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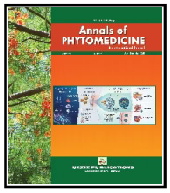

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From seedlings to greens: Unravelling the phytochemical and nutritional transformation in Mungbean (*Vigna radiata* (L.) Wilczek)

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Abstract

With the global population growing at an unprecedented rate, food security and hidden hunger have been the major challenges that require immediate attention. Sprouts and microgreens, being concentrated sources of phytochemicals and minerals, may help mitigate these issues. In our study, five different mungbean (*Vigna radiata* (L.) Wilczek) varieties were analysed at different developmental stages (sprouts and microgreens at 7 and 10 days after germination) for various parameters including: proximate analysis (moisture, ash, crude fibre, crude fat, total carbohydrate and protein content); antioxidant activity; phenolics; flavonoids; ascorbic acid content; minerals (iron and zinc) and antinutrients (phytic acid and tannin). Our results indicated that sprouts were nutritionally superior in terms of carbohydrate (6.05% to 7.23%), protein content (5.30% to 6.83%), antioxidant activity (60.26% to 89.57%), phenolic (3.26 mg/g to 5.10 mg/g) and flavonoid content (0.74 mg/g to 1.33 mg/g), whereas microgreens at 7 days after germination were higher in ascorbic acid (1.77 mg/g to 2.10 mg/g) and mineral content (iron and zinc: 0.61 mg/100g to 1.86 mg/100 g; 0.30 mg/100 g to 0.64 mg/100 g, respectively). The phytic acid and tannin content in our findings were lowest for microgreens at 7 days after germination and then increased in microgreens at 10 days after germination and sprouts. Germination reduces the antinutrients in sprouts and microgreens due to higher enzymatic activity, which enhances the bioavailability of nutrients.

1. Introduction

In recent years, there has been a remarkable shift in consumer preference towards fresh, organic and nutrient-dense food sources. This growing awareness has led to increased interest in sprouts and microgreens, which are recognized as functional foods. Their superior nutritional composition, rapid growth cycle and ease of cultivation year-round make them suitable as sustainable food sources. Sprouts represent the first stage of germination and are the fastest-growing foods, usually in dark, humid conditions with good ventilation. In contrast, microgreens are young, immature plants requiring proper light and growth media, harvested upon 7 to 21 days after germination (Wojdylo *et al.*, 2020). The edible portion of microgreens is only the stem and cotyledons, unlike sprouts. A wide range of species, such as cereals, vegetables, herbs, legumes and oil seeds can be utilized to produce sprouts or microgreens. Due to their outstanding nutritional properties, microgreens are also known as “superfoods” or “functional foods” (Galieni *et al.*, 2020).

The nutritional composition of sprouts and microgreens depends on several factors, such as the species, growing media, environmental conditions, harvest time, storage and processing (Ebert, 2022; Heena and Riar, 2025). Due to their unique color, texture and aroma, microgreens are utilised as innovative culinary ingredients in drinks, desserts, soups, sandwiches and salads. During seed germination, various physiological and biochemical transformations occur, such as enzyme activation, enhanced respiration and macromolecule breakdown (such as fats and polysaccharides). These changes lead to improved digestibility and mineral absorption due to reduced antinutrients such as trypsin inhibitors, phytic acid, pentosan, tannin and cyanides (Farooq *et al.,* 2022; Tuncel *et al.*, 2025). Numerous studies have reported the nutritional superiority of microgreens and sprouts relative to their mature plant counterparts, with higher concentrations of phytochemicals (such as polyphenols, carotenoids, flavonoids), minerals and vitamins. These bioactive compounds contribute to their potent antioxidant capacity and health benefits, including anti-inflammatory, anti-hyperglycemic, anti-carcinogenic, anti-obesogenic and anti-atherosclerotic properties (Galieni *et al.*, 2020; Zhang *et al.*, 2021).

Globally, billions of people suffer from micronutrient deficiencies and chronic malnutrition, contributing to the prevalence of diet-based disorders such as diabetes, cardiovascular disease, hypertension, stroke, cancer and obesity (Chaudhary *et al.*, 2022; WHO, 2021).To address these issues, nutrient-rich foods such as fruits and vegetables are essential; however, their availability is limited by seasonal constraints. In this context, microscale vegetables, particularly sprouts and microgreens, have become increasingly popular (Seth *et al.*, 2025). Given their short life cycle, minimal resource input (fertilizers and pesticides) and adaptability to limited space, microgreens are suitable for urban and controlled environment farming (Singh *et al.*, 2024). Moreover, the choice of growing media has a significant impact on their growth and nutritional quality, with systems including hydroponics, aeroponics, aquaponics and various soil and peat-based media being widely

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used (Dalal *et al.*, 2022). Consequently, microgreens have emerged as a promising resource-efficient strategy to combat global concerns of food security, hidden hunger and environmental sustainability. Given their nutritional richness and potential to enhance dietary diversity, the global microgreens market is estimated to rise from 7.5% (2021 to 2026) to 13.1% (2020 to 2028) (Lone and Pandey, 2024). Despite their concentrated nutrient profile and sensory appeal, microgreens present certain challenges. One of the major concerns with microgreens is food safety, as they are susceptible to microbial contamination due to high moisture content. Hence, the post-harvest packaging methods and storage conditions are important for maintaining their quality (Turner *et al.*, 2020).

Pulses occupy an important place in vegetarian diets across the globe, as they have exceptional nutritional value. Among the various pulses consumed globally, mungbean (*Vigna radiata* (L.) Wilczek) stands out as a rich, affordable plant-based protein source. It is also rich in minerals, vitamins, essential amino acids and dietary fibre that aid digestion (Singh *et al.*, 2025). However, mungbean seeds have antinutrients (phytic acid, tannins, saponins, trypsin inhibitors) that hinder nutrient absorption and digestibility. Mungbean microgreens offer a promising solution to mitigate antinutritional factors associated with mature mungbean due to high enzymatic activity during germination (Narale *et al.*, 2024). Therefore, the objective of the present study was to conduct a comparative analysis of biochemical parameters, including proximate analysis, antioxidant activity, phenolic and flavonoid concentrations, ascorbic acid content, mineral composition (iron and zinc), as well as antinutritional factors (phytic acid and tannins) in sprouts and microgreens, to elucidate their potential as natural, health promoting functional foods.

2. Materials and Methods

2.1 Authentication of plant material

The mungbean varieties used in the present study were procured from the Pulses Section, Chaudhary Charan Singh Haryana Agricultural University, Hisar. The varieties are officially notified by the Government of India under the respective Gazette Notification and S.O. Number.

