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Original Article

Evaluation of the Aromarker allele and quality attributes of Tai Phuan rice landrace (variety Kai Noi) from northern Laos

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Abstract

Glutinous rice (var. Kai Noi) is a staple food in Laos and it is more expensive than other rice cultivars grown in Laos. This study evaluated the quality traits and antioxidant properties of Kai Noi. The results showed that the Aromarker for fragrance allele badh 2.1 was absent, indicating that Kai Noi is a non-fragrant rice. The pasting and textural properties indicated that most parameters were similar to those of a *japonica* type. Total protein, fat, and amylose contents were found at very low levels. Total phenolic content and total flavonoid content of brown and white rice grains ranged from 31.7 to 32.1 mg gallic acid equivalents and 1.9-2.0 mg rutin equivalents/g, respectively. The main phenolic acids found were the hydrocinnamic acids, while the major flavonoids were rutin and myricetin. Our findings provide fundamental data and valuable information on the Kai Noi variety that can serve as a basis for application in food or beverage processing.

Keywords: glutinous rice, nutritional value, bioactive compounds, antioxidants, pasting properties

1. Introduction

Asian cultivated rice (*Oryza sativa* L.) is grown worldwide. These accessions consist of traditional rice landraces preserved by indigenous farmers. Modern rice cultivars developed by breeding programs of each country around the world exhibit genotypic and phenotypic diversity resulting in about 120,000 different accessions (Khush, 1997). In terms of crop germplasm, landraces have significant characteristics, including higher genetic variability among the different groups of germplasm, due to adaption to the natural and cultural environment (Lanteri & Barcaccia, 2006). Rice landraces or local rice varieties or both have particular properties or characteristics such as grain with aromatic characteristics (Roy *et al.*, 2015) with good cooking and eating quality (Luo *et al.*, 2014) and with submergence tolerance (Bailey-Serres *et al.*, 2010). Trends in rice research indicate the need for a better understanding of the factors that contribute to the overall grain quality of rice. This knowledge will lay the foundation for the development of new breeding and selection strategies to combine high quality with high yield (Fitzgerald *et al.*, 2009). The South and South-east Asian regions consist of rice landraces with genetic diversity (Roy *et al.*, 2015). Some of these show good grain quality, such as appearance, nutritional value, cooking, and sensory properties. In addition, some varieties have been used as materials in rice genetic improvement such as Manawthukha and Paw San Hmwe in Myanmar (Oo *et al.*, 2015; Yi *et al.*, 2009).

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Kai Noi is a glutinous rice landrace from the northern region of Laos. The growth of this rice landrace is limited to the northern part of Laos in the provinces of Xiangkhoang and Houaphan due to its adaptation to highland environmental conditions (>1,200 meters above sea level). It was originally cultivated in rain-fed lowland areas in the two provinces of Laos (Figure 1). The topography of Xiangkhoang Province is one of nature's marvels bringing together the peaks of the Laos mountains and the ecological hot spots of the northern region. Most people living in this province belong to an ethnic group called "Tai Phuan" (the "Tai-speaking people" of mainland Southeast Asia). They have consumed glutinous rice as a staple food since ancient times. Kai Noi is usually a staple food of people in Laos and it is more expensive than other rice cultivars grown in Laos. The price indicates the quality and appeal of the product, for example, a beer-production factory in Laos uses the grain of this landrace for its premium grade of beer. Japanese and Koreans prefer japonica cooked rice because of its moderate elasticity and stickiness and because this type of rice has a lower amylose content (Kang *et al.,* 2006). In addition, glutinous rice is used in ready-to-eat products such as rice crackers and steamed rice cakes. The physicochemical and texture properties of rice grains determine the basic food quality and palatability of the cooked products (Fitzgerald *et al.,* 2009; Kang *et al.,* 2006).

Even though most of the cultivation of Kai Noi is limited to two provinces, it is accepted by the Lao people throughout the country. However, the genetic identification of this rice variety, as well as its quality attributes, has never been previously reported. We aimed to elucidate the genetics of Kai Noi and evaluate its pasting properties, cooking time, and texture properties. Furthermore, the bioactive components and their biological activities were investigated. Keeping the above in mind, the present study also aimed to characterize the aroma of Kai Noi using DNA marker(s).

