



Phytochemical and Nutrient Composition of Rice Husks

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ABSTRACT

This study deals with the determination of possible phytochemicals, minerals, vitamins and nutritional constituents present in Rice husk (RH). Dried rice husk samples were collected from a local rice mill at Awgu Town in Enugu, South-East Nigeria. The proximate, phytochemical, vitamin and mineral composition of the ground sample were investigated using standard methods of analysis, while a Gas chromatography-mass spectrometry (GC-MS) device was employed for the characterization of bioactive components of the sample. The proximate analysis revealed the presence of carbohydrate (37.04%), crude protein (1.85%), moisture (7.93%), crude fat (3.76%), fibre (25.74%), and ash content (23.39%). The phytochemical screening high amount of flavonoids, carotenoid, and glycosides in the RH sample. Vitamins such as A, C, D, E, K and B-complexes (B₁, B₂, B₃, B₅, B₆, B₇, B₉, and B₁₂) as well as minerals (calcium, magnesium, potassium, sodium, zinc, iron, selenium, cobalt, manganese, copper and phosphorus) were also identified in the sample. Meanwhile, the GC-MS analysis of the sample revealed the presence of cynarin, demecolcine, penta-triacontene, hyoscyamine, kheltin, acetyldigoxin, quinoline-3-methyl, coumaric acid, copaene, benzyl benzoate, caryophyllene, danthron, lupenon, beta-amyrin, ephedrine, alpha-ergosterol, galanthamine, kawain, agrimophol, eugenol, anabesine, benzyloquinoline, carpaine, phytol, phytic acid, myrcene, lobeline, beta-caryophyllene, sparteine, and emetine. This study established that the high nutritional value and bioactive composition of rice husk indicates the rice milling by-product to be a promising food formulate with numerous health benefits.

Keywords: GC-MS, Minerals, Phytochemicals, Rice husk and Vitamins

Introduction

Rice husk (RH) is a substantial by-product of rice milling generated globally in high quantity. Its utilization as prospective raw material plays significant role in waste management in milling industries. According to Singh,¹ RHs are the hard-outer part of rice grains that are removed from the grains during milling process. Chen *et al.*² reported that about 150 million tons of rice husks are generated globally per annum. Sani *et al.*³ stated that rice is a major cash crop cultivated for commercial purposes and its industries contributes significantly in employment of workforce. Omolehin *et al.*⁴ reported that rice is an essential food consumed by majority of Nigeria population. According to Soltani *et al.*⁵ RH is a natural fiber applied in composites. These natural fiber composites play significant roles in reducing dependence on non-renewable energy/material sources, reducing pollutant emissions and greenhouse gas discharges, and improving energy recovery. According to Varzakas *et al.*⁶ the inedible parts of plants such as the peel and husk rich in polyphenolics has been applied as food supplements. Currently, approximately two-third of global populace depends on plant-based treatments for health benefits.⁷

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Thus, utilization of phyto-nutrients from agro-wastes should be advocated as some by-products especially from milling industries might possess numerous and significant bioactive substances which can serve as intrinsic precursor for drug production. RH and bran are the major by-products of rice milling and accounts for two-third of the total rice milling products. Parichart *et al.*⁸ posited that extract of rice husk extract generated in some parts of the globe has shown anti-inflammatory, antidiabetic, antihyperlipidemic, hepatoprotective, anticancer, and antioxidant effects. It is worthy to note that limited benefits of RH from Southeast Nigeria has been documented, thus entailed this study. These benefits if identified will be attributed to the bioactive components of the plant's by-product. In most scientific report, the plant's secondary metabolites predict its therapeutic proclivity, though the development of plants extracts with potential medical benefits is hindered by poor biopharmaceutical processes, insufficient scientific data and resources. According to Hatfield & Marita,⁹ plant's intrinsic bioactive compounds are mainly attached to cell walls via polysaccharide, lignin, and perhaps silicon, and may be extracted under mild conditions using various extraction methods and solvents notwithstanding the available concentrations. The most preferred technique for evaluating the bioactive composition of long chain hydrocarbons, alcohols, acids, esters, alkaloids, steroids, amino and nitro chemicals in medicinal plants is via gas chromatography-mass spectroscopy (GC-MS). However, combination of GC and MS with specific identification procedures have advanced into a refined method for analysing plant constituents¹⁰. The proximate, vitamin, mineral and GC-MS analyses of RH investigated in this study is part of the search for plants by-products with bioactive compounds as to establish their possible nutritional and medicinal potentials.

