

# The Relationship between Color Values in Rice to Phenolic Acids, Flavonoids, and Antioxidants

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Thai rice is diverse. The seed coat manifests as multiple colors: white, yellow, brown, red, dark red, purple, dark purple, and black; dark-colored rice usually has a significant amount of antioxidants, far higher than those found in white or lighter variants. The aimed of this study was to find a simple, quick and economical way to assess phenolic acids, flavonoids, and antioxidants through color parameters, evaluating 12 varieties of various colors. The results showed L\* values having strong negative correlations with almost all antioxidant parameters, except the IC<sub>50</sub> value by DPPH assay. Antioxidant contents and antioxidant capacities demonstrated low correlation with a\*, and b\* had moderate negative correlations. Thus, L\* values are the best data to assess other significant substances in rice, followed by b\*. These results could be used to examine other compounds present in rice and may aid in the selection of varieties for cultivation.

**Keywords:** Rice, Pigment, Phenolic acid, Flavonoid, Anthocyanin, Antioxidant

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Rice is the staple food of Thais, providing 70 percent of all starch. This grain also contains many other nutrients, including proteins, fats, minerals, vitamins, and fiber. Hemicellulose, a type of fiber, in rice and rice bran extract can reduce cancer from chemical and environmental exposure<sup>(1-3)</sup> as well as chronic inflammation<sup>(4-7)</sup>. Many antioxidant compounds, such as phenolic acids, flavonoids, and anthocyanins, are also present in rice: these act as a barrier against the continuous reactions of free radicals, capturing oxygen and chelating metals<sup>(8)</sup>. Rice bran substances appear to prevent diabetic complications<sup>(9)</sup> as well.

Thai rice is diverse. The seed coat, rice skin or bran, manifests as multiple colors: yellow, red to dark red, purple to dark purple, and to almost black. Black rice has a significant amount of antioxidants, far higher than those found in white rice<sup>(10-12)</sup>. Two other notable rice bran extracts are gamma-oryzanol and tocopherols. The colors found in red rice are from  $\beta$ -carotene, lutein, zeaxanthin, and lycopene, which are able to reduce cancer risk, cardiovascular disease, and prevent cataracts<sup>(13)</sup>. Purple-black rice contains 4 kinds of antioxidants and anti-inflammatory anthocyanins which produce its unique color: cyanidin dihexoside, cyanidin 3-glucoside, cyanidin hexoside and peonidin 3-glucoside<sup>(14,15)</sup>. Extracts from purple-black rice inhibit aldose reductase<sup>(9)</sup>,

reduce blood sugar levels<sup>(16)</sup>, decrease inflammation<sup>(11)</sup>, and prevent cardiovascular disease by plasma antioxidation. Various compounds in black rice have been shown to be toxic to leukemia, inhibit the growth of lung cancer cells<sup>(17)</sup>, and slow the growth of tumors<sup>(18)</sup>.

Reports confirm measuring color parameters as a simple, quick and economical way to evaluate the amount of chemical constituents in grains such as rice. In waxy purple corn varieties, factors such as the varieties themselves, harvest times, and interactions between other varieties all affected total anthocyanin content and color values (L\*, a\* and b\*)<sup>(19)</sup>. Antioxidant contents and capacities of pigmented rice varieties are much higher than those of white rice with the relationships between color and amount of antioxidants appearing to be clear<sup>(20)</sup>.

The purpose of the present study was to measure antioxidants via colors in 12 different-colored rice varieties, both glutinous and non-glutinous. These results could be used as a method for assessing other significant substances in rice and may aid in the selection of varieties for cultivation. Other unknown health benefits from colored rice may also come to light through further research. The authors determined the amounts of 7 phenolic acids and 3 flavonoids using high-performance liquid chromatography (HPLC). Levels of antioxidant activity/free radical scavenging were measured using 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay or 2,2'-azino-bis 3-ethylbenzothiazoline-6-sulphonic acid (ABTS) assay as required. pH differential method was used to calculate monomeric anthocyanin contents and 4-dimethylaminocinnamaldehyde (DMAC) assay for proanthocyanidins.

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## Materials and Methods

### Rice sampling and milling

The 12 samples of normal and glutinous rice consisted of purple to black (n = 3), orange to deep red (n = 3), and white pigmentation (n = 6). All were collected from various rice research centers throughout Thailand and half-milled at Pathumthani Rice Research Center.

