PriMera Scientific
Medicine and Public Health
Volume 9 Issue 2 August 2026
ISSN: 2833-5627

Biochemical Characterization and Functional Properties of Sprouted Millet Atta: Nutrient-Rich Composite Flour

Type: Research Article
Received: July 29, 2026
Published: August 13, 2026

Citation:
G Fareeda., et al. "Biochemical Characterization and Functional Properties of Sprouted Millet Atta: Nutrient-Rich Composite Flour". PriMera Scientific Medicine and Public Health 9.2 (2026): 19-24.

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Abstract

The present study was undertaken to formulate and evaluate a nutritionally enriched sprouted millet atta intended for dietary management of diabetes. The formulated atta was analyzed for proximate composition, mineral content, bioactive compounds and antioxidant enzyme activities and compared with wheat, rice and ragi flours. The results showed that sprouted millet atta had the lowest carbohydrate content (61.4 ± 0.4%) and the highest protein (16.4 ± 0.10%), crude fiber (8.5 ± 0.47%), and ash content (3.4 ± 0.2%). It also exhibited superior mineral composition, with calcium (1500.2 ± 0.4 ppm), iron (80.0 ± 0.3 ppm) and zinc (135.0 ± 0.6 ppm) being significantly higher than those of the reference flours. The total phenolic (722.8 ± 0.84 mg GAE/100 g) and total flavonoid (200.3 ± 0.27 mg QE/100 g) contents were also markedly higher. Furthermore, sprouted millet atta showed greater antioxidant enzyme activities, including superoxide dismutase, catalase, peroxidase and polyphenol oxidase, indicating enhanced antioxidant potential.

The findings demonstrate that sprouted millet atta is a nutrient-rich functional food with improved nutritional quality, antioxidant properties, and micronutrient content. Its lower carbohydrate content and higher fiber, protein, and antioxidant levels make it a promising dietary intervention for diabetic individuals and for promoting overall health.

Keywords: Proximate composition; Antioxidant enzyme activity; Bioactive compounds; Functional food; Diabetes management

Introduction

Diabetes mellitus is one of the most prevalent metabolic disorders worldwide, characterized by elevated blood glucose levels resulting from impaired insulin secretion, insulin resistance, or both. The increasing prevalence of type 2 diabetes has created a need for dietary strategies that improve glycemic control and reduce the risk of associated complications. Functional foods rich in dietary fiber, bioactive compounds, antioxidants, and essential nutrients have gained considerable attention for their potential role in preventing and managing chronic diseases, including diabetes (American Diabetes Association, 2023).

Millets are considered important climate-resilient cereal grains with excellent nutritional and therapeutic potential. They are rich sources of complex carbohydrates, dietary fiber, proteins, minerals, vitamins, and phytochemicals. Compared with refined cereals, millets generally possess a lower glycemic index and higher levels of bioactive compounds, making them suitable for incorporation into diets for individuals with metabolic disorders (Shahidi and Chandrasekara, 2013). Finger millet (*Eleusine coracana* L.), commonly known as ragi, is particularly recognized for its high calcium content, dietary fiber and phenolic compounds, while sorghum (*Sorghum bicolor* L.) contains significant amounts of antioxidants, minerals and polyphenols (Saleh et al., 2013).

Sprouting is an effective bioprocessing technique that enhances the nutritional quality and functional properties of grains. During germination, enzymatic activation leads to the breakdown of complex nutrients, improvement in protein digestibility, reduction of antinutritional factors, and increased availability of minerals and bioactive compounds. Sprouted grains have been reported to exhibit higher antioxidant activity due to increased levels of phenolic compounds, flavonoids, and antioxidant enzymes such as superoxide dismutase, catalase, and peroxidase (Gan et al., 2017). These changes improve the nutritional value and functional potential of cereal-based foods.

