

Changes in Nutritional Components throughout Germination in Paddy Rice and Brown Rice

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Abstract

The aim of this study was to investigate changes in 7 nutritional components (fatty acid, protein, fat, ash, total dietary fiber (TDF), γ -aminobutyric acid (GABA), and γ -oryzanol) of paddy rice (PR) and brown rice (BR) throughout the germination process, as measured at different shoot lengths (10 mm, 20 mm, and 30 mm). With the increase of shoot length, the nutritional components' concentrations increased, as compared to the concentrations measured before germination. Moreover, BR exhibited higher GABA, γ -oryzanol, and protein than PR. Among the components, TDF, GABA, and γ -oryzanol showed significant concentration differences throughout germination, while the others exhibited only slight variations. In particular, GABA and γ -oryzanol were predominantly increased in grains of 10 mm shoot length. These compounds might prove to be important factors from germinated rice. Additionally, the germinated cultivar 'Keunnun' might also prove to be a very important food source, owing to its high GABA and γ -oryzanol contents. These results suggest that variations in nutritional components related to the increase of shoot length may prove to be important when considering the beneficial aspects of rice on human health.

Key words: germination, shoot length, GABA, γ -oryzanol, paddy rice, brown rice

INTRODUCTION

Rice (*Oryza sativa* L.) is consumed as a staple food around the world because of its many nutritional functions. Numerous components in rice can be improved by biofortification through cultivar production and selective breeding, which are important approaches to enhancing the quality and quantity of food (1). Moreover, scientific technologies of genetic modification make it possible to enhance rice's nutritional values. In spite of the developed methods concerning increase of biofunctional components, rice is not considered suitable because of its safety. Additionally, many researchers have reported that changes in functionalities were associated with processing technologies (2). It is well established that germination is a processing method by which the quality of cereal can be improved for physiological and nutritional functions (3,4). During germination, hydrolytic enzymes and biological components are activated, while starch, polysaccharide, and amino acid are decomposed (3,5,6). The decomposition of polymers in germinated cereal leads to the generation of functional materials (7). Although several reports in the literature demonstrate the presence of functional components during germination (4,8), there is a lack of information on changes in various nutritional components through germination stage.

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Rice has many nutritional components, including carbohydrates, protein, oil, dietary fiber, vitamin, γ -amino butyric acid (GABA), and γ -oryzanol (9-11). GABA is involved in the regulation of certain cardiovascular functions, such as blood pressure and heart rate (12) as well as physiological functions, such as neurotransmission, diuretic, and tranquilizing effects (13,14). γ -Oryzanol is comprised of 10 ferulate esters of triterpene alcohol including 3 main components, 24-methylene cycloartenyl ferulate, cycloartenyl ferulate, and campesteryl ferulate (15) (Fig. 1) and shows biological functionalities such as inhibition of tumor promotion, reduction of serum cholesterol levels and antioxidant properties (16). Total dietary fiber (TDF) has received a lot of attention in the past few years due to its potential beneficial effects on glucose metabolism and reducing the risk of colorectal cancer (17). For these reasons, rice is receiving a renewed interest as a potential dietary supplement.

In the present research, we elucidated changes in 7 nutritional components of rice, including fatty acid, protein, fat, ash, TDF, GABA, and γ -oryzanol, at three dif-

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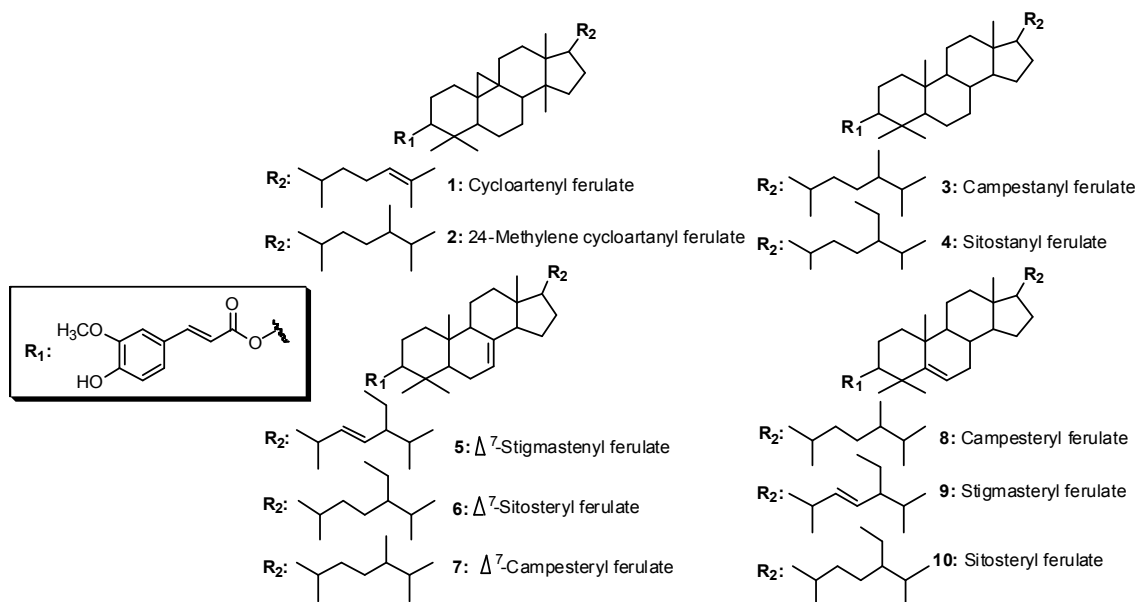


Fig. 1. Chemical structures of γ -oryzanol in rice.

ferent shoot lengths during germination. The variation of these nutrients within different Korean cultivars was also investigated.

