Physicochemical and Functional Properties of Bario Rice Varieties as Potential Gluten-Free Food Ingredients

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ABSTRACT

This study aimed to analyse the proximate composition, physical characteristics, and functional attributes of flour of four selected varieties of Bario rice (*Oryza sativa* L. Opaceae.): Bario Adan, Bario Padan, Bario Hitam, and Bario Kulit Merah. The rice flour was prepared using a semi-wet grinding method. The results on proximate composition showed that the moisture content ranged from 12.56% to 13.87%. All flours were high in crude fibre and low in fat content ranged 23.07 to 25.30% and 2.46 to 2.69%, respectively. Bario Padan exhibited the highest amount of crude fibre and protein (p<0.05). There were significant differences ($p<0.05$) observed for L, a*, and b* values, Bario Hitam shows the lowest L^* (5.29) and b^{*} (4.96) value (p<0.05). The functional properties among flour variety differ significantly $(p<0.05)$ with a range of 1.160 to 1.257 g/g for water absorption capacity and oil holding capacity ranged from 0.970 to 1.158 g/g. Highest swelling power was obtained in Bario Adan (5.594 g/g) while Bario Padan possessed highest water solubility index (0.099%). In conclusion, Bario rice varieties showcased favourable nutritional and functional traits, indicating their potential as gluten-free ingredients in the formulation of food products.

Keywords: bario rice, functional properties, proximate

INTRODUCTION

Gluten-related disorders, such as celiac disease, are increasing globally. Celiac disease impacts around 0.5% to 1% of the world's population (Hosseini *et al.* 2018). Gluten-free diets are the primary treatment for celiac disease, sparking interest from researchers and consumers. Rice flour has become a popular gluten-free alternative to wheat flour due to its availability, neutral effectiveness, hypoallergenic properties, neutral taste, and minimal impact on final products (Ronie *et al.* 2022; Burešová *et al.* 2023).

Rice (*Oryza sativa* L.) is the second most important crop after wheat, and Asia takes up to 90.6% of the consumer region, making it the world's largest producer and consumer (Rajamoorthy *et al.* 2015). Indigenous crops of Malaysia hold significant promise for the future, playing a vital role in ensuring both food security and health. Research indicate that the composition and properties of rice vary based on variety and location of rice production (Abera *et al.* 2021). Among the local potential crops are the Bario rice varieties. According to Khazanah Research Institute (2018), these rice varieties,

known for their fine and elongated grains, gentle and pleasant fragrance, and exceptional flavour. Bario rice is primarily grown in the Bario highland of Sarawak, cultivated by the local ethnic community using traditional methods and without artificial fertilisers (Thomas *et al.* 2014a). Bario Adan, Bario Padan, Bario Kulit Merah, Bario Merah, and Bario Hitam are notable varieties of Bario rice. Bario Adan, Bario Padan, and Bario Kulit Merah are white rice with 10‒18% amylose content, while Bario Merah and Bario Hitam are pigmented rice with red and black bran layers, respectively (Nicholas *et al.* 2014). The diverse attributes of Bario rice offer valuable benefits for future use.

Currently, there is limited new scientific research regarding the nutritional value and functional properties of these cultivars. Therefore, the objective of this paper is to determine the physicochemical and functional properties of four different types of Bario rice. Additionally, it is envisaged that the results generated from this study will be useful to popularise these exotic rice varieties as an alternative for glutenfree ingredients for better marketability at international levels.

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METHODS

Design, location, and time

The research was conducted in the Food Analysis Laboratory, Faculty of Applied Sciences, Universiti Teknologi MARA and Paddy and Rice Research Centre, Malaysian Agricultural Research and Development Institute, Selangor, Malaysia. The study was carried out from November 2022 to June 2023.

Material and tools

Bario Adan, Bario Padan, Bario Kulit Merah, and Bario Hitam were obtained from local farmers in Pa' Dalih, Bario, Sarawak. These rice varieties were harvested at the ripening stage. The paddy was dried until it reached below 14% of moisture content and dehulled using Satake Rice Machine THUS35B (Satake Engineering Co. Ltd, Japan).

The tools used in this study were a Kjedahl digestion and distillation unit (C. Gerhardt GmbH & Co. KG, Germany), Soxhlet extractor unit (Favorit S.p.A Hamelin Group, Italy), water bath, muffle furnace, drying oven, tabletop centrifuge, and colour spectrophotometer. Kjedahl selenium catalyst tablets, boric acid, sulphuric acid, sodium hydroxide, phenolphthalein, and petroleum ether used in this research were analytical grade.