### Grain size and shape

Grain size and shape were measured by a random 10-grain sample from the seed batch. A Vernier caliper was used to measure grain dimensions.

### Color of half-milled rice

The Konica Minolta Chroma Meter (Model CR-400, Konica Minolta Sensing Americas, Inc., USA) with 8-mm aperture, CIE D65 Illuminant was used to determine the color of half-milled rice. The chrome meter was calibrated with white calibration tiles, and measured in the CIE L\* a\* b\* color space using 15 g of half-milled rice<sup>(21)</sup>.

### Rice samples extraction

All half-milled rice samples were mashed by blender and sieved through a 28-mesh screen. Half-milled flour (100 g) was extracted twice with 200 mL of hexane. The defatted flour was extracted twice by shaking with 250 mL of 1% hydrochloric acid in 95% ethanol. The samples were then centrifuged at 10,000 g for 20 min, at 20°C; the supernatants were collected, pooled, evaporated to the crude extract and stored at -20°C.

### DPPH radical scavenging assay

The DPPH scavenging capacity assay<sup>(22)</sup> was performed by pipet 100 µL different concentrations of sample extracts in ethanol, and added to 100 µL of DPPH solution (2.4 mg/100 mL). Each mixture was stored in the dark for 30 min. The absorbance (A) of each reaction mixture at 517 nm was measured against a blank of ethanol using a Biotek microplate reader (Model: PowerWave XS, Vermont, USA). The percent inhibition for each reaction was calculated as:

$$\% \text{ inhibition} = [(A_{\text{blank}} - A_{\text{sample}})/A_{\text{blank}}] \times 100$$

The IC<sub>50</sub> value (the half-inhibitory concentration that can decrease of DPPH radicals) of each samples were also calculated.

### ABTS radical cation decolorization assay

Free radical scavenging activity of samples was determined by ABTS radical cation decolorization assay<sup>(23)</sup>. ABTS<sup>+</sup> cation radical was formed by 7 mM ABTS (1: 1) 2.45 mM potassium persulfate, and then stored in the dark for 14 h at room temperature before use. The solution was diluted with methanol to achieve an absorbance of 0.700 at 734 nm. The diluted ABTS<sup>+</sup> solution was pipet 3.995 mL to test tube and then added 5 µL of extract, the absorbance was measured

at 734 nm after 30 min. The percent inhibition was calculated using the formula:

$$\text{Percent inhibition (\%)} = [(AB - AA)/AB] \times 100$$

AB = ABTS radical + methanol

AA = ABTS radical + sample/standard

Trolox was used as the standard, and calculated for trolox equivalent capacity (TEAC, µg TE/g extract).

### Monomeric anthocyanin content

Monomeric anthocyanin content was determined using the pH differential method<sup>(24)</sup>. The solution of extract was diluted using pH 1.0 and pH 4.5 (sodium acetate) buffers. Absorbance was read on 1 cm path length cuvettes at 510 nm (absorbance should between 0.100 to 1.200) and 700 nm using a UV-visible Spectrophotometer (UV2550, Shimadzu, Japan). Total monomeric anthocyanin content of each extraction was calculated as follow.

$$\text{Monomeric anthocyanin (mg/liter)} = (A \times \text{MW} \times \text{DF} \times 1,000)/(\epsilon \times 1)$$

$$A = (A_{\lambda_{\text{vis-max}}} - A_{700 \text{ pH } 1.0}) - (A_{\lambda_{\text{vis-max}}} - A_{700 \text{ pH } 4.5})$$

MW = molecular weight

DF = dilution factor

ε = molar absorptivity

### Total proanthocyanidins (PAs) content

Total extractable PAs [microgram catechin equivalent (CE)/g grain] were determined using 4-dimethylaminocinnamaldehyde (DMAC) assay with some modifications<sup>(21)</sup>. Briefly, 50 µL of extracts, standards, or blank were added to 96-well microplates. Then each well received 150 µL of 0.1% DMAC in acidified ethanol [6:1:1 (v: v: v) of ethanol: water: HCl]. The color reaction was read at 640 nm every 1.5 min for 45 min, at 25°C. The maximum absorbance of the sample extract and standards was used to calculate the concentration of total PAs. Catechin was the standard used ranging from 1.25 µg/mL to 50 µg/mL.