S.No.	Variety	Gazette Notification No.	S.O. No.
1.	IPM 02-3	1341	2187 (E)
2.	MH 1-25	171	211 (E)
3.	MH 2-15	51	72 (E)
4.	MH 1762	1479	1560 (E)
5.	MH 1772	1479	1560 (E)

2.2 Plant material

Five mungbean varieties (IPM 02-3, MH 1-25, MH 2-15, MH 1762, MH 1772) were selected to cultivate sprouts and microgreens (Figure 1). Mungbean sprouts at 24 h and microgreens were sampled at two developmental stages: 7 and 10 days after germination (DAG). Sprouts were grown in petri plates by uniformly spreading the seeds and maintained in the dark under moist conditions to support germination. In contrast, microgreens were cultivated in trays (52 cm × 28 cm × 5 cm), watered daily and exposed to a photoperiod of 8 to 10 h to ensure proper growth. Both sprouts and microgreens were cultivated under controlled conditions at a temperature range of 30 to 40°C.

Figure 1: Microgreens and sprouts of mungbean (*Vigna radiata* (L.) Wilczek).

2.3 Proximate analysis

The moisture, ash, crude fibre, crude fat and crude protein content (calculation: N × 6.25) were determined following the AOAC methods (Association of Analytical Chemists) and total carbohydrate content was calculated by their difference (AOAC, 2019).

2.4 Sample extraction: Antioxidant activity

The samples (500 mg) were homogenized in 85% ethanol and centrifuged (12,000 rpm) for 15 min. The resulting pellet was then washed twice with fresh 85% ethanol.

2.4.1 Antioxidant activity

Antioxidant activity was evaluated using the DPPH free radical scavenging assay (Gunjal *et al.*, 2024). 500 µl of supernatant was mixed with 0.002% (w/v) of 2,2-diphenyl-1-picrylhydrazyl solution (freshly prepared using methanol as solvent) and incubated for 30 min in the dark at room temperature. The absorbance was then recorded at a wavelength of 515 nm using a spectrophotometer. The results were calculated as follows:

$$\text{DPPH free radical scavenging activity (\%)} = \frac{Ac - As}{Ac}$$

where,

Ac = Absorbance of control

As = Absorbance of sample

2.4.1.1 Phenolic content

The phenolic content was measured following the method adapted from Gunjal *et al.* (2024).The sample extract (1 ml) was mixed with 500 µl of freshly prepared Folin-Ciocalteu's reagent (0.1 N). After vortexing, 1 ml of saturated sodium bicarbonate was added to the tubes. The absorbance was recorded at a wavelength of 725 nm after 1 h incubation at room temperature. The phenolic content in samples was measured against gallic acid standard (mg GAE/g).

2.4.1.2 Flavonoid content

The flavonoid content was measured based on Gunjal *et al.* (2024) with slight modifications. A 300 µl aliquot was mixed with 0.15 ml of 5% sodium nitrite and 0.15 ml of 10% aluminium chloride. After 6 min, 4% sodium hydroxide was added and the absorbance was noted at a wavelength of 510 nm. The flavonoid content was measured using quercetin standard and expressed as mg QE/g.

2.5 Ascorbic acid content

Ascorbic acid content was estimated following the method of Singh *et al.* (2011) with some modifications. The sample extraction was done using metaphosphoric acid (5%), followed by centrifugation (5,000 rpm) for 15 min. An aliquot (500 µl) was mixed with 1 ml of phosphate buffer (150 mM, pH=7.4). The color was developed after the addition of different reagents, including 0.6 ml of 10% TCA (trichloroacetic acid), 0.6 ml of ortho-phosphoric acid (44%), 0.6 ml of 2, 2-bipyridine (4%) and 0.3 ml of 0.3% $FeCl_3$ (ferric chloride), followed by incubation (40°C) for 40 min. The absorbance was then recorded at a wavelength of 525 nm and ascorbic acid content was determined using a standard curve (mg/g).

2.6 Minerals

About one gram of dried samples was digested in nitric acid and perchloric acid (4:1) mixture using high performance microwave digestor (ETHOS UP). After digestion, the samples were cooled at room temperature and the final volume was made up to 50 ml using deionized water. The samples were filtered twice (using Whatman filter) before analysis by flame atomic absorption spectrophotometer (thermoscientific iCE FiOS). The respective standards solutions of iron and zinc were used for calibration and thereby determining the final concentration (mg/100 g) in samples (Fuente *et al.*, 2019).

2.7 Phytic acidcontent

Phytic acid content was determined following the method of Haug and Lantzsch (1983) with some modifications. The samples were homogenized in 0.2 N HCl and a 200 µl aliquot of the extract was reacted with 1 ml of ferric solution (0.02% ammonium ferrous sulphate in 2N HCl). The mixture was boiled for 30 min, then cooled at room temperature and mixed with 2 ml of bipyridine solution (prepared by dissolving 10 g of 2,2-bipyridine in 10 ml thioglycolic acid and making up the volume using deionized water). Absorbance was recorded immediately at a wavelength of 519 nm and phytic acid concentration (mg/g) was calculated using a standard curve.

2.8 Tannin content

The samples (500 mg) were homogenized in methanol and incubated at 30°C for 14 to 16 h. The homogenate was centrifuged (3000 rpm, 10 min) and 1 ml of supernatant was mixed with 3 ml of vanillin (4%) and 2 ml of concentrated HCl. After proper mixing, the tubes were kept at room temperaturefor about 20 min and absorbance was measured at a wavelength of 525 nm. Tannin content in the samples was quantified using catechin (mg/g) standard (Burns, 1971).

2.9 Statistical analysis

Statistical analysis was done using SPSS (version 23.0, USA) and PCA (Principal Component Analysis) was performed using Origin Pro 2024.

3. Results

3.1 Proximate analysis

Proximate analysis of five different varieties of mungbean sprouts and microgreens at the two growth stages (7 and 10 DAG) included: moisture, ash, crude fibre, crude fat, total carbohydrate and crude protein content, as shown in Tables 1, 2 and 3. The moisture content of sprouts varied between 83% to 87% with the maximum being for MH 1762 microgreens at 10 DAG (86.62%). Moisture content ranged between 81-85% and 83-87% in microgreens harvested at 7 and 10 DAG, respectively. The ash content in sprouts and microgreens ranged between 0.83% to 2.59%, being higher for microgreens at the 10 DAG (1.82% to 2.59%) and 7 DAG (1.45% to 2.49%), while lower for sprouts (0.83% to 1.70%), depicting the influence of species and developmental stage (Table 1). Significant differences in crude fibre and crude fat content were observed among the studied varieties, with microgreens (at the 10 DAG) depicting the highest fibre and fat content in the range of 3.15% to 4.58% and 0.24% to 0.36%, respectively (Table 2). For fat content, MH 1762 and MH 1-25 had the highest levels in microgreens at the 10 DAG (0.36%; 0.34%), followed by microgreens at the 7 DAG (0.34%; 0.33%) and sprouts (0.23%; 0.26%), respectively.

