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### Research Article

### Nutritional and antioxidant profile of black rice-based (Poireiton Chak-hao and Chak-hao Amubi) traditional foods of Manipur

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### **Abstract**

Black rice becomes a potential source of phytonutrients among the grains. Traditional food is crucial for food and nutrition security in the present food system. It is necessary to have scientific information on the nutritional profile of black rice-based traditional foods to promote the consumption of black rice. Hence, the present study aimed to evaluate the nutritional and antioxidant content of black rice-based traditional foods. In Manipur's Andro Village, black rice is consumed in traditional foods such as kheer, thaotan, tanphut, kabok matum, etc. Two black rice cultivars Poireiton Chak-hao (PC) and Chak-hao Amubi (CA) were used to prepare black rice-based traditional foods such as PC-Kheer, CA-Kheer, PC-Thaotan, CA-Thaotan, PC-Tanphut, CA-Tanphut, PC-Kabok matum. The foods were analyzed for proximate, mineral, polyphenol content and antioxidant activity using the standard procedure. Results showed significant differences (p<0.05) in moisture, protein, fat, carbohydrate, iron, zinc, polyphenol content and antioxidant activity among the black rice-based traditional foods. The values are presented on dry weight basis. PC-kheer had the highest antioxidant activity (51.21 % DPPH), protein (13.49 %), fat (5.08%) and polyphenol content (55.56 mg GAE/100 g) among the traditional foods. PC-Kabok matum recorded the highest iron (4.53 mg/100g) and carbohydrate content (84.82%), whereas PC-Tanphut had the highest zinc content (2.7 mg/100 g). Thus, black rice-based traditional foods have a promising role in providing food and nutrition security to black rice consumers.

**Keywords**: Black rice-based traditional food, Antioxidant, Nutritional, Polyphenol

### INTRODUCTION

Traditional food plays a substantial role in preserving culture and promoting food security in today's world (Schuster et al., 2011; Ghosh et al., 2023). Food security within indigenous communities depends on ensuring access to traditional food resources (Sidig et al., 2023). Rice-based cuisine is one such traditional foods which embodies the present food system. Rice is the staple diet of the people of Southern and Eastern parts of India. Different types of rice, such as white rice, pigmented rice, parboiled rice, and brown rice, are consumed worldwide. Pigmented rice includes red rice and black rice.

In India, black rice is not generally cultivated for commercial purposes except in a few parts of India such as Manipur, Assam, Odisha and Tamil Nadu (Devi and Pathak, 2023). Black rice is called Chak-hao meaning

'delicious rice' in Manipuri. In Manipur, black rice is being cultivated in traditional practices by the farmers covering only a few areas (7-10 %) of total cultivated land as it has low yield and requires cultivation for three consecutive years as a cultural practice (Borah et al., 2018; Devi and Pathak, 2023). Commercially, most of Manipur's black rice farmers grow it in less than 1 hectare of the land holding (Borah et al., 2018). Despite this, the farmers of Manipur have started cultivating black rice for commercial purposes by successfully exporting one metric ton of organic black rice from Manipur to Europe through Manipur Organic Mission Agency. Black rice has a substantial role in promoting the culture and traditions of Manipur, considering its place in the religious ceremonies of the people (Borah et al., 2018; Asem et al., 2019). Manipur got the Geographical Indication (G.I) tag for black rice (Chak-hao) in 2020 (Devi and Pathak, 2023). The different genotypes of black rice of Manipur include Chak-hao Amubi, Chak-hao Poireiton, Wairi Chak-hao, Khurkhul Chak-hao, Pong Chak-hao, Chak-hao sempak, Khunou Chak-hao and Chetemo (Asem et al., 2019). Black rice from Manipur has scented and nutty flavours, which turn a deep purple colour when cooked (Asem et al., 2019; Saha et al., 2022). Black rice is not consumed as a daily or regular meal but as a delicacy in religious and cultural ceremonies among 'Meitei community of Manipur' (Borah et al., 2018; Asem et al., 2019).