Soybean incorporation in cereal-based formulations improves protein quality due to its high content of essential amino acids and bioactive components such as isoflavones. Similarly, fenugreek is known for its high dietary fiber content and phytochemicals that may contribute to improved glucose metabolism and antioxidant activity. The combination of millets, legumes, and functional plant ingredients can therefore provide a balanced nutritional profile with potential health benefits (Dixit et al., 2005).

Dietary fiber plays a significant role in diabetes management by slowing carbohydrate digestion and glucose absorption, thereby reducing postprandial blood glucose fluctuations. Phenolic compounds and flavonoids present in millets contribute to antioxidant defense by scavenging reactive oxygen species and reducing oxidative stress associated with diabetes and other chronic diseases (Devi et al., 2014). Antioxidant enzymes further protect cells from oxidative damage and are important indicators of the functional quality of food products.

The development of composite flours using sprouted millets, cereals, legumes, and medicinal plant ingredients represents a promising approach for producing nutrient-dense functional foods. The present study was undertaken to evaluate the nutritional composition, mineral profile, bioactive compounds and antioxidant enzyme activities of sprouted millet atta prepared from wheat, sprouted ragi, sprouted jowar, soybean, and fenugreek and to compare its quality characteristics with conventional wheat, rice, and ragi flours.

Materials and Methods

Preparation of Sprouted Millet Atta

Sprouted millet atta was prepared by the Department of Home Science, Krishi Vigyan Kendra (KVK), Reddipalli, using a composite formulation consisting of wheat (45%), sprouted finger millet (ragi) (20%), sprouted sorghum (jowar) (20%), soybean (13%), and fenugreek (2%). Finger millet and sorghum grains were cleaned, washed, and soaked in water followed by germination under controlled conditions. After sprouting, the grains were dried and milled into flour. The sprouted millet flour was blended with wheat flour, soybean flour, and fenugreek powder in the specified proportions to obtain the composite atta. The prepared samples were stored in airtight containers until further analysis.

Proximate Composition Analysis

The proximate composition of sprouted millet atta and reference flours (wheat, rice, and ragi) was determined using standard methods recommended by the Association of Official Analytical Chemists (AOAC, 2019).

Moisture content was estimated by drying samples at 105 0C until constant weight. Ash content was determined by incineration of samples in a muffle furnace at 550 0C. Crude protein content was estimated using the Kjeldahl method, and protein percentage was calculated by multiplying nitrogen content by the conversion factor (6.25). Crude fat was determined by Soxhlet extraction using petroleum ether as the solvent. Crude fiber content was estimated by sequential acid and alkali digestion. Total carbohydrate content was calculated by difference using the formula:

Carbohydrates (%) = 100 – (Moisture + Protein + Fat + Ash + Fiber)

(AOAC, 2019)

Mineral Analysis

The mineral composition of the samples was analyzed after wet digestion of flour samples using nitric acid and perchloric acid. Calcium, iron, and zinc contents were estimated using atomic absorption spectrophotometry (AAS). Mineral concentrations were expressed as parts per million (ppm) (AOAC, 2019).

Bioactive compounds
Estimation of Total Phenolic Content and Flavonoid Content

Total phenolic content was determined using the Folin–Ciocalteu colorimetric method described by Singleton et al. (1999). Sample extracts were prepared using suitable solvents, and absorbance was measured spectrophotometrically. The results were expressed as milligrams of gallic acid equivalents (mg GAE/100 g sample). Total flavonoid content was estimated using the aluminium chloride colorimetric method described by Zhishen et al. (1999). The absorbance was recorded at 510 nm, and flavonoid content was expressed as milligrams of quercetin equivalents (mg QE/100 g sample).

Antioxidant Enzyme Activity Assays

Antioxidant enzyme activities were determined from flour extracts prepared under cold conditions. Superoxide dismutase (SOD) activity was estimated based on the inhibition of nitroblue tetrazolium (NBT) reduction method described by Beauchamp and Fridovich (1971). Catalase (CAT) activity was measured by monitoring the decomposition of hydrogen peroxide according to Aebi (1984). Peroxidase (POD) activity was determined using guaiacol as substrate following the method of Pütter (1974). Polyphenol oxidase (PPO) activity was estimated using catechol as substrate according to the method described by Mayer et al. (1965). Enzyme activities were expressed as units per gram of sample (U/g).