Figure 1. Location of Xiangkhoang province in Lao PDR and a rice field of rice landrace Kai Noi.

2. Materials and Methods

2.1 Rice samples

Samples of rice seed (Kai Noi variety) were collected from the paddy field of a local farmer at harvesting time in October 2015. The harvested seeds were sun-dried until the moisture was less than 15% and stored for two weeks. The rice samples were milled using a MC-90A polisher (Toyo, Japan).

2.2 Analysis of pasting properties

Starch was isolated following the alkaline steeping method described by Wang and Wang (2001) and rice flour was used for the analyses. Pasting properties of the rice flour were analyzed using a rapid visco analyzer (RVA) (RVA-4, Newport Scientific, Warriewood, Australia). The procedure was as follows. Starch slurries (8.3% w/w dsb, 3 g of total weight) were equilibrated at 50 °C for 1 min, heated at 12 °C /min to 95 °C, held at 95 °C for 3 min, cooled at 6 °C/min to 50 °C, and held at 50 °C for 2 min. A constant rotating speed of the paddle (0.35 g) was used.

2.3 Cooking time determination

Sixty milled whole rice kernels were selected for use in this analysis. Rice samples were cooked in 20 ml of distilled water in a boiling water bath at around 98 °C. The cooking time was determined by removing three cooked kernels at one-minute intervals during the cooking and pressed between two glass slides until no white core was left.

2.4 Texture analysis of cooked rice

The textural properties of the cooked rice were determined using the procedure described by Amornsin and Siriamornpun (2004), using a texture analyzer (TA-XT2, Stable Micro System, UK). The parameters obtained from the test were hardness (N) and stickiness (N).

2.5 DNA extraction, PCR amplification, and DNA sequencing

Seed samples were germinated on moistened filter paper in a Petri dish. Genomic DNA was extracted from the fresh young leaves of one-month old seedlings (0.5 g of leaf tissue) following the protocol of Doyle and Doyle (1990). In this study, the Aromarker for fragrance allele badh2.1 (Prathepha, 2009; Yi *et al.,* 2009) was used to genotype rice landrace Kai Noi and to compare it with fragrant rice (i.e. KDML105) as a positive control. Aromarker primer pairs (Forward: 5'-TGCTCCTTTGTCATCACACCC-3' and Reverse: 5'-TTTCCACCAAGTTCCAGTGAA-3') were developed based on an 8 bp deletion in the exon7 of the *Os2AP* or *badh2* gene, which encodes a putative beta in aldehyde dehydrogenase 2. PCR amplification of the *badh2* gene followed the protocol of Prathepha (2009). The PCR products were separated on 2% agarose gel electrophoresis and stained with ethidium bromide. After the PCR products were electrophoresed, the DNA fragments were cut from the gel and purified using a gel extraction kit. Sequences were separated on an ABI 3730 automated sequencer (Applied Biosystems, USA) at the Macrogen DNA Sequencing Service (Macrogen Korea, Seoul, Republic of Korea).

2.6 Nutritional composition analysis

All nutritional value determinations were measured using standard methods. Protein content was determined according to the Kjeldahl method using the factor of 5.95×N for conversion (Association of Official Analytical Chemists

[AOAC], 1990). Total fat was measured using a Soxhlet extractor with petroleum ether as the extraction solvent (American Association of Cereal Chemists [AACC], 1987). Amylose content of the starches was analyzed in triplicate with an amylose/amylopectin assay kit (Megazyme International Ireland Ltd., Bray, Co. Wicklow, Ireland).

2.7 Determination of bioactive compounds

2.7.1 Sample extraction

Samples were extracted using a modified method reported previously from our laboratory (Butsat *et al.*, 2011). White rice and brown rice grains were ground into fine powders and sieved (100 mesh) to a uniform size. Five grams of each sample were extracted with 50 ml of 80% ethanol (1:10), shaken at 150 rpm in an incubator at 37 °C for 2 h, and then filtered (Whatman No. 4). The extracts were kept at 4 °C for further analysis of total phenolic content, total flavonoid content, and 1,1-diphenyl-2-picrylhydrazyl (DPPH) and ferric reducing ability of plasma (FRAP) assays.