Black rice has many health benefits, such as antiinflammatory properties, anti-atherogenic, cancer prevention, improving iron deficiency anaemia and antidiabetic properties (Rahim et al., 2020). Black rice has a lower glycaemic index than white rice (Andriania et al., 2020; Ou et al., 2023). There are proposed potentials for black rice in treating diabetics, celiac diseases with positive outcomes (Andriania et al., 2020; Piazza et al., 2023). Chak-hao of Manipur has a total anthocyanin content of 692 to 740 mg/kg cyanidin 3-glucoside/kg of dried powdered samples of black rice (Asem et al., 2015). However, most of the Indian population consumes white polished rice even though there is enough evidence of the importance of brown rice and pigmented rice. Thus, the inclusion of black rice in regular meals is still far less. The strategy to improve utilization of black rice is enhancing the consumption of the traditional foods of black rice.

In Manipur, black rice is also used for the preparation of different types of rice-based traditional foods such as Chak-hao kheer, thaotan, tanphut, chak-hao kabok, utong chak and yu (black rice beer) (Borah et al., 2018). Chak-hao kheer is the black rice kheer prepared with milk and consumed as dessert in Manipur households. Thaotan is a flatbread of black rice fried in oil. Tanphut is a flatbread of black rice wrapped in banana leaves and steamed. Chak-hao kabok is a popped black rice and Chak-hao kabok matum is the black rice laddoo prepared from popped black rice with jaggery. Utong chak is cooking rice by putting it inside a bamboo stick. These black rice-based traditional foods have been consumed for ages and passed on from generation to generation. However, due to busy lifestyles, changing food patterns, and negligence of the importance of these foods, traditional food consumption is less, although it is slowly reviving. Promoting traditional food also requires knowing the nutrient content of the consumed foods. Thus, the present investigation was conducted to study traditional foods' nutritional profile and phytonutrient content.

### **MATERIALS AND METHODS**

### Preparation of black rice-based traditional foods

Preparation methods for black rice-based traditional foods were collected from Andro Village, Imphal East District, Manipur (24.7480°N, 94.0420°E). Andro is one of the Scheduled Caste Meitei Village of Manipur,

where Meitei's distinct cultures, religions, traditions and food habits are preserved and continue to this day. A total of 45 households (who cultivate and consume black rice) were selected through "Snowball Sampling" method (Harris *et al.* 2009) to elicit knowledge associated with the preparation methods of black rice-based traditional recipes. The respondents were interviewed using a self-structured schedule; thus, information of traditional black rice-based recipes was obtained.

Two black rice varieties Chak-hao Amubi and Poireiton Chak-hao were used to prepare the black rice based traditional foods. Chak-hao Amubi and Poireiton Chak-hao were procured from Seijang village, Manipur and Krishi Vigyan Kendra, Andro, Imphal East District, Manipur, respectively. The black rice-based traditional foods such as *Thao-tan*, *Tanphut*, *Chak-hao Kheer* and *Chak-hao Kabok matum* were prepared as per the methods collected from the households of Andro (Fig.1.). The preparation methods of traditional recipes are explained below.

#### Thaotan

Thaotan (PC-Thaotan and CA-Thaotan) is a fried flatbread prepared from black rice varieties Poireiton chakhao (PC) and Chak-hao Amubi (CA). Black rice was washed and soaked overnight in a container, drained the water the next day and ground into a smooth paste using a mixer grinder. Then refined flour and sugar were added and mixed properly to make a dough that was then made into small balls. After that greased the hand with a little oil, made the balls into flatbread with fingers, and flipped them from palm to palm. Then oil was added to heated tawa and fried the thaotan till it becomes crispy.

### **Tanphut**

Tanphut (PC-Tanphut and CA-Tanphut) is a steamed black rice flatbread. Black rice was soaked in a container overnight, drained the water and ground into fine rice flour. Then sugar and little water were added to the rice flour, mixed properly, and kneaded into a dough, making the shape of flatbread with the greased hands. Then, the banana leave was greased with oil and the flatbread was kept in the banana leaves. Then banana leave was folded and tied it properly. A known quantity of water was added to an open pan, the folded banana leaves were kept, and the lid was covered. The mixture was boiled till it was cooked.

