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Yield and nutritional properties of improved red pericarp Thai rice varieties

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ABSTRACT: Value can be added to rice varieties by developing ones that have high yield and high nutritional value. Value-added rice varieties contribute to high yield and high nutritional value. Eight Thai rice cultivars were evaluated for their yield and nutritional properties: black coloured rice (one cultivar and one mutant line), red coloured rice (two cultivars and two promising red rice lines), and two non-coloured rice cultivars (brown rice). Pathum Thani 1 had the highest yield (4.87 ton/ha) followed by R684-13-15, Mali Komain Surin #3 and RD69 with yield of 4.13, 3.59 and 3.35 ton/ha, respectively. The protein and lipid contents were highest in the black rice followed by the red rice and the brown rice. The highest total phenolic content (TPC), the 2,2-diphenyl-1-picrylhydrazyl (DPPH), and the ferric reducing antioxidant power (FRAP) were found in the red rice followed by the black rice and the brown rice. A promising red rice line, R2535-8-10, had the highest TPC and FRAP values of 320.78 mg GAE/100 g and 791.83 mg $FeSO_4$/100 g, respectively. The two mutant red rice lines derived from Pathum Thani 1, R684-13-15 and R2535-8-10, showed high yield, TPC, and antioxidant activity values; and hence, they could be recommended for cultivation due to their high nutritional properties.

KEYWORDS: nutritional properties, mutant red rice line, proximate analysis, antioxidant activity

INTRODUCTION

Rice (*Oryza sativa* L.) is one of the most important staple foods, providing energy for nearly half the world's population. Brown rice is an unpolished whole grain obtained after removing the husk, and its colour can be light brown, reddish, purple, or black [1]. Whole rice grains are believed to contribute positively to human health due to their polyphenols, minerals, fibre, vitamins, and other phytochemicals, which are mostly found in the bran layer and the embryo fraction [2]. Rice is classified as either brown or white, depending on whether the bran has been removed or not. Generally, brown rice consists of 6–7% bran, 2–3% embryo, and 90% endosperm by weight [3]. Consumption of coloured rice is becoming more popular in recent years because of its higher antioxidant content than white rice. The health benefits of coloured rice are partly attributed to its phytochemicals strong antioxidant properties [1]. Pigmented rice has been reported to have a higher phenolic content and stronger antioxidant activity than white rice [4]. Phenolic compounds, such as flavonoids, phenolic acids, and tannins, possess potent antioxidant activity and provide health benefits associated with a lower risk of chronic diseases [5] and prevention of cell destruction caused by free radicals [6].

Coloured rice bran (yellow, brown, purple, red, and black) have been extensively studied. They contain an abundance of naturally occurring phytochemicals such as tocopherols, tocotrienols, oryzanols, flavonoids, and phenolic compounds in the bran layer [7]. Rice bran obtained from different varieties of rice has different chemical compositions and nutritional properties [8]. Black rice is rich in anthocyanins and other polyphenolic compounds at greater levels than white rice [9]. However, the nutritional composition of rice depends on its cultivation, and the content of bioactive compounds also depends on the genetic characteristics of the rice variety [10].

More than one hundred rice varieties in Thailand are brown pericarp rice. But some varieties produce grains with purple-black and red pericarps. Both local and improved rice varieties are available in those groups. Traditional rice breeding has mainly focused on improving agronomic traits such as the yield, the resistance to disease and insect, milling quality, grain appearance, and cooking quality. Recently, there has been interest in the development of new rice varieties which are rich in one or more phytochemical fractions to improve human health and create new market opportunities. Therefore, the objective of the present study was to evaluate the yield and the nutritional properties of new promising red pericarp rice varieties from Thailand.

MATERIALS AND METHODS

Plant materials

Eight rice varieties were selected based on their pericarp colour; namely, four red rice varieties/lines (RD69, Mali Komain Surin #3, R684-13-15, and

R2535-8-10), two black rice varieties/lines (Riceberry and Riceberry mutant line), and two brown rice varieties (Pathum Thani 1 (PTT1) and RD43). All rice varieties were evaluated in September 2018 at the Pra Nakon Sri Ayutthaya Rice Research Center, Rice Department, using a randomized complete block design with four replications. Individual varieties were planted in plots of 6 rows and 5 m long at a spacing of 20 × 20 cm. The four rows in the middle of each plot were harvested and analyzed for characteristics, yield, and yield component. The paddy rice samples were put in a hot-air oven at 50 °C to reduce the moisture content to approximately 12% (wb). The rice samples were de-husked to obtain the unpolished rice using a small milling mechanical machine. The unpolished samples were ground to a fine powder and kept at 4 °C.