MATERIALS AND METHODS

Materials

Four rice cultivars, 'Keunnun', 'Heugkwang', 'Ilpum', and 'Chucheong' were used for this study. These cultivars were developed in the National Institute of Crop Science (NICS), Rural Development Administration (RDA). All cultivars were harvested in the NICS, RDA, Suwon, Korea in 2006.

Chemicals and reagents

A 37-fatty acid methyl ester (FAME) standard was purchased from Supelco (USA) and GABA was prepared from Tokyo Kasei (Kogyo Co., Japan). Hexane and petroleum ether (HPLC grade) were obtained from J.T. Baker (Phillipsburg, NJ, USA). γ -Oryzanol and sodium methoxide were purchased from Sigma Aldrich (St. Louis, MO, USA). All chemicals and solvents were of the highest commercial grade and used without further purification.

Sample preparation

After harvesting, rice was stored in airtight containers for 7 days at 30°C. PR (100 g) was dehulled with Satake Grain Testing Mill (Satake Engineering Co., Higashi-Hiroshima-Shi, Japan) to BR. PR and BR (each 50 g) were decontaminated with 1% sodium hypochlorite solution for 2 hr. The sterilized grains were put in a petri dish covered with filter paper (Advantec No. 2, 90 mm),

and then 10 mL distilled H₂O was added. The petri dish was germinated under dark conditions in a controlled growth chamber (25°C with 50% air humidity, PCE-22MF, Shizuoka Seiki Co., Ltd.). In this study, the shoot lengths of germinated rice were considered when the coleoptiles were 10 mm (3 days), 20 mm (3.5 days), and 30 mm (4 days).

Measurement of fatty acid composition

Fatty acid methyl esters were prepared from rice oil by methylation (18) and subjected to gas chromatography, which was performed using a Hewlett-Packard instrument (model 5890, PaloAlto, CA, USA) equipped with a flame ionization detector, an auto sampler (model 7673, Hewlett-Packard), a fused silica capillary column SPTM-2380 (30 m \times 0.25 mm, 0.25 μ m, Supelco, Bellefonte, PA), and Chemstation software system (version A.09, Hewlett-Packard). The carrier gas was helium at a flow rate of 1.0 mL/min and injection volume was 1 μ L at a split ratio of 30%. The injector and detector temperatures were 250°C. The temperature was programmed as follows: hold at 45°C for 4 min, increase by 13°C/min to 175°C, hold again at 175°C for 27 min, increase at 4°C/min for 4 min to 250°C, and then hold at 250°C for 35 min with a final hold time of 10 min. Individual fatty acid methyl esters were identified by comparing the gas chromatography retention time with those of standards and the amount was expressed as a percent of total fatty acids.

Measurement of protein, fat, ash, and total dietary fiber (TDF)

Protein content was determined by the micro-Kjeldahl

method (19). The percentage of protein was calculated as the percentage of nitrogen determined, multiplied by 6.25, and expressed on a dry basis. The pulverized rice (5 g) was extracted in approximately 70 mL hexane for 3 hr at 105°C via a Soxhlet apparatus (Buchi B-811). Hexane was evaporated using a Buchi Rotovapor R-200 (Flawil, Switzerland) at a reduced process. The remaining fat was weighted and calculated (20). Total fat content was represented on a dry matter basis of rice. Ash was by the AOAC method 923.03 (19). Briefly, ash was incineration for 5 hr at 600°C in an electric furnace (DS-84E-1, Dasol, Korea) and soxhelt (DongHa-Tech., Seoul, Korea). TDF content was measured according to the AOAC enzymatic gravimetric method (21).

Measurement of GABA

GABA content was extracted according to the method of Oh and Oh (22) with a slight modification. Briefly, the mixture of organic solution (CH₃OH 5 mL : CHCl₃ 10 mL : H₂O 5 mL) was added to the pulverized grains (1.0 g). The aqueous solution layer containing GABA was obtained through centrifugation (2,800×g, 4°C, 10 min), and then the supernatant was freeze dried. GABA was measured by a 1.0 mL/assay system in spectrophotometric assay at 340 nm (23).

Measurement of γ -oryzanol

γ -Oryzanol content was measured according to the methods of Rogers et al. (10) and Chotimarkorn et al. (24). Fat was weighed to exactly 100 mg and dissolved in 1.0 mL of propanol before being filtered through a 0.45- μ m syringe filter. The HPLC analysis was performed by Agilent 1100 series equipped with Hypersil ODS column (4.0×250 mm, 5 μ m, Agilent Technologies, Palo Alto, CA, USA) at 330 nm. The mobile phase, MeOH : CH₃CN : CH₂Cl₂ : CH₃COOH (50:44:3:3), was used at a flow rate 1.0 mL/min. γ -Oryzanol was identified by the retention time of the standard and calculated by comparing the peak area of sample with the standard calibration curve.

Statistical analysis

All measurements were repeated 3 times and the results were the mean \pm standard deviation (SD). Results were analyzed by using Sigma Plot 2001 (SPSS Inc., Chicago, IL, USA).