### Determination of phenolic acids content

The content of phenolic acids in half-milled rice were extracted by 1% HCl in 95% ethanol. The extracts were characterized by HPLC (Spectra System P-4000, Thermo Separation Products-TSP, Riviera Beach, CA, USA) using a RP-C18 (150 x 4.6 mm, Luna 5 µm, Phenomenex) column. Data acquisition and processing was performed by ChromQuest. The solvents were 1% acetic acid (A) and acetonitrile (B). The gradient elution was performed from 0 to 45 min, linear gradient from 5% to 30% solvent B; from 45.1 to 52 min, isocratic at 60% solvent B and re-equilibration period of 8 min with 5% solvent B were used between individual runs. The flow rate was 1 mL/min. Detection was performed with a UV detection at 280 nm<sup>(25)</sup>.

### Determination of flavonoids content

The content of flavonoids in half-milled rice were extracted by 1% HCl in 95% ethanol. The extracts were characterized by high performance liquid chromatography (Spectra System P4000, Thermo Separation Products-TSP, Riviera Beach, CA, USA) using a RP-C18 (150x4.6 mm, Luna 5  $\mu$ m, Phenomenex) column. Data acquisition and processing performed by Chrome quest software. The solvents were formic acid solution pH 2.5 (A) and methanol (B). The gradient elution was performed from 0 to 20 min, linear gradient from 80% to 30% solvent A; from 20.1 to 22 min, isocratic at 90% solvent B and re-equilibration period of 8 min with 80% solvent A were used between individual runs. The flow rate was 1 mL/min. Detection was performed with UV detection at 280 nm<sup>(26)</sup>.

### Data analysis

Data was analyzed using ANOVA post hoc tests or Duncan's new multiple range test (DMRT) for multiple comparisons and Pearson's coefficient for correlation analysis:  $p < 0.05$ .

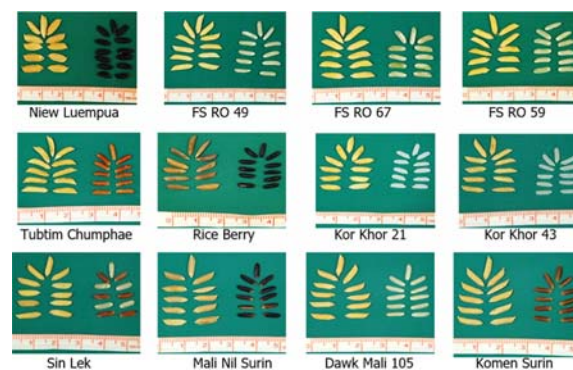
### Results

Rice physical properties are shown in Table 1. The grain weights range from 17.1 to 28.2 mg and differed due to rice variety. Those with the biggest size and shape, were white rice, FS RO 67 and FS RO 59, also the heaviest weight 28.2 and 26.7 mg, respectively. The smallest were two kinds of non-glutinous black rice, Rice Berry and Mali Nil Surin also being the lightest at 17.1 and 17.8 mg, respectively.

The color parameters of half-milled rice samples included  $L^*$ ,  $a^*$  and  $b^*$  values shown in Table 2. Whiteness ( $L^*$ ) was more dominant in the white rice group ( $L^* > 58.5$ ) followed by red rice ( $L^* 44$  to 58). Black rice logically had the lowest ( $L^* < 44$ ) values. The redness ( $a^*$ ) value were, as expected, the highest for a red rice variety (Tubtim

Chumphae) due to its reddish external layers ( $a^* = 12.62$ ). Yellowness ( $b^*$ ) was more dominant in the white rice group ( $b^* 8.41$  to 20.58) followed by red, with black having the lowest ( $b^* < 4.5$ ) values. Komen Surin visually appeared green but when measured by the CIE  $L^* a^* b^*$  color space  $L^* = 52.69$ ,  $a^* = 8.21$  and  $b^* = 9.72$ , thus truly being pale brown grey.

The antioxidant capacities of the varieties are given as DPPH and ABTS assay, shown in Table 2. The  $IC_{50}$  values from DPPH assay for black rice varied from 6.94 to 19.20  $\mu$ g (extract)/ml, while red and white rice were in the range of 26.54 to 45.13 and 41.12 to  $>100$   $\mu$ g (extract)/ml, respectively. The lowest  $IC_{50}$  value occurred in Niew Luempua, corresponding to it having the highest levels of phenolic acids, flavonoid, and monomeric anthocyanin content. These results are similar to those of Pramai and Jiamyangyeun, 2016<sup>(20)</sup>. The TEAC values of the crude extracts were performed using ABTS assay. For black rice ranged from 31.46 to 45.23  $\mu$ g TE/g (extract), higher than those of red and white varieties. Red rice TEAC values ranged from 5.46 to 26.22  $\mu$ g TE/g (extract). White rice displayed the lowest TEAC values, from