The carbohydrate content of mungbean sprouts (6.05% to 7.23%) was found to be higher in comparison to microgreens at the 7 (4.25% to 6.57%) and 10 (3.85% to 5.73%) DAG (as shown in Table 3). The carbohydrate content for sprouts varied in the order: MH 1-25>IPM 02-3 >MH 1772>MH 1762>MH 2-15. There was significant variation between the carbohydrate content of sprouts and microgreens at both growth stages (7 and 10 DAG). The highest crude protein content was found in MH 2-15 (6.83%) sprouts, which decreased in microgreens at the 7 DAG (3.62%) and 10 DAG (3.60%) (Table 3). Similarly, for other varieties, the protein content was maximum in sprouts (5.30% to 6.83%), followed by microgreens at the 7 DAG (2.35% to 4.25%) and 10 DAG (2.33% to 4.23%). The protein content significantly differed between sprouts and microgreens at the 7 DAG; however, there was no significant difference in crude protein content between microgreens at the 7 and 10 DAG.

Table 1: Moisture and ash content (mean ± SE) in mungbean sprouts and microgreens (at 7 and 10 DAG)

S.No.	Variety	Moisture content (%)			Ash content (%)		
		Sprouts	7 DAG microgreens	10 DAG microgreens	Sprouts	7 DAG microgreens	10 DAG microgreens
1	IPM 02-3	85.25 ± 5.17^{b}	84.29 ± 1.28^{b}	85.38 ± 2.15^{ab}	1.28 ± 0.31^{ab}	2.28 ± 0.36^{ab}	2.52 ± 0.14^{b}
2	MH 1-25	83.76 ± 4.39^{a}	81.69 ± 1.07^{a}	83.30 ± 1.40^{a}	1.45 ± 0.28^{ab}	1.61 ± 0.21^{ab}	1.82 ± 0.33^{a}
3	MH 2-15	84.98 ± 3.96^{ab}	84.76 ± 2.51^{ab}	85.84 ± 1.02^{ab}	1.05 ± 0.22^{ab}	1.88 ± 0.15^{ab}	2.56 ± 0.19^{b}
4	MH 1762	85.14 ± 3.21^{ab}	83.59 ± 1.96^{ab}	86.62 ± 3.10^{b}	1.70 ± 0.29^{b}	2.49 ± 0.36^{b}	2.59 ± 0.24^{b}
5	MH 1772	85.37 ± 2.64^{b}	83.61 ± 1.10^{ab}	84.53 ± 2.74^{ab}	0.83 ± 0.14^{a}	1.45 ± 0.33^{a}	1.93 ± 0.18^{a}

Values are mean ± standard error (SE) (n = 3). Means followed by different superscript letters differ significantly at $p<0.05$ according to Duncan's Multiple Range Test.

Table 2: Crude fibre and crude fat content (mean ± SE) in mungbean sprouts and microgreens (7 and 10 DAG)

S.No.	Variety	Crude fibre (%)			Crude fat (%)		
		Sprouts	7 DAG microgreens	10 DAG microgreens	Sprouts	7 DAG microgreens	10 DAG microgreens
1	**IPM 02-3**	0.91 ± 0.21^{b}	2.29 ± 0.24^{ab}	3.63 ± 0.32^{ab}	0.18 ± 0.08^{a}	0.24 ± 0.12^{a}	0.27 ± 0.06^{ab}
2	**MH 1-25**	0.79 ± 0.11^{ab}	3.29 ± 0.31^{c}	4.58 ± 0.48^{c}	0.26 ± 0.09^{b}	0.33 ± 0.05^{b}	0.34 ± 0.08^{b}
3	**MH 2-15**	0.84 ± 0.36^{ab}	2.86 ± 0.42^{b}	3.91 ± 0.28^{b}	0.25 ± 0.06^{b}	0.21 ± 0.04^{a}	0.24 ± 0.04^{a}
4	**MH 1762**	0.66 ± 0.14^{a}	1.99 ± 0.28^{a}	3.15 ± 0.18^{a}	0.23 ± 0.04^{ab}	0.34 ± 0.02^{b}	0.36 ± 0.02^{b}
5	**MH 1772**	0.81 ± 0.20^{ab}	3.18 ± 0.33^{bc}	4.22 ± 0.18^{bc}	0.18 ± 0.02^{a}	0.26 ± 0.06^{ab}	0.27 ± 0.04^{ab}

Values are mean ± SE (n = 3). Means followed by different superscript letters differ significantly at $p<0.05$ according to Duncan's Multiple Range Test.

Table 3: Total carbohydrate and crude protein (mean ± SE) in mungbean sprouts and microgreens (at 7 and 10 DAG)

S.No.	Variety	Total carbohydrate content (%)			Crude protein (%)		
		Sprouts	7 DAG microgreens	10 DAG microgreens	Sprouts	7 DAG microgreens	10 DAG microgreens
1	IPM 02-3	7.08 ± 0.96^{ab}	5.82 ± 0.71^{ab}	4.96 ± 0.52^{ab}	5.30 ± 0.34^{a}	3.26 ± 0.58^{ab}	3.24 ± 0.52^{ab}
2	MH 1-25	7.23 ± 1.05^{b}	6.57 ± 1.24^{b}	5.73 ± 0.71^{b}	6.51 ± 0.45^{b}	4.25 ± 0.88^{c}	4.23 ± 0.41^{c}
3	MH 2-15	6.05 ± 1.08^{a}	4.25 ± 0.64^{a}	3.85 ± 0.58^{a}	6.83 ± 0.27^{b}	3.62 ± 0.76^{b}	3.60 ± 0.39^{b}
4	MH 1762	6.33 ± 0.84^{a}	5.68 ± 0.42^{ab}	4.95 ± 0.63^{ab}	6.04 ± 0.53^{ab}	2.35 ± 0.91^{a}	2.33 ± 0.22^{a}
5	MH 1772	7.05 ± 0.63^{ab}	6.20 ± 0.61^{b}	5.36 ± 0.30^{b}	5.76 ± 0.21^{ab}	3.71 ± 0.67^{b}	3.69 ± 0.47^{b}

Values are mean ± SE (n = 3). Means followed by different superscript letters differ significantly at $p<0.05$ according to Duncan's Multiple Range Test.