RESULTS AND DISCUSSION

Changes in fatty acid composition content

Changes in sugar, vitamin, and γ -oryzanol contents of germinated rice are well documented (8). However, changes in fatty acid composition over different stages

of germination have not been reported in the literature. Although fatty acid composition showed no significant differences according to the shoot lengths, the main components in all samples were palmitic acid, oleic acid, and linoleic acid (C16:0, C18:1, and C18:2, respectively), with relative concentrations of C18:1>C18:2>C16:0 (Table 1). These compositions exceeded 80% of total fatty acid content. Among them, C18:1 showed considerable fluctuations in comparison with C16:0 and C18:2. As shown in Table 1, C18:1 showed the highest content in 'Keunnun' and 10 mm shoot length in germinated PR was observed the highest content (45.4%). Moreover, 'Chucheong' exhibited lower content than other cultivars with the lowest content being 28.6% in 20 mm shoot length of germinated BR. The minor fatty acid profiles were in the order of C18:3 (<8.0%), C18:0 (<3.0 %), C14:0 (<1.0%), C20:0 (<0.8%), and C22:0 (<0.7%). These components were not influenced by germination and showed no significant differences between PR and BR (Table 1). SFA content (C14:0, C16:0, C18:0, C20:0, and C22:0) slightly increased according to the shoot length of germination, with the average content of BR exhibiting slightly higher concentrations than that of PR (BR: 26.3% and PR: 24.1%). On the contrary, USFA content (C18:1, C18:2, and C18:3) was slightly reduced. The maximum content of the average USFA was observed in 'Keunnun' (77.2%) of PR, while the minimum was observed in 'Chucheong' (72.1%) of BR. Generally, chemical and functional components significantly increased after germination because of biosynthesis (8). Therefore, our results suggest that fatty acid composition may be significantly affected by lengths of germination and cultivars.

Changes in protein, fat, ash, and TDF contents

With the increase of shoot length, protein concentration slightly increased in PR and BR (Table 2). Also, the total average protein of BR was more than that of PR (Fig. 2A). It seems that changes in protein at different shoot lengths during germination were similar to those of phytochemicals in rough rice (12). In this study, 'Heugkwang' exhibited the highest average content (PR: 7.3% and BR: 9.1%), followed by 'Keunnun' (PR: 6.8% and BR: 8.7%), 'Ilpum' (PR: 6.6% and BR: 7.6%), and 'Chucheong' (PR: 6.3% and BR: 7.5%).

As shown in Table 2, with an increase of shoot length, fat content also slightly increased. Moreover, total average fat content of PR was higher than that of BR (Fig. 2B). Among the 5 cultivars, 'Heugkwang' showed the highest average content (8.6%) in PR, while 'Keunnun' was observed to have the highest average content (5.5%) in BR (Table 2). Fat levels did not display significant

Table 1. Changes in fatty acid composition through different shoot lengths from PR and BR during germination

