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FULL PAPER

The effect of drying temperature on the physical and antioxidant qualities of MARDI Warna 98 rice

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Abstract

Drying is an important activity after the rice is harvested in order to minimize quality deterioration. Generally, drying temperature ranging from 42-45°C is known worldwide to preserve rice physical quality, mainly for white rice and it is widely practised by all rice millers. A study was conducted to determine the optimum drying temperature of the new coloured rice variety, MARDI Warna 98 (MW 98) in the preservation of physical as well as antioxidant qualities. The rice was dried at ambient temperature as control and oven-dried at 40-45°C, 46-50°C and 51-55°C until moisture content reached 13-14%. Physical qualities that have been determined were head rice and broken rice, while the antioxidant properties evaluation measured were total phenolic content (TPC), total flavonoid content (TFC), total anthocyanin content (TAC) and 2, 2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activities. Antioxidant parameters were measured spectrophotometrically. The results showed that the head rice recovery was significantly lower at ambient temperature and 51-55°C drying temperature (90.59% and 91.23%, respectively) and this was directly proportional to a higher percentage of broken rice (9.41% and 8.77%, respectively). The results also showed no significant difference in head rice recovery either drying at 40-45° or 46-50°C, which were 95.50% and 95.93%, respectively. The rice was then evaluated for its antioxidant activities and results showed there was no significant difference in the content of total phenolic, total flavonoid, total anthocyanins and DPPH radical scavenging activities at different drying temperatures except for ambient temperature. Rice dried at ambient temperature showed significantly lower content of total phenolic, total flavonoid, total anthocyanins and DPPH inhibition. In conclusion, drying temperature does affect the physical and antioxidant qualities of MW 98. Therefore, we suggested that drying at 46-50°C was the best for MW98 to preserve rice's physical and antioxidant qualities.

1. Introduction

Rice (*Oryza sativa* L.) in the Asia-Pacific region produces 95% of the total world rice production and is a staple food for half of the world's population. It is mainly consumed as white rice and is the main source of carbohydrates, which contributes 55-80% to the total calorie intake of people (Bhattacharjee *et al*., 2002). The high prevalence of non-communicable diseases (NCD) such as cardiovascular disease, cancer and diabetes among Asians has gained a lot of attention among researchers, and studies suggested healthy diet intake and lifestyle management can help to reduce the severity of NCD. This has encouraged people to switch from traditional white rice consumption to brown (unpolished) or pigmented rice (red, black and purple rice), which constitutes high proteins, dietary fibre, essential minerals, vitamins and antioxidants which are located in the rice bran (Butsat and Siriamornpun, 2010; Laokuldilok *et al*., 2013) due to its tremendous health benefits. The pigmented rice also contains bioactive compounds such as total phenolic and flavonoid as well as radical scavenging activity (Thitipramote *et al*., 2016). The pigment present in the coloured rice is water-soluble flavonoids, called anthocyanins, which the most abundant anthocyanin reported are cyanidine-3-glucoside, peonidin-3-glucoside and cyanidin diglucoside (Ling *et al*., 2001).

Malaysia Agricultural Research and Development Institute (MARDI) has been mandated to develop rice for local use and to date 50 rice varieties were launched since 1964. Out of 50 rice varieties released, six (6) were

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speciality rice, which includes aromatic, jasmine, basmati and coloured. The development of speciality rice especially aromatic either jasmine or basmati is mainly to substitute imported aromatic rice, while coloured rice was purposely to cater to health-conscious consumers. In 2018, a second red rice variety was launched, namely MARDI Warna 98 (MW 98), a single cross of varieties Hijau Manis and Jaya, followed by pedigree breeding. Besides its healthy properties, the varietal development was also for breeding improvement on traditional coloured varieties in East Malaysia, which typically have a longer maturation time (more than 150 days), this could only be cultivated once a year (Zaki *et al*., 2019). Introducing the new coloured rice cultivated twice a year would offer an alternative variety, as practised in West Malaysia.