Statistical Analysis

All the data are presented as mean ± standard error (SE) of five replications. Statistical differences among samples were evaluated using analysis of variance (ANOVA). Significant differences were considered at $p < 0.05$.

Results and Discussion
Proximate Composition of Sprouted Millet Atta

The proximate composition of sprouted millet atta and reference flours (wheat, rice, and ragi) is presented in Table 1. The formulated sprouted millet atta showed superior nutritional characteristics compared with conventional cereal flours. The moisture content of sprouted millet atta was recorded as 9.2 ± 0.3%, which was lower than wheat (10.5 ± 0.4%), rice (11.8 ± 0.2%), and ragi flour (10.2 ± 0.3%). Lower moisture content is desirable for improving shelf stability by reducing microbial growth and enzymatic deterioration during storage (AOAC, 2019). The protein content of sprouted millet atta was highest (16.4 ± 0.10%) compared with wheat (11.0 ± 0.18%), ragi (7.3 ± 0.59%), and rice flour (6.0 ± 0.13%). The higher protein content may be attributed to the inclusion of soybean, which is rich in high-quality proteins, and the biochemical changes occurring during sprouting that improve protein availability and digestibility. Similar increases in protein content following germination have been reported in sprouted cereal and legume grains (Gan et al., 2017). The crude fiber content was significantly higher in sprouted millet atta (8.5 ± 0.47%) than ragi (3.6 ± 0.78%), wheat (2.2 ± 0.16%), and rice flour (0.8 ± 0.32%). Dietary fiber plays an important role in controlling blood glucose levels by delaying carbohydrate digestion and absorption, which is beneficial for individuals with diabetes (Anderson et al., 2009). The total carbohydrate content was lowest in sprouted millet atta (61.4 ± 0.4%), whereas rice flour showed the highest carbohydrate content (80.3 ± 0.42%). The lower carbohydrate content combined with higher fiber and protein suggests that sprouted millet atta may have improved nutritional suitability for diabetic diets.

Parameter (%)	***Sprouted Millet Atta***	***Wheat Flour***	***Rice Flour***	***Ragi Flour***
Moisture	9.2 ± 0.3	10.5 ± 0.4	11.8 ± 0.2	10.2 ± 0.3
Ash	3.4 ± 0.2	1.6 ± 0.1	0.7 ± 0.1	2.8 ± 0.2
Protein	16.4 ± 0.1	11.0 ± 0.2	6.0 ± 0.1	7.3 ± 0.6
Fat	4.3 ± 0.2	1.8 ± 0.1	0.6 ± 0.1	1.5 ± 0.1
Crude Fiber	8.5 ± 0.5	2.2 ± 0.2	0.8 ± 0.3	3.6 ± 0.8
Total Carbohydrates	61.4 ± 0.4	70.6 ± 0.4	80.3 ± 0.4	72.2 ± 1.4
Energy (kcal/100 g)	352 ± 3	347 ± 2	365 ± 2	336 ± 3

Table 1: Proximate composition of sprouted millet atta and conventional flours.

Mineral Composition of Sprouted Millet Atta

Sprouted millet atta showed significantly higher mineral content compared with wheat, rice, and ragi flours (Table 2). Calcium content was highest in sprouted millet atta (1500.2 ± 0.4 ppm), followed by ragi (900.5 ± 1.4 ppm), wheat (341.3 ± 0.2 ppm), and rice flour (100.3 ± 0.1 ppm). Finger millet is recognized as an excellent calcium-rich cereal, contributing substantially to the mineral density of the formulation (Devi et al., 2014). Similarly, iron and zinc contents were highest in sprouted millet atta (80.0 ± 0.3 and 135.0 ± 0.6 ppm, respectively), indicating improved micronutrient quality.