Cultivar (shoot length, mm)	Fatty acid composition (% ¹⁾)														
	C14:0 ²⁾		C16:0 ²⁾		C18:0 ²⁾		C18:2 ²⁾		C18:3 ²⁾		C20:0 ²⁾		C22:0 ²⁾		
	PR ³⁾	BR ⁴⁾	PR	BR	PR	BR	PR	BR	PR	BR	PR	BR	PR	BR	
Keunmun	0.5±0.0 ^a	0.6±0.0 ^a	19.7±1.5 ^a	19.4±0.7 ^b	3.6±0.3 ^a	2.8±0.6 ^a	42.2±2.2 ^b	41.5±1.9 ^a	32.3±1.1 ^c	34.3±1.4 ^a	1.3±0.1 ^b	1.3±0.1 ^b	0.4±0.0 ^a	ND	ND
Keunmun (10) ⁵⁾	0.4±0.0 ^a	0.7±0.1 ^a	17.6±1.9 ^b	19.4±1.0 ^b	0.9±0.0 ^c	2.6±0.5 ^a	45.4±2.6 ^b	40.0±2.3 ^b	33.0±0.7 ^b	34.6±1.6 ^a	1.6±0.2 ^b	2.0±0.2 ^a	0.8±0.1 ^a	0.7±0.0 ^a	0.5±0.0 ^a
Keunmun (20) ⁶⁾	0.7±0.1 ^a	1.0±0.1 ^a	19.9±1.3 ^a	20.1±0.9 ^b	2.4±0.6 ^b	2.7±0.3 ^a	38.5±3.0 ^{ab}	37.8±2.5 ^c	34.5±0.4 ^{ab}	34.5±0.6 ^b	3.3±0.5 ^a	2.2±0.3 ^a	0.6±0.1 ^a	0.6±0.0 ^a	0.2±0.0 ^a
Keunmun (30) ⁸⁾	0.7±0.0 ^a	1.0±0.1 ^a	20.0±1.2 ^a	22.0±1.2 ^a	2.6±0.4 ^b	2.7±0.5 ^a	38.1±1.5 ^a	36.9±1.4 ^c	35.1±1.5 ^a	34.3±2.1 ^a	3.6±0.6 ^a	2.5±0.2 ^a	ND	0.6±0.0 ^a	ND
Heugkwang	0.3±0.0 ^a	ND ⁵⁾	19.4±1.0 ^c	21.1±0.4 ^c	2.3±0.7 ^a	2.0±0.2 ^a	40.5±2.5 ^a	37.3±1.9 ^a	35.6±1.9 ^a	38.4±2.5 ^a	1.2±0.0 ^d	1.3±0.0 ^b	0.5±0.0 ^a	ND	0.2±0.0 ^a
Heugkwang (10)	0.2±0.0 ^a	0.6±0.0 ^a	20.2±1.2 ^b	22.1±0.9 ^b	1.5±0.2 ^b	2.6±0.5 ^a	33.6±2.0 ^a	33.5±1.3 ^b	37.2±2.0 ^b	38.3±1.9 ^a	7.4±0.6 ^a	2.5±0.4 ^{ab}	ND	0.5±0.0 ^a	ND
Heugkwang (20)	0.4±0.0 ^a	0.7±0.0 ^a	20.2±0.7 ^b	23.9±1.3 ^a	0.3±0.0 ^c	2.7±0.4 ^a	36.7±0.9 ^b	31.7±0.9 ^c	37.2±1.4 ^b	37.9±1.5 ^a	4.2±0.1 ^b	3.2±0.4 ^a	0.5±0.0 ^a	ND	0.4±0.0 ^a
Heugkwang (30)	0.5±0.1 ^a	0.7±0.0 ^a	22.2±0.9 ^a	23.9±1.9 ^a	1.8±0.2 ^b	2.8±0.7 ^a	33.7±1.2 ^c	31.5±0.5 ^c	38.7±1.2 ^a	37.3±1.3 ^a	3.1±0.3 ^c	3.2±0.6 ^a	ND	0.6±0.0 ^a	ND
Ilpum	0.4±0.0 ^a	0.7±0.1 ^a	20.3±1.9 ^b	21.1±0.6 ^c	2.2±0.6 ^a	2.2±0.2 ^a	36.8±0.9 ^b	37.0±0.8 ^a	38.6±2.2 ^a	37.7±1.7 ^a	1.8±0.0 ^c	1.3±0.2 ^b	ND	ND	ND
Ilpum (10)	0.6±0.1 ^a	0.9±0.1 ^a	22.2±2.1 ^a	22.9±1.3 ^b	2.4±0.3 ^a	2.0±0.3 ^a	34.4±1.4 ^b	32.9±1.5 ^b	37.8±1.5 ^b	38.5±0.9 ^a	2.0±0.3 ^b	2.4±0.1 ^a	0.4±0.0 ^a	0.4±0.0 ^a	0.2±0.0 ^a
Ilpum (20)	0.6±0.1 ^a	1.0±0.1 ^a	23.0±1.3 ^a	23.6±1.2 ^b	1.7±0.2 ^b	2.3±0.5 ^a	34.3±1.1 ^b	33.2±1.3 ^b	37.4±1.4 ^b	37.6±2.3 ^a	2.6±0.3 ^b	2.4±0.3 ^a	0.5±0.1 ^a	ND	ND
Ilpum (30)	0.6±0.1 ^a	1.1±0.1 ^a	22.6±1.9 ^a	25.5±0.9 ^a	2.0±0.6 ^a	2.2±0.6 ^a	33.7±1.8 ^b	31.1±0.7 ^c	36.8±0.8 ^b	36.9±2.0 ^b	3.5±0.5 ^a	2.6±0.3 ^a	0.5±0.0 ^a	0.5±0.1 ^a	0.3±0.0 ^a
Chucheong	0.4±0.0 ^a	0.6±0.0 ^a	21.1±1.0 ^c	22.4±0.4 ^c	2.8±0.4 ^a	1.8±0.2 ^c	35.6±1.6 ^a	35.3±0.6 ^a	38.7±1.6 ^a	38.6±1.6 ^a	1.5±0.1 ^d	1.4±0.2 ^b	ND	ND	ND
Chucheong (10)	0.6±0.0 ^a	0.9±0.0 ^a	22.2±1.6 ^b	24.0±0.9 ^b	2.0±0.3 ^b	2.3±0.5 ^a	33.6±1.8 ^b	32.2±0.9 ^b	36.9±0.9 ^c	38.4±2.7 ^a	4.0±0.4 ^b	2.3±0.2 ^a	0.4±0.0 ^a	ND	0.3±0.0
Chucheong (20)	0.6±0.1 ^a	1.0±0.0 ^a	22.7±2.3 ^b	26.9±2.3 ^a	2.0±0.7 ^b	3.2±0.6 ^a	31.8±1.1 ^c	28.6±1.6 ^d	37.2±1.6 ^b	37.6±1.8 ^a	5.4±0.5 ^a	2.7±0.3 ^a	0.4±0.0 ^a	ND	ND
Chucheong (30)	0.5±0.0 ^a	0.8±0.1 ^a	24.0±1.7 ^a	24.3±1.5 ^b	1.7±0.4 ^c	2.5±0.3 ^b	32.6±1.6 ^c	30.3±1.2 ^c	38.7±1.1 ^a	38.8±1.9 ^a	2.5±0.4 ^c	2.3±0.3 ^a	ND	0.6±0.0 ^a	ND

¹⁾The values indicate the mean±SD (*n*=3) for fatty acid composition contents of each sample; ²⁾C14:0 myristic acid, C16:0 palmitic acid, C18:0 stearic acid, C18:1 oleic acid, C18:2 linoleic acid, C18:3 linolenic acid, C20:0 arachidic acid, C22:0 behenic acid; ³⁾PR, paddy rice; ⁴⁾BR, brown rice; ⁵⁾ND: not detected; ⁶⁾germination during 3 days; ⁷⁾germination during 3.5 days; ⁸⁾germination during 4 days.