Procedures

Semi-wet grinding of rice grains. The rice samples were grounded through the application of a semi-wet grinding method, which was adapted from Asmeda *et al.* (2016). The rice grains were soaked in water for 8 hr at a 1:1 rice-to-water ratio (w/v). Then, the excess water was drained, and the rice grains were allowed to dry at 30° C for removal of surface moisture before being ground using the multifunctional grinder (Ormismart Multifunctional Grinder Machine, ALX Sdn.Bhd, Malaysia).

Proximate analysis. The proximate composition includes total carbohydrates, moisture, protein, crude fat, crude fibre, and ash. Moisture, ash, and crude fibre were determined using standard AOAC methods (AOAC 2000). According to AOAC International (2000), the determination of total carbohydrates in foods can be performed using the enzymatic gravimetric method (Method 985.29).

Colour analysis. The colour profile of the rice was assessed using Chromameter (CR400, Konica Minolta, Japan). A 5g of rice flour sample was placed in a petri dish and pressed firmly to ensure complete coverage of the dish's bottom surface. The colour profiles were represented by L^* , a^* , and b^* values. The L^* value corresponds to the brightness level $(L=100)$ or darkness level $(L=0)$, the a^{*} value signifies the presence of redness (+a) or greenness (−a), and the b^* value indicates the presence of yellowness (+b) or blueness (−b).

Bulk density. A 5 g sample was filled into a 10 mL graduated cylinder and their weight was noted. The cylinder was tapped continuously until there was no further change in volume. The weight and final volume of the starch in the cylinder were noted, and the difference in weight and volume was determined. Bulk density was calculated as gram per millilitre (g/mL) .

Functional properties analysis. Water and oil absorption, swelling capacity, and water solubility index were determined according to Ronie *et al.* (2022) and Falade *et al.* (2014) with slight modifications.

Approximately 1.00±0.02 g of the sample was weighed and recorded as w_0 . The sample was then placed into a 15 mL centrifugal tube and reweighed, recorded as w_1 . Subsequently, 10 mL of distilled water or refined oil was introduced into the centrifugal tube. The mixtures were vortexed for 30 seconds at 5-mins intervals over a 30 mins period at room temperature. Following this, centrifugation was performed for 10 mins at 2,000 rpm. The resulting supernatant was weighed and recorded as w_2 . The quantity of water or oil absorbed by the flour was calculated as the difference and presented as the weight of water absorbed by 100 g of dry flour. Water and oil absorption capacity was calculated as below:

Water / Oil absorption capacity= $(\mathbf{w}_2 - \mathbf{w}_1)$ **/** \mathbf{w}_0

For swelling properties, 1 $g(w_1)$ of the rice flour sample was initially moistened with 30 mL of distilled water in a centrifuge tube. Subsequently, the tubes containing the sample were subjected to continuous shaking and heat at 80° C in the water bath. Following the cooling phase, the samples underwent centrifugation at 2,200 rpm for 15 mins. The supernatant was then removed from the centrifuge tube, and the residue weight was recorded (w_2) . The swelling capacity (g/g) was determined using below equation:

Swelling power (g/g)= w_2 / w_1

The supernatant obtained after the swelling power procedure was employed to assess the rice flour's water solubility index. Initially, the empty crucibles were weighed and noted as w_4 . Subsequently, the supernatant was cautiously transferred into the pre-weighed crucible. Following this, the supernatant was subjected to oven drying at 105°C for an overnight period. On a subsequent day, the desiccated supernatant was cooled to room temperature within a desiccator and then weighed as w_3 . The water solubility index was calculated using below equation:

Water solubility index, $\% = ((w_3 - w_4) / w_1) x$ **100**

Data analysis

All experimental data were performed in triplicate and analysed using IBM SPSS Statistics Version 28.0 (SPSS Inc., Chicago, USA) in a completely randomised study design. All experimental values were presented as mean±standard deviation (mean±SD). In general, a one-way analysis of variance (ANOVA) was employed to determine the significant differences in data among the experimental units. Duncan's Multiple Range test was utilised for multiple comparisons. Statistical significance was comparisons. Statistical significance was established at p<0.05.

RESULTS AND DISCUSSION

Table 1 outlines the physicochemical properties of rice flour, showing variability in proximate composition due to factors like rice variety, growth conditions, and post harvest procedures. Notably, Bario Kulit Merah stood out with the highest carbohydrate concentration, accounting for 52.75%. It is observed that there is a significant difference $(p<0.05)$ in carbohydrate levels between Bario Kulit Merah and Bario Padan (48.77%). According to prior research by Oppong *et al.* (2021), brown rice typically features a lower carbohydrate content compared to white rice. This discrepancy can be attributed to the fact that brown rice encompasses the bran and germ, which are the most nutrient-rich segments of the grain. Consequently, brown rice boasts a more comprehensive nutritional profile than its white counterpart, encompassing attributes such as fibre, magnesium, and a spectrum of other vital nutrients (Oppong *et al.* 2021; Ronie *et al.* 2022; Nicholas *et al.* 2014).