2.7.2 Total phenolic content

Total phenolic content was determined using the Folin–Ciocalteau method reported by Abu Bakar *et al.* (2009). Briefly, the extract (300 µl) was mixed with 2.25 ml of Folin–Ciocalteu reagent (previously diluted 10-fold with distilled water) and allowed to stand at room temperature for 5 min. Then 2.25 ml of sodium carbonate solution (60 g/l) were added to the mixture. After 90 min at room temperature, absorbance was read at 725 nm using a spectrophotometer. Results were expressed as mg gallic acid equivalents per gram of dried sample (mg GAE/g dry weight).

2.7.3 Total flavonoid content

Total flavonoid content was determined using the colorimetric method described by Kaisoon *et al.* (2011). Briefly, the extract (0.5 ml) was mixed with 2.25 ml of distilled water in a test tube followed by addition of 0.15 ml of 5% $NaNO_2$ solution. After 6 min, 0.3 ml of a 10% $AlCl_3.6H_20$ solution was added and allowed to stand for another 5 min before 1.0 ml of 1 M NaOH was added. The mixture was mixed well by vortex. The absorbance was measured immediately at 510 nm using a spectrophotometer. Results were expressed as mg rutin equivalents per gram of dried sample (mg RE/g dry weight).

2.7.4 Analysis of phenolic acids and flavonoids by reverse phase high performance liquid chromatography (RP-HPLC)

1) Sample extraction

The extraction was performed using a modified method of Butsat *et al.* (2009). White rice and brown rice grains were ground into a fine powder and sieved (100 mesh) to a uniform size. Five grams of each sample were extracted with 50 ml of absolute methanol with 1% HCl (1:10) then shaken at 150 rpm in an incubator at 37 °C for 12 h and filtered (Whatman No. 4). The extract was evaporated in a rotary evaporator at 45 °C to dryness and reconstituted to 5 mL with absolute methanol. The extract was filtered (0.45 µm) using a membrane filter prior to HPLC analysis**.**

2) HPLC analysis

The content and composition of phenolic acids and flavonoids of Kai Noi were analyzed using HPLC as previously described by Butsat and Siriamornpun (2010). The composition and content of phenolic acids and flavonoids from the crude ethanol extract and its fractions were determined using RP-HPLC (LC-20AC, Shimadzu, Japan). Each extract was dissolved in ethanol, filtered through a 0.45 µm membrane filter and injected onto an Inetsil ODS-3C18 column (4.6×250 mm, 5 m; Hichrom Limited, Berks, U.K.) with an injection volume of 20 µl. The mobile phases were 1% acetic acid (mobile phase A) and acetonitrile (mobile phase B) at a flow rate of 0.8 ml/min. The components of the extract were separated using gradient elution at 38 °C. The eluted bioactive compounds were detected at 280 nm for phenolic acids and 370 nm for flavonoids with a UV-diode array detector (SPD-M20A, Shimadzu, Japan). Identification of the phenolic compounds in the samples was done by comparison of retention times and UV spectra of authentic compounds. The content of phenolic compounds in the samples was detected using the external standard methods reported by Butsat and Siriamornpun (2010).

2.8 Determination of antioxidant activity

2.8.1 DPPH assay

The DPPH radical-scavenging activity was measured according to a previously published method (Kubola & Siriamornpun, 2008) for DPPH radical-scavenging activity. The hydrogen atom or electron-donation ability of the corresponding extracts and some pure compounds were measured from the bleaching of a purple-colored methanol solution of DPPH (Gulluce *et al.*, 2007). The antioxidant activity of the extracts, on the basis of the scavenging activity of the stable DDPH free radical, was determined by the method described by Braca *et al.* (2001). Aqueous extract (0.1 ml) was added to 3 ml of 0.001 M DPPH in methanol. Absorbance at 517 nm was determined after 30 min and the percent inhibition of activity was calculated as [(AoAe)/ Ao]100 (Ao=absorbance without extract, Ae=absorbance with extract).

