}. Column temperature was arranged at 40°C and the injection volume was 20 ml. The fluorometric was arranged at a wave length of 520 nm. Vitamin B1 content was calculated with formula as follows:

$$\text{Vitamin B1} = \frac{\text{Dilution factor x sample reading/standard reading x standard concentration x 100\%}}{\text{Sample weight (g)}}$$

Folic acid was analysed using HPLC (Waters 1525). A total of 10 g of sample was added in a buffer solution containing 50 ml of 0.2 M K_3PO_4 and Na-ascorbate 0.25%. The solution was shaken for 30 minutes at 150 RPM, then filtered and pH of the filtrate was set to 4.5 with HCl solution. Then, the solution was centrifuged at 5.000 RPM at 4°C. Supernatant was taken and injected into the HPLC. The mobile phase consisted of 0.08 M K_3PO_4 and 10% acetonitrile at pH 3, and a C18 column (250 mm, 4.6 mm i.d. and 5 mm particle size). An isocratic elution was chosen with a flow rate of 1 ml minute^{-1}. Column temperature was arranged at 40°C and the injection volume was 20 ml. The UV detector was operated at a wave length of 280 nm. Folic acid concentration was calculated with the following formula:

Folic acid (µg 100 g^{-1}) = sample concentration from standard curve x dilution factor x 100

Data Analysis

The data were analysed for their variance followed with DMRT. The correlation between physical characteristics and vitamin B1-folic acid content was analyzed with Pearson correlation using SPSS 11.5 software.

RESULTS AND DISCUSSION

Vitamin B1 Content

Result of the experiment showed that the vitamin B1 content in brown rice of 15 promising lines varied from 0.23 to 3.03 mg 100 g^{-1} (Table 2). Line B10267-4-PN-6-2-3-2-2-2-3-3-2 had the highest vitamin B1 content (3.03 mg 100 g^{-1}) followed by B10876H-MR-2 (2.13 mg 100 g^{-1}), B10531E-KN-1-2-PN-1-4-2 (1.33 mg 100 g^{-1}) and B12411E-RS*-1-2-1 (1.21 mg 100 g^{-1}), much higher than that of Ciherang (0.33 mg 100 g^{-1}). But after milling, B12411E-RS * -1-2-1 (1.14 mg 100 g^{-1}) and B10531E-KN-1-2-PN-1-4 -2 (0.66 mg 100 g^{-1}) had higher vitamin B1 contents than the others, i.e. 1.14 and 0.66 mg 100 g^{-1}, respectively.

After milling, vitamin B1 content from brown rice to milled rice (80% milling degree) decreased significantly; it ranged from 34.5 to 46%, with an average of 41.8%. This value was higher than the study of Indrasari (2011) who reported that the average loss of thiamine content due to milling process was 24.6%. Previous study showed that complete milling and polishing that converted brown rice into white rice removed 80% of the vitamin B1 (Morita *et al*. 2004). Das *et al*. (2008) also reported that the mechanical milling caused 65% loss of thiamine. Most of the vitamins were contained in the outer layer of rice, therefore, removing outer layer during milling decreases significantly thiamine content (Lebiedzinska and Szefer 2006).

The study revealed that four rice lines had a unique character on vitamin B1, namely B12411E-RS*-1-2-1,

Table 2. Vitamin B1 content of 15 promising lowland rice lines and the control Ciherang.