The moisture content of rice flour in the current study falls within the range of 12.56% to 13.87%, consistent with findings by Nicholas *et al*. (2014) for Bario cultivars. As outlined by the United States Department of Agriculture (USDA) Foreign Agricultural Service (2017),

Table 1. Physicochemical composition of Bario Rice

Parameter	Variety			
	B.Adan	B.Padan	B.Kulit Merah	B.Hitam
Proximate composition $(\%)$				
Carbohydrate	51.00 ± 0.73 ^b	48.77 ± 0.96 ^c	52.75 ± 0.74 ^a	50.16 ± 0.10^b
Moisture	$13.87 \pm 0.17^{\mathrm{a}}$	13.16 ± 0.05^{ab}	12.56 ± 0.62^b	13.58 ± 0.58 ^a
Protein	7.46 ± 0.08 ^c	9.68 ± 0.25 ^a	7.58 ± 0.27 °	8.65 ± 0.30^b
Crude fat	2.56 ± 0.08 ^{*b}	2.46 ± 0.20 ^{ab}	2.69 ± 0.04 ^a	2.26 ± 0.32^b
Crude fiber	23.90±0.60 ^b	25.30 ± 0.50 ^a	$23.10\pm0.55^{\rm b}$	$24.00 \pm 0.50^{\circ}$
Ash	1.20 ± 0.04^a	$0.64\pm0.18b$	$1.35 \pm 0.03^{\text{a}}$	$1.36 \pm 0.03^{\text{a}}$
Physical properties colour				
L^*	82.76 ± 0.13^b	84.20 ± 0.11 ^a	80.85 ± 0.09 ^c	60.47 ± 0.30 ^d
a^*	1.75 ± 0.10^b	1.30 ± 0.03 ^c	$1.79 \pm 0.05^{\rm b}$	5.29 ± 0.04 ^a
h^*	12.22 ± 0.17 ^b	11.74 ± 0.08 ^c	12.62 ± 0.07 ^a	4.96 ± 0.05 ^d
Bulk density (g/mL)	0.81 ± 0.00^b	0.83 ± 0.01 ^a	0.81 ± 0.00^b	0.84 ± 0.01 ^a

Data is expressed as means \pm standard deviations (n=3)

Means carrying different alphabets in a row differ significantly $(p<0.05)$

rice flour production standards indicate that the moisture content of rice can fall within the range of over 10% and under 15%. In general, moisture content below 14% is considered optimal for extended storage, particularly when dealing with cereals and cereal products (Hamel *et al.* 2020). Maintaining a moisture level below 14% serves to mitigate the risk of insect infestation and the proliferation of microorganisms, both of which can lead to a decrease in the shelf life of food products (Nicholas *et al.* 2013).

The protein content in all the Bario varieties was in the range of the average value of protein in unmilled rice (Juliano & Tuaño 2019), except in Bario Padan and Bario Hitam. Notably, the protein content in question meets the criteria to be classified as a noteworthy protein source, exceeding 5 g per 100 g, as stipulated by the Nutrient Reference Value in the 18C Act, Table II of the Food Regulations 1985 (MOH 2023). The highest crude protein content was 9.68% in Bario Padan, followed by Bario Hitam (8.65%). Ronie *et al.* (2022) investigated that milled Bario rice flour's protein content was between 6% to 9%. On the contrary, Nicholas *et al.* (2014) reported that the protein content of milled Bario rice flour ranges from 5.85% to 7.30%. Variations in crude protein content are primarily attributed to external factors, encompassing environmental parameters and storage conditions. Incorporating high-protein rice flour into food development endeavours can yield benefits by diminishing carbohydrate content, consequently reducing the glycaemic load during the absorption and digestion processes within the human body.

The fat content of Bario rice variaties ranged from 2.26% to 2.69%. This contrasts with findings by Ronie *et al.* (2022) suggesting that black-pigmented rice typically has higher fat content. The rationale behind this phenomenon lies in the outer bran layer, which is known to contain around 20% lipids or fats on a dry basis (Rathna Priya *et al.* 2019). The fat composition can exhibit slight variations depending on factors like specific rice variety, growth conditions, and processing techniques, including the presence of oils within the bran layer and the extent to which the bran layer is retained during processing (Ronie *et al.* 2022).