Proximate analysis

The moisture, protein, lipid, ash, fibre, and carbohydrate contents were determined according to the Association of Official Analytical Chemists (AOAC) standard methods. Determination was done using different methods: moisture content, gravimetrically measured by drying the samples in an oven at 100 °C to a constant weigh; crude protein content, Kjeldahl method [11]; crude lipid, Soxhlet extraction method with petroleum ether as the extracting agent [12]; crude ash content, burning the samples in a muffle furnace at 550 °C [13]; crude fibre content, using a standard method for water [14]; and carbohydrate content, using the equation: % carbohydrate = 100−(% moisture + % crude fibre + % protein + % lipid + % ash).

Determination of total phenolic content (TPC)

Each rice flour sample (1.5 g) was weighed accurately and extracted at room temperature using 85% aqueous methanol with stirrer agitation for 30 min. Each mixture was centrifuged at 2500×*g* for 10 min, and the supernatant was collected. The residue was re-extracted twice under the same conditions, resulting finally in 50 ml of combined crude extract. The extracts were, then, used to determine the TPC and antioxidant capacity. The TPC was determined using the Folin-Ciocalteau reagent [15]. An appropriate 120 ml of diluted extract was added to 600 ml of freshly tenfold diluted Folin-Ciocalteau reagent; and 2 min later, 960 µl of sodium carbonate solution (75 g/l) were added, and the mixture was left at 50 °C for 5 min for a reaction to take place. Then, the absorbance of the resulting blue colour mixture was measured at 760 nm. Ferulic acid (FA) was used as the standard, and the TPC was expressed as milligrams of FA equivalent per 100 g flour.

Determination of antioxidant properties

The reducing antioxidant power test of iron (FRAP) is based on the reduction of the Fe^{3+} 2,4,6-Tris(2-pyridyl)-s-triazine (TPTZ) complex to the ferrous form at low pH. This reduction is monitored by following the absorbance change at 595 nm [16]. Briefly, 200 ml of extract was mixed with 1.3 ml of the FRAP reagent. The absorbance of the mixture was measured at 595 nm after 30 min incubation at 37 °C. The FRAP reagent was prepared daily and consisted of 0.3 M acetate buffer (pH 3.6), 10 mM TPTZ in 40 mM HCl, and 20 mM $FeCl_3$ in a volume ratio of 10:1:1, respectively. The FRAP values were determined by comparing the changes of absorbance in the test mixture with doses obtained from increasing concentrations of Fe^{3+} and expressed as millimole Fe^{2+} equivalents per 100 g flour.

The determination of the DPPH radical scavenging activity of rice extracts was carried out according to the method described by Brand-Williams et al [17]. The reaction mixture contained 1.5 ml of DPPH working solution (4.73 mg DPPH in 100 ml ethanol HPLC-grade) and 300 ml of rice extract. The mixture was shaken and incubated in the dark at room temperature for 40 min. Absorbance was measured using a UV-Vis spectrophotometer (U-1100, Hitachi, Japan) at 515 nm against a blank (100%). The ability to scavenge DPPH free radicals was calculated using the following formula:

$$\text{Scavenging ability (\%)} = \frac{(\text{A}_{\text{control}} - \text{A}_{\text{sample}})}{A_{\text{control}}} \times 100$$

Statistical analyses

All data were analyzed by the variance procedure (ANOVA) using R and package agricolae [18]. Differences were considered statistically significant at $p < 0.05$. When significant differences were found, means were separated by least significant difference (LSD) at the 5% probability level. Associations between trait values were estimated using the Pearson correlation coefficient.

RESULTS

Yield and yield components

The paddy rice yield was determined based on the yield, plant height, number of panicles per plant, and 1000-grain weight (Table 1). PTT1 had the highest yields of 4.87 ton/ha followed by R684-13-15 and R2535-8-10 with yields of 4.13 and 3.35 ton/ha, respectively. Mutant line R2535-8-10 had the highest plant height of 123.9 cm, followed by Mali Komain Surin #3 (120.35 cm). There were no significant differences in the number of panicles per plant among the eight varieties. The RD43 had the highest 1000-grain weight (28.49 g), while the 1000-grain weights of the two mutant rice lines R684-13-15 and R2535-8-10 were 22.32 and 21.23 g, respectively.

Table 1 Yield and yield components in coloured pericarp Thai rice varieties.