Promising lines	Vitamin B1 (mg 100 g^{-1})		
	Brown rice (BR)	Milled rice (80% milling degree)	Decrease (%)
B12411E-MR-9-1-3	0.43abcd	0.24ab	43.5abc
B12411E-RS*-1-2-1	1.21e	1.14d	4.0a
B10531E-KN-1-2-PN-1-4-2	1.33e	0.66c	43.5abc
B10544E-KN-73-3-PN-2-2-3	0.57abcd	0.35ab	40.5abc
B10876H-MR-2	2.13f	0.35ab	45.5abc
B10267-4-PN-6-2-3-2-2-2-3-3-2	3.03g	0.23ab	91.5d
B11822-MR-2-22-1-1-3-Si-1-1	0.79cd	0.39abc	44.0abc
B12411E-MR-9-5-1	0.51abcd	0.22a	56.5bcd
B11823-MR-1-27-2-3-1-Si-1-1-MR-2	0.62abcd	0.31ab	46.0abc
B11824E-MR-1-54-1-2-2-Si-2-3	0.41abcd	0.29ab	29.0ab
B11742-RS*-2-3-MR-5-5-1-Si-1-3	0.81d	0.53bc	34.5abc
B11143D-MR-1-PN-3-MR-3-Si-2-3-PN-2	0.69bcd	0.17a	75.0cd
B11143D-MR-1-PN-3-MR-3-Si-2-3-PN-3	0.38abc	0.21a	44.0abc
BP400G-PN-12-3-6	0.63abcd	0.40abc	36.5abc
B10533F-KN-11-1	0.23a	0.17a	27.5ab
Ciherang	0.33ab	0.30ab	8.5a
Average		0.37	41.8

Means in the same column followed with the same letters are not significantly different at the 0.05 level DMRT.

B10531E-KN-1-2-PN-1-4-2, B10876H-MR-2, and B10267-4-PN-6 -2-3-2-2-2-3-3-2. Vitamin B1 content in the brown rice of B12411E -RS*-1-2-1 line was high (1.21 mg 100 g^{-1}), almost four times than that of Ciherang as a standard variety. After milling, this line contained the highest vitamin B1 (1.14 mg 100g^{-1}), therefore the vitamin B1 loss was the least (4%) compared to that in Ciherang variety (8.5%). The other two lines, i.e. B10531E-KN-1-2-PN-1-4-2 and B10876H-MR-2 although had higher vitamin B1 in the brown rice (1.33 and 2.13 mg 100 g^{-1}, respectively), vitamin B1 losses after milling were higher (43.5-45.5%) than that of B12411E -RS*-1-2-1. Therefore, the final vitamin B1 that can be consumed was low (0.66 and 0.33 mg 100 g^{-1}, respectively). B10267-4-PN-6-2-3-2-2-2-3-3-2 line had the highest vitamin B1 in brown rice (3.03 mg 100 g^{-1}), but had the highest percentage of vitamin B1 loss (91.5%). This means that vitamin B1 loss significantly reduces the availability of the vitamin for consumption. It is speculated that vitamin B1 loss in milled rice is related with the removal of outer layer which rich in nutrient, such as vitamin B1. Therefore, nutritional intake of vitamin B1 from milled rice is lower that the recommended rate for adult men and women with moderate levels of job categories, i.e. 0.9 mg and 1.1 mg per capita per day, respectively (Departemen Kesehatan RI 2004). If the average amount of vitamin B1 in rice is 0.37 mg 100 g^{-1} (Table 2) and the average consumption of rice per day is 250 g, the nutritional adequacy for vitamin B1 for adult men with moderate levels of job categories has been fulfilled.

Hoshikawa (1967) *in* Juliano (2003) showed that rice varieties differed in the thickness of the outer layer; coarser or bolder; short-grain rice have more cell layers than slender, long-grain rice. According to Ardhiyanti *et al*. (2012), the rice lines B12411E-RS*-1-2-1, B10531E-KN-1-2-PN-1-4-2, and B10267-4-PN-6 -2-3-2-2-2-3-3-2 had slender type of grain (length x width was 7.03-7.09 mm x 2.18 mm; the ratio was 3.36), whereas B10876H-MR-2 had medium type (length x width was 5.49 mm x 2.67 mm; and its ratio was 2.06). This means that grain size did not correlated with the degree of vitamin B1 loss during milling.

Folic Acid Content

The study showed that folic acid content in brown rice of the 15 promising lines ranged from 20.0 to 158.5 mcg 100 g^{-1} (Table 3). Three lines had highest folic acid content in brown rice, namely BP400G-PN-12-3-6 (158.5 µg 100 g^{1}), B10876H-MR-2 (152.0 mcg 100 g^{-1}), and B11742-RS*-2-3-MR-5-5-1-Si-1-3 (100.0 µg 100g^{-1}).

After milling, B10876H-MR-2 had the highest folic acid (52.0 µg 100 g^{-1}), much higher than that of Ciherang as a control. This was likely due to the difference in the thickness of the aleurone and the

Table 3. Folic acid content of 15 promising lowland rice lines and the control Ciherang.