Primary processing especially the drying of red rice might be different from widely consumed white rice. The drying temperature is important to be controlled or monitored because it would affect the physicochemical properties of rice, especially head rice yield and taste due to the hygroscopic characteristic of rice grains (Xian-Zhe *et al.,* 2011). A study conducted by Inprasit and Noomhorn (2001) showed that drying temperature at more than 60°C caused the starch granules of white rice (KDML -105 and Suphanburi I varieties) to gelatinized and without tempering could decrease head rice (Figure 1 and Figure 2). There was a positive relation between drying temperature with the hardness of cooked rice. Rapid drying also induced cracks and chalkiness to the grain. However, there was no significant effect that occurred at lower drying temperatures (below 45°C). A study by Akowuah *et al.* (2012) demonstrated that white rice, Jasmine 85 variety can be dried by using a mechanical dryer at higher temperatures of 45°C and 50° C without affecting its milling qualities. The existing standard practice is mainly to achieve a safe storage level of moisture content (13 - 14%), in order to inhibit fungi growth and production of mycotoxins and prevent broken rice. However, in red rice, the preservation of healthy traits is the main concern, as it possesses health benefits for consumers. A suitable drying temperature would retain as much as possible antioxidant qualities of coloured rice. A study by Junka *et al*. (2015) showed that high-temperature drying using fluidized bed dryers in the range of 100 - 150°C did not affect the anthocyanins content nor the antioxidant activity of purple rice (Kum Doi Saket variety). This might be because of the very short time exposure of rice at a higher temperature. Studies on drying in relation to antioxidant properties in red rice are still lacking. Therefore, the objective of the present study was to compare different drying temperatures of MW 98 i.e., 40 -45°C, 46°C-50°C, 51°C -55°C as well as the ambient temperature in terms of physical quality and antioxidant capacity.

Figure 1. The head rice

Figure 2. The broken rice

2. Materials and methods

2.1 Rice samples

Red rice variety namely MARDI Warna 98 (MW 98) was planted for the experimental study located at MARDI Headquarters, in Serdang, Selangor, Malaysia.

2.2 Drying procedure

Rice was dried at ambient temperature (served as control at temperature ranges 30 - 35°C; T1) and using an oven (Memmert) at 40 -45°C (T2), 46°C-50°C (T3), 51°C-55°C (T4), in triplicate. The selected range for drying temperature was based on the standard practice of commercial rice miller at 42-45°C and as a previous study done by Hanisa *et al*. (2014) with some modifications. Drying was stopped when moisture content reached a safe storage level at 13 - 14%.

2.3 Physical quality analyses

In the rice drying experiment, crack grain was the main effect to be observed. Rice kernels that are lesser than 3/4 of the actual kernel size are called broken rice (BR) (Karim *et al*., 2002). Broken rice percentage was calculated as formula given below:

$$\%\ BR\ = Weight\ of\ BR\ (g) \div Weight\ of\ paddy\ (g) \times 100$$

Broken rice was directly proportionate to head rice (HR). Head rice refers to the milled rice of 3⁄4 or more of an actual kernel size (Karim *et al*., 2002). Head rice recovery of dried sample was calculated using the formula below:

$$\%\ HR\ = Weight\ of\ HR\ (g) \div Weight\ of\ paddy\ (g) \times 100$$

2.4 Preparation of rice extract for antioxidant analyses

One gram of rice sample was ground to superfine flour before being extracted with 25 mL methanol absolute containing 1% HCl according to the method described by Shen *et al*. (2009) with slight modifications. The extract was shaken for 24 hrs by using an orbital shaker at 200 rpm (WiseShake, SHO-2D) in the dark. The mixtures were then centrifuged at 4000×g for 15 mins and the supernatant was filtered by using filter paper (Whatman No. 1) and collected into a 10 mL amber bottle. The extracted solution was kept at 4°C

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until further analysis.

2.5 Total phenolic content

The total phenolic content was assayed using the Follin-ciocalteu calorimetric method with slight modification (Shen *et al.*, 2009). Briefly, an aliquot (300 µL) of the extract was introduced into a measuring cylinder containing freshly diluted (2.25 mL) 10-fold Folin-Ciocalteu reagent (Merck) and allowed standing at room temperature for 5 mins. Then, the reaction was neutralized with 2.25 mL saturated sodium carbonate (60 g/L) before being kept in dark for 90 mins at ambient temperature until the sample turned blue. Absorbance was measured at 765 nm using a UV-visible spectrophotometer (Cary 60 UV-VIS). The total phenolic content was calculated on the basis of the calibration curve of gallic acid and expressed as mg of gallic acid equivalent per 100 g of the rice flour (mg GAE/100g) of dry weight.