3.2 Bioactive compounds

The antioxidant activity of mungbean sprouts ranged between 60% to 90%, which significantly differed from those in microgreens at the 7 (57% to 86%) and 10 DAG (55% to 82%). The antioxidant activity of sprouts and microgreens at both growth stages varied as: MH1-25>MH2-15>MH1762>MH1772>IPM02-3 (Figure 2). Antioxidants (phenolics, flavonoids and ascorbic acid) have health-promoting properties as they reduce oxidative stress and improve immune defense. The phenolic content varied between 3.26 mg/g to 5.10 mg/g; 2.34 mg/g to 4.31 mg/g and 1.29 mg/g to 3.64 mg/g in sprouts and microgreens (at the 7 and 10 DAG), respectively (Figure 3). There was a notable decrease in phenolic content of microgreens in comparison to sprouts. A highly positive correlation was found between phenolic and antioxidant activity of the sprouts and microgreens across the varieties.

Flavonoid content varied significantly between the five different mungbean varieties. The flavonoid content was found to be highest for sprouts (0.74 mg/g to 1.33 mg/g), thereafter decreasing in microgreens at the 7 (0.62 mg/g to 0.94 mg/g) and 10 DAG (0.47 mg/g to 0.73 mg/g) (Figure 4). For ascorbic acid, the values ranged from 0.57 mg/g to 0.81 mg/g for mungbean sprouts, exhibiting a marked increase in microgreens at the 7 DAG (1.77 mg/g to 2.10 mg/g), followed by a subsequent decline at the 10 DAG (1.50 mg/g to 1.86 mg/g). The ascorbic acid content was found higher in MH 1-25 (0.81 mg/g; 2.10 mg/g and 1.86 mg/g), MH 2-15 (0.74 mg/g; 1.99 mg/g and 1.61 mg/g) and MH 1762 (0.68 mg/g; 2.09 mg/g and 1.85 mg/g) in sprouts and microgreens (at 7 and 10 DAG), respectively (Figure 5).

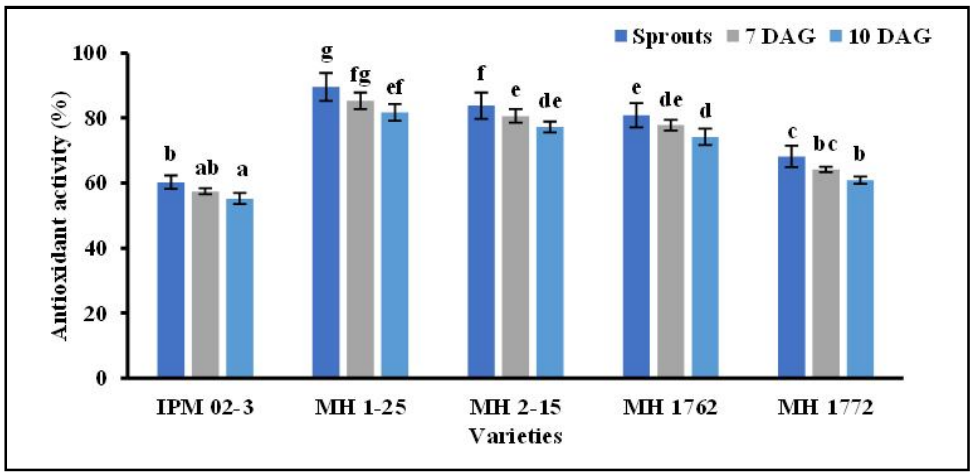

Figure 2: Antioxidant activity in mungbean sprouts and microgreens (at 7 and 10 DAG). A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

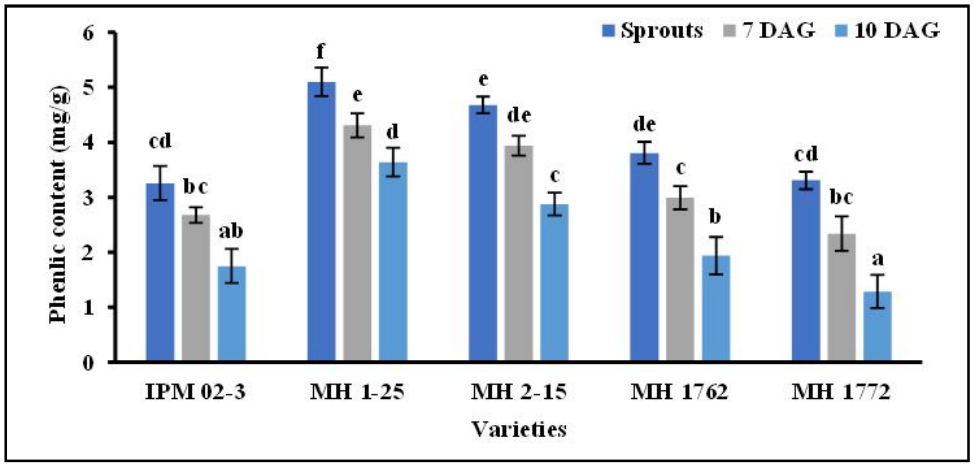

Figure 3: Phenolic content in mungbean sprouts and microgreens (at 7 and 10 DAG).A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

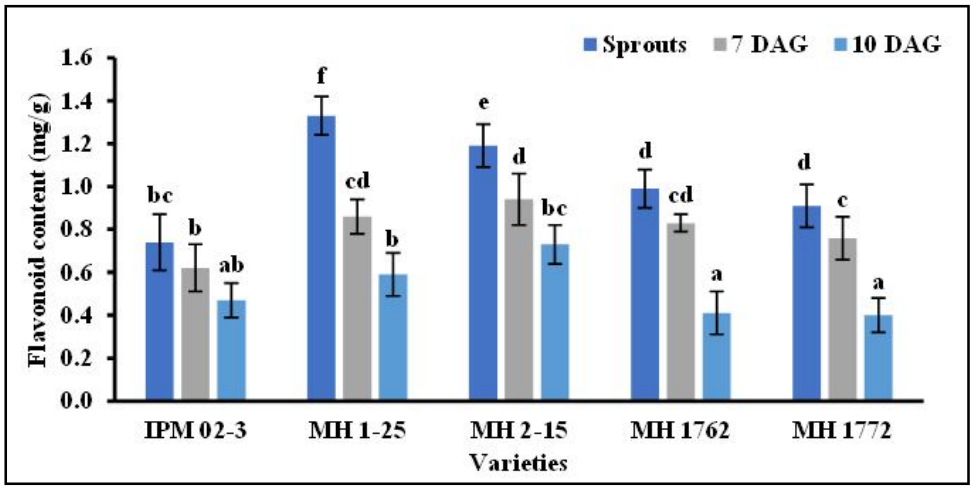

Figure 4: Flavonoid content in mungbean sprouts and microgreens (at 7 and 10 DAG). A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