Promising lines	Folic acid (mcg 100g^{-1})		
	Brown rice	Milled rice (80% milling degree)	Decrease (%)
B12411E-MR-9-1-3	32.00a	18.50abc	41.5bcde
B12411E-RS*-1-2-1	29.50a	16.00ab	45.0bcdef
B10531E-KN-1-2-PN-1-4-2	44.00ab	11.50a	74.0gh
B10544E-KN-73-3-PN-2-2-3	20.00a	14.00ab	29.0abc
B10876H-MR-2	152.00d	52.00f	65.0efgh
B10267-4-PN-6-2-3-2-2-2-3-3-2	24.00a	16.00ab	33.0abcd
B11822-MR-2-22-1-1-3-Si-1-1	35.50a	11.50a	59.5defgh
B12411E-MR-9-5-1	27.00a	15.00ab	43.5bcde
B11823-MR-1-27-2-3-1-Si-1-1-MR-2	43.00ab	19.00abc	56.0cdefg
B11824E-MR-1-54-1-2-2-Si-2-3	34.50a	28.00cde	18.5ab
B11742-RS*-2-3-MR-5-5-1-Si-1-3	100.00c	26.50cde	72.0fgh
B11143D-MR-1-PN-3-MR-3-Si-2-3-PN-2	34.00a	20.00abc	40.0bcde
B11143D-MR-1-PN-3-MR-3-Si-2-3-PN-3	66.50b	35.00e	46.0bcdefg
BP400G-PN-12-3-6	158.50d	24.00bcd	84.5h
B10533F-KN-11-1	31.00a	20.00abc	35.0abcd
Ciherang	34.00a	30.00de	10.0a
Average of percentage decreasing		47.0	

Means in the same column followed with the same letters are not significantly different at the 0.05 level DMRT.

shape of rice. The grain of rice line BP400G-PN-12-3-6 was medium shape, B11742-RS*-2-3-MR-5-5-1-Si-1-3 was slender, while the B10876H-MR-2 was medium shape.

Processing and cooking conditions cause significant loss of vitamins. The average loss in folic acid due to milling to the milled rice milling degree was 47.0%. This value was higher than the study of Indrasari *et al.*(2011) who reported that the average loss of folic acid content of brown rice into milled rice (80% milling degree) was 10.5%. This was probably due to the difference in the thickness of the aleurone layer of each line. Most of the vitamins are contained in the outer layer of the grain. The study indicates the importance of the milling process to minimize loss of aleurone layer containing high vitamin B and folic acid.

The Indonesian recommended dietary allowance figure shows that men and women adult need 400 µg of folic acid per day (Departemen Kesehatan RI 2004). Compared to these figures, the milled rice from rice line samples were not a good source of folic acid, except the BP400G-PN-12-3-6 in the form of brown rice that contain 158 mg 100 g^{-1} folic acid (Table 3). Therefore, education or promotion to consumers to consume brown rice needs to be intensified.

This study suggests that consumption of brown rice which has a higher vitamin B1 content is recommended for health rather than the white rice, in addition of developing milling equipment which can minimize loss of rice nutrient, especially vitamin B and folic acids.

CONCLUSION

Four out of fifteen rice lines showed unique characteristics on vitamin B1 content in the brown rice and milled rice. Rice lines which have highest vitamin B1 in the brown rice were B10267-4-PN-6-2-3-2-2-2-3-3-2 (3.03 mg 100 g^{-1}), B10876H-MR-2 (2.13 mg 100 g^{-1}), B10531E-KN-1-2-PN-1-4-2 (1.33 mg 100 g^{-1}), and B12411E-RS*-1-2-1 (1.21 mg 100 g^{-1}). However, after milling, 43-92% of their vitamin B1 were lost; the least was on line B12411E-RS*-1-2-1 (1.21 mg 100 g^{-1}) which was only as much as 4%.