2.6 Total flavonoid content

The total flavonoid content was determined by a colourimetric method (Shen *et al.*, 2009) with minor modifications. An aliquot (0.5 mL) of the appropriately diluted extract was pipetted into test tubes containing 2.25 mL of deionized water and reacted with 0.15 mL of 5% sodium nitrite ($NaNO_2$). After incubation for 6 mins, 0.3 mL 10% aluminium chloride hexahydrate ($AlCl_3.6H_2O$) solution was added, and the mixture was allowed to stand for another 5 mins before 1 mL 1M sodium hydroxide (NaOH) was added. The reaction solution was mixed thoroughly by using a vortex (Vortex Mixer VM-300) and measured immediately at the absorbance of 510 nm with a UV-visible spectrophotometer (Cary 60 UV-VIS). Total flavonoid content was calculated using the standard catechin curve prepared using (+)-catechin hydrate (SIGMA) and expressed as mg catechin equivalent (CAE) per 100 g dry weight of the sample.

2.7 Total anthocyanin content

The total anthocyanin content was measured by the pH differential method which is based on the structural changes in anthocyanin chemical forms and absorbance at pH 1.0 and 4.5 (Lee *et al.*, 2005). Briefly, 0.5 mL extracted solution was transferred into a test tube containing 3.5 mL of 0.025 M potassium chloride (KCl) buffer at pH 1.0 and kept in dark for 15 mins at room temperature. The absorbance was measured at 515 nm and 700 nm respectively. The extracts then followed the same procedure with 0.025 M sodium acetate (CH_3COONa) buffer at pH 4.5 against solvent extraction. The absorbance was measured using a UV-visible spectrophotometer (Cary 60 UV-VIS). The difference in absorbance between pH values and wavelength was calculated as follow:

$$A = (A520\,nm - A700\,nm)\,pH1.0 - (A520\,nm - A700\,nm)\,pH\,4.5$$

The total anthocyanin concentration was calculated and expressed as cyanidin-3-glucoside equivalents as follows:

$$\text{Anthocyanin (mg/L)} = [A \times MW \times DF \times 1000] / \varepsilon \times 1$$

Where A = absorbance, MW = molecular weight for cyanidin-3-glucoside (449.2 g/mol), DF = dilution factor, ε = molar extinction coefficient (26,900 L/cm mol), 1 = path length in cm, and 1000 is the factor for conversion from g to mg.

2.8 DPPH radical scavenging activity

The free-radical scavenging capacity of each extract was evaluated according to the procedure of (Somaratne *et al.*, 2017) with some modifications. One hundred microlitter of extract solution was added to the freshly prepared 0.1 mM 2, 2-diphenyl-1-picrylhydrazyl (DPPH) solution (2.9 mL), and the mixture was kept at room temperature in dark for 30 mins. The absorbance was read at 517 nm using methanol absolute as the blank with a UV-visible spectrophotometer (Cary 60 UV-VIS). The percentage of DPPH radical scavenging activity was expressed as follows:

$$DPPH\ Scavenging\ Activity = (Abs\ Control - Abs\ sample)/Abs\ control \times 100$$

All experiments were conducted in triplicates (n = 3) and the results are expressed as mean ± standard error mean (SEM).

2.9 Statistical analysis

The experimental design was Randomised Complete Block Design (RCBD). Analysis of variance was performed using Statistical Analysis Software (SAS 9.4). Treatment means were compared based on the LSD test at $p \leq 0.05$.