**Figure 1.** Rice varieties, paddy and half-milled (grain).

**Table 1.** Rice varieties and physical properties

Varieties	Grain color	Source by province	Grain weight (mg)	Grain size and shape (mm)		
				Length	Width	Thickness
Niew Luempua	Black	Phitsanulok	24.9±2.8 <sup>abc</sup>	7.13±1.61 <sup>bcd</sup>	2.70±0.10 <sup>a</sup>	1.70±0.10 <sup>cd</sup>
Mali Nil Surin	Brown-black	Surin	17.8±0.8 <sup>g</sup>	7.17±0.06 <sup>bcd</sup>	2.27±0.06 <sup>bcd</sup>	1.57±0.06 <sup>d</sup>
Rice Berry	Black	Kanjanaburi	17.1±2.4 <sup>fg</sup>	6.60±0.20 <sup>de</sup>	2.00±0.00 <sup>g</sup>	1.60±0.00 <sup>d</sup>
FS RO 49	White	Phitsanulok	24.6±1.5 <sup>bcd</sup>	8.27±0.12 <sup>a</sup>	2.20±0.00 <sup>cde</sup>	1.87±0.06 <sup>ab</sup>
FS RO 67	White	Phitsanulok	28.2±1.6 <sup>a</sup>	7.77±0.21 <sup>abc</sup>	2.30±0.10 <sup>bc</sup>	2.00±0.10 <sup>a</sup>
FS RO 59	White	Phitsanulok	26.7±1.8 <sup>ab</sup>	7.93±0.21 <sup>ab</sup>	2.20±0.00 <sup>cde</sup>	1.97±0.06 <sup>a</sup>
Dawk Mali 105	White	Surin	22.1±1.4 <sup>cde</sup>	7.67±0.12 <sup>ac</sup>	2.17±0.06 <sup>def</sup>	1.80±0.00 <sup>bc</sup>
Kor Khor 21	White	Kanjanaburi	22.0±0.7 <sup>cde</sup>	6.07±0.06 <sup>e</sup>	2.37±0.06 <sup>b</sup>	1.70±0.10 <sup>cd</sup>
Kor Khor 43	White	Kanjanaburi	21.6±1.6 <sup>def</sup>	6.93±0.12 <sup>cde</sup>	2.07±0.06 <sup>fg</sup>	1.77±0.06 <sup>b</sup>
Sin Lek	Light red	Phitsanulok	20.6±1.5 <sup>efg</sup>	6.97±0.06 <sup>cde</sup>	2.17±0.06 <sup>def</sup>	1.87±0.06 <sup>ab</sup>
Tubtim Chumphae	Red	Khon Kaen	19.5±2.7 <sup>fg</sup>	7.33±0.06 <sup>bcd</sup>	2.10±0.00 <sup>efg</sup>	1.57±0.12 <sup>d</sup>
Komen Surin	Red	Surin	20.9±1.4 <sup>efg</sup>	7.27±0.40 <sup>bcd</sup>	2.13±0.06 <sup>ef</sup>	1.67±0.12 <sup>cd</sup>

Mean values superscripted in column with differing letters are significantly different ( $p < 0.05$ , DMRT)