### Chak-hao Kheer

Chak-hao Kheer (PC-Kheer and CA-Kheer) is a black rice kheer. The preparation method involved washing and soaking the black rice overnight. A known quantity of water was taken in an open pan. Rice was added to boiling water and cooked it until it was done. Chopped cashew nut, shredded coconut, liquid and powdered milk were added and mixed properly. Then, sugar was added, stirred occasionally, and the kheer cooked until

it was thick and soft.

### Chak-hao Kabok matum

It is a black rice laddoo (PC-Kabok matum) prepared from the variety Poireiton Chak-hao. Black rice paddy grain was popped in a vessel. The husk was removed and kept it aside. In a karahi, jaggery was melted and poured into the popped black rice, then bound into a round shape like laddoo.

## Nutrient analysis of black rice-based traditional foods

The prepared foods were dehydrated using the hot air oven at 45-50°C till the favourable moisture content was achieved. Mixer grinder was used to grind the dehydrated product and the powdered samples were used for proximate analysis, mineral estimation and phytonutrient content analysis (polyphenol content and antioxidant activity). Moisture, ash, crude fibre, protein, fat and total carbohydrate contents were analyzed according to the standard procedures of the Association of Analytical Chemists (2000).

# Proximate analysis of black rice-based traditional foods

The moisture content of black rice-based traditional foods was estimated using oven-drying. The crude protein was estimated by Kjeldahl distillation method with the digestion of organic nitrogen using sulphuric acid, liberated as ammonia and distilled into a known volume of acids, which is then titrated back. The obtained nitrogen value was multiplied by factor 5.95 and reported as a percentage. Fat content was estimated using petroleum ether as a solvent in Soxhlet apparatus and reported as crude ether extract. Total ash content was estimated by heating the samples in a muffle furnace at 600°C for 4 to 5 hours till the ash colour turned grey or whitish grey. Crude fibre was estimated by the acidalkali digestion method in Pelican's Fibre Estimation System. Fat free sample was treated with sulphuric acid (0.255 N) and sodium hydroxide (0.313 N). The residue obtained after acid and alkali wash was dried in a crucible and its weight was taken (W<sub>1</sub>). The dried residue was then kept in a muffle furnace at 600°C for 2 hours and its weight was recorded (W2). The total carbohydrate content was obtained after deducting the sum of the value for moisture, protein, fat, ash, and crude fibre from 100.

### **Estimation of mineral content**

Iron, zinc, copper and manganese content in traditional foods were analyzed following the standard procedures (AOAC, 2000) by Atomic Absorption Spectrophotometry. Samples were ground finely and wet digested with triacid mixtures (nitric acid, perchloric acid and sulphuric acid) and minerals were estimated as mentioned.

### **Estimation of phytonutrient content**

Total phenol content was estimated using the Spectrophotometric method with Folin-Ciocalteau Reagent (Singleton and Rossi, 1965). Powdered samples were defatted before the analysis and dried in a hot air oven at 60°C. The defatted samples (1g) were treated with ethanol in Pestle and mortar, and the homogenate was centrifuged for 20 minutes at 10000 rpm. The supernatant was evaporated to dryness and the residue was dissolved in a known volume of distilled water. Aliquots (0.2-2ml) were taken, and the volume (3 ml) was made up with distilled water and 0.5 ml of Folin Ciocalteau Reagent was added to each tube. Then two ml of 20% Na<sub>2</sub>Co<sub>3</sub> solution was added after 3 minutes and mixed thoroughly. Test tubes were placed in boiling water bath for exactly one minute, cooled and absorbance was measured at 650 nm against a reagent blank. Polyphenol content was expressed as mg of Gallic Acid Equivalents per q of extract.

Antioxidant activity was estimated using DPPH (2, 2-diphenyl-1 picryl-hydrazyl) as a free radical method (Yu et al.,2002). 10g of defatted sample was taken and refluxed for half an hour using 100 ml of methanol and the extract was centrifuged at 500 rpm for 10 minutes and supernatant was used for estimation. Freshly prepared DPPH solution (0.1 ml) was added in to methanol solution (3.9 ml) to individual sample (0.1 ml), kept for reaction at ambient temperature for 60 minutes, then absorbance was taken at 517 nm against a blank of pure methanol. Radical scavenging power of DPPH was determined from the difference in absorbance with or without antioxidants and expressed as per cent DPPH.