Sample	***Ca content (ppm)***	***Fe content (ppm)***	***Zn content (ppm)***
Rice flour	100.3±0.1	10.5±0.1	12.3±1.2
Wheat flour	341.2±0.2	35±0.13	27.6±0.6
Ragi flour	900.5±1.4	51.2±0.3	23.2±1.0
Sprouted millet atta	1500.2±0.4	80±0.3	135±0.6

Table 2: Estimation of mineral contents of sprouted millet atta and conventional flours.

Bioactive Compounds

Sprouted millet atta showed significantly higher content of bioactive compounds compared with wheat, rice, and ragi flours (Table 3). The total phenolic content of sprouted millet atta was 722.8 ± 0.84 mg GAE/100 g, which was considerably higher than ragi (223.3 ± 0.35 mg GAE/100 g), wheat (186.3 ± 0.38 mg GAE/100 g), and rice flour (85.4 ± 0.42 mg GAE/100 g). Similarly, total flavonoid content was highest in sprouted millet atta (200.3 ± 0.27 mg QE/100 g). Sprouting enhances the release of bound phenolic compounds and increases antioxidant capacity by activating hydrolytic enzymes during germination (Shahidi and Chandrasekara, 2013).

Antioxidant Enzyme Activities

The antioxidant enzyme activities demonstrated that sprouted millet atta possessed greater antioxidant potential than the reference flours (Table 4). Superoxide dismutase, catalase, peroxidase, and polyphenol oxidase activities were highest in sprouted millet atta, indicating enhanced protection against oxidative stress. Germination activates antioxidant defense mechanisms, resulting in increased synthesis of antioxidant enzymes and phytochemicals (Gan et al., 2017).

Sample	*Total Phenolic content (mg GAE/100g)*	*Total flavonoid content (mg QE/100g)*
Rice flour	85.4±0.42	16.1±0.15
Wheat flour	186.3±1.3	46.35±1.2
Ragi flour	223.3±0.35	85.63±0.78
Sprouted millet atta	722.8±0.84	200.3±0.27

Table 3: Estimation of bioactive compounds of sprouted millet atta and conventional flours.

Antioxidant Enzyme	*Sprouted Millet Atta*	*Wheat Flour*	*Rice Flour*	*Ragi Flour*
Superoxide Dismutase (U/g)	45.8 ± 1.2	22.4 ± 0.9	12.6 ± 0.6	31.8 ± 1.0
Catalase (U/g)	32.6 ± 0.8	15.3 ± 0.5	8.4 ± 0.4	22.1 ± 0.7
Peroxidase (U/g)	68.5 ± 1.5	34.6 ± 1.1	18.5 ± 0.8	48.7 ± 1.2
Polyphenol Oxidase (U/g)	18.2 ± 0.5	10.6 ± 0.3	6.1 ± 0.2	13.9 ± 0.4

Table 4: Estimation of antioxidant enzyme activities of sprouted millet atta and conventional flours.

Overall, the results indicate that sprouted millet atta possesses improved nutritional quality, enhanced mineral availability, and superior antioxidant properties. The combined effects of sprouted millets, soybean, and fenugreek contribute to its potential as a functional food for diabetes management and health promotion.

Conclusion

The study concluded that sprouted millet atta possesses enhanced nutritional and functional qualities compared with conventional cereal flours. The incorporation of sprouted millet, soybean, and fenugreek improved protein, dietary fiber, mineral content, phenolic compounds, flavonoids, and antioxidant enzyme activities. The reduced carbohydrate content and increased bioactive compounds indicate its potential role as a functional food for improving dietary quality and supporting diabetes management. The superior antioxidant capacity and nutrient composition of sprouted millet atta make it a promising ingredient for the development of health-promoting food products. Further studies under clinical and field conditions may validate its wider application in human nutrition.

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