3. Results and discussion

3.1 Physical qualities

The results of physical qualities are summarized in Table 1. The head rice recovery was found significantly lowest ($p<0.05$) when drying at T1 and T4. The finding was significantly ($p<0.05$) inclined with the highest percentage of broken rice in both samples. Drying at open areas would usually face fluctuation in temperature, compared to mechanical drying which temperature could be controlled efficiently as planned in the study. Fluctuation of temperature causes fissuring to rice grain due to moisture adsorption and leads to crack grain

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during the milling process, while higher drying temperature causes a higher rate of water removal from the centre of rice endosperm to the outer surface, resulting in crack grain (Inprasit and Noomhorm, 2001). There was no significant difference($P>0.05$) of head rice recovery at T2 and T3, but significantly ($p<0.05$) decreased at T4. In the rice milling industry, head rice recovery generates higher income for the millers. The price of milled rice is based on the percentage of broken rice. The milled rice with 15% broken (or called Super Tempatan 15; ST15) or contains 85% head rice is sold at RM 2,100/tonne, while at 10% broken (ST10) or contains 90% head rice and 5% broken (ST5) or 95% head rice is sold at RM 2,300/tonne and RM 2,500 - 2600/tonne, respectively. The broken rice has a lower price at RM 1,450 – 1,500/tonne. The ceiling prices at retailers are also different based on ST categories. The price for ST 15%, 10% and 5% are RM 1.80/kg, RM 2.40/kg and RM 2.60/kg, respectively. Therefore, at each step of the primary processing of rice, the millers work toward a higher percentage of head rice recovery.

Table 1. Head rice recovery and broken rice of red rice MARDI Warna 98 at different drying temperatures

Drying Treatment	Head Rice (%)	Broken Rice (%)
Ambient Temperature	90.59±0.60^{b}	9.41±1.17^{a}
40-45°C	95.50±0.38^{a}	4.50±0.38^{b}
46-50°C	95.93±0.47^{a}	4.07±0.47^{b}
51-55°C	91.23±0.63^{b}	8.77±0.63^{a}

Values are presented as mean±SEM of triplicates (n = 3). Values with different superscript within the same column are significantly different ($p<0.05$).

3.2 Antioxidant qualities

The antioxidant capacities of MW 98 (TPC, TFC and TAC) at different drying temperatures are shown in Table 2. Gallic acid served as the standard for determining TPC and its calibration curve is shown in Figure 3. The TPC of MW 98 at T1, T2, T3 and T4 were found to be 278.64±2.22, 286.84±2.90, 275.85±1.90 and 242.93±1.67 mg GAE/g dry weight, respectively. This result showed that TPC at ambient temperature was significantly ($p<0.05$) lowest than using a mechanical dryer. Similar findings were shown in TAC, as Cyanidine-3-glucoside equivalent of MW 98 and TFC, in which catechin was served as the standard and its calibration curve is shown in Figure 4. Drying under ambient temperature took a longer time to reach 13 – 14%% moisture content than using an oven, which was 8 days. The loss of phenolic compounds with the traditional practice of drying may have been caused by enzymatic processes. This process could not inactivate the degradative enzymes such as polyphenol oxidases, thus they can degrade phenolic compounds during long time drying procedures (Lim and Murtijaya, 2007). This might suggest quality deterioration of MW 98, thus leading to decreased TPC value. Besides, drying at an ambient temperature usually protects against the degradation of the active constituents, but it is slow and the metabolic process may continue longer, thus may lead to quality loss (Orphanides *et al.,* 2013). The highest TPC was found in MW 98 dried at T3, but not significantly ($p<0.05$) higher than the other two temperatures. Drying at T4 showed decreased TPC than T3 and T2. The TPC values decreased might be attributed to the degradation of heat-sensitive phenolics at high drying temperatures (Orphanides *et al*., 2013). The finding was similar to TFC. In addition to that, the TFC of MW 98 was found higher than reported by Ghazemsadeh *et al*. (2018), which showed TFC of red rice ranged from 40.15 to 823.88 mg QE/100 g. In the study, quercetin was used as the standard. Quercetin and apigenin was the most abundant flavonoid compound in black rice, whereas catechin and myricetin were the most abundant in red rice (Zhou *et al*., 2004). However, the highest TAC was found in MW 98 dried at T4, but not significantly ($p<0.05$) higher than the other two temperatures. The major constituents in red rice were proanthocyanidins such as catechins and epicatechins (Nawa and Ohtani, 1992), which have higher thermal stability (Goto-Yamamoto *et al*., 2010), thus might explain higher TAC at the highest drying temperature

Table 2. Total phenolic contents (TPC), total flavonoid contents (TFC), total anthocyanin contents (TAC) and DPPH radical scavenging activity of red rice MARDI Warna 98