Materials and Methods

Collection of plant materials

Dried rice husks sample was collected from a local rice mill at Awgu town, Enugu State, South-East, Nigeria, in December 2021.

Preparation of extract

The husks were ground and samples filtered through a mesh sieve size of 48. Extraction was done with appropriate solvents for the specific analysis as stated by the AOAC¹¹ method of analysis.

Methodology

Dried RH was assayed for proximate to evaluate the carbohydrate (CHO), moisture, ash, fibre, crude protein and lipid content using Association of Official Analytical Chemists.¹¹ Moisture content was evaluated by measuring the change in the weight of 5g portion of the rice husk after it was dried in an electric oven set at 105°C for 6 hours. Ash content, the inorganic residue that remains after the organic matter has been dried was quantified by incinerating 5g of the sample in a muffle furnace at 500°C for about 240 minutes. Crude protein was evaluated using Kjeldahl procedure.¹² The procedure involves degradation of sample with conc. H₂SO₄ in the presence of a metallic catalyst with further reduction of nitrogen to ammonia (NH₃) which is retained in the solution as ammonium sulfate [(NH₄)₂SO₄], followed by distillation and titration. Crude fibre was obtained using acid and alkaline digestion method.¹³ Lipid value was evaluated by continually extracting the sample with non-polar organic solvent using a Soxhlet extractor. CHO content was quantified by the difference method by subtracting the total sum of moisture, ash, crude protein, crude fat and crude fibre content from 100. The energy level was calculated by totalling the product of the mean values of crude protein, crude fat and total carbohydrate and the factors 4, 9, and 4 respectively, and result expressed in kilocalories per 100g sample.¹⁴

The mineral component of rice husk such as calcium, iron, zinc, selenium, copper, magnesium, manganese, cobalt, potassium, sodium and phosphorus were obtained with an atomic absorption spectrophotometry (AAS) (Solar Thermos Elemental Flame AAS; Model: S4 = 71096) following the procedure of Długaszek and Kaszczuk.¹⁵

The sample's bioactive components was obtained with the procedure of Balamurugan *et al.*,¹⁶ using a GC-MS device (Agilent 6890N Gas Chromatograph with Agilent 5975 Mass Selective Detector, and a 30 µm x 0.25 mm ID fused-silica capillary column chemically attached with SE-54 (DB-5), and 1-µm film thickness). The unknown components in the mass spectrum of the GC-MS were identified using a database from the National Institute of Standards and Technology (NIST) library.

For the quantitative phytochemical analysis, the standard techniques of Harborne¹⁷ was adopted for alkaloid estimation; Obadoni and Ochuko method,¹⁸ was used for saponin estimation; Awah *et al.* method,¹⁹ was adopted for tannin estimation; Kumaran and Karunakaran procedure,²⁰ with slight modification by Awah *et al.*,¹⁹ was used for total phenol content and total flavonoid content; while Munro and Bassir guideline,²¹ was adopted for the estimation of oxalate, glycosides, phytate, and cyanogenic glycosides.

The vitamins composition was quantified using UV-Visible spectrophotometer at 512nm for various vitamins, based on their standard calibration curves as stated by AOAC.¹¹

Ethical approval

The research was duly approved by the University of Port Harcourt Research Ethics Committee with Approval No: UPH/CEREMAD/REC/MM79/026.

Statistical analysis

Recorded data were analysed with T-Test of SPSS version 25.0, and results expressed as Mean ± SD of three replicates of analysis.