2.8.2 FRAP assay

The FRAP assay was conducted as described by Benzie and Strain (1996). The anti-oxidant potential of the extract was determined against a standard curve of ferrous sulfate (FeII, 0, 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0 mM) in Milli-Q water or methanol with 0.1% (v/v) HCl. The FRAP reagent was freshly prepared by mixing 100 ml of acetate buffer (300 mM, pH 3.6), 10 ml 2,4,6-Tri(2'-pyridyl)-1,3,5-triazine (TPTZ) solution (10 mM TPTZ in 40 mM HCl), 10 ml $FeCl_3$ $6H_2O$ (20 nM) in a ratio of 10:1:1, and 12 ml distilled water at 37 °C. To perform the assay, 1.8 ml of FRAP reagent, 180 µl Milli-Q water, and 60 µl sample, standard or blank were then added to the same test tubes and incubated at 37 °C for 4 min.

The absorbance was measured at 593 nm using the FRAP working solution as a blank. The antioxidant potential of the sample was determined from a standard curve plotted using the $FeSO_4 7H_2O$ linear regression equation to calculate the FRAP values of the sample (Benzie and Strain, 1996).

3. Results and Discussion

3.1 Genotyping of Kai Noi using Aromarker

The gene for fragrance in rice was surveyed in a rice gene pool world-wide to look for allelic variants at this locus. The results indicated the involvement of other genetic loci in controlling the fragrance trait (Sakthivel *et al.,* 2009), including a 7 bp insertion in exon8, a 7 bp deletion in exon2, and 8 bp deletion in exon7. Based on evidence of grain morphology and a negative reaction with phenol solution, rice landrace Kai Noi was classified as *tropical japonica* (Prathepha, 2016). Evidence from previous studies on fragrance genes in rice have reported that most of the rice accessions of *indica* and *tropical japonica* type of fragrant rice accessions had the *badh 2.1* allele (8 bp deletion in exon7) (Kovach *et al.,* 2009). Therefore, in this study the Aromarker developed by Vanavichit *et al.* (2007) was selected to identify the genotype of the Kai Noi landrace. It was found that the DNA sequence of the seventh exon of the Kai Noi landrace had a non-aromatic allele of *badh2 gene* (without the 8 bp deletion), while KDML105 showed the aromatic allele (with the 8 bp deletion) (Figure 2). This result supports the sensory test for aroma from leaf tissue. It was found that when a solution of 1.7% KOH with leaf tissue was subjected to smell, the absence of aroma could be scored. Based on all this evidence, we suggest that the Kai Noi landrace is non-fragrant rice. The DNA sequences were aligned using GeneStream ALIGN home page (Pearson *et al.,* 1997).

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KDML105   GTGAACGCTGGTAGCTATTCTCCTGTAATCATGTATACCCCATCAATGGAAATGATATTC
          : :  : ::: : :::::::::::::: ::::::::::::::::::::::::::::::::
Kai Noi   GAG--CCCTG-TCGCTATTCTCCTGTA-TCATGTATACCCCATCAATGGAAATGATATTC
                 10        20         30        40        50
               70        80        90       100       110       120
KDML105   CTCTCAATACATGGTTTATGTTTTCTGTTAGGTTGCATTTACTGGGAGTTATGAAACTGG
          ::::::::::::::::::::::::::::::::::::::::::::::::::::::::::::
Kai Noi   CTCTCAATACATGGTTTATGTTTTCTGTTAGGTTGCATTTACTGGGAGTTATGAAACTGG
           60        70        80        90       100       110

                        130       140       150       160       170
KDML105    TATATA--------TTTCAGCTGCTCCTATGGTTAAGGTTTGTTTCCAAATTTCTGTGGA
           :: : :         :::::::::::::::::::::::::::::::::::::::::::
Kai Noi    TAAAAAGATTATGGCTTCAGCTGCTCCTATGGTTAAGGTTTGTTTCCAAATTTCTGTGGA
           120       130       140       150       160       170