Table 2. Changes in protein, oil, ash, and TDF contents at different shoot lengths of PR and BR during germination

Cultivar (shoot length, mm)	Overall composition (% ¹⁾)									
	Protein		Oil		Ash		TDF ²⁾			
	PR ³⁾	BR ⁴⁾	PR	BR	PR	BR	PR	BR		
Keunmun	6.3±0.2 ^b	6.7±0.4 ^d	6.5±0.3 ^d	3.1±0.1 ^c	5.0±0.5 ^a	1.4±0.0 ^b	15.2±1.8 ^c	5.2±0.5 ^d		
Keunmun (10) ⁵⁾	6.6±0.3 ^b	7.8±0.5 ^c	7.2±0.2 ^c	4.0±0.3 ^b	5.0±0.4 ^a	1.5±0.1 ^b	15.8±1.9 ^c	9.2±0.4 ^c		
Keunmun (20) ⁶⁾	7.0±0.3 ^a	9.4±0.8 ^b	8.2±0.4 ^b	4.8±0.2 ^b	5.2±0.2 ^a	1.8±0.1 ^b	18.5±1.1 ^b	13.2±1.1 ^b		
Keunmun (30) ⁷⁾	7.2±0.2 ^a	11.0±1.1 ^a	9.1±0.4 ^a	7.7±1.0 ^a	5.2±0.4 ^a	2.3±0.3 ^a	21.5±1.0 ^a	19.9±1.0 ^a		
Heugkwang	6.9±0.4 ^a	7.6±0.2 ^d	7.2±0.2 ^c	2.7±0.2 ^c	4.4±0.3 ^a	1.2±0.2 ^b	14.8±1.7 ^c	5.3±0.3 ^d		
Heugkwang (10)	7.3±0.6 ^a	8.5±0.3 ^c	7.9±0.2 ^{bc}	2.8±0.2 ^c	4.3±0.2 ^a	1.5±0.2 ^{ab}	15.3±0.9 ^c	10.3±0.4 ^c		
Heugkwang (20)	7.6±0.5 ^a	9.6±0.7 ^b	8.6±0.6 ^b	3.7±0.3 ^b	4.4±0.4 ^a	1.7±0.0 ^a	18.1±1.6 ^b	18.4±0.8 ^b		
Heugkwang (30)	7.2±0.3 ^a	10.6±1.0 ^a	9.2±0.5 ^a	4.7±0.7 ^a	4.7±0.3 ^a	1.9±0.3 ^a	20.8±1.9 ^a	23.6±1.3 ^a		
Ilpum	6.2±0.3 ^a	6.4±0.3 ^c	6.3±0.1 ^b	2.3±0.2 ^b	3.7±0.2 ^a	1.2±0.2 ^a	12.1±0.9 ^d	4.8±0.4 ^d		
Ilpum (10)	6.6±0.2 ^a	7.3±0.3 ^b	6.9±0.2 ^{ab}	2.6±0.0 ^b	3.7±0.3 ^a	1.2±0.1 ^a	13.0±0.9 ^c	6.0±0.3 ^c		
Ilpum (20)	6.8±0.3 ^a	7.9±0.7 ^b	7.3±0.4 ^a	2.8±0.3 ^b	3.8±0.5 ^a	1.3±0.2 ^a	15.5±1.0 ^b	8.5±0.7 ^b		
Ilpum (30)	6.8±0.1 ^a	8.8±0.8 ^a	7.8±0.3 ^a	3.7±0.6 ^a	4.0±0.4 ^a	1.6±0.2 ^a	17.6±0.6 ^a	12.1±1.2 ^a		
Chucheong	6.0±0.3 ^a	6.3±0.4 ^c	6.2±0.0 ^b	2.4±0.2 ^b	4.1±0.3 ^a	1.1±0.1 ^a	11.3±0.7 ^d	4.2±0.1 ^d		
Chucheong (10)	6.1±0.3 ^a	7.2±0.3 ^{ab}	6.7±0.3 ^{ab}	2.9±0.1 ^b	4.1±0.4 ^a	1.2±0.2 ^a	12.8±0.4 ^c	7.2±0.3 ^c		
Chucheong (20)	6.4±0.4 ^a	7.8±0.6 ^b	7.1±0.4 ^a	3.3±0.3 ^b	4.2±0.6 ^a	1.3±0.2 ^a	16.0±0.6 ^b	9.1±0.9 ^b		
Chucheong (30)	6.7±0.3 ^a	8.5±0.9 ^a	7.6±0.5 ^a	6.0±0.7 ^a	4.3±0.4 ^a	1.4±0.2 ^a	19.1±1.2 ^a	16.6±1.1 ^a		

¹⁾The values indicate the mean±SD (*n*=3) for protein, oil, ash, and TDF contents of each sample; ²⁾TDF, total dietary fiber; ³⁾PR, paddy rice; ⁴⁾BR, brown rice; ⁵⁾germination during 3 days; ⁶⁾germination during 3.5 days; ⁷⁾germination during 4 days.