**Table 2.** Rice grain color (CIE L\* a\* b\*) measurement DPPH assay and ABTS assay

Varieties	Grain color (CIE L* a* b*)			DPPH assay, IC <sub>50</sub> [μg (extract)/ml]	ABTS assay TEAC [μg TE/g (extract)]
	L*	a*	b*		
Niew Luempua	35.62±0.01 <sup>k</sup>	1.20±0.01 <sup>i</sup>	0.96±0.01 <sup>i</sup>	6.94±2.22 <sup>a</sup>	45.23±7.57 <sup>a</sup>
Mali Nil Surin	45.74±0.00 <sup>h</sup>	6.62±0.03 <sup>c</sup>	4.27±0.01 <sup>j</sup>	13.83±0.86 <sup>b</sup>	31.85±3.08 <sup>b</sup>
Rice Berry	42.39±0.00 <sup>j</sup>	6.41±0.02 <sup>d</sup>	2.63±0.01 <sup>k</sup>	19.20±3.86 <sup>b</sup>	31.46±5.65 <sup>b</sup>
FS RO 49	68.12±0.01 <sup>a</sup>	0.02±0.01 <sup>j</sup>	9.32±0.01 <sup>h</sup>	>100 <sup>e</sup>	nd
FS RO 67	66.47±0.01 <sup>b</sup>	2.33±0.02 <sup>f</sup>	20.58±0.02 <sup>a</sup>	>100 <sup>e</sup>	0.61±2.93 <sup>de</sup>
FS RO 59	63.18±1.73 <sup>c</sup>	-0.02±0.02 <sup>k</sup>	8.41±0.01 <sup>i</sup>	41.22±3.11 <sup>d</sup>	nd
Dawk Mali 105	63.28±0.00 <sup>c</sup>	-0.07±0.02 <sup>l</sup>	9.37±0.02 <sup>g</sup>	44.14±4.06 <sup>d</sup>	5.07±4.07 <sup>cd</sup>
Kor Khor 21	58.75±0.01 <sup>e</sup>	2.19±0.00 <sup>h</sup>	13.32±0.01 <sup>e</sup>	27.59±4.31 <sup>c</sup>	nd
Kor Khor 43	60.80±0.00 <sup>d</sup>	2.27±0.02 <sup>g</sup>	14.46±0.02 <sup>b</sup>	31.68±7.52 <sup>c</sup>	nd
Sin Lek	55.74±0.00 <sup>f</sup>	3.48±0.02 <sup>e</sup>	13.46±0.01 <sup>d</sup>	45.13±5.70 <sup>d</sup>	5.46±4.67 <sup>cd</sup>
Tubtim Chumphae	44.86±0.02 <sup>i</sup>	12.62±0.02 <sup>a</sup>	14.01±0.02 <sup>c</sup>	26.54±3.40 <sup>c</sup>	26.22±4.67 <sup>b</sup>
Komen Surin	52.69±0.01 <sup>g</sup>	8.21±0.01 <sup>b</sup>	9.72±0.01 <sup>f</sup>	>100 <sup>e</sup>	11.09±5.08 <sup>c</sup>

Mean values superscripted in column with differing letters are significantly different ( $p < 0.05$ , DMRT)

nd = non-detected

non-detected (nd) to 5.07 μg TE/g (extract).

Phenolic acids, the singular flavonoid, monomeric anthocyanin and proanthocyanidin contents are shown in Table 3. Phenolic acids in pigmented varieties were significantly higher than in white ones. As previously mentioned, the highest phenolic acids contents were found in black rice, specifically Niew Luempua, with the main phenolic acids being protocatechuic and ferulic acids (Figure 2). Some varieties of red rice also showed high levels of phenolic acids. However, almost all rice had very low levels of rutin: only flavonoid was detectable, with black rice being significantly higher than red. All white rice exhibited very low flavonoids. Anthocyanins were only found in purple or black rice, the highest being Niew Luempua.

Relationships among the color parameters and antioxidant compounds are found in Table 4. The highest significant correlations were between TEAC (ABTS assay) and rutin contents ( $r = 0.893$ ), while monomeric anthocyanin and PCA also demonstrated highly significant correlations with the ABTS assay ( $r = 0.797$  and  $0.741$ , respectively). Nonetheless, proanthocyanidin contents exhibited a lower correlation coefficient with other parameters.

Negative correlations between L\* and b\* value with antioxidants were detected in all parameters, except for DPPH activity; this presented a positive correlation. The L\* value had negative correlations with almost all antioxidant parameters. This is with the exception of DPPH radical scavenging ability, meaning white rice varieties with a high L\* values had lower antioxidant contents and capacities. The results from DPPH assay are expressed as IC<sub>50</sub> values, indicating the half-inhibitory concentration of antioxidants that decrease DPPH radicals: lower IC<sub>50</sub> values indicate higher antioxidant efficiency. The b\* values also had moderate negative correlations with most antioxidant parameters: those with anthocyanin and rutin contents demonstrated

the strongest negative correlations ( $r = -0.783$  and  $-0.657$ , respectively). However, b\* had positive correlations with DPPH radical scavenging ability ( $r = 0.568$ ). Antioxidant contents and capacities had very low correlation with a\* color values, with the exception of proanthocyanidin demonstrating significant correlations ( $r = 0.641$ ). The results show L\* values are likely to be the best data in assessing other significant substances in rice, followed by b\*.