Drying Treatment	TPC (mg GAE/ 100 g)	TFC (mg CAE/ 100 g)	TAC (mg CYE/100 g)	DPPH (% inhibition)
Ambient Temperature	242.93±1.67^{b}	967.61±4.00^{b}	49.71±1.30^{b}	83.39±0.33^{b}
40-45°C	278.64±2.22^{a}	1129.98±1.18^{a}	68.62±3.66^{a}	98.36±0.22^{a}
46-50°C	286.84±2.90^{a}	1173.0±1.17^{a}	69.48±4.65^{a}	98.14±0.38^{a}
51-55°C	275.85±1.90^{a}	1107.49±1.14^{a}	70.30±5.00^{a}	98.41±0.24^{a}
				Gallic Acid: 90.69±0.27
				BHT: 90.85±0.13

Values are presented as mean±SEM of triplicates (n = 3). Values with different superscript within the same column are significantly different ($p<0.05$).

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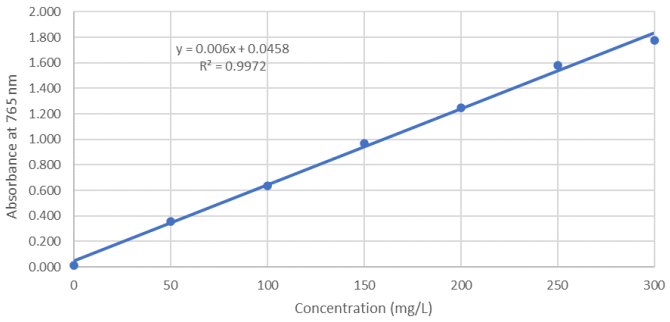

Figure 3. The calibration curve of gallic acid used to estimate TPC in red rice of MARDI Warna 98

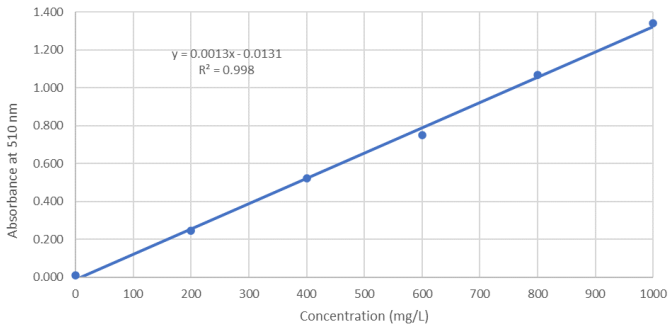

Figure 4. The calibration curve of catechin used to estimate TFC in red rice of MARDI Warna 98

tested. Although the TAC was found at T4, however, the temperature could not be recommended to dry MW 98, as the percentage of broken rice was significantly ($p<0.05$) highest at that temperature. The TAC of MW 98 was found at a similar level as found by Ghasemzadeh *et al*. (2018), at 77.87 mg Cy_3-GE/100 g.

Then, the antioxidant activity of MW 98 was determined since total phenolics and flavonoids have always been reported to have a close correlation with antioxidant activity in coloured rice. Besides, there was also a strong correlation between antioxidant activity and the TAC in the red pericarp grains (Oki *et al*., 2002). The DPPH assay was used to determine antioxidant properties, based on its ability to intercept free radicals as shown in Table 2. The MW 98 dried using a mechanical dryer scavenged DPPH radicals significantly ($p\leq0.05$) higher, which was more than 98% compared to ambient temperature at 83%. The results were directly proportionate with the lowest value of TPC, TFC and TAC in MW 98 drying at ambient temperature. There was no significant ($p>0.05$) difference in DPPH inhibition among the three drying temperatures but showed slightly higher in sample dried at 51-55°C.

4. Conclusion

Drying temperature affects physical qualities of MARDI Warna 98 variety, which head rice recovery decreased with higher temperature. However, the antioxidant activities of the coloured rice were not affected at the tested temperature; 40-45°C, 46-50°C and 51-55°C but significantly decreased at ambient temperature due to quality deterioration as a result of delayed drying. Thus, drying temperature for MW 98 is recommended at 46 - 50°C, to preserve not only antioxidant capacities but also to ensure good head rice recovery.

Conflict of interest

The authors declare no conflict of interest.

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