Varieties	Pericarp color	Yield (ton/ha)	Plant height (cm)	No. of parnicles/plant	1000-grain weight (g)
RD43	brown	3.04^{ab}	101.60^{c}	9.7	28.49^{a}
PTT1	brown	4.87^{a}	106.40^{c}	9.6	23.53^{d}
Riceberry	purple	3.34^{ab}	107.30^{c}	9.7	24.80^{c}
Riceberry mutant line	purple	2.89^{b}	109.65^{bc}	9.5	26.32^{b}
RD69	red	2.58^{b}	105.95^{c}	9.0	26.66^{b}
R684-13-15	red	4.13^{ab}	102.30^{c}	9.9	22.32^{e}
R2535-8-10	red	3.35^{ab}	123.95^{a}	9.1	21.23^{f}
Mali Komain Surin #3	red	3.59^{ab}	120.35^{ab}	10.5	25.18^{c}
Means		3.47	109.68	9.59	24.82
F-test		**	**	ns	**
% CV		18.54	4.34	18.1	1.05

Different small letters in the column (agronomic traits) are significantly different at $p < 0.05$.
** = significantly different at $p < 0.05$, ns = non-significant.

Table 2 Proximate analysis of coloured pericarp Thai rice varieties.

Varieties	Proximate analysis (% dry weight)					
	Moisture	Protein	Lipid	Ash	Fiber	Carbohydrate
RD43	10.39 ± 0.16^{a}	9.27 ± 0.02^{e}	1.77 ± 0.26^{de}	1.94 ± 0.21	1.40 ± 0.05^{a}	83.96 ± 0.40^{bc}
PTT 1	9.61 ± 0.07^{ab}	9.06 ± 0.04^{f}	2.29 ± 0.07^{bcd}	2.06 ± 0.04	0.48 ± 0.06^{b}	85.16 ± 0.15^{a}
Riceberry	9.99 ± 0.22^{ab}	8.56 ± 0.03^{g}	2.77 ± 0.29^{ab}	1.82 ± 0.04	1.55 ± 0.36^{a}	83.25 ± 0.45^{cd}
Riceberry mutant line	10.18 ± 0.23^{ab}	9.48 ± 0.08^{d}	2.92 ± 0.11^{a}	1.82 ± 0.23	1.79 ± 0.03^{a}	81.86 ± 0.03^{e}
RD69	5.96 ± 0.34^{c}	9.13 ± 0.05^{f}	1.48 ± 0.23^{e}	1.50 ± 0.57	0.72 ± 0.04^{b}	85.16 ± 0.33^{a}
R684-13-15	9.72 ± 0.42^{ab}	9.96 ± 0.02^{a}	2.10 ± 0.25^{cd}	1.63 ± 0.14	1.55 ± 0.25^{a}	84.69 ± 0.41^{ab}
R2535-8-10	10.28 ± 0.12^{ab}	9.61 ± 0.04^{c}	1.98 ± 0.23^{cde}	2.05 ± 0.09	1.79 ± 0.13^{a}	83.29 ± 0.27^{cd}
Mali Komain Surin #3	9.58 ± 0.33^{b}	9.76 ± 0.10^{b}	2.39 ± 0.08^{abc}	1.85 ± 0.11	1.39 ± 0.21^{a}	82.76 ± 0.32^{de}
Means	9.47	9.35	2.21	1.81	1.31	85.76
F-test	**	**	**	ns	**	**
% CV	2.9	0.61	9.03	13.59	14.58	0.38

Different small letters in the column (agronomic traits) are significantly different at $p < 0.05$.
** = significantly different at $p < 0.05$; ns = non-significant.

Proximate analysis

The results of the proximate analysis were presented in Table 2 showing significant differences in the proximate compositions among the different rice varieties. The moisture contents range between 5.96% and 10.39% for all the rice cultivars. While the highest protein content of 9.96% was found in the R684-13-15, the highest lipid content of 2.92% was observed in the Riceberry mutant line. There were no significant differences in ash content among the eight varieties. Crude fibre content was the highest in the R2535-8-10 and the Riceberry mutant line (both of 1.79%). The highest carbohydrate content of 85.16% was found the in both the PTT1 and the RD69, followed by the R684-13-15 (84.69%) and the RD43 (83.96%).

TPC content

The result showed highly significant differences ($p < 0.001$) in the TPC among the eight varieties (Table 3). Coloured rice had a higher TPC than brown rice with the highest value of 320.78 mg GAE/100 g sample in the R2535-8-10, followed by the Riceberry mutant line and R684-13-15 with 237.89 and 227.69 mg GAE/100 g sample, respectively. The lowest value of 94.43 mg GAE/100 g sample was found in RD43 (brown rice bran).

