



An Assessment of Different Phytochemical Screening and Analysis of Polyphenol Compounds in Gamma-Oryzanol

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Article History	Abstract
Received: 06 June 2023 Revised: 05 Sept 2023 Accepted: 17 Oct 2023	<p>The current research work's concentration is on mineral content analysis and polyphenol compound analysis. Using proximate analytical techniques, the chemical composition (ash, fat, proteins, fiber, and gross energy) was identified. Utilizing mass spectrometry and liquid chromatography, the phenolic profile was identified. It was discovered that they contain Gallic Acid, Caffeic Acid, Catechin, Epicatechin, Epigallocatechin Gallate, P-coumaric Acid, O-coumaric Acid, Naringin, Rosmarinic Acid, Quercetin, Naringenin, Rutin, Phytic Acid, Ferulic Acid, Chlorogenic Acid, Kaempferol, and Hydroxybenzoic Acid. The results showed great variations in each parameter of the nutritional and bioactive components. It was discovered during the phytochemical screening of the Gamma Oryzanol that they contain a variety of minerals, including Potassium, Phosphorous, Phosphorous, Magnesium, Calcium, Iron, Zinc, Sodium, and Copper.</p>
CC License CC-BY-NC-SA 4.0	<p>Keywords: Rice bran, phenolic chemical, and nutritional value, Gamma Oryzanol, Cholorgenic acid.</p>

1. Introduction

One of the most often consumed staple foods worldwide is rice (*Oryza sativa* L.). Based on its hue, the caryopsis can be distinguished as brown, white (polished brown rice), red, purple, or black. Around 755 million tons of rice, or 504 million tons of milled rice, were produced globally between 2019 and 2020 (1). More than 5,000 different types of rice are known to exist in Thailand, which is regarded as one of the origins of rice diversity in the world (2, 3). At the moment, specialized kinds of rice are being produced that pay attention to Thai consumers' needs. Consequently, a type of rice has emerged that is advantageous in a variety of fields with diverse conditions. Paddy is the name for rice gathered from the field. By removing the inedible husk, the rice grain is changed into a shape that is acceptable for human eating during the essential process of milling. Treatment of menopausal problems and a rise in muscle mass are the main goals of a milling procedure [3]. The husk, straw, and bran are the three main byproducts of the manufacturing of rice. About 8–10% of the rice grain makes up rice bran, a significant and underused byproduct of the rice milling process (9, 10). As a result, rice milling generates more than 80 million tons of bran globally (11).

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In recent years, research has focused on rice bran's possible biological applications. than the other fractions used for grinding rice (16). About 20% of rice bran is made up of lipids. Due to its distinct fatty acid composition and nutraceutical benefits, rice bran oil has garnered interest (17). However, bran is underappreciated as a source of human diet and has historically been used as animal feed (18, 19). The biological effects of rice bran, including its putative anti-inflammatory, antimutagenic, anticarcinogenic, antibacterial, and antioxidant activities, have been examined recently (20).

Since rice bran is becoming more popular as a source of nutraceuticals that promote health and new variations of these raw materials have been created, It is important to look at the profile of Thai rice bran variants with unique grain characteristics for practical uses.

2. Materials And Methods

The Plant of *Oryza Sativa* was collected from Bilaspur Chhattisgarh Resion and its botanical identity was authenticated & confirmed by Department of Botany Semi Govt, Kalyan Post Graduate Autonomous College, Bhilai Nagar, Distt- Durg (C.G.) & dried under shade. A voucher specimen has been deposited in Department of Botany, Semi Govt, Kalyan Post Graduate Autonomous College, Bhilai Nagar, Distt- Durg (C.G.) for future reference.

Evaluation of Physico-chemical properties:

Organoleptic Properties and different physicochemical parameters, such as total ash, extractive value, and loss on drying, Residue on Ignition, Residual Solvent, Melting Point. were calculated as per the Indian Pharmacopoeia in order to determine the evaluation of physicochemical properties. Following the guidelines of the Association of Official Analytical Chemists (AOAC), the proximate composition of various rice bran kinds was assessed, including dry matter content, ash content, crude fiber, crude protein, crude fat, gross energy, and mineral content (22).

In a nutshell, ash content was calculated using method 942.05, crude fiber was calculated using method 962.09, and moisture content was assessed using method 934.01. The Kjeldahl technique was used to determine the crude protein, which was computed as nitrogen 6.25 (method 2001.11). Soxhlet extraction was used to quantify the amount of crude fat (method 2003.06).

An oxygen bomb calorimeter was used to measure gross energy in duplicate. Mineral concentrations were measured using an Agilent Following the guidelines of the Association of Official Analytical Chemists (AOAC), the proximate composition of various rice bran kinds was assessed, including dry matter content, ash content, crude fiber, crude protein, crude fat, gross energy, and mineral content (22).