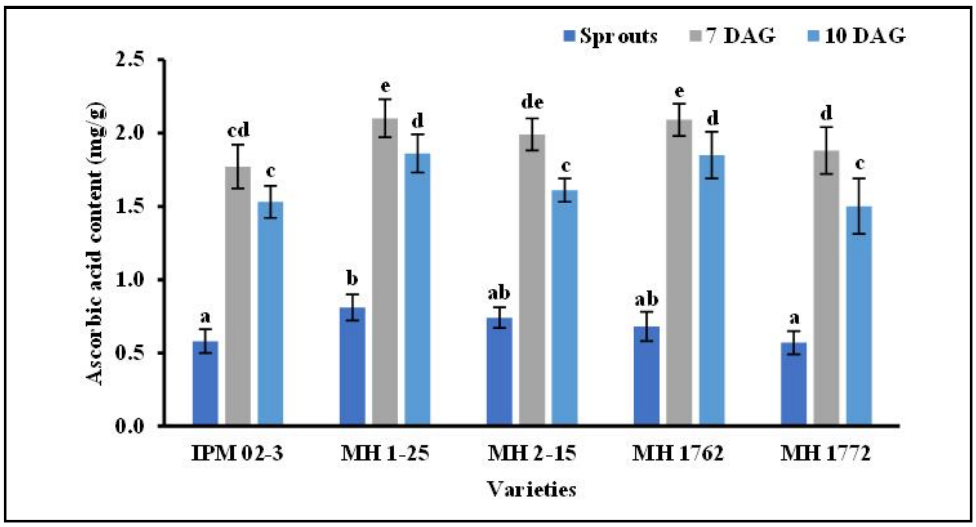

Figure 5: Ascorbic acid content in mungbean sprouts and microgreens (at 7 and 10 DAG). A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

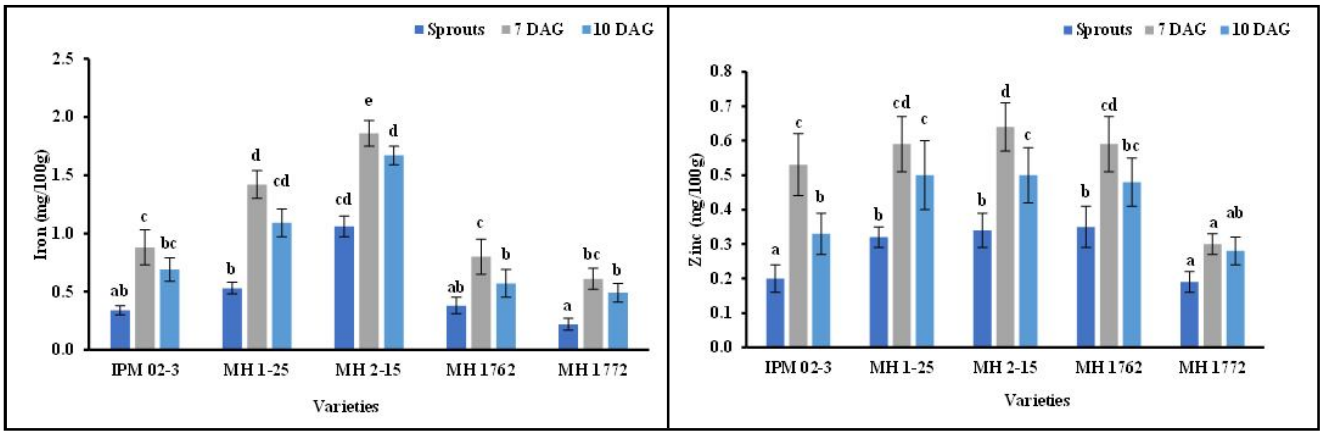

Figure 6: Mineral content (iron and zinc) in mungbean sprouts and microgreens (at 7 and 10 DAG). A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

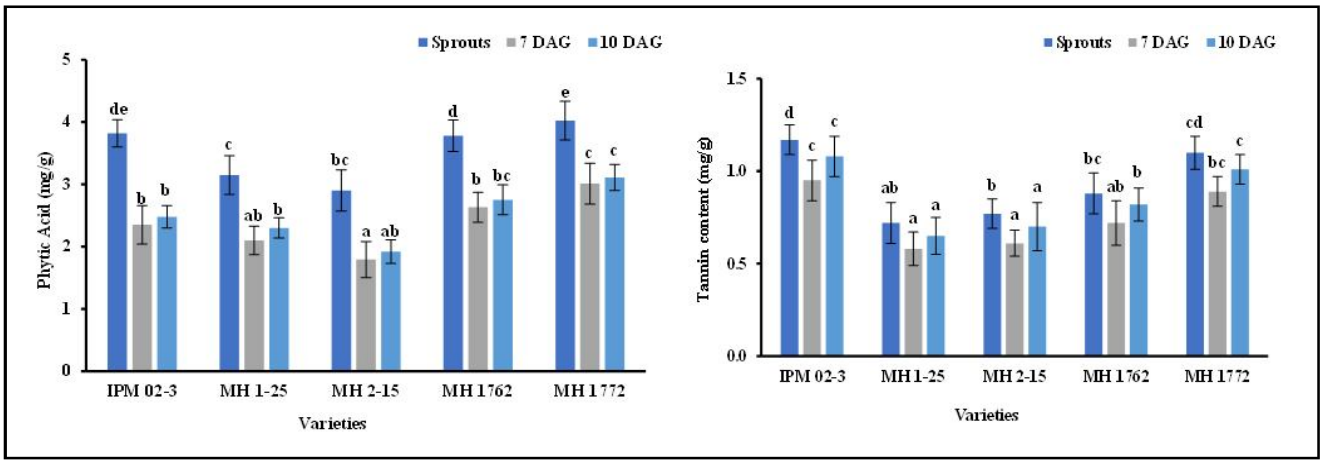

Figure 7: Antinutrient content (phytic acid and tannin) in mungbean sprouts and microgreens (at 7 and 10 DAG). A column and bar are used to illustrate the mean and standard error. Different letters on distinct bars denote significant differences ($p<0.05$) based on Duncan Multiple Range Test.