Results and Discussion

According to Dey *et al.*,²² RH is an agro-waste of rice milling with an appreciable fibre content. RH is believed to contain many bioactive compounds like phytochemicals, proximate nutrients, vitamins and minerals. The current study revealed the presence of numerous bioactive components of RH. The phytochemical screening revealed the presence of important phytochemicals such as alkaloid, flavonoid, steroid, carotenoid, tannin, and terpenoid. Table 1 showed some phytochemicals such as alkaloid (13.07 mg/100g), phytate (8.93 mg/100g), glycoside (7.92 mg/100g), cardiac glycoside (6.57 mg/100g), flavonoid (5.64 mg/100g), oxalate (0.63 mg/100g), carotenoid (0.05 mg/100g), cyanogenic glycoside (0.05 mg/100g) and steroid (0.03 mg/100g). As per Altemimi *et al.*,²³ phytochemicals such as phenols, flavonoids, carotenoid, and phenols have been previously reported to be responsible for the therapeutic potentials of some plant extracts. Thus, Amirkia & Heinrich,²⁴ posited that the plants' therapeutic effects are solely dependent on their secondary metabolites, though these therapeutic metabolites exist in few plant species. According to Varzakas *et al.*,⁶ inedible plant portions such as the peel and husk, have recently been adopted as food supplements. Butsat & Siriamornpun²⁶; Wanyo *et al.*,²⁵ reported that bioactive components from RH have shown resistance against oxidative damage. Studies have also reported biological benefits of phytate despite its anti-nutrient property.²⁷ Phytate intake has been reported to inhibit development of kidney stone through complex formation with calcium (Ca).²⁸ According to Lee *et al.*,²⁹ phytic acid inhibited free radicals and increased antioxidant potentials against alcohol-related liver impairment. Tannin containing plants have been used to treat intestinal disorders such as diarrhoea and dysentery.³⁰ Tannins has been reported to aid in managing inflamed/ulcerated tissues.³¹ Flavonoids are antioxidants and free radical scavengers that shield cells from oxidative damage. According to Okwu & Iroabuchi,³² flavonoids possess strong anticancer activity and protect cells from all stages of carcinogenesis. Thus, this study is in tandem with that of Irakli *et al.*,³³ which recorded significant polyphenol and flavonoids content. RH was also screened for its biological components using a GC-MS. The analysis revealed cynarin, demecolcine, penta-triacontene, hyoscyamine, khelthin, acetyldigoxin, quinoline-3-methyl, coumaric acid, copaene, benzyl benzoate, caryophyllene, danthron, lupenon, beta-amyrin, ephedrine, alpha-ergosterol, galanthamine, kawain, agrimophol, eugenol, anabasine, benzylisoquinoline, carpaine, phytic acid, myrcene, lobeline, beta-caryophyllene, sparteine, and emetine as presented in Table 2.

Table 1: Quantitative phytochemical constituents of rice husk

Phytochemical Constituent	Relative amount (mg/100g)
Alkaloid	13.07 ± 0.11
Flavonoid	5.64 ± 0.02
Saponin	0.29 ± 0.02
Tannin	1.73 ± 0.01
Oxalate	0.63 ± 0.01
Phytate	8.93 ± 0.10
Glycoside	7.92 ± 0.07
Cyanogenic glycoside	0.05 ± 0.00
Cardiac glycoside	6.57 ± 0.05
Steroid	0.03 ± 0.00
Phenol	2.79 ± 0.02
Anthraquinone	4.83 ± 0.03
Terpenoid	1.83 ± 0.01
Carotenoid	0.05 ± 0.00

Values are means ± standard deviations of triplicate determinations.

The GC-MS spectrum confirmed the presence of various components with different retention times as illustrated in Figure 1. The mass spectrometer examines the molecules eluted at various periods to estimate their type. The big compound splits into little compounds, causing peaks with varying mass-to-charge (m/z) ratios to appear. These mass spectra are the compound's fingerprint, which are discovered using the data library.

The proximate screening of RH revealed substantial amount of carbohydrate (37.04 g/100g), fibre (25.74 g/100g), and ash (23.39 g/100g); while moisture (7.93 g/100g), lipid (3.76 g/100g), and protein (1.85 g/100g) were in small amount with an energy value of 189.4 Kcal

(Table 3). High ash content was recorded in this study. Reports by Pivie and Butler,³⁴; Onyeike and Nwideezia,³⁵ had suggested that human food sources should contain about 3.0% ash. RH recorded over 3.0% ash and therefore should be considered for human consumption. The low crude protein recorded indicates that RH cannot contribute significantly to the day-to-day protein requirement of 23 to 56 g for humans as recommended by the National Research Council.³⁶ Furthermore, the low crude lipid indicates that the sample may be recommended to people suffering from hyperlipidemia. This research also established that RH has higher fibre content than wheat husk. According to Heshu *et al*,³⁷ wheat husk has a fibre content of 9.97 g/100g.