After calibrating the apparatus with the appropriate standard solutions in accordance with the official procedure established by the Association, moisture content was determined using method 934.01, 240 Table No.1 and Table No. 2 15,16,20

Preliminary phytochemical screening:

Plants are biosynthesized in laboratories to produce different metabolites that have physiological and therapeutic effects in addition to primary metabolites like carbohydrates, proteins, and lipids that are used as food by humans. The mother extract obtained through the process extraction was then tested for the presence of several qualitative mineral components, including Potassium, Phosphorous, Magnesium, Calcium, Iron, Manganese, and Sodium, among others. on Table no.3 21.

Analysis of Polyphenol Compounds:

The samples were extracted using the Arribas et al. (25) technique using ethanol/water (95:5, v/v) as the solvent. To obtain a residue, the ethanolic portion of the extracts was evaporated using a rotary evaporator at reduced pressure and 40°C. The Mighri et al. method (30) was then uscd to examine the phenolic chemicals. An Agilent 1260 Infinity II series chromatography system and an Agilent 6130 electrospray ionization quadrupole mass spectrometer were used for the analytical liquid chromatography. An Ultra C18 column (5m 4.6mm 250mm; Restek, Bellefonte) was used for separation. A and B (0.2% acetic acid in 95% water and 5% MeOH, respectively) made up the mobile phase. 50% acetonitrile and 50% water) using a linear elution gradient: 0-45 min, 10-20% B; 45-85 min, 20-55% B; 85-97 min, 55-100% B; 97-110 min, 100% B; the initial conditions were maintained

for 10 min as a re-equilibration step. The column temperature was kept at 40°C, the mobile phase flow rate was 0.5 mL/min, and the injection volume was 20 L. Table No:4.

3. Results and Discussion

Physicochemical parameter:

Utilized to calculate the herbs' moisture content in accordance with standard. A measure of a herb's quality and purity is its ash value. Some people use ash values as a guide to estimate the moisture content of herbs in accordance with standards. A measure of a herb's quality and purity is its ash value. A natural drug's foreign matter content and the degree of care taken in its collection and preparation for market are both indicated by its ash values. Ashing is used to get rid of any organic material that can interfere with an analysis of the inorganic components. The amount of acid-insoluble ash in a sample can be used to evaluate the amount of adhering sand, dirt, and calcium oxalate variation. Typically, silicates, silica, carbonates, and phosphates make up the majority of the ash value. an acid Silica-based insoluble ash is a sign of contamination with earthy material. To calculate the amount of inorganic components, present in medications, water soluble ash is utilized. The extractive values are helpful in identifying the chemical components of a crude medication and in estimating which components are likely to be soluble in a given solvent. The appearance of food, One of the most important factors influencing consumer choice is plant matrices. The early report by Wisetkomolmat et al. (32) employed multivariate PCA to assess the variables of the bioactive components in indigenous Thai plants in order to discover the link between biological parameters. This approach, which has been utilized in numerous domains of biology and chemistry, is the most basic methodology in chemometrics. It can reduce the data dimension and visually cluster variants using principal components.

Table 1. Organoleptic evaluation of *Oryza Sativa*

S.No	Parameters:	Practical Yield:	Standard:
1.	Colour	White	Yellow /White
2.	Odour	Odourless	Odourless/Characterless
3.	Assay	98%	NLT- 98.0%
4.	Melting Point	88C	NLT- 120C
5.	Loss of drying	0.01%	NMT-0.5%
6.	Residue on Ignition	0.02%	NMT- 0.1%
7.	Residual Solvents	Methanol, Hexane	Methanol, Hexane

The germ, pericarp, aleurone, and sub-aleurone layers make up the majority of the rice bran, a by-product of the dry milling of rice kernels. (Some storage polysaccharides and hemicelluloses) in rice (16).

Crude fiber content has an impact on rice digestibility, with a high crude fiber content in rice reducing its digestion (35). Ash concentration is crucial in revealing the mineral components of a food sample. The experimental results for the amount of ash and protein were consistent with those of Amagliani et al. (16), who reported that these substances were more prevalent in the bran layer than the other layers.

