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Original research

Evaluation of Nutritional Profile, Phytochemical Content, and Antioxidant Potential of Finger Millet Landraces from the Tribal Belt of Chhattisgarh, India

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ABSTRACT

With global food demand rising and dietary deficiencies widespread, nutrient-dense, climate-resilient crops such as finger millet offer promising alternatives to conventional staples. This study investigates the nutritional composition, phytochemical constituents, and antioxidant potential of 8 finger millet [*Eleusine coracana* (L.) Gaertn.] landraces cultivated in the tribal regions of Chhattisgarh, India. Our analysis revealed that the landrace F4 possessed the highest amount of protein (10.13 ± 0.12%) and dietary fibre (14.88 ± 0.61%) with low carbohydrate content (56.77 ± 0.86%) in comparison to other landraces. Similarly, calcium content (1477.73 ± 0.37 mg/kg) was found to be higher in F5, whereas the highest iron content (121.38 ± 0.23 mg/kg) was recorded in F7. High levels of phenolics (26.03 ± 0.19 mg GAE/g dw) and flavonoids (35.39 ± 0.76 mg QE/g dw) were observed in F2 landrace, underscoring the potent nutraceutical attributes of this traditional variety. Furthermore, the antioxidant potency was evaluated using DPPH and ABTS radical scavenging assays, with the lowest IC_{50} values recorded for F2 landrace. The findings highlight the potential of indigenous finger millet landraces to not only combat nutrient deficiencies but also support metabolic health through their antioxidant and functional properties. This also underscores the importance of promoting and conserving traditional millet varieties to enhance nutrition and food security, particularly in underserved tribal communities.

1. Introduction

Global population growth, projected to reach 9.8 billion by 2050, is expected to significantly increase food demand, with estimates exceeding 14,886 million tons (Islam, 2019). Current global food systems are heavily dependent on a limited number of staple crops such as rice, wheat, and maize, which have led to a lack of dietary diversity and widespread micronutrient deficiencies. Despite the availability of thousands of edible plant species, less than 1% are widely cultivated and consumed (Bouis et al., 2011). This overreliance contributes to an imbalance in global nutrition and is associated with the rising prevalence of non-communicable diseases such as diabetes, obesity, and malnutrition (Prasad et al., 2020). Addressing these nutritional challenges requires a shift toward sustainable, nutrient-dense crops that can thrive under changing climatic conditions.

Finger millet [*Eleusine coracana* (L.) Gaertn.], commonly known as ragi, is a hardy, climate-resilient cereal crop predominantly cultivated in the semi-arid and rainfed regions of Asia and Africa. It possesses a short growth cycle, exceptional drought tolerance, and the ability to grow in marginal soils, making it particularly suitable for cultivation in resource-limited and ecologically vulnerable regions. Traditionally grown by tribal communities in India, finger millet holds significant nutritional and cultural importance and contributes to local food security (Behera, 2017; Baduni et al., 2024). Nutritionally, finger millet is rich in carbohydrates (65–75%), proteins (5–8%), and dietary fibre (15–20%), and is a notable source of essential micronutrients such as calcium (344 mg/100 g), iron, magnesium, and vitamins (Maharajan et al., 2022). In addition to its macronutrient profile, finger millet contains a wide array of secondary metabolites, including phenolic acids, flavonoids, tannins, and lignans, which are known for their antioxidant, antidiabetic, antimicrobial, and anti-inflammatory properties (Durairaj et al., 2019; Maharajan et al., 2022). High-performance liquid chromatography (HPLC) analyses have revealed that finger millet seeds contain several bioactive phenolic compounds, including ferulic acid (32.8%), proto-catechuic acid (15.3%), p-hydroxybenzoic acid (17.9%), gallic acid (12.6%), quercetin (5.6%), vanillic acid (3.8%), p-coumaric acid (4.4%), syringic acid (4.0%), and trans-cinnamic acid (3.6%) (Chethan et al., 2008). These compounds have demonstrated therapeutic potential in managing chronic health conditions such as type 2 diabetes and cardiovascular diseases (Ci & Indira, 2016). Recent studies have also shown promising cytotoxic activity of finger millet extracts against cancer cell lines. For example, Pears et al. (2023) reported in-vitro cytotoxic effects of methanolic extracts of finger millet against HepG2 liver cancer cells, with significant activity observed at 250 µg/mL.

Despite numerous health benefits and environmental adaptability, finger millet remains underutilized in mainstream agriculture due to limited research investment, lack of high-yielding varieties, and post-

harvest processing challenges (Wambi et al., 2021). However, the United Nations' declaration of 2023 as the International Year of Millets has revitalized global interest in these ancient grains, positioning them as strategic crops for achieving nutritional security and climate resilience.