3.3 Mineral content

Mineral analysis revealed notable levels of iron and zinc, both of which are crucial for normal growth and development. Mineral content is lower in sprouts and microgreens compared to their mature counterparts; however, bioavailability of minerals shows an increment with germination (Bhaswant *et al.*, 2023). Across the different varieties, our results depicted higher iron (Fe) and zinc (Zn) content in microgreens at the 7 DAG (1.86 mg/100 g to 0.61 mg/100 g; 0.64 mg/100 g to 0.30 mg/100 g, respectively) and 10 DAG (1.67 mg/100 g to 0.49 mg/100 g; 0.50 mg/100 g to 0.28 mg/100 g, respectively) compared to sprouts (1.06 mg/100 g to 0.22 mg/100 g; 0.35 mg/100 g to 0.19 mg/100 g, respectively) (Figure 6).

3.4 Antinutrients

Antinutrients (phytic acid and tannin) analysis helps determine the nutritional quality of sprouts and microgreens, as they reduce mineral bioavailability by forming insoluble complexes. The phytic acid content was found to be higher for sprouts (4.02 mg/g to 2.90 mg/g) with varietal order as: MH 1772>IPM 02-3>MH 2-15>MH 1-25>MH 1762. Further, the phytic acid content reduced in microgreens at the 7 (3.01 mg/g to 1.79 mg/g) and 10 DAG (3.11 mg/g to 1.92 mg/g) (Figure 7). In our study, the tannin content varied significantly in sprouts and microgreens of the two growth stages (1.17 mg/g to 0.58 mg/g). Among the five different varieties, MH 1-25, MH 2-15 and MH 1762 showed the lowest tannin content (Figure 7).

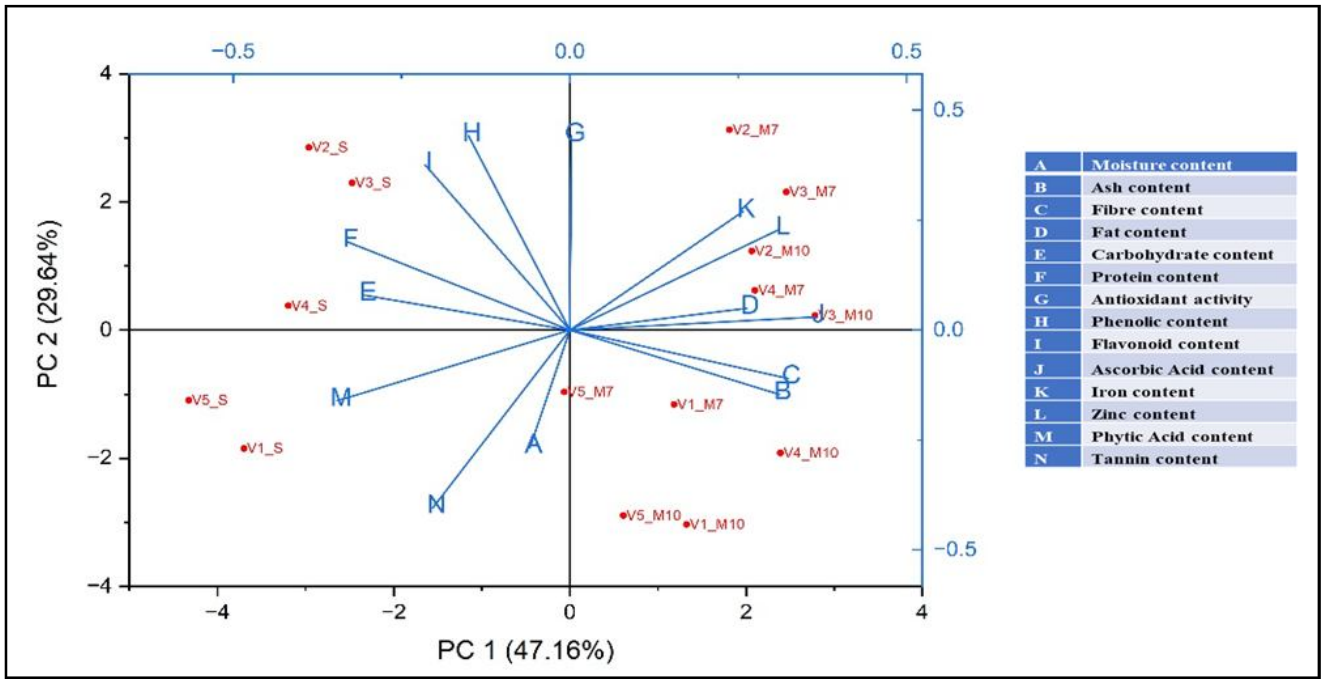

Figure 8: Principal component analysis of five mungbean varieties at three stages (namely sprouts and microgreens at 7 and 10 DAG). PC, principal component; V1, IPM 02-3; V2, MH 1-25; V3, MH 2-15; V4, MH 1762; V5, MH 1772; S: Sprouts; M7: Microgreens (at 7 DAG), M10: Microgreens (at 10 DAG). The blue arrow represents variable and the red dots represent 5 different mungbean varieties at 3 stages.

3.5 Principal component analysis (PCA)

PCA was performed to analyse the multivariate relationship between 11 different parameters (moisture, ash, crude fibre, crude fat, total carbohydrate, crude protein content, antioxidant activity, phenolic, flavonoid, ascorbic acid, iron and zinc content, phytic acid and tannin content) among different varieties (V1 to V5) and growth stages (sprouts and microgreens at 7 and 10 DAG). In the total variance (76.80%) of the dataset, PC1 and PC2 contributed 47.16% and 29.64%, respectively. Both V1-S and V5-S are strongly associated with phytic acid and tannin as they are clustered on the negative side of PC1 (Figure 8). Sprouts retain higher levels of these antinutrients, which tend to decrease as the growth period extends. Parameters such as ash, crude fibre, crude fat, minerals (Fe and Zn) and ascorbic acid showed strong positive loadings, highlighting the distinction between sprouts and microgreens. Antioxidant activity, phenolic and flavonoid content varied significantly between different developmental stages, particularly between sprouts and microgreens (at 7 DAG), as they were moderately positioned along PC2, as shown in Figure 8. V2-S and V3-S varieties were in proximity with these parameters, indicating relatively higher levels of antioxidant activity, phenolic and flavonoid content at the sprouting stage. Overall significant influence of developmental stage and varieties was observed on the various quality parameters of microgreens. With sprouts containing higher antinutrients, carbohydrates, protein, phenolic, flavonoid content and antioxidant activity, microgreens were comparatively higher in fibre, ascorbic acid and mineral content (Fe and Zn). These findings offer a scientific base and guidance to target specific mungbean varieties at optimal growth stages for their nutritional potential as functional food in the human diet.