               180       190       200       210       220       230
KDML105   -ATTTTTTGTTCTCTTTCTACTAACTCTCTATTATCAATTCTCAATGTTGTCCTTTTCTT
           ::::::::::::::::::::::::::::::::::::::::::::::::::::::::::::
Kai Noi   TATTTTTTGTTCTCTTTCTACTAACTCTCTATTATCAATTCTCAATGTTGTCCTTTTCTT
           180       190       200       210       220       230

               240       250       260       270       280       290
KDML105   TTAACTCCTTTACTTTTTAGAATTGTGATCAAGACACTTTGAGCATCATTCTAGTAGCCA
          ::::::::::::::::::::::::::::::::::::::::::::::::::::::::::::
Kai Noi   TTAACTCCTTTACTTTTTAGAATTGTGATCAAGACACTTTGAGCATCATTCTAGTAGCCA
           240       250       260       270       280       290

           300       310       320       330       340       350

KDML105   GTTCTATCCTGTTTCTTACCTTTTTATGGTTCGTCTTTTCTTGACAGCCTGTTTCACTGG
          ::::::::::::::::::::::::::::::::::::::::::::::::::::::::::::
Kai Noi   GTTCTATCCTGTTTCTTACCTTTTTATGGTTCGTCTTTTCTTGACAGCCTGTTTCACTGG
           300       310       320       330       340       350

           360
KDML105   AACTTGG-TGGAAAA
          ::::::: :::::::
Kai Noi   AACTTGGGTGGAAAA
           360       370
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Figure 2. Comparison of DNA sequences of exon7 of the *badh2* gene in aromatic rice cultivar KDML105 which carried 8 bp deletion mutation (GATTATGG) and rice land race Kai Noi. The exon7 showed no 8 bp deletion.

3.2 Pasting, cooking, and textural properties of rice flour

Pasting behavior has been reported to influence the cooking and eating qualities and the functional properties of rice (Ye *et al.,* 2016). The pasting parameters of rice flour obtained from Kai Noi glutinous rice, as analyzed by an RVA, are presented in Table 1. Pasting temperature is the temperature at which the starch paste begins to rise. The pasting temperature of Kai Noi rice flour was approximately 67.8 ℃. This temperature was consistent with the findings of Jang *et al.* (2016) who previously reported a pasting temperature range of 67-72 ℃ for glutinous *japonica* rice from Korea. Peak viscosity is the maximum viscosity during heating with water and is used to study the water-binding capacity of starch granules (Shimelis *et al.,* 2006; Wani *et al.,* 2012). The peak viscosity of the rice flour was approximately 223.8 rapid visco units (RVU). Sang *et al.* (2008) reported that amylose inhibits the swelling of starch granules by forming complexes with lipids which results in a lower peak viscosity and a higher pasting temperature. The low peak viscosity may be due to the concentration of amylose which inhibits the swelling of starch granules and maintains the swollen granules. The values of trough viscosity, break viscosity, and final viscosity were approximately 123.0, 100.8, and 146.4 RVU, respectively. The breakdown viscosity was used to measure the starch paste resistance to heat and shear during cooking. The higher breakdown viscosity value indicated lower ability to withstand heating (Adebowale *et al.,* 2005). The final viscosity represents the stability to the swollen granule structure and the high final viscosity value may be due to linear amylose reducing the swelling of granules.

Table 1. Pasting properties measured with a rapid visco analyzer, cooking time and textural properties of rice flours of a glutinous rice landrace, Kai Noi

Rice flour property	Parameter	Value
Viscosity properties	Pasting temperature (°C)	67.75±0.00
	Peak viscosity (RVU)	223.84±3.66
	Trough viscosity (RVU)	123.00±0.59
	Break viscosity (RVU)	100.83±4.24
	Final viscosity (RVU)	146.42±0.47
	Setback (RVU)	23.42±1.06
Cooking property	Cooking time (min)	15
Textural properties	Hardness (N)	93.59±4.12
	Stickiness (N)	24.60±1.67

All values are expressed as mean±standard deviation (S.D) of duplicate measurements.