Three out of fifteen rice lines had highest folic acid in brown rice, i.e. BP400G-PN-12-3-6 (158.5 µg 100 g^{-1}), B10876H-MR-2 (152.0 µg 100 g^{-1}), and B11742-RS*-2-3-MR-5-5-1-Si-1-3 (100.0 µg 100 g^{-1}). After milling, B10876H-MR-2 had the highest folic acid content of 52.0 µg 100 g^{-1}, much higher than that of Ciherang.

This study indicates the importance of having a suitable milling instrument that could minimize the loss of vitamin B1 and folic acid. The study also suggests the importance of creating new rice lines which have high nutrient and resistant to the milling process.

ACKNOWLEDGEMENT

The authors thank to Prof. Dr. Djoko S. Damardjati, Prof. Supriadi and Dr. Sri Yuliani for their valuable comments and suggestions on the manuscript.

REFERENCES

Amissah, J.G.N., W.O. Ellis, I. Oduro and J.T. Manful. 2003. Nutrient composition of bran from new rice varieties under study in Ghana. Food Control 14: 21-24.

Ardhiyanti, S.D., S.D. Indrasari, dan B. Abdullah. 2012. Mutu fisik dan mutu giling 30 galur harapan padi sawah. hlm 149-158. *Dalam* O.D. Hajoeningtijas, A. Shoviayani, dan D. Watemir (Ed.). Prosiding Seminar Nasional Pengembangan Teknopreneur Berbasis Pertanian untuk Meningkatkan Pemberdayaan Masyarakat. Universitas Muhammadiyah Purwokerto.

Arella, F., S. Lahely, J.B. Bourguignon and C. Hassellmann. 1996. Liquid chromatographic determination of vitamins B1 and B2 in foods. A collaborative study. Food Chem. 56(1): 81-86.

Babu, P.D., R.S. Subhasree, R. Bhakyaraj and R. Vidhyalaksmi. 2009. Brown rice-beyond the colour reviving a lost health food - a review. American-Eurasian J. Agron. 2(2): 67-72.

Balittan Sukamandi (Balai Penelitian Tanaman Pangan Sukamandi). 1989. Evaluasi dan pengembangan metode penetapan derajat sosoh beras giling. Laporan Penelitian. Kerjasama Balai Penelitian Tanaman Pangan Sukamandi dan Badan Urusan Logistik.

Das, M., R. Banerjee and S. Bal. 2008. Evaluation of physicochemical properties of enzyme treated brown rice (Part B). Food Sci. Technol. 41: 2092-2096.

Deepa, G., V. Singh and K.A. Naidu. 2008. Nutrient composition and physicochemical properties of Indian medicinal rice – Njavara. Food Chem. 106: 165-171.

Departemen Kesehatan RI. 2004. Tabel Angka Kecukupan Gizi 2004 bagi Orang Indonesia. http://gizi.depkes.go.id/download/AKG2004.pdf. (28 Oktober 2011).

Eggum, B.O. 1979. The nutritional value of rice in comparison with other cereals. Workshop on Chemical Aspects of Rice Grain Quality. International Rice Research Institute, Los Banos, Laguna, Philippines.

Goffman, F., S. Pinson and Bergman. 2003. Genetic diversity for lipid content and fatty acid profile in rice bran. J. Am. Oil. Chem. Soc. 80: 485-490.

Indrasari, S.D., Jumali, dan A.A. Daradjat. 2006. Pengaruh metode dan tingkat penyosohan terhadap kualitas beras giling dan nilai duga persentase derajat sosoh. Penelitian Pertanian Tanaman Pangan 25(3): 194-199.

Indrasari, S.D. 2011. Pengaruh penyosohan gabah dan pemasakan terhadap kandungan vitamin B beras merah. Jurnal Penelitian Pertanian Tanaman Pangan 30(3): 182-188.

Indrasari, S.D., P. Wibowo, dan S.D. Ardhiyanti. 2011. Evaluasi mutu gizi dan kandungan asam folat beberapa galur beras merah. Prosiding Seminar Ilmiah Hasil Penelitian Padi Nasional 2010 “Variabilitas dan Perubahan Iklim: Pengaruhnya terhadap Kemandirian Pangan Nasional”. Buku 3. Balai Besar Penelitian Tanaman Padi, Sukamandi. hlm. 1.293-1.300.