Antioxidant properties

The DPPH and the FRAP activities of all samples ranged from 32.91 to 49.21 mg Trolox/100 g and 46.87 to 791.83 mg $FeSO_4$/100 g, respectively (Table 3). The red coloured rice had the highest DPPH radical scavenging activity. Although the RD69 had the highest DPPH activity of 49.21 mg Trolox/100 g, the value was not significantly different from the values for the R684-13-15, the R2535-8-10, and the Mali Komain Surin #3, which were 48.56, 48.04, and 48.86 mg Trolox/100 g, respectively. The R2535-8-10 had the highest FRAP content of 791.83 mg $FeSO_4$/100 g sample, followed by the Riceberry mutant line and the R684-13-15 with 525.31 and 523.36 mg $FeSO_4$/100 g, respectively.

Table 3 Total Phenolic compound and antioxidant activity of coloured pericarp Thai rice varieties.

Varieties	TPC (mg GAE/100 g sample)	DPPH (mg Tolox/100 g sample)	FRAP (mg $FeSO_4$/100 g sample)
RD43	94.43 ± 12.55^e	32.91 ± 0.36^e	46.87 ± 5.03^e
PTT 1	102.76 ± 13.63^e	35.30 ± 1.37^d	64.15 ± 6.03^e
Riceberry	199.43 ± 7.53^c	46.58 ± 0.07^b	406.69 ± 2.89^c
Riceberry mutant line	237.89 ± 5.31^b	44.87 ± 0.06^c	525.31 ± 13.27^b
RD69	164.78 ± 5.57^d	49.21 ± 0.11^a	346.29 ± 5.99^d
R684-13-15	227.69 ± 6.59^b	48.56 ± 0.26^a	523.36 ± 6.03^b
R2535-8-10	320.78 ± 3.67^a	48.04 ± 0.06^a	791.83 ± 8.63^a
Mali Komain Surin #3	162.15 ± 3.75^d	48.86 ± 0.07^a	324.31 ± 7.70^d
Means	188.74	44.29	378.59
F-test	**	**	**
% CV	4.92	1.1	2.26

Different small letters in the column (agronomic traits) are significantly different at $p < 0.05$.
** = significantly different at $p < 0.05$.

Notably, the red rice's FRAP value was 1.4 times higher than the black rice and 9.7 times higher than the brown rice.

DISCUSSION

This study focused on yield and nutritional properties of different coloured pericarp rice cultivars from Thailand. There were eight varieties or lines used in the study. Four rice varieties: PTT1, RD43, RD69, and Mali Komain Surin #3 were developed by the Rice Department (RD). Riceberry was developed by the Rice Science Center, Kasetsart University (KU). The PTT1 had a high yield (4.06 to 4.84 ton/ha) and low amylose content (15 to 19%); while the RD43 was a short-duration rice variety (95 days) with an average yield of 3.5 ton/ha, a low amylose content (18.8%), and a low glycemic index. RD69 with red pericarp had a yield of 3.74 ton/ha. Mali Komain Surin #3 showed the highest sensitivity to photoperiod and has an average yield of 2.86 ton/ha despite its low amylose content of 15.5% [19]. The highest yield of PTT1 variety with an average of 4.87 ton/ha was similar to the value reported by RD and Tongmark et al [20] of 4.8 ton/ha when planted at Chainat Rice Research Center in 2016. However, PTT1 had a yield of 6.3 and 9.3 ton/ha when planted at the Pathum Thani Rice Research Center and the National Center for Genetic Engineering and Biotechnology in 2015, respectively [20]. RD43 and RD69 had a yield of 3.04 and 2.58 ton/ha, respectively. The values were below the average yield reported by the RD. On the other hand, Mali Komain Surin #3 had a yield of 3.59 ton/ha, which was higher than the RD value. Riceberry has a green stem, purple leaf and panicle, and a deep purple whole rice grain with an average yield of 1.88–3.13 ton/ha, and it is enriched with both water-soluble (mainly anthocyanins) and fat-soluble (such as carotenoids, gamma-oryzanol, and vitamin E) antioxidants [21]. The Riceberry mutant line was a purple stem variety derived from Riceberry by spontaneous mutation. In the current study Riceberry and Riceberry mutant line had a yield of 3.34 and 2.89 ton/ha, respectively, which were higher than those reported by KU. The R684-13-15 and the R2535-8-10) were two promising red rice mutant lines derived from PTT1 by acute gamma radiation. Based on 20 single sequence repeat markers, the genetic similarities of the R684-13-15 and the R2535-8-10 with the PTT1 were 85 and 95%, respectively. While the amylose content of the PTT1 was 17.28%, the amylose contents of the R684-13-15 and the R2535-8-10 were 16.67 and 17.35%, respectively [22]. From the current study, the yields of the R684-13-15 (4.13 ton/ha) and the R2535-8-10 (3.35 ton/ha) were lower than the wild type PTT1 (4.87 ton/ha), but they were rich in antioxidant properties.