## Conclusion

The color parameters of half-milled rice samples showed whiteness (L\*) more dominant in the white rice group followed by red rice. Black rice logically had the lowest (L\*) values. The lowest IC<sub>50</sub> value from DPPH assay occurred in Niew Luempua, corresponding to it having the highest levels of phenolic acids, flavonoid, monomeric anthocyanin content and the TEAC values by ABTS assay. Black rice had higher antioxidant compounds than those of red and white varieties. Positive correlations between color and antioxidants were detected in all parameters, except for DPPH activity; this presented a negative correlation. The L\* value had negative correlations with almost all antioxidant parameters, except DPPH assay. Antioxidant contents and antioxidant capacities have low correlation with a\* color value, and b\* value also had moderate negative correlations.

As mentioned before, the L\* value should be the most appropriate data for assessing other significant substances in rice, followed by b\*. Color parameters may be useful as a method to aid in the future selection of varieties for cultivation. More research is required into the use of color measurements in determining other beneficial substances in rice.

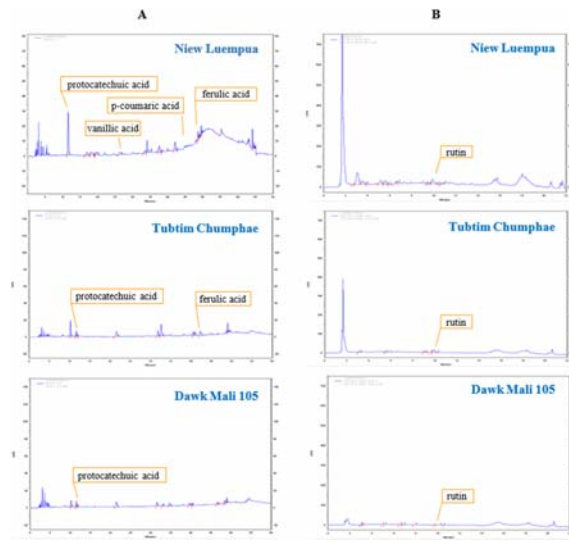
## What is already known on this topic?