### Statistical analysis

One-way Analysis of Variance and Duncan's Multiple Range Test (DMRT) were performed to test the mean's significance differences. IBM SPSS software version 23 was used to perform statistical analysis. All the experiments were done in triplicates and data are shown as mean values with standard deviation.

### **RESULTS AND DISCUSSION**

# Proximate content of the black rice-based traditional food

The proximate composition of black rice-based traditional food is shown in Table 1. Significant differences (p<0.05) were observed among the moisture, protein, fat, and total carbohydrates samples. Results indicated that CA-*Thaotan* (9.14%) had the highest moisture content followed by CA-Tanput (8.99%), PC-*Tanphut* (8.58%), PC-*Thaotan* (7.70%), PC- *kabok matum* (7.24%), CA-kheer (6.84%) and PC-Kheer (6.61%). Sharma *et al.* (2024) found a moisture content of 4.84% to 25.30% in traditional cereal-based recipes. Protein content among the traditional foods was significantly different and ranged from 4.77% to 13.49%.

Table 1. Proximate composition of black rice-based traditional foods (%, DW)

Traditional black rice foods	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Crude fibre (%)	Total carbohydrate (%)
PC- Thaotan	7.70 ±0.00°	7.10 ± 0.19°	4.49 ± 0.01 <sup>b</sup>	1.57 ± 0.01	1.73 ±0.13	78.97 ± 0.33 <sup>d</sup>
PC-Tanphut	8.58 ±0.06 <sup>b</sup>	5.86 ±0.21de	$2.00 \pm 0.05^{\circ}$	1.24 ± 0.11	1.70 ± 0.26	81.85 ± 0.41 °
PC-Kheer	6.61 ±0.01 <sup>d</sup>	13.49 ±0.24 <sup>a</sup>	$5.08 \pm 0.03^{a}$	1.58 ± 0.01	1.88 ± 0.24	72.92 ± 0.51 <sup>e</sup>
PC-kabok matum	7.24 ±0.17 <sup>cd</sup>	$4.77 \pm 0.19^{f}$	$2.03 \pm 0.02^{c}$	1.51 ± 0.13	1.13 ± 0.91	84.82 ± 0.80 <sup>a</sup>
CA-Thaotan	9.14 ± 0.15 <sup>a</sup>	6.19 ± 0.21 <sup>d</sup>	$1.81 \pm 0.00^{d}$	1.72 ± 0.12	1.63 ± 0.03	81.26 ± 0.17°
CA-Tanphut	8.99± 0.12 <sup>ab</sup>	$5.43 \pm 0.19^{e}$	1.76 ± 0.05 <sup>d</sup>	1.79 ± 0.19	1.72 ± 0.05	82.03 ± 0.29 <sup>b</sup>
CA-kheer	6.84 ± 0.41 <sup>d</sup>	$8.54 \pm 0.20^{b}$	$4.32 \pm 0.12^{b}$	1.52 ± 0.17	1.51 ± 0.03	78.78 ± 0.66 <sup>d</sup>

PC: Poireiton Chak-hao, CA: Chak-hao Amubi, DW-Dry weight basis, Values are mean of three replications ± Standard Deviation, Different superscripts in a column indicate significance of difference (P<0.05) using Duncan's Multiple Range Test

PC-kheer possessed the highest protein content (13.49 %) followed by CA-Kheer (8.54 %) and the lowest was observed in PC-kabok matum (4.77 %). Higher protein content was linked to the role of milk, and coconut was included in the recipe preparation of kheer, as well as the varietal difference of black rice in protein content. Das and Samantaray (2023) reported that the protein content of rice-based traditional cuisines of Odisha was between 1.1 to 52 g per 100 g. Lavanya and Pinky (2019) found that the protein content of different ricebased traditional cuisines was 3.45 % to 15.48 %. Sharma et al. (2014) reported that cereal-based ethnic foods had a 5.1 to 6.2 % protein content. In addition to this, black rice is rich in protein. As per previous findings (Chanu et al., 2016), protein content in Poireiton chak-hao (PC) and Chak-hao Amubi (CA) was 10.4% and 9.05 %, respectively. Among the cereals, rice protein quality is better than that of other cereals, such as maize and wheat (Jayaprakash et al., 2022). These traditional foods can be promoted and included in people's daily diet.