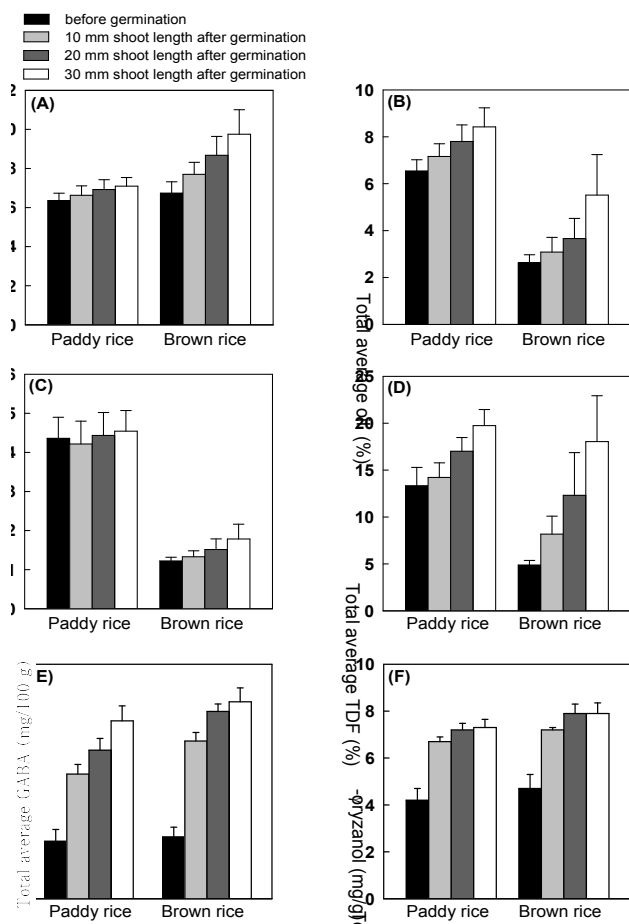


Fig. 2. Changes in the average contents of 6 nutritional components at different shoot lengths of PR and BR during germination. (A) The average protein content, (B) The average oil content, (C) The average ash content, (D) The average TDF content, (E) The average GABA content, (F) The average γ -oryzanol content.

changes during germination. Ash showed significant differences among cultivars (25), but no significant differences were observed in this study (Table 2). Ash content of PR was higher than that of BR (total average content before germination, PR: 4.3%, BR: 1.3%). This result suggests that ash content progressively decreases with an increase in milling duration (26). With the increase of shoot length during germination, total average ash content showed nearly the same content as shown in Fig. 2C. Thus, ash levels in germinated rice are not affected by increased germination time. In this study, ‘Keunnun’ rice exhibited the highest average content (PR: 5.1% and BR: 1.8%), while ‘Ilpum’ showed the lowest (PR: 3.8% and BR: 1.3%).

Recently, it was reported that TDF in rough rice increased 1.1~1.9 times throughout germination (8). As can be seen in Table 2, TDF showed significant differences between PR and BR. Total average TDF content

of PR before germination was observed to be 13.4% and in BR was 4.9%. With the increase of shoot length during germination, the total average content of TDF in PR (17.0%) was higher than that of BR (12.8%) (Fig. 2D). However, the rate of increase of TDF in BR was much higher than that in PR. Also, this component showed significant differences in comparison with protein, oil, ash, and fatty acid. In the 4 cultivars used for this study, ‘Keunnun’ showed the highest content of TDF in PR and ‘Heugkwang’ exhibited the highest content in BR. It is well established that TDF promotes beneficial biological effects, including a laxative effect, lowered blood cholesterol, and glucose attenuation (8). Therefore, germinated rice has the potential to be used as healthy and functional food ingredient.

Changes in GABA content

GABA is enhanced in the germination state (4), so allowing time for germination during processing can help improve rice quality (3). Numerous researchers studied the effects of pretreatment conditions on GABA, but changes in GABA throughout the different germination stages have not been as thoroughly investigated.

As shown in Table 3, changes in GABA between cultivar and shoot length during germination showed the most differences. With the increase of shoot length, GABA levels significantly increased. GABA in the 3 different shoot lengths of germinated rice showed higher total average contents in BR (290.5→39.6→78.7→23.7 mg/100 g) than those of PR (270.0→84.7→97.1→34.5 mg/100 g) (Fig. 2E). In the 4 cultivars used for this study, ‘Keunnun’ showed the highest content and variation (458.4→324.6 mg/100 g), followed by ‘Heugkwang’ (346.8→32.4 mg/100 g), ‘Chucheong’ (185.5→50.7 mg/100 g), and ‘Ilpum’ (89.4→30.4 mg/100 g). This phenomenon is likely the result of ‘Keunnun’s’ large embryo, where these physiological activities take place (8). Based on the above results, the germinated ‘Keunnun’ may be prove to be an important contribution for human health owing to its high GABA content. Interestingly, the grains of 10 mm shoot length showed the most significant changes and ratios, with the highest GABA content in all cultivars (Fig. 2E). Thus, GABA showed the highest variation in the early stage of germination and appears to be significantly influenced by germination time.