Antioxidant contents and capacities of pigmented

**Table 3.** Phenolic acids, flavonoid, monomeric anthocyanin and proanthocyanidin contents in rice

Varieties	Phenolic acids [ $\mu\text{g/g}$ (grain)]				Flavonoid (rutin) [ $\mu\text{g/g}$ (grain)]	Monomeric anthocyanin [ $\mu\text{g/g}$ (grain)]	Proanthocyanidin [ $\mu\text{g/g}$ (grain)]
	PCA	VA	CA	FA			
New Luempua	5.37 $\pm$ 0.41 <sup>a</sup>	1.72 $\pm$ 0.10 <sup>c</sup>	1.91 $\pm$ 0.25 <sup>a</sup>	2.55 $\pm$ 0.10 <sup>a</sup>	18.33 $\pm$ 1.13 <sup>a</sup>	1.55 $\pm$ 0.27 <sup>a</sup>	28.33 $\pm$ 1.28 <sup>de</sup>
Mali Nil Surin	4.97 $\pm$ 0.52 <sup>a</sup>	2.95 $\pm$ 0.14 <sup>a</sup>	1.83 $\pm$ 0.06 <sup>a</sup>	0.62 $\pm$ 0.01 <sup>e</sup>	9.34 $\pm$ 0.96 <sup>c</sup>	0.66 $\pm$ 0.13 <sup>b</sup>	31.20 $\pm$ 0.62 <sup>c</sup>
Rice Berry	3.31 $\pm$ 2.33 <sup>bc</sup>	1.53 $\pm$ 0.16 <sup>d</sup>	1.88 $\pm$ 0.41 <sup>a</sup>	1.20 $\pm$ 0.46 <sup>cd</sup>	14.24 $\pm$ 2.76 <sup>b</sup>	0.78 $\pm$ 0.18 <sup>b</sup>	30.08 $\pm$ 2.94 <sup>cd</sup>
FS RO 49	nd	1.23 $\pm$ 0.04 <sup>e</sup>	1.25 $\pm$ 0.28 <sup>bc</sup>	0.75 $\pm$ 0.23 <sup>e</sup>	1.80 $\pm$ 0.04 <sup>d</sup>	nd	24.99 $\pm$ 0.88 <sup>fg</sup>
FS RO 67	1.28 $\pm$ 0.07 <sup>de</sup>	1.17 $\pm$ 0.07 <sup>e</sup>	0.77 $\pm$ 0.00 <sup>e</sup>	1.14 $\pm$ 0.03 <sup>c</sup>	2.17 $\pm$ 0.42 <sup>d</sup>	nd	21.18 $\pm$ 3.02 <sup>b</sup>
FS RO 59	nd	nd	1.71 $\pm$ 0.06 <sup>ab</sup>	0.66 $\pm$ 0.05 <sup>e</sup>	1.85 $\pm$ 0.15 <sup>d</sup>	nd	23.11 $\pm$ 1.58 <sup>gh</sup>
Dawk Mali 105	0.18 $\pm$ 0.11 <sup>ef</sup>	1.31 $\pm$ 0.03 <sup>e</sup>	1.12 $\pm$ 0.61 <sup>cde</sup>	0.65 $\pm$ 0.27 <sup>e</sup>	1.93 $\pm$ 0.18 <sup>d</sup>	nd	23.78 $\pm$ 3.44 <sup>g</sup>
Kor Khor 21	1.50 $\pm$ 0.24 <sup>cd</sup>	1.15 $\pm$ 0.01 <sup>e</sup>	1.64 $\pm$ 0.01 <sup>ab</sup>	0.62 $\pm$ 0.01 <sup>e</sup>	2.45 $\pm$ 0.73 <sup>d</sup>	nd	26.80 $\pm$ 1.69 <sup>ef</sup>
Kor Khor 43	0.15 $\pm$ 0.03 <sup>ef</sup>	1.24 $\pm$ 0.04 <sup>e</sup>	1.64 $\pm$ 0.00 <sup>ab</sup>	0.79 $\pm$ 0.01 <sup>de</sup>	1.67 $\pm$ 0.07 <sup>d</sup>	nd	26.30 $\pm$ 0.86 <sup>ef</sup>
Sin Lek	0.02 $\pm$ 0.07 <sup>ef</sup>	1.24 $\pm$ 0.09 <sup>e</sup>	1.48 $\pm$ 0.01 <sup>abc</sup>	0.60 $\pm$ 0.01 <sup>e</sup>	1.71 $\pm$ 0.14 <sup>d</sup>	nd	26.84 $\pm$ 1.58 <sup>ef</sup>
Tubtim Chumphae	0.71 $\pm$ 0.03 <sup>def</sup>	nd	1.69 $\pm$ 0.14 <sup>a</sup>	2.05 $\pm$ 0.15 <sup>b</sup>	9.92 $\pm$ 2.66 <sup>c</sup>	nd	48.85 $\pm$ 2.08 <sup>b</sup>
Komen Surin	2.85 $\pm$ 0.06 <sup>b</sup>	2.29 $\pm$ 0.26 <sup>b</sup>	0.83 $\pm$ 0.03 <sup>de</sup>	0.70 $\pm$ 0.09 <sup>e</sup>	9.60 $\pm$ 0.22 <sup>c</sup>	nd	90.74 $\pm$ 7.32 <sup>a</sup>

Mean values superscripted in column with differing letters are significantly different ( $p < 0.05$ , DMRT) PCA = protocathechuic acid, VA = vanillic acid, CA = p-coumaric acid, FA = ferulic acid, nd = non-detected



**Figure 2.** HPLC chromatogram of A = phenolic acid; B = flavonoid determination in black (New Luempua), red (Tubtim Chumphae) and white (Dawk Mali 105) rice.

rice varieties are much higher than white rice. The color values ( $L^*$ ,  $a^*$  and  $b^*$ ) are correlate with the amount of antioxidants

### What this study adds?

The  $L^*$  color value should be the most appropriate data for assessing other significant substances in rice, follow by  $b^*$ . Antioxidant contents and antioxidant capacities have low correlation with  $a^*$ .

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### Potential conflicts of interest

The authors declare no conflict of interest.