Tribal populations in Chhattisgarh, India, continue to cultivate and consume diverse landraces of finger millet, preserving their genetic variability and adaptation to local agroecological conditions. These traditional varieties not only support the nutritional needs of marginalized communities but also offer valuable genetic resources for future crop improvement and climate adaptation.

The present study aims to investigate the nutritional composition, phytochemical constituents, and antioxidant activity of finger millet landraces from tribal regions of Chhattisgarh. By evaluating the nutraceutical potential of these underutilized varieties, the study seeks to emphasize their role in promoting metabolic health and supporting sustainable agricultural practices in resource-constrained environments.

2. Materials and Methods

2.1. Collection and morphological characterization of finger millet landraces

Finger millet grains were collected from various tribal regions of the Indian state of Chhattisgarh to explore nutritional diversity among local landraces. These landraces are cultivated across an extensive geographical area (Table 1). Although these landraces are not listed as endangered or threatened species, prior informed consent was obtained from the seed-donor farmers involved in their cultivation. Seed collection for this study was carried out in full compliance with institutional guidelines. Nutritional values were recorded to assess potential variations across these landraces, which are designated as F1 to F8. The collected seeds were identified and authenticated by a Krishi Vigyan Kendra, affiliated with Indira Gandhi National Tribal University (IGNTU), Amarkantak, Madhya Pradesh. Voucher specimens for each landrace were deposited in the Department of Botany, Central Tribal University of Andhra Pradesh, with the following accession numbers, as presented in Table 1. Seed size analysis was conducted for all eight landraces using a Magnus microscope (Model TZM6). Each seed was measured for average diameter, revealing notable differences among the samples.

Table 1: Collection area of finger millet landraces from Chhattisgarh state of India.

S. No.	Finger millet	Sample collection location	Voucher No.
1	F1	Ganiyari, Bilaspur	CTUAP/BOT/FM-EC/GAN-BIL/25042022/001
2	F2	Koliyari, Nigari, Dhamtari	CTUAP/BOT/FM-EC/KOL-NGR-DMT/22052022/002
3	F3	Korar, Kanker	CTUAP/BOT/FM-EC/KOR-KKR/20052022/003
4	F4	Bhaismundi, Kanker	CTUAP/BOT/FM-EC/BHM-KKR/21052022/004
5	F5	Shahwada, Kanker	CTUAP/BOT/FM-EC/SHW-KKR/21052022/005
6	F6	Bhiragaon, Kondagaon	CTUAP/BOT/FM-EC/BHR-KDN/17052022/006
7	F7	Chipawan, Kondagaon	CTUAP/BOT/FM-EC/CHP-KDN/18052022/007
8	F8	Kasoli, Geedam, Dantewada	CTUAP/BOT/FM-EC/KSL-GDM-DTW/28052022/008

2.2. Proximate analysis

Proximate analysis is a technique for determining the macronutrient composition and nutritional value of food samples. Proximate analysis includes crucial constituents such as protein, fat, moisture, ash, and carbohydrates, and the technique for determining them is known as proximate analysis.

The grains were ground in a cyclone mill (UDY Corporation, USA) for estimating the proximate composition. Proximate composition of the finger millet landraces was determined as per the Association of Official Analytical Chemists (AOAC) methods (Gaithersburg, 2016). Protein content of the grains was determined by the Kjeldahl method. Fat content was estimated by Soxhlet extraction using hexane. Ash content was analysed in a muffle furnace at 650˚C. The total dietary fibre content was determined by the enzymatic-gravimetric method using the total dietary fibre assay kit (Megazyme, Ireland) as per the manufacturer's instructions. The carbohydrates were obtained by the difference method as given in the following formula.

Carbohydrate = 100 – (moisture % + protein % + dietary fibre % + fat % + ash %).

2.3. Mineral analysis by acid digestion method

As per AOAC (Horwitz & Latimer, 2000; Palma et al., 2015), the acid digestion method is used to determine the metal content in the sample. First, 1.0 gram of the material was inserted in a glass tube with 25 millilitres of a di-acid solution (4:1 solution of nitric acid and perchloric acid). When the temperature was raised to 50–350°C for 2 hours, the brown fume began to dissipate, indicating that digestion was complete. After cooling to room temperature (RT), 50 ml of DDW was added and filter paper was used to remove the dust particles. Subsequently, it was stored at 4°C for future use. An ICP-MS Triple Quad (Agilent 8900) was used to measure heavy metal levels. The nebulizer aspirates the aqueous sample into the flame atomizer to measure analyte concentrations with high precision at the