The setback of Kai Noi rice flour was approximately 23.4 RVU. The setback shows the tendency of starch pastes to retrograde, which is an index of starch retrogradation and is usually related to the amylose content of starch. The amylose component within starch retrogrades more readily than amylopectin due to its essentially linear structure. The straight chain structure of amylose helps it to form hydrogen bonds between molecules, contributing to firm gels (Gani *et al.,* 2013). High values in breakdown viscosity and low values in setback are indicative of high cooking quality since the cooked rice neither retrogrades nor becomes stiff upon cooling (Asante *et al.,* 2013).

The textural parameters of rice flour from Kai Noi glutinous rice, such as hardness and stickiness, were determined as these are important attributes with respect to sensory properties (Table 1). The values of hardness and stickiness of Kai Noi rice flour were 93.6 N and 24.6 N, respectively. High hardness values in rice flour are due to high amylose content and are mainly caused by starch gel retrogradation associated with the crystallization of amylose in a short time that leads to harder gels (Mir and Bosco, 2014). Previous studies have reported that the amylose content of rice flour is positively related to gel hardness (Bao *et al.,* 2006; Wang *et al.,* 2010). Starches that exhibit harder gels tend to have higher amylose content and longer amylopectin chains (Wang *et al.,* 2010).

3.3 Nutritional composition, total phenolic content, total flavonoid content, and antioxidant activity

The compositions of protein, fat, amylose, total phenolic, and total flavonoid, as well as the antioxidant activity of brown and white rice grains of Kai Noi cultivar are listed in Table 2. Protein and fat are possibly important components to determine the processing and nutritional quality of rice. Table 2 shows that the protein content ranged between 0.5% for white rice and 0.9% for brown rice, which was appreciably low (<7%) for Kai Noi glutinous rice. The protein content was similar to that of different cultivars of glutinous *japonica* rice from Korea (0.1-1.8%) as previously reported by Jang *et al.* (2016). Additionally, Kai Noi glutinous rice contained low total fat content that varied from 0.8% for white rice to 3.2% for brown rice.

Table 2. Nutritional characterization, total phenolic content, total flavonoid content, and antioxidant activity

Components	Content	
	Brown rice	White rice
Protein (%)	0.93±0.03a	0.45±0.01b
Fat (%)	3.18±0.10a	0.84±0.01b
Amylose (%)	2.51±0.32a	1.63±0.13b
Total phenolics (mg GAE/g dw)	31.74±0.78a	32.13±1.16a
Total flavonoids (mg RE/g dw)	1.95±0.12a	1.99±0.11b
Antioxidant activities		
DPPH radical scavenging (%)	47.03±1.31a	33.46±0.44a
FRAP activity (mmol Fe(II)/g dw)	3.94±0.50a	5.39±0.51a

All data are expressed as mean±standard deviation (S.D) of triplicate measurements. Different letters indicate significant difference between the results at $P<0.05$).

The amylose content of rice is one of the most important criteria of rice quality in terms of cooking and pasting properties (Adu-Kwarteng *et al.,* 2003). The amylose content of Kai Noi glutinous rice was in the range of 1.6 to 2.5% which classified it as very low amylose rice. These values were in the range of those reported for glutinous rice (0-2%). Kong *et al.* (2015) reported the amylose content of different rice genotypes ranged from 1.5 to 31.6%. The differences in amylose content are dependent on genotype, botanical source, and conditions during rice development (Wang *et al.,* 2010). The low amylose content of Kai Noi rice flour indicated an increase in gel stickiness, a decrease of gel

firmness, a lower pasting temperature, and a tendency to retrograde.

Phenolic compounds, such as phenolic acids and flavonoids are considered to be an important bioactive compound found in cereals due to their various potential biological activities, such as antioxidant, anti-cancer, and anti-inflammatory activities (Shahidi & Chandrasekara, 2013). No significant difference was observed in the total phenolic content of brown rice (31.7 mg GAE/g) and white rice (32.1 mg GAE/g). The total flavonoid content of brown and white rice grains showed similar trends with respect to total phenolic content, which was approximately 2.0 mg RE/g for Kai Noi glutinous rice.