Table 2: Characterization of phytochemical constituents of rice husk with using GC-MS

Name of compound/ Peak Name	Molecular Formula	M.W (g/mol)	R/T (min)	Conc. (mg/100g)	Reported Biological Activity
Cynarin	C ₂₅ H ₂₄ O ₁₂	516.45	8.67	7.73	Antioxidant and Antichoke activity ⁵¹
Demecolcine	C ₁₂ H ₂₀ N ₂ O ₃	371.43	9.19	9.42	Anti-inflammatory activity ⁵²
Penta-triacontene	C ₃₅ H ₇₀	236.00	9.85	1.98	Antibacterial activity ⁵³
Hyoscyamine	C ₁₇ H ₂₃ NO ₃	289.38	10.44	12.13	Anticholinergic agent and Antioxidant activity ⁵⁴
Kheltin	C ₁₄ H ₁₂ O ₅	260.24	10.61	5.57	Anti-inflammatory, Antimicrobial, Antipyretic and cholesterol effects ⁵⁵
Acetyldigoxin	C ₄₃ H ₆₆ O ₁₅	300.20	12.95	3.16	Positive inotropic effect on cardiac muscle ⁵⁶
Quinoline, 3-methyl	C ₁₀ H ₉ N	143.00	13.53	1.74	Antimicrobial, Antibacterial and Cytotoxic activity ⁵⁷
Coumaric acid	C ₉ H ₈ O ₃	164.00	14.72	8.40	Antioxidant and Anti-inflammatory activities ⁵⁸
Copaene	C ₁₅ H ₂₄	204.36	15.70	3.49	Anti-inflammatory, Antioxidant, Antimicrobial activities and Antinociceptive effect ⁵⁶
Benzyl Benzoate	C ₁₄ H ₁₂ O ₂	212.25	16.21	1.53	Anti-inflammatory, Antimicrobial activity and Antinociceptive effect ⁵⁹
Caryophyllene	C ₁₅ H ₂₄	204.30	16.78	9.18	Anti-inflammatory, Anticancer, Antioxidant, Antimicrobial and Analgesic ⁶⁰
Danthron	C ₁₄ H ₈ O ₄	240.21	18.36	3.72	Antidiabetic, suppress autophagy and pancreatic cancer ⁶¹
Lupenon	C ₃₀ H ₄₈ O	424.71	18.76	15.18	Antimicrobial, Antibacterial and Antifungal ⁶²
Beta-Amyrin	C ₃₀ H ₅₀ O	410.00	19.43	11.13	Antihyperglycemic and hypolipidemic effect ⁶³
Ephedrine	C ₁₀ H ₁₅ NO	165.24	19.69	9.08	Decongestant stimulant, bronchodilator, Ergogenic supplement ⁶⁴
Alpha-ergosterol	C ₂₈ H ₄₄ O	398.00	19.85	2.42	Antimicrobial, Antifungal activity ⁶⁵
Galanthamine	C ₁₇ H ₂₁ NO ₃	287.35	20.27	4.19	Competitive acetylcholinesterase inhibitor ⁶⁶
Kawain	C ₁₄ H ₁₄ O ₃	230.26	20.61	9.43	Anticancer and Anticonvulsive activity ⁶⁷

Agrimophol	$C_{26}H_{34}O_8$	478.70	21.25	7.18	Antihelmintic effect and Antitumor activity ⁶⁸
Eugenol	$C_{10}H_{12}O_2$	164.20	21.65	5.72	Anti-inflammatory and Antioxidant activities ⁶⁹
Anabasine	$C_{10}H_{14}N_2$	162.30	22.40	2.58	Anti-inflammatory activity ⁷⁰
Benzylisoquinoline	$C_{16}H_{13}N$	219.28	22.70	3.44	Antiprotozoal natural product ⁷¹
Carpaine	$C_{28}H_{50}N_2O_4$	478.70	22.95	1.75	Anti-thrombocytopenic, and Antioxidant activities ⁷²
Phytol	$C_{20}H_{40}O$	296.53	23.37	18.16	Antinociceptive, Antioxidant, Anti-inflammatory, Anti-cancer, Antimicrobial, and Cytotoxic activity ⁷³
Phytic acid	$C_6H_{18}O_{24}P_6$	660.04	23.64	24.28	Antioxidant and Antinutrient activity ^{74, 75}
Myrcene	$C_{10}H_{16}$	136.23	23.91	8.65	Sedative, Antidiabetic, Antioxidant, Anti-inflammatory, Antibacterial and Anticancer activity ⁷⁶
Lobeline	$C_{22}H_{27}NO_2$	337.46	24.53	2.70	Antimicrobial, Antidepressant, Anticancer and Antiepileptic activity ⁷⁷
Beta-caryophyllene	$C_{15}H_{24}$	204.36	24.86	6.69	Anti-inflammatory and Antinociception activity ^{78,79}
Sparteine	$C_{15}H_{26}N_2$	234.39	26.21	5.52	Anti-inflammatory activity, Antibacterial activity, Diuretic effect and Antiarrhythmic effect ⁸⁰
Emetine	$C_{29}H_{40}N_2O_4$	480.64	26.80	2.76	Antiviral, Anticancer, Antiparasitic and contraceptive activities ⁸¹