Changes in γ -oryzanol content

Many researchers have been focused on the increase of γ -oryzanol, because of its activities as an antioxidant, as well as its effects on plasma lipids, and high density lipoprotein-cholesterols (27). In particular, the germination effect concerning γ -oryzanol has been described in

Table 3. Changes in GABA and γ -oryzanol contents at different shoot lengths in PR and BR during germination

Cultivar (shoot length, mm)	Changes in content			
	GABA (mg/100 g) ¹⁾		γ -Oryzanol (mg/g) ¹⁾	
	PR ²⁾	BR ³⁾	PR	BR
Keunnnun	458.4 ± 10.9 ^d	465.2 ± 7.8 ^c	4.6 ± 0.4 ^b	4.9 ± 0.3 ^c
Keunnnun (10) ⁴⁾	1020.8 ± 39.4 ^c	1237.4 ± 32.7 ^b	7.2 ± 0.2 ^{ab}	7.5 ± 0.2 ^b
Keunnnun (20) ⁵⁾	1240.4 ± 48.2 ^b	1394.6 ± 30.9 ^a	7.9 ± 0.3 ^a	8.2 ± 0.3 ^a
Keunnnun (30) ⁶⁾	1324.6 ± 45.7 ^a	1405.1 ± 44.9 ^a	8.0 ± 0.1 ^a	8.3 ± 0.2 ^a
Heugkwang	346.8 ± 14.7 ^c	392.4 ± 8.2 ^d	3.7 ± 0.3 ^b	4.0 ± 0.3 ^b
Heugkwang (10)	785.1 ± 25.8 ^b	886.9 ± 13.6 ^c	7.2 ± 0.1 ^{ab}	7.8 ± 0.4 ^{ab}
Heugkwang (20)	828.9 ± 20.4 ^a	980.1 ± 17.2 ^b	7.9 ± 0.3 ^a	8.4 ± 0.2 ^a
Heugkwang (30)	832.4 ± 19.0 ^a	1043.7 ± 20.5 ^a	7.9 ± 0.3 ^a	8.6 ± 0.5 ^a
Ilpum	89.4 ± 5.7 ^d	105.6 ± 2.8 ^d	4.5 ± 0.3 ^d	5.2 ± 0.4 ^b
Ilpum (10)	104.7 ± 4.2 ^c	346.7 ± 6.8 ^c	5.4 ± 0.2 ^c	6.2 ± 0.1 ^{ab}
Ilpum (20)	128.7 ± 3.8 ^b	469.3 ± 7.9 ^b	5.8 ± 0.1 ^b	6.9 ± 0.3 ^a
Ilpum (30)	530.4 ± 14.2 ^a	537.5 ± 12.3 ^a	6.1 ± 0.2 ^a	6.9 ± 0.2 ^a
Chucheong	185.5 ± 5.1 ^d	198.6 ± 4.2 ^d	4.0 ± 0.4 ^b	4.7 ± 0.2 ^c
Chucheong (10)	428.1 ± 11.9 ^c	487.4 ± 7.9 ^c	6.8 ± 0.2 ^{ab}	7.4 ± 0.1 ^b
Chucheong (20)	590.4 ± 17.4 ^b	670.7 ± 8.1 ^b	7.1 ± 0.3 ^a	7.9 ± 0.3 ^a
Chucheong (30)	650.7 ± 21.3 ^a	708.5 ± 11.5 ^a	7.2 ± 0.3 ^a	7.9 ± 0.3 ^a

¹⁾The values indicate the mean ± SD ($n=3$) for GABA and γ -oryzanol contents of each sample; ²⁾PR, paddy rice; ³⁾BR, brown rice; ⁴⁾germination during 3 days; ⁵⁾germination during 3.5 days; ⁶⁾germination during 4 days.

previous reports (8). However, there has been no report on variations of γ -oryzanol through different shoot lengths of germinated rice. We proceeded to examine γ -oryzanol in PR and BR and these results are shown in Table 3. During germination, γ -oryzanol was observed in varying amounts between cultivars and shoot lengths. Furthermore, this component showed higher content as of shoot length increased. As shown in Table 3, BR exhibited more γ -oryzanol content than PR. Moreover, 'Keunnnun' and 'Heugkwang' showed higher content than other cultivars. These cultivars may be very important sources in processing and functional supplements through germination. Interestingly, the grains of 10 mm shoot length showed the predominant variations as shown in Fig. 2F (total average content; PR: 4.2→6.7 and BR: 4.7→7.2 mg/g). As a result, γ -oryzanol showed the highest variation in the early stage of germination, similar to what was observed with GABA content.

CONCLUSION

The results of this study provide a basic understanding of some of the functional properties of germinated rice. This research evaluated the profile changes of 7 nutritional components in four different cultivars of germinating rice, in both the PR and BR forms. These components include: fatty acid, protein, fat, ash, TDF, GABA, and γ -oryzanol. The germination stages were identified by shoot lengths of 10 mm, 20 mm, and 30 mm. All nutritional components increased with an increase of shoot length. Most notably, GABA and γ -oryzanol

showed the predominant differences between cultivars and shoot lengths during germination. These two nutritional components were most highly expressed in the early stage of germination (10 mm). Their high concentrations indicate they might play a key role in the nutritional benefits of rice. Additionally, the data from these studies suggest that germinated 'Keunnnun' may be a very important nutrition because of its high GABA and γ -oryzanol contents.