Rice grain consists of moisture, lipid, protein, crude fibre, ash, and carbohydrates. Its protein content is only 7% and very digestible (93%) with excellent biological value (74%) and high protein efficiency (2.02 to 2.04%) due to the high lysine content (about 4%) [23]. Moisture content always affects the quality and palatability of rice grains. In this study, the moisture contents were in the range of 5.96 to 10.39%. The differences in moisture content among rice varieties could be due to the variations in the paddy moisture content after harvesting [24]. Variation in the lipid contents may be due to oxidation of lipid, since most of the lipid contents in rice grains are unsaturated and easily oxidized by the atmospheric oxygen [25]. The percentages of lipid contents in this study were in the range of 1.48 to 2.92%, higher than the value (0.06 to 0.92%) reported by Verma and Srivastav [26]. However, the current results were in agreement with those reported (0.5 to 3.5%) by Oko and Ugwu [27]. The differences in the lipid content could also be attributed to the degree of milling, as the process removes the outer layer of the rice grain where most of the fats are concentrated [28]. Proteins in rice are very important because they form the basic building blocks for cells and tissue repair in the body [29]. The protein content affects the nutritional quality of rice. In the present study, the protein contents were in the range 8.56 to 9.96%, consistent with the results reported by Vunain et al [30]. Brown rice usually provides more dietary fibre than white rice. Fibre is best known for relieving constipation, and it also provides a number of other health benefits, including lowering cholesterol level, controlling blood sugar level, lowering diabetes risk, reducing heart disease risk, nourishing gut bacteria, and helping weight control by providing the feeling of being full rapidly [31]. The dietary fibre contents in the current study were in the range of 0.48 to 1.79%, which were lower than the range of 1.93 to 4.3% reported by Oko et al [32]. The ash content is generally accepted as a quality measure for evaluating the functional properties of foods [29]. The amount of ash in food plays an important role in determining the essential mineral content and may affect the sensory quality of rice, especially its colour and flavour. Differences in the ash contents could be due to differences in the mineral contents of the soil and the water used for irrigation [26]. The ash contents in the current study were in the range of 1.5 to 2.06%, close to those reported by Mbatchou and Dawda [29] but higher than those of Thomas et al [33]. Both white rice and brown rice are rich in carbohydrates. Brown rice is a whole grain and contains more overall nutrients than its lighter counterpart. Whole grain products may help lower cholesterol and reduce the risk of stroke, heart disease, and type-2 diabetes [31]. The carbohydrate contents in the current study were 81.86 to 85.16%, which were similar to the values reported by Vunaia et al [32], but lower than those reported by Verma and Srivastav [26].

Phenolic compounds are secondary metabolites thought to have evolved to protect plants from biotic and abiotic stresses [34]. However, much research has been conducted on phenolics due to the health benefits for human. The concentration of total phenolics in the grain has been positively associated with the antioxidant activity [9], with potential beneficial effects on health, such as reduction of oxidative stress [33], prevention of cancer [34], control of blood lipids and related diseases, which may ultimately help in the prevention of cardiovascular problems [35]. In the current study, the red rice brans had more than twice higher TPC than the black and the brown rice brans. These results were in agreement with Oki et al [36] who reported that the red-hulled rice varieties had higher radical scavenging activity than the black- and the white-hulled rice varieties. In addition, Tian et al [37] consistently found that rice grains with red and black pericarp colours had higher total phenolic contents than light brown grains. The colour of rice are related to the quantity of phenolic compounds, proanthocyanidins (redness), anthocyanin (blueness and purpleness), and antioxidant activity in rice [38, 39]. DPPH and FRAP antioxidant activities differed significantly among different coloured rice, and black rice had the highest level followed by red rice and brown rice [40]. In the current report, the highest levels of DPPH and FRAP were found in the red rice followed by the black rice and the brown rice.

CONCLUSION

PTT1, a brown rice bran, had the highest yield, followed by R684-13-15. The black rice varieties had the highest protein and lipid contents, followed by the red and the brown rice varieties. In addition, the red rice varieties had the highest antioxidant activity, followed by the black and the brown rice varieties. The mutant red rice line, R2535-8-10, had the highest values of both TPC and FRAP, as well as high DPPH radical scavenging activity; while high yield, TPC, FRAP, and DPPH values were found in R684-13-15. Thus, both red rice varieties (R2535-8-10 and R684-13-15) should be recommended for cultivation due to their high nutritional value.

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