4. Discussion

Our study evaluated the proximate and phytochemical analysis of different mungbean varieties to optimize the harvest time for superior nutritional quality in mungbean sprouts and microgreens. Moisture content is an important parameter that affects the quality, shelf life and ultimately consumer acceptability of fresh produce. The moisture

content in sprouts and microgreens (at 7 and 10 DAG) varied between 81% to 87%. The variation may be attributed to genetic makeup, which determines the water-holding capacity and transpiration rates. Similar findings were observed by Eswaranpillai *et al.* (2023), who observed moisture content of (90% to 91%) in sprouts of six different species, with corresponding soil-grown microgreens showing slightly higher values (90.34% to 92.21%). These values are considerably higher than those observed in the present study, where the moisture content was lower than the reported average value of 90.60% for mungbean sprouts and microgreens, indicating the possible varietal or environmental effects. In another study conducted on six diverse microgreens species, the moisture content of mungbean microgreens ranged between 85% to 86% which closely aligns with the present findings (Dhaka *et al.*, 2023).The ash content represents non-combustible materials (mineral oxides) reflecting the total mineral content (such as calcium, phosphorus, magnesium, manganese) and possible contaminants (Babayemi *et al.*, 2010). Our findings depicted higher ash content in microgreens at the 7 DAG (3% to 6%), followed by microgreens at the 10 DAG (1% to 3%). At the early stage, seedlings have a high metabolic rate and active nutrient mobilization; however, as the growth advances, metabolic processes slow down, depleting the overall biomass, hence lower ash content (Gunjal *et al.*, 2024). However, Kowitcharoen *et al.* (2021) reported lower ash content (0.64%) in mungbean microgreens than the values reported in the present study, while Elobuike *et al.* (2021) depicted an increase in ash content from 4.32% in mungbean sprouts (at 24 h) to 5.16% (at 5 DAG). The increment of ash content with advancing growth stages suggests a shift in physiological priorities from nutrient mobilization to biomass development, reflecting the dynamic nutrient patterns in sprouts and microgreens (Ebert, 2022).

The fibre content in sprouts and microgreens ranged between 0.66% to 5.00%. The fibre content of sprouts and microgreens is more available, hence aids to digestion by stimulating bowel movement and maintaining healthy gut microbiota (Ebert, 2022; Megat *et al.*, 2019). Hassan and Divakar (2024) reported fibre levels between 1.73% to 2.14% in mungbean microgreens (at 9 DAG), which were lower than those observed in the present study. While another study reported higher fibre (3.41%; 3.74%) and fat content (2.73%; 1.58%) in mungbean sprouts and microgreens (5 DAG), respectively, thereby highlighting the impact of variety and growing conditions. Furthermore,Wang *et al.* (2021) reported higher levels of fat (0.59% to 1.26%) in 22 different mungbean varieties compared to our results.

Sprouts (6% to 8%) exhibited higher total carbohydrates compared to the microgreens (3% to 6%) at both growth stages (7 and 10 DAG). The difference in carbohydrate content between sprouts and microgreens may be attributed to varietal differences and different growth stages. Enzyme activity is enhanced when the seed develops into sprouts and complex carbohydrates are broken down into simpler sugars that are utilised in microgreens to generate complex structures (leaves and stem) and develop other nutrients (such as vitamins), which explains lower carbohydrate levels in microgreens (at 7 DAG and 10 DAG) compared to sprouts (Wojdylo *et al.*, 2020). The lower carbohydrate content in microgreens (at 7 and 10 DAG) makes them suitable for a low-calorie diet and highlights their potential as functional foods for weight management and glycemic regulation. Kowitcharoen *et al.* (2021) studied 14 different microgreens and the carbohydrate content ranged between 2.32% to 7.16%, among which mungbean microgreens showed higher levels (7.16%) of carbohydrate than those reported in our study. Another study conducted in India compared sprouts and microgreens of six different species, reporting a decline in carbohydrate content, with values declining from 7.27 mg/g in mungbean sprouts to 5.13 mg/g in mungbean microgreens (Eswaranpillai *et al.*, 2023).

Mungbean microgreens and sprouts are an excellent addition to vegan diet as they offer a nutritious and low-calorie food option with significant protein content (Kaur *et al.*, 2022). In our study, sprouts showed higher protein content in comparison to microgreens, depicting the seed reserves. As the growth progresses, there is higher enzyme activity leading to protein hydrolysis, rearrangements and mobilization of amino acids for various metabolic processes, resulting in the evident decline of protein accumulation in microgreens. Ebert *et al.* (2017) reported protein content (3.38% to 5.19%) in sprouts of four different mungbean varieties that closely align with our findings. Similarly, a study in Thailand reported 4.55% of protein content in mungbean microgreens, comparable to the present results (Kowitcharoen *et al.*, 2021). Consistent with our observation, Eswaranpillai *et al.* (2023) also reported lower protein content in mungbean microgreens compared to sprouts.

In our study, the antioxidant activity of sprouts and microgreens ranged between 50% to 90%. The enhancement in antioxidant activity during germination can be attributed to the enhanced enzymatic activity and biosynthesis of diverse bioactive compounds such as phenolics, flavonoids, ascorbic acid and other metabolites that contribute to oxidative stress mitigation. Another study reported by Chon *et al.* (2013) analysed DPPH free radical scavenging activity in cowpea (44%), mungbean (42%) and soybean sprouts (25%). The antioxidant activity reported in the mungbean sprouts of this study was much lower compared to our findings. Dhaka *et al.* (2023) reported the antioxidant activity of six different microgreens, varying between 72% to 87%. Collectively, the higher metabolic activity during germination and early growth stages promotes the synthesis of various phytochemicals, thereby strengthening their functional and nutraceutical value.

Microgreens are recognized for their elevated phenolic content, which contributes significantly to their antioxidant capacity and potential health benefits. The high phenolic content in sprouts (3% to 5%) may be attributed to upregulation of phenolic biosynthetic pathways (shikimic and phenylpropanoid) as influenced by germination. Studies have shown that the total phenolic content in microgreens varies widely depending on species and development stage (Hollman, 2011; Ho, 1992). Dhaka *et al.* (2023) reported phenolic content ranging from 52 µg/100 g to 1136 µg/100 g across six different microgreens species, with mungbean microgreens containing 840 µg/100 g, which is comparatively lower than the levels obtained in the present study. Conversely, Sikora and Sweica (2018) reported phenolic content between 7 mg/g to 10 mg/g in mungbean sprouts, almost twice as observed in our study. Our results revealeda stage-dependent decline in phenolic content from sprouts to microgreens, indicating metabolic transition across the developmental stages. A similar trend was observed by Huang *et al.* (2014), who reported higher phenolic content in sprouts (1.2 mg/g), followed by a decline at 5 DAG (0.65 mg/g). This decline may be attributed to the transformation of phenolic compounds as the plant shifts its metabolic focus to growth and photosynthesis, requiring other phytochemicals (such as chlorophyll, carotenoids).