Jordan, F. 2003. Current mechanistic understanding of thiamine diphosphate-dependent enzymatic reactions. Nat. Prod. Rep. 20: 184-201.

Juliano, B.O. 2003. Rice Chemistry and Quality. Phil Rice, Manila. 480 pp.

Kementerian Pertanian. 2011. Pusat Data dan Informasi Pertanian. http://aplikasi.deptan. go.id/bdsp/hasil_kom.asp. (28 Oktober 2011).

Lebiedzin´ska, A. and P. Szefer. 2006. Vitamins B in grain and cereal-grain food, soy-products and seeds. Food Chem. 95: 116-122.

Lonsdale, D. 2006. A review of the biochemistry, metabolism and clinical benefits of thiamine andits derivatives. ECAM 3: 49-59.

Lucock, M. 2000. Folic acid: nutritional biochemistry, molecular biology, and role in disease processes. Mol. Genet. Metab. 71: 121-138.

Maclean, J.L., D.C. Dawe, B. Hardy and G.P. Hettel. 2002. Rice almanac. International Rice Research Institute, Los Banos, Philippines; West Africa Rice Development Association, Bouake, Cote D’Ivoire, Cali, Colombia; International Center for Tropical Agriculture, Food and Agriculture Organization. Rome, Italy. 253 pp.

Menon, M.V. 2004. The healing touch. Sci. Rep. Feb. 28-30.

Morita, H., Y. Uno, T. Umemoto, C. Sugiyama, M. Matsumoto, Y. Wada and T. Ishizuka. 2004. Effect of gamma-aminobutyric acid-rich germinated brown rice on indexes of life style related diseases. Nippon Ronen Igakkai Zasshi. March 41(2): 211-216.

Nosaka, K. 2006. Recent progress in understanding thiamine biosynthesis and its genetic regulation in *Saccharomyces cerevisiae*. Appl. Microbiol. Biotechnol. 72: 30-40.

Roche, H.A. 1976. Vitamin Compendium: The properties of vitamin and their importance in human and animal nutrition. Hoffmann-La Roche, Basle, Switzerland.

Rukmini, C. and T.C. Raghuram. 1991. Nutritional and biochemical aspects of the hypolipidemic action of rice bran oil: a review. J. Am. Coll. Nutr. 10: 593-601.

Scott, J., F. Rebeille and J. Fletcher. 2000. Folic acid and folates: the feasibility for nutritional enhancement in plant foods. J. Sci. Food Agric. 80: 795-824.

Settembre, E., T.P. Begley and S.E. Ealick. 2003. Structural biology of enzymes of the thiamine biosynthesis pathway. Curr. Opin. Struct. Biol. 13: 739-747.

Shen, Y., L. Jin, P. Xiao, Yan Lu and J.S. Bao. 2009. Total phenolics, flavonoids, antioxidant capacity in rice grain and their relations to grain colour, size and weight. J. Cereal Sci. 49: 106-111.

Slavin, J.L., M.C. Martini, D.R. Jacobs and L. Marquart. 1999. Plausible mechanisms for the protectiveness of whole grains. Am. J. Clin. Nutr. 70: 459S-463S.

Sotelo, A., V. Sousa, I. Montalvo, M. Hernandez and L. Hernandez-Aragon. 1990. Chemical composition of different fractions of 12 Mexican varieties of rice obtained during milling. Cereal Chem. 67(2): 209-212.

Stork, C.R., L.P. da Silva and C.A.A. Fagundes. 2005. Categorizing rice cultivars based on differences in chemical composition. J. Food Composition Analysis 18: 333-341.

Swain, E.W., H.L.Wang and C.W. Hesseltine. 1978. Note on vitamin and minerals of wild rice. Cereal Chem. 55: 412-414.

Villareal, C. and B.O. Juliano. 1989. Variability in the contents of thiamine and riboflavin in brown rice, crude oil in brown rice and bran-polish, and silicon in the hull of IR rices. Plant Foods for Human Nutrition 39: 287-297.

Wang, G., B. Parpia and Z. Wen. 1997. The composition of Chinese foods. Institute of Nutrition and Food Hygiene, Chinese Academy of Preventive Medicine. ILSI Press, Washington, DC.