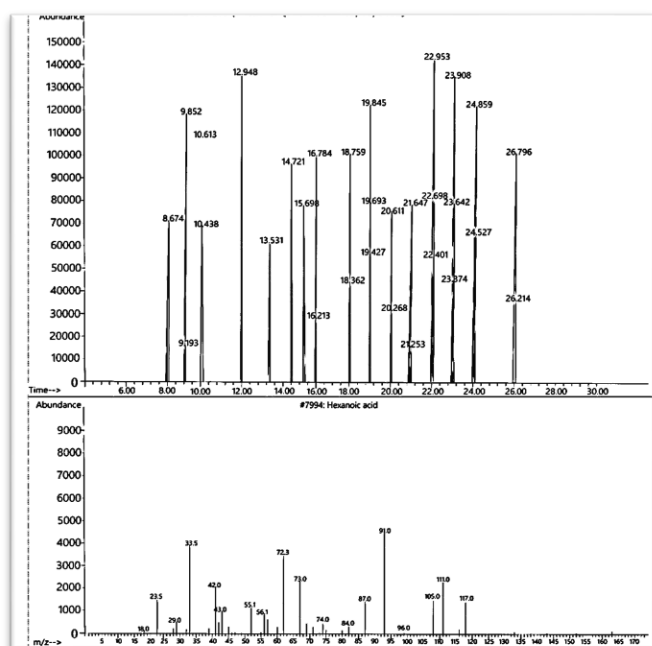


Figure 1: GC-MS chromatogram of rice husk methanolic extract

Meanwhile, review report by Elleuch *et al.*,³⁸ stated that wheat husk and corn husk have high dietary fibre content more than RH. Studies presented that dietary fibre may affect cation exchange properties which enable them to bind easily with calcium (Ca), zinc (Zn), iron (Fe) and other cations,³⁹ and it can also affect trace mineral absorption, gastric filling and emptying including gastrointestinal transition time.⁴⁰ As per the Institute of Medicine (IOM), 14 grams of total dietary fibre per 1000 calories is recommended to minimize the risk of developing chronic illnesses.⁴¹ Dietary fibre has been identified to be deficient in America diet by the USDA Dietary Guidelines Advisory Committee.⁴² Consuming foods enriched with RH fibre can therefore contribute to increase fibre consumption. Thus, food and/or by-products rich in dietary fibre should be encouraged in food processing industries or company as they contains various bioactive chemicals and dietary fibres. The high CHO content of RH is linked to the low crude lipid and crude protein concentrations. CHO has been reported to contribute greatly to the calorific value than crude protein and crude fat.³⁵ The low moisture content may be due to loss of water during storage and sun drying before milling. This signifies that RH could be stored for a very long time without spoilage.

Vitamins and minerals are vital nutrition components required in adequate proportions for healthy living. They serve as critical cofactors and coenzymes for various physiological and metabolic functions, including regulation of some important reactions required for energy generation metabolic functions.⁴³ The vitamin composition showed that vitamin A (8.93 mg/100g), vitamin C (2.40 mg/100g) and vitamin E (1.29 mg/100g) are present in significant amount; while other vitamins such as B7 (0.02 mg/100g), B5 (0.03 mg/100g), B9 (0.03 mg/100g), B3

(0.05 mg/100g), B2 (0.12 mg/100g), D (0.16 mg/100g), B1 (0.28 mg/100g), B12 (0.35 mg/100g), K (0.41 mg/100g) and B6 (0.49 mg/100g) existed in trace amount as shown in Table 4. The significant amount of ascorbic acid reveals that the sample possess antioxidant potentials. Vitamin C also aids in the transport and uptake of non-haeme iron through the mucosa, including decrease in folic acid intermediates and cortisol formation.⁴⁴