The crude fibre content exhibited variation from 23.07% to 25.30%, with Bario Padan having the highest amount and Bario Kulit Merah the lowest. The quantity of crude fibre in brown rice. Brown rice generally contains 1 to 3 g more fibre per cup than white rice, signifying its high dietary fibre content. While previous research (Ronie *et al.* 2022; Nicholas *et al.* 2014) reported the crude fibre content of polished Bario rice ranging from 0.21% to 2.05%, this study reveals that unmilled Bario rice possesses more than tenfold the amount of crude fibre, ranging from 23.07% to 25.30%, potentially du e to the bran fraction retained on unmilled rice (Nicholas *et al.* 2014; Ronnie *et al.* 2022).

According to the Food Regulation of Malaysia 1985, the ash content shall not yield more than 1.5%. Bario Padan has significantly lower ash content (0.637%) and conversely, Bario Hitam has higher ash content compared to other Bario rice varieties (p>0.05). These results are consistent with findings by Oppong *et al.* (2021) indicating higher ash content in pigmented rice flour compared to white rice flour. The ash concentration in different rice flours is dependent on the chemical composition within the bran layers of the caryopsis, and variations within commodities are further shaped by agricultural factors like soil and irrigation sources.

Food colour enhances visual appeal, influencing consumer preference (Phimolsiripol *et al.* 2012; Popov-Raljić *et al.* 2013). The degree of brightness of rice varieties in this study spans from 60.743 to 84.203 (Table 1). Among these, Bario Padan stands out with the highest brightness, followed by Bario Adan and Bario Kulit Merah. These varieties exhibit elevated L^{*} values owing to their pristine white appearance (Figure 1). Conversely, Bario Hitam rice flour had the lowest L^* values, attributable to the purple-hued outer layers of its rice grains. A positive a* value represents redness, whereas a positive b* value represents yellowness. The a* and b* values ranged between 1.303 and 5.290 and between 4.96 and 12.62, respectively. The a* values, with all measurements above zero, confirmed that the red tone dominates over the green in the rice flour. Additionally, the positive a* value for Bario Hitam was significantly high $(p<0.01)$ compared to the other varieties due to the colour pigmentation of the rice bran. On the other hand, the b^* value was significantly high ($p<0.01$) in Bario Kulit Merah. The colour characteristics of rice flour samples are linked to the pigments found in the rice bran (Loan *et al.* 2022). Rice

Figure 1. Colour diversity of Bario rice flour: (a) Bario Adan; (b) Bario Padan; (c) Bario Kulit Merah; (d) Bario Hitam

grains with a brownish-red pigment are termed red rice and are characterized by the presence of proanthocyanidins or condensed tannins, whereas purple pigmentation is identified as black rice is known for containing anthocyanins (Limtrakul *et al.* 2019).

The bulk densities of the rice flours ranged from 0.81 g/mL to 0.84 g/mL. Bulk density plays a crucial role in determining compressibility and cohesiveness, thereby preventing the formation of air gaps. The composition and particle size of flour are commonly recognised as influential factors impacting bulk density (Nkurikiye *et al.* 2023). Notably, both starch and fat contents display a positive correlation with the bulk density of flour. Bario Padan and Bario Hitam exhibit higher bulk densities, signifying their potential suitability as effective thickening agents in food preparations.

The Water Absorption Capacity (WAC) of food products refers to the ability of the flour to

interact with water (Awuchi *et al.* 2019). WAC holds a pivotal role in the advancement of cerealbased food products, as it effectively enhances cohesiveness and consistency, and imparts body to the food (Atuna *et al.* 2022). As outlined in Table 2, the WAC of all samples ranged from 1.16 g/g to 1.26 g/g , with significant differences among samples ($p<0.05$). Rice flour derived from Bario Adan and Bario Kulit Merah demonstrated the highest WAC value, each registering at 1.26 g/g. The levels observed in this investigation are below those observed by Thomas *et al.* (2014b). However, results from Ronie *et al.* (2022) corroborated current findings, reporting WAC contents within the range of 1.21 g/g to 1.26 g/g, with the peak value found in Bario Adan. The substantial presence of hydrophilic groups within starch molecules contributes to the velvety texture, pliancy, and viscosity of the resulting food product. Rice flour with lower WAC levels is well-suited for making rice bread, while those with high WAC can be used in bakery products, effectively averting moisture loss, and preserving the delectability of bread, cakes, sausages, and even serving as a soup thickener (Han *et al.* 2012).