Table 2. Physico-chemical parameters of *Oryza Sativa*

S.No	Parameters:	Practical Yield:
1.	Dry matter	85.17 ±0.09
2.	Ash Value	8.64 ± 0.10
3.	Crude fiber	5.18±0.01
4.	Crude Protein	14.41± 0.03
5.	Crude fat	13.85± 0.48
6.	Gross energy, cal/g	4243.20 ± 4.13

Proximate Analysis and Mineral Composition:

Calcium was the mineral with the highest abundance in the rice bran (225.53 – 10.57 mg/kg), followed by manganese (47.35 – 8.34 mg/kg), iron (35.01-1.15 mg/kg) zinc(32.96-1.96 mg/kg) and sodium (20.41 - 0.00 mg/kg) . Potassium, copper, phosphorus, and magnesium were minerals detected in the minor contents (2.32-0.04 mg/kg , 3.57-0.45 mg/kg, 1.14-0.06 mg/kg, and 0.36-0.03 mg/kg, respectively). It is a fantastic natural source of protein (14.41± 0.03%), fat (13.85± 0.48%), crude fiber (5.18 ± 0.01%), carbs, and other bioactive substances, all of which are proven to have positive effects on health (13). It has been demonstrated that the mineral content of various rice types varies (16, 31). Both macro and microminerals, such as Na, K, Mg, Ca, Zn, Fe, Cu, and Mn, are acknowledged for their significance and importance in biological processes.

Table 3. Physico-chemical parameters of *Oryza Sativa* (Mineral content)

S.No	Mineral:	Abbreviation:	Practical Yield:
1.	Potassium	K (g/100g)	2.32 ± 0.04
2.	Phosphorous	P (g/100g)	1.14 ± 0.06
3.	Magnesium	Mg(g/100g)	0.36 ± 0.03
4.	Calcium	Ca (mg/kg)	225.53 ±10.57
5.	Iron	Fe (mg/kg)	35.01 ±1.15
6.	Manganese	Mn(mg/kg)	47.35 ± 8.34
7.	Znic	Zn (mg/kg)	32.96 ±1.96
8.	Sodium	Na (mg/kg)	20.41 ±0.00
9.	Copper	Cu (mg/kg)	3.57±0.45

Analysis of Phenolic Substances:

The quantity of polyphenolic chemicals found in various types of Table 4 lists the results of the rice bran samples. By comparing their mass spectra and retention times, 17 compounds were found. The rice bran samples' elevated concentrations of some chemicals included Quercetin (0.25-0.65 mg/100 g sample), phytic acid (0.03-0.60 mg/100 g sample) chlorogenic acid, p-coumaric acid, ferulic acid, epigallocatechin gallate, and Additionally, o-coumaric acid and naringin were found in rice samples of bran. Phenolic compounds are secondary metabolites that are present in Plants that can remove free radicals and lower oxidative stress shielding biological macromolecules against potential stress risk (44). The properties of phenolic compounds as antioxidants are responsibility for chronic illness prevention, including Atherosclerosis, cancer, diabetes, obesity, and cardiovascular illnesses (12, 44). The phenolic compounds' makeup was little different between samples of rice bran, colored, and non-colored. Phenolics are present in rice bran in three distinct forms: insoluble bound, soluble free, and soluble conjugated. The bound phenolics have a greatly increased antioxidant capability. greater than that of soluble or unconjugated free forms with the main phenolic component found is ferulic acid (43). In opposition we discovered that phytic acid was the most prevalent phenolic chemical, may be as a result of ferulic acid's rice's cell wall components after being esterified dietary fiber that is present as an insoluble binding component form (45).

Table:4 Phenolic contents in different varieties of rice bran (mg/100g sample).

S.No	Phenolic contents	Yield	Standard
1.	Gallic Acid	ND	0.12µg/ml
2.	Caffeic acid	0.08 ± 0.00	0.06µg/ml
3.	Catechin	ND	0. .30µg/ml
4.	Epicatechin	0.09 ±0.00	01µg/ml
5.	Epigallocatechin gallate	0.18 ± 0.02	6110µg/ml
6.	p-coumaric acid	0.250 ± 0.05	0.10 µg/ml
7.	o-coumaric acid	0.15 ± 0.08	0.96 µg/ml
8.	Naringin	0.15 ± 0.01	1.06 µg/ml
9.	Rosmarinic acid	0.03 ± 0.05	0. .88µg/ml,
10.	Quercetin	0.65 ± 0.25	958.14 mg
11.	Naringenin	ND	0. .88µg/ml,

12.	Rutin	0.06 ± 0.03	1.07 µg/ml,
13.	Phytic acid	0.60 ± 0.03	14.68 µg/ml,
14.	Ferulic acid	0.15 ± 0.02	5.75 µg/ml
15.	Chlorogenic acid	0.42 ± 0.05	2.81 µg/ml
16.	Kaempferol	ND	0.92 µg/ml,
17.	Hydroxybenzoic acid	0.29 ± 0.02	0.95 µg/ml.

Where: ND meaning not detected.

4. Conclusion

As people's understanding of nutrition and health has grown, focus has shifted to developing functional food products with bioactive ingredients. The goal of this study was to describe the nutritional profile and bioactive component profile of rice brans from Chhattisgarh with unique characteristics. In addition to several nutrients including fiber, vitamins, and minerals, rice bran also contains bioactive phytochemicals that are good for your health, like g-oryzanol, and phenolic compounds. uncolored rice bran has a distinct composition of bioactive ingredients.

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