Table 3: Proximate composition of rice husk

Parameter	Concentration (g/100g)
Ash	23.39 ± 0.19
Carbohydrate	37.04 ± 0.82
Crude Fibre	25.74 ± 0.42
Crude Lipid	3.76 ± 0.42
Moisture	7.93 ± 0.02
Crude Protein	1.85 ± 0.10
Energy value (kcal/100g sample)	189.40

Values are means ± standard deviations of triplicate determinations

Table 4: Vitamins composition of rice husk

Parameter	Amount (mg/100g)
Vitamin A	8.93 ± 0.19
Vitamin B1	0.28 ± 0.01
Vitamin B2	0.12 ± 0.02
Vitamin B3	0.05 ± 0.01
Vitamin B5	0.03 ± 0.01
Vitamin B6	0.49 ± 0.04
Vitamin B7	0.02 ± 0.00
Vitamin B9	0.03 ± 0.00
Vitamin B12	0.35 ± 0.01
Vitamin C	2.40 ± 0.08
Vitamin D	0.16 ± 0.03
Vitamin E	1.29 ± 0.07
Vitamin K	0.41 ± 0.01

Values are means ± standard deviations of triplicate determinations.

Table 5: Mineral composition of rice husk

Mineral	Concentration of rice husk (mg/kg)
Calcium	29.49 ± 1.77
Iron	14.01 ± 0.17
Zinc	6.13 ± 0.12
Selenium	1.25 ± 0.01
Copper	9.85 ± 0.08
Magnesium	19.32 ± 0.19
Manganese	11.54 ± 0.13
Cobalt	1.85 ± 0.02
Potassium	48.28 ± 0.46
Sodium	2.81 ± 0.03
Phosphorus	4.02 ± 0.02

Values are means ± standard deviations of triplicate determinations.

Result of the mineral composition showed that RH contained appreciable amounts of minerals as shown in Table 5. Minerals, according to Iniahe *et al.*,⁴⁵ are daily essential nutrients required for optimal tissue function. From the analysis, RH contains good amount of potassium (48.28 mg/kg), calcium (29.49 mg/kg), magnesium (19.32 mg/kg), iron (14.01 mg/kg), and manganese (11.54 mg/kg), while other minerals such as copper (9.85 mg/kg), zinc (6.13 mg/kg), phosphorus (4.02 mg/kg), sodium (2.81 mg/kg), cobalt (1.85 mg/kg), and selenium (1.25 mg/kg) existed in lower concentrations. Minerals perform various important functions in humans. Ca is required for normal growth, muscle activity, and skeletal development; Cu and Fe are necessary for cellular activities and oxygen transport; chemical reactions in the body and intestinal absorption are enhanced by the presence of Mg; while effective fluid balance and nerve transmission requires Na and K.⁴⁶ Abbaspour *et al.*⁴⁷ also reported that Fe is required for the formation of haem-enzymes and other iron-containing enzymes involved in electron transfer and redox reactions, including the synthesis of oxygen-carrying proteins such as haemoglobin and myoglobin. Mg is a known activator in enzyme systems that maintain electrical potential in nerves, whereas Na and K influence osmotic pressure and contribute to normal pH balance.⁴⁸ According to Muhammad *et al.*,⁴⁹ Mg is important in energy production and immune system support. Ca, along with P, is an important component of body fluid that enhances bone formation.⁴⁶ In fact, the roles of minerals in human diet cannot be over emphasized as they serve multiple purposes, including provision of building elements for our bones, influencing muscle and neuron function, and maintaining body water balance.⁵⁰ Thus, nutrient-dense foods are essential for both adults and children, as deficiencies can cause human dysfunction and diseases. With the significant presence of these important vitamins and minerals, it is obvious that RH can be adopted to combat mineral and vitamin deficiencies.

Conclusion

This study revealed that rice husks is rich in various essential compounds. RH has shown to contain low amount of protein and thus, not a good source of protein. RH also contains significant amount of fibre which can supplement fibre intake since no adverse effect has been recorded in the use RH as animal feeds. This study has revealed that rice husk has high nutritional value which indicates the plant to be a promising food formulate with numerous health benefits.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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