Oil Absorption Capacity (OAC) represents the physical entrapment of oil by protein through its non-polar side chains (Lira *et al.* 2023). The OAC of the rice flours ranged from 0.97 g/g to 1.16 g/g. The highest OAC is presented in Bario Padan, followed by Bario Adan and Bario Kulit Merah which has no significant difference $(p>0.05)$ from each other but has a significant difference (p<0.05) from Bario Hitam. OAC is a key feature that elevates the shelf life and mouthfeel while preserving flavour in food products (Atuna *et al.* 2022). The notable high OAC value indicates its suitability for incorporation into lipid-rich foods, such as cakes. More pronounced hydrophobic

Parameter	Variety				
	B. Adan	B. Padan	B. Kulit Merah	B. Hitam	
Water absorption capacity (g/g)	1.26 ± 0.02^a	1.17 ± 0.02^b	$1.26 \pm 0.02^{\text{a}}$	1.16 ± 0.02^b	
Oil absorption capacity (g/g)	$1.11 \pm 0.02^{\circ}$	1.16 ± 0.10^a	1.05 ± 0.07 ^{ab}	0.97 ± 0.06^b	
Swelling power (g/g)	5.59 ± 0.07 ^a	$4.40\pm0.15^{\circ}$	5.20 ± 0.12^b	5.21 ± 0.06^b	
Water solubility index (%)	0.05 ± 0.00^b	0.10 ± 0.01 ^a	0.05 ± 0.02^b	0.04 ± 0.00^b	

Table 2. Water absorption capacity, swelling power, and water solubility index of Bario rice

Data is expressed as means \pm standard deviations (n=3)

Means carrying different alphabets in a row differ significantly $(p<0.05)$

proteins tend to expose a greater number of nonpolar amino acids to fats, consequently enhancing the efficacy of lipid binding processes (Oppong *et al.* 2021).

The swelling of starch granules occurs during gelatinisation because the hydroxyl groups within the starch granules formed new hydrogen bonds with water molecules due to the disruption of hydrogen bonds between the hydroxyl group in the double helices of starch molecules (Zhou *et al.* 2021). The swelling capacity range in this study was between 4.40 g/g and 5.59 g/g. Generally, Bario Adan exhibited the highest swelling capacity value whilst Bario Padan presented the lowest value. The outer bran of the pigmented rice grains is said to delay water penetration into the starch granules, consequently lowering the swelling capacity of the flour. Hence, the post-harvest operations of this rice play a crucial part in managing the swelling capacity (Awuchi *et al.* 2019). Swelling capacity is depicted as an important functional property that can impact the quality of food products such as hydration capacity which measures the ability of the substance to absorb water and form a gel-like substance. This property is important in many food products, such as bread, cakes, and pastries, where the dough or batter needs to hold its shape and texture during baking (Huang *et al.* 2019). Besides that, swelling capacity can also affect the emulsification properties of flour, which is essential in many food products, such as sauces, dressings, and mayonnaise. The flour's ability to absorb water and form a gel-like substance can help stabilise the emulsion and prevent separation.

The Water Solubility Index (WSI) is a crucial parameter indicating the amount of solute dissolved in a solvent at equilibrium, often used to measure starch conversion during extrusion and the release of soluble polysaccharides from the starch granule (Taverna *et al.* 2012). Low WSI in flour implies lower adhesiveness and stickiness in food products but a higher structural preservation (Kraithong *et al.* 2018). Bario Padan (0.10%) exhibited a significantly higher WSI value $(p<0.05)$ compared to other varieties, influenced by compounds like starch and fibre. Research by Ronie *et al.* (2022) stated that pigmented rice usually elucidated higher WSI but lower SP than white rice flour due to the leached phenolic compounds during processing. The inverse

relationship between swelling power and WSI in rice flour is influenced by the ratio of soluble to insoluble amylose content, molecular weight and shape of amylopectin, and granule size of starch (Lan *et al.* 2015, Zhang *et al.* 2023). Although amylose and amylopectin ratios influence swelling and solubility, stronger intermolecular interactions and higher hydrophobicity contribute significantly to the variance in swelling power and solubility (Aidoo *et al.* 2022).

CONCLUSION

Bario rice flour demonstrates notable protein and fibre content. The pigmentation of the rice bran influences the colour profiles of Bario rice flour. The swelling capacity displayed a high level across all Bario rice flour types. Notably, Bario Padan rice flour exhibited a high WSI, potentially attributed to elevated phenolic compound concentrations released during processing. However, further research should focus on investigating Bario rice's nutrient bioavailability and compatibility in various food processing methods. Such investigations would deepen our understanding of Bario rice flour varieties and encourage the utilisation of indigenous crops to create gluten-free products.

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DECLARATION OF CONFLICT OF INTERESTS

The authors state that they have no conflict of interest.

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