The antioxidant activities of brown and white rice grains determined by DPPH and FRAP assays are also shown in Table 2. The DPPH radical scavenging activity ranged from 33.5 to 47.0%. The FRAP value ranged between 3.9 mmol Fe(II)/g for brown rice and 5.4 mmol Fe(II)/g for white rice. Previous studies reported that the potential for antioxidant activity of rice is dependent on the actual composition of milling fractions (Butsat & Siriamornpun, 2010). Brown rice contains a greater amount of some bioactive compounds than white rice such as gamma oryzanol and tocopherols (Butsat & Siriamornpun, 2010).

3.4 Phenolic acid and flavonoid compositions

Phenolic compounds in cereals have received significantly attention in recent years due to their potent antioxidant capacities and ability to reduce the risk of diseases caused by oxidative stress (Butsat & Siriamornpun, 2010). In the present study, phenolic acids were identified and quantified in Kai Noi glutinous rice by HPLC analysis (Table 3) and compared with nine authentic standards available in our laboratory. From these, five were hydroxybenzoic acids (gallic, protocatechuic, p-hydroxybenzoic, vanillic, and syringic acids) and four were hydroxycinnamic acids (chorogenic, caffeic, *p*-coumaric, ferulic, and sinapic acids). However, only three hydroxycinnamic acids were found in both brown and white rice grains, namely *p*-coumaric acid, ferulic acid, and sinapic acid. In contrast, none of the hydroxybenzoic acids were found in either the brown or white rice grains. Previous research showed that the concentration of individual phenolic acids is higher in brown rice than in milled rice (Butsat & Siriamornpun, 2010; Zhou *et al.,* 2004). Flavonoids are one of the most important polyphenolic compounds with human health benefits due to their potent antioxidant and pharmacological effects (Khanam *et al.,* 2012; Miean & Mohamed, 2001). The content and composition of flavonoids in brown and white rice grains are also shown in Table 3. Rutin and myricetin were found to be dominant in both the brown and white rice grains. However, the concentrations of phenolics and flavonoids can vary in accordance with variety and environmental effects (Tegelberg *et al.,* 2004). Epidemiological studies showed that dietary intake of flavonoids reduces the risk of many cancers such as breast and lung cancers (Romagnolo & Selmin, 2012).

Table 3. Content values of phenolic acids and flavonoids in brown and milled rice grains of Kai Noi

Compounds		Contents (mg/100g dw)	
		Brown rice	White rice
Phenolic acids	Gallic acid	n.d.	n.d.
	Protocatechuic acid	n.d.	n.d.
	p-Hydroxybenzoic acid	n.d.	n.d.
	Vanillic acid	n.d.	n.d.
	Syringic acid	n.d.	n.d.
	Caffeic acid	n.d.	n.d.
	p-Coumaric acid	0.97±0.01	0.92±0.01
	Ferulic acid	1.19±0.04	0.95±0.01
	Sinapic acid	1.09±0.02	0.98±0.02
	Total	3.25±0.07	2.85±0.04
Flavonoids	Rutin	2.86±0.51	12.30±0.90
	Myricetin	1.61±0.01	1.50±0.01
	Quercetin	n.d.	n.d.
	Apigenin	n.d.	n.d.
	Total	4.47±0.53	13.80±1.02

Values are expressed as mean±standard deviation (S.D) of three replicates. n.d.= not detected.and amylose, at very low levels.

4. Conclusions

Our results show that Kai Noi glutinous rice contains nutritional components, such as crude protein, fat, and amylose, at very low levels. The Aromarker for fragrance allele badh 2.1 was absent, which indicated that Kai Noi is non-fragrant rice. The pasting and textural parameters of Kai Noi were similar to those of the *japonica* type. Kai Noi showed good antioxidant capacity determined by DPPH and FRAP assays. Hydrocinnamic acids, including *p*-coumaric acid, ferulic acid, and sinapic acid, were the most dominant phenolic acids in brown and white rice grains from Kai Noi, while the major flavonoids were rutin and myricetin. This study provides valuable information of Kai Noi variety that can serve as a basis for application in food or beverage processing.

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