Flavonoids have a significant role in neutralizing free radicals, reducing oxidative stress and thereby supporting cellular metabolism (Kumar and Pandey, 2013; Panche *et al.*, 2016). In our study, flavonoid content ranged between 0.74 mg/g to 1.33 mg/g for sprouts, while for microgreens at the two different growth stages, it ranged between 0.40 mg/g to 0.94 mg/g. The relatively higher flavonoid accumulation in sprouts depicts active secondary metabolite synthesis during germination, which subsequently shifted to growth and development (Bhaswant *et al.*, 2023). On the other hand, Priti *et al.* (2021) studied the flavonoid content (2.8 mg/100 g to 3.71 mg/100 g) in 20 different varieties of mungbean microgreens grown in Delhi, which were higher compared to our findings. This difference may be attributed to the genetic makeup, which affects their biosynthetic pathways. Another study in Korea reported flavonoid content between 0.54 mg/g to 0.91 mg/g for mungbean sprouts, which was lower than the values obtained in our study (Jeon *et al.*, 2023). Hu *et al.* (2024) reported an increase in flavonoid content during germination, being maximum at 3 DAG (1000 µg/g) and thereafter decreasing on the 5 DAG (600 µg/g). Moreover, the bioactive compounds such as phenolic, flavonoids and other phytochemicals not only enhance the nutritional value of microgreens but also contribute to their distinct flavor, aroma and taste (Rawat *et al.*, 2024).

Ascorbic acid is a potent antioxidant that mitigates oxidative stress and supports immune function, though its concentration varies depending upon the species, harvest time and environmental factors such as light and water availability (Wu *et al.*, 2024). In the present study, microgreens exhibited higher ascorbic acid compared to sprouts, which may be attributed to the higher photosynthetic capacity and longer growth time under light, promoting the biosynthesis of vitamins, including ascorbic acid. These observations were consistent with findings by Masood *et al.* (2014), who reported an increase in ascorbic acid from 4.83 mg/100 g (in sprouts) to 28.50 mg/100 g (in microgreens). On the other hand, Gan *et al.* (2016) reported a higher level of ascorbic acid (123 mg/100 g) on the 3 DAG and thereafter the content gradually reduced by 5 DAG (90 mg/100 g), indicating the stage-dependent variation during growth.

Based on the mineral analysis, iron was more prevalent compared to zinc among the various mungbean varieties at different growth stages. The higher bioavailability of iron and zinc in sprouts and microgreens helps combat micronutrient deficiencies such as anaemia and impaired immune functions, highlighting their role as functional foods for overall human health (Bhaswant *et al.*, 2023). A study from Taiwan reported that mungbean sprouts contain zinc levels ranging between 0.49 mg/100 g to 0.67 mg/100 g and iron levels ranging between 0.36 mg/100 g to 0.81 mg/100 g (Ebert *et al.*, 2017). While our result for iron was consistent with their findings; however, zinc levels were comparatively lower. Similarly, Vayupharp (2013) showed an increment in iron content of mungbean cultivars after germination. Microgreens accumulate higher levels of minerals in comparison to sprouts, as the extended growth period allows for continuous mineral mobilization from seed reserves. On the other hand, a study conducted in Leh reported comparatively narrow ranges for iron and zinc content, with values ranging between 0.62 mg/100 g to 0.79 mg/100 g and 0.21 mg/100 g to 0.32 mg/100 g, respectively, in 20 different cultivars of mungbean microgreens (Priti *et al.*, 2021).

Although mungbean is protein, vitamins and mineral-rich legume, they contain antinutrients that inhibit the bioavailability of nutrients. However, studies suggest that germination leads to partial breakdown of phytic acid and other mineral-binding antinutrients, making the nutrients more available for absorption (Idris *et al.*, 2025; Thakur *et al.*, 2021). Thus, encouraging the consumption of microgreens is beneficial in addressing specific micronutrient deficiencies. Our study revealed notably lower antinutrients (phytic acid and tannin). Dhaka *et al.* (2023) also determined the phytic acid in microgreens of six different species (1.56 mg/g to 3.04 mg/g), among which mungbean microgreens content was 2.38 mg/g, closely aligning with our findings. While Tajjodin *et al.* (2011) reported a reduction in phytic acid content by 60% to 73% due to germination in nine cultivars of mungbean.Tannins are phenolic compounds that impede protein digestion by deactivating various digestive enzymes (de Melo *et al.*, 2023). The degree of antinutrients (phytic acid and tannin content) reduction depends upon species, germination stage and growing media (Elliott *et al.*, 2022). For example, a study conducted in India reported tannin content of 7 different microgreens in the range 0.48 mg/g to 1.36 mg/g that aligned with our results (Gunjal *et al.*, 2024), while Kurian *et al.* (2020) found a reduction in tannin content in mungbean sprouts and microgreens (0.47% and 0.42%, respectively). Tuncel *et al.* (2025) studied the phytic acid and tannin content of raw and germinated pulses (including mungbean), reporting phytic acid and tannin content of 1.05 mg/g and 0.03 mg/g in germinated mungbean, which deviated from our findings, highlighting the influence of cultivars and germination conditions.

5. Conclusion

As there is growing demand for healthy, ready-to-eat foods with functional benefits, fresh, organic and nutrient-dense sprouts and microgreens have the potential to meet these demands. The distinct nutrient profile of microgreens influences the organoleptic properties (color, texture, aroma and taste) of microgreens and thereby consumer palatability and perception. Although, microgreens have been popular as functional foods, comparative studies analysing multiple species and developmental stages remain scarce (specifically for mungbean). Our study indicated a significant effect of varieties and growth stages (sprouts, microgreens at 7 and 10 DAG) on the nutritional quality of mungbean. Based on our analysis, microgreens at 7 DAG, followed by 10 DAG, are preferred as a potential dietary component due to their high ascorbic acid, fibre, mineral content and lower antinutrients, while sprouts are preferred for their high carbohydrate, protein, phenolic, flavonoid content and antioxidant activity. Further studies should focus on exploring nutrient bioavailability *in vivo* for validating their health benefits, while addressing challenges related to post-harvest storage, shelf life and microbial contamination to ensure their safe and effective utilization as functional foods.

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Author contributions

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Tokas. All authors have read and agreed to the final version of the manuscript for publication.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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