Fat content in rice affects the taste of the cooked rice and rice rich in fat is tastier. Crude fat content in black rice-based traditional foods was significantly different (p<0.01) and found to be highest in PC-Kheer (5.08 %), followed by PC-Thaotan (4.49 %) and CA-Kheer (4.32%). CA-Thaotan (1.81 %) and CA-Tanphut (1.76 %) possessed low-fat content and were almost on par with the fat content in brown rice of Chak-hao Amubi (1.91 %), as reported by Chanu et al. (2016). The significantly higher fat content in PC-Thaotan from the rest of the black rice-based traditional food was due to the inclusion of oil in the preparation method and the higher fat content in raw Poireiton chak-hao cultivar (2.94%). Further, these traditional foods had crude fibre and ash content of 1.13 to 1.88 % and 1.24 to 1.79 %, respectively. PC-kabok matum had the highest total carbohydrate content (84.82 %), followed by CA-Tanphut (82.03 %) and PC-Tanphut (81.85%), whereas, PC-Kheer (72.92%) had the lowest total carbohydrate content. Thus, traditional black rice-based foods are rich in protein, carbohydrates and crude fibre and can provide nutrition security for the black rice consumer when included in a regular meal.

## Mineral content of black rice-based traditional foods

The mineral content of black rice-based traditional food is revealed in Fig. 2. Iron, zinc and manganese content were statistically different among the black rice-based traditional foods (p<0.05). PC-kabok matum exhibited the highest iron content (4.53 mg/100 g) followed by PC -Kheer (3.7 mg/100 g) and CA-Kheer (3.57 mg/100 g). Zinc content was significantly higher in PC-Tanphut (2.7 mg/100g) followed by CA-Tanphut (1.97 mg/100 g), PC -Kheer (1.93 mg/100 g), PC-Thaotan (1.93 mg/100g), CA-kheer (1.67 mg/100 g) and PC-Kabok matum (1.33 mg/100 g). Manganese content ranged from 0.11 mg/100 g in CA-Tanphut to 1.27 mg/100 g in PC-Thaotan. Copper content was not significantly different among the black rice-based traditional foods and ranged from 0.19 mg to 0.39 mg/100 g. Black rice has higher iron, zinc, manganese and calcium than white rice and is necessary for promoting and maintaining health (Sampong et al., 2011; Chanu et al., 2016; Rahim et al., 2022). Iron in black rice is important in maintaining normal levels of haemoglobin and supporting cognitive ability (Thanuja and Parimalavalli, 2018), while zinc is required for normal growth, prevention of infection and improving the immune system (Mawouma et al., 2023). Established evidence suggests the potential benefit of black rice in reducing iron deficiency anaemia. The present study showed that traditional black rice-based foods had a good amount of iron and zinc even though the values were lower than those in uncooked black rice. Thus, black rice can be promoted to combat the hidden hunger among the people.

# Phytonutrient content of black rice-based traditional foods

Black rice is known for its high anthocyanin, phenolic compounds and antioxidant activities. Two black rice cultivars, i.e. Poireiton chak-hao (PC) and Chak-hao amubi (CA) of Manipur are also rich in phenolic and anthocyanins compounds, which offers high radical scavenging activity (Bhuvaneswari et al., 2020). Free