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**Table 4.** Correlation coefficients between color, phenolic acids, flavonoid, anthocyanin and proanthocyanidin (PAs) contents in rice

	L*	a*	b*	DPPH	ABTS	PCA	VA	CA	FA	Rutin
a*	-0.559 <sup>a</sup>									
b*	0.629 <sup>a</sup>	0.071								
DPPH	0.649 <sup>a</sup>	-0.134	0.568 <sup>a</sup>							
ABTS	-0.917 <sup>a</sup>	0.473 <sup>a</sup>	-0.648 <sup>a</sup>	-0.541 <sup>a</sup>						
PCA	-0.74 <sup>a</sup>	0.26	-0.56	-0.393 <sup>b</sup>	0.741 <sup>a</sup>					
VA	-0.306	0.065	-0.353 <sup>b</sup>	0	0.332 <sup>b</sup>	0.671 <sup>a</sup>				
CA	-0.532 <sup>a</sup>	0.105	-0.552 <sup>a</sup>	-0.787 <sup>a</sup>	0.435 <sup>a</sup>	0.34 <sup>b</sup>	-0.062			
FA	-0.638 <sup>a</sup>	0.262	-0.262	-0.335 <sup>b</sup>	0.652 <sup>a</sup>	0.44 <sup>a</sup>	-0.169	0.389 <sup>b</sup>		
Rutin	-0.918 <sup>a</sup>	0.456 <sup>a</sup>	-0.657 <sup>a</sup>	-0.43 <sup>a</sup>	0.893 <sup>a</sup>	0.791 <sup>a</sup>	0.339 <sup>b</sup>	0.391 <sup>b</sup>	0.702 <sup>a</sup>	
PAs	-0.274	0.641 <sup>a</sup>	0.1	0.317	0.176	0.226	0.254	-0.295	0.028	0.33 <sup>b</sup>
Anthocyanin	-0.78 <sup>a</sup>	0	-0.783 <sup>a</sup>	-0.542 <sup>a</sup>	0.797 <sup>a</sup>	0.764 <sup>a</sup>	0.413 <sup>b</sup>	0.459 <sup>a</sup>	0.6 <sup>a</sup>	0.818 <sup>a</sup>

PCA = protocatechuic acid, VA = vanillic acid, CA = p-coumaric acid, FA = ferulic acid

<sup>a</sup> Correlation is significant at 0.01, <sup>b</sup> Correlation is significant at 0.05

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## ความสัมพันธ์ระหว่างค่าสีของข้าวกับกรดฟีนอลิก ฟลาโวนอยด์และสารต้านอนุมูลอิสระ

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ข้าวไทยมีความหลากหลายทางพันธุกรรม เชื้อหอมเมล็ดข้าวกล่อมมีหลายสี ได้แก่ ขาว, เหลือง, น้ำตาล, แดง, แดงเข้ม, ม่วง, ม่วงเข้มและสีดำ ข้าวที่มีเชื้อหอมเมล็ดขาวสีเข้ม จะมีสารต้านอนุมูลอิสระมากกว่าข้าวที่มีเชื้อหอมเมล็ดขาวหรือจางกว่า การศึกษาครั้งนี้มีวัตถุประสงค์เพื่อหาวิธีที่ง่าย รวดเร็วและประหยัดในการประเมินปริมาณความสัมพันธ์ระหว่างค่าพารามิเตอร์สีของเชื้อหอมเมล็ดข้าว โดยการศึกษาครั้งนี้ ใช้ข้าว 12 พันธุ์ ที่มีสีแตกต่างกัน ผลการวิจัยพบว่า ค่าสี  $L^*$  มีความสัมพันธ์เชิงลบกับปริมาณกรดฟีนอลิกฟลาโวนอยด์และสารต้านอนุมูลอิสระ ยกเว้นค่า  $IC_{50}$  จากการทดสอบฤทธิ์ต้านอนุมูลอิสระด้วยวิธี DPPH ปริมาณกรดฟีนอลิกฟลาโวนอยด์และสารต้านอนุมูลอิสระมีความสัมพันธ์เพียงเล็กน้อยกับค่า  $a^*$  และค่า  $b^*$  ก็มีความสัมพันธ์เชิงลบในระดับปานกลาง ดังนั้นวิธีวัดค่าสีโดยใช้ค่า  $L^*$  จึงเป็นข้อมูลหลักสำหรับใช้เป็นวิธีการในการพิจารณาประเมินปริมาณกรดฟีนอลิก ฟลาโวนอยด์และสารต้านอนุมูลอิสระต่างๆ ในข้าว และค่า  $b^*$  อาจใช้เป็นข้อมูลรองในการพิจารณาค่ายวิธีนี้จึงเป็นวิธีที่ง่าย รวดเร็วและประหยัดในการประเมินสารสำคัญต่างๆ ในข้าวและช่วยในการเลือกพันธุ์เพื่อการเพาะปลูกและขยายพันธุ์ต่อไป

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