REFERENCES

- Zhang L, Hu P, Tang S, Zhao H, Wu O. 2005. Comparative studies on major nutritional components of rice with a giant embryo and a normal embryo. *J Food Biochem* 29: 653-661.
- Kallithraka S, Salacha MI, Tzourou I. 2009. Changes in phenolic composition and antioxidant activity of white wine during bottle storage: accelerated browning test versus bottle storage. *Food Chem* 113: 500-505.
- Yang F, Basu TK, Ooraikul B. 2001. Studies on germination and antioxidant contents of wheat grain. *J Food Sci Nutr* 52: 319-330.
- Komatsuzaki N, Tsukahara K, Toyoshima H, Suzuki T, Shimizu N, Kimura T. 2007. Effect of soaking and gaseous treatment on GABA content in germinated brown rice. *J Food Eng* 78: 556-560.
- Rimsten L, Haraldsson AK, Anderson R, Alminger M, Sandberg AS, Aman P. 2003. Effects of malting on beta-glucanase and phytase activity in barley grain. *J Sci Food Agric* 82: 904-912.
- Mikola K, Brinck O, Jones BL. 2001. Characterization of oat endoproteinases that hydrolyze oat avenins. *Cereal Chem* 78: 55-58.
- Subba Rao MV, Muralikrishna G. 2002. Evaluation of the antioxidant properties of free and bound phenolic acids

- from native and malted finger millet (ragi, *Eleusine coracana* Indaf-15). *J Agric Food Chem* 50: 889-892.
8. Lee YR, Kim JY, Woo KS, Hwang IG, Kim KH, Kim KJ, Kim JH, Jeong HS. 2007. Changes in the chemical and functional components of Korean rough rice before and after germination. *Food Sci Biotechnol* 16: 1006-1010.
 9. Chung YM, Lee JC, Kim KS, Eun JB. 2001. Chemical compositions of 26 varieties of Korean rice straw. *Food Sci Biotechnol* 10: 267-271.
 10. Rogers EJ, Rice SM, Nicolosi RJ, Carpenter DR, McClelland CA, Romanczyk Jr LJ. 1993. Identification and quantitation of γ -oryzanol components and simultaneous assessment of tocopherols in rice bran oil. *J Am Oil Chem Soc* 70: 301-307.
 11. Woo SM, Jeong YJ. 2006. Effect of germinated brown rice concentrates on free amino acid levels and antioxidant and nitrite scavenging activity in *kimchi*. *Food Sci Biotechnol* 15: 351-356.
 12. Tran TU, Suzuki K, Okadome H, Ikezaki H, Homma S, Ohtsubo K. 2005. Detection of changes in taste of *japonica* and *indica* brown and milled rice (*Oryza sativa* L.) during storage using physicochemical analyses and a taste sensing system. *J Agric Food Chem* 53: 1108-1118.
 13. Ramezanzadeh FM, Rao RM, Windhauser M, Prinyawiwatkul W, Tulley R, Marshall WE. 1999. Prevention of hydrolytic rancidity in rice bran during storage. *J Agric Food Chem* 47: 3050-3052.
 14. Genkawa T, Uchino T, Inoue A, Tanaka F, Hamanaka D. 2008. Development of a low-moisture-content storage system for brown rice: storability at decreased moisture contents. *Biosystems Eng* 99: 515-522.
 15. Xu Z, Godber JS. 1999. Purification and identification of compounds of γ -oryzanol in rice bran oil. *J Agric Food Chem* 47: 2724-2728.
 16. Xu Z, Hua N, Godber JS. 2001. Antioxidant activity of tocopherols, tocotrienols, and γ -oryzanol components from rice bran against cholesterol oxidation accelerated by 2,20-azobis(2-methylpropionamidine) dihydrochloride. *J Agric Food Chem* 49: 2077-2081.
 17. Ang JF, Crosby GA. 2005. Formulating reduced-calorie foods with powered cellulose. *Food Technol-Chicago* 59: 35-38.
 18. Yu L, Adams A, Watkins B. 2009. Comparison of commercial supplements containing conjugated linoleic acid. *J Food Compos Anal* 114: 20-27.
 19. AOAC. 1990. *Official methods of analysis*. 16th ed. Method 923.03. Association of Official Analytical Communities, Arlington, VA, USA.
 20. Kim SL, Berhow MA, Kim JT, Chi HY, Lee SJ, Chung IM. 2006. Evaluation of soyasaponin, isoflavone, protein, lipid, and free sugar accumulation in developing soybean seeds. *J Agric Food Chem* 54: 10003-10010.
 21. AOAC. 1997. *Official methods of analysis*. 16th ed. Method 923.03 and 991.43. Association of Official Analytical Communities, Gaithersburg, MD, USA.
 22. Oh SH, Oh CH. 2003. Brown rice extracts with enhanced levels of GABA stimulate immune cells. *Food Sci Biotechnol* 12: 248-252.
 23. Zhang G, Brown AW. 1997. The rapid determination of γ -aminobutyric acid. *Phytochemistry* 44: 1007-1009.
 24. Chotimarkorn C, Benjakul S, Silalai N. 2008. Antioxidant effects of rice bran extracts on refined tuna oil during storage. *Food Res Int* 41: 616-622.
 25. Singh N, Singh H, Kaur K, Bakshi MS. 2000. Relationship between the degree of milling, ash distribution pattern and conductivity in brown rice. *Food Chem* 69: 147-151.
 26. Tsugita T, Kurata T, Kato H. 1980. Volatile components after cooking rice milled to different degrees. *Agric Biol Chem* 44: 835-840.
 27. Wennermark B, Ahlmen H, Jagerstad M. 1994. Improved vitamin E retention by using freshly milled whole-meal wheat flour during drying. *J Agric Food Chem* 43: 1348-1351.

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