Fig. 1. Black rice-based traditional foods of Manipur

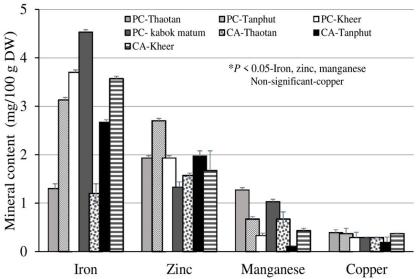


Fig. 2. Mineral content of black rice-based traditional foods

radicals are produced in the body and cause a lot of health disorders such as cancer, atherosclerosis, cataract, cardiovascular disease, diabetes mellitus, and inflammation (Bhuvaneswari et al., 2020; Zheng et al., 2020; Tyagi et al., 2022). Dietary antioxidants found in pigmented rice, fruits, and vegetables may curb the effect of high levels of reactive oxygen species. The black rice-based traditional foods for their phytonutrient content are shown in Fig.3. They had shown significant differences in antioxidant activity and polyphenol content (p<0.05). The polyphenol content ranged from 20.87 mg GAE/100g in CA-Thaotan to 55.56 mg GAE/100g in PC-kheer. A similar trend of antioxidant

activity was also found and ranged from 10.35 % DPPH (CA-*Thaotan*) to 51.21 % DPPH (PC-*Kheer*) in these traditional foods. PC-*kheer* recorded the highest polyphenol content (55.46 mg GAE/100g) and antioxidant activity (51.21 % DPPH) among these foods.

Furthermore, it exhibited that CA-kheer had a lower content of polyphenol (34.47 mg GAE/100 g) and antioxidant activity (30.77 % DPPH) when compared to PC -Kheer. Differences in polyphenol content and antioxidant activity in the uncooked black rice cultivars were also observed in previous studies (Asem et al., 2015; Chanu et al., 2016). Poireiton chak-hao (PC) had an antioxidant activity of 72.52 % DPPH and polyphenol

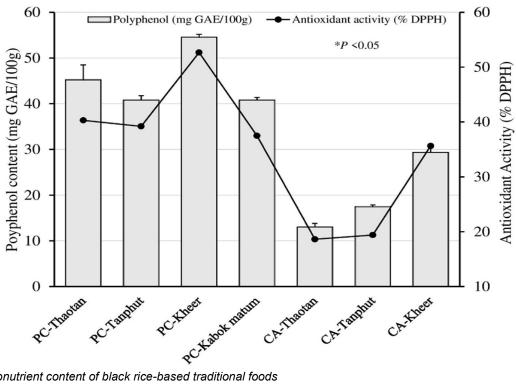


Fig. 3. Phytonutrient content of black rice-based traditional foods

content of 62.33 mg GAE/100 g, whereas Chak-hao amubi (CA) had 61.59 % DPPH antioxidant activity and 19.75 mg GAE of polyphenol (Chanu et al., 2016). Further, adding certain ingredients in these traditional foods and preparation method could influence these foods antioxidant activity and polyphenol content. The results further indicate that the higher the polyphenol content, the higher the antioxidant activity in all the black rice-based traditional foods.

Phenolic compounds present in grains, beverages, fruits and vegetables provide health benefits when consumed regularly. Black rice has a similar cellular antioxidant activity to ascorbic acid (Tyagi et al., 2020). Cyanidin -3-Glucoside of black rice can improve diabetic nephropathy through antioxidative inflammatory properties (Zheng et al., 2020). Black rice contains natural antioxidants such as tocopherols, tocotrienols, gamma oryzanol and thus is claimed to prevent destruction of pancreatic beta cells, indicating black rice's antidiabetic role (Verma and Srivastav, 2020). Therefore, black rice-based traditional foods could be a source of nutraceuticals for indigenous people who consume black rice in their regular meals.

### Conclusion

The present study showed the nutritional and phytochemical content of black rice-based traditional foods. They are rich in iron, protein, carbohydrates, fibre, polyphenols and antioxidants. Black rice is a potential source of dietary antioxidants and other bioactive substances. Black rice kheer is a good source of protein and antioxidant. Kabok matum, a form of laddoo (recipe) prepared from black rice, is also a good source of iron among black rice-based traditional foods. Therefore, promoting black rice-based traditional food can be one of the strategies to enhance the utilization of black rice in daily meals in the existing food system.

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### **Conflict of interest**

The authors declare that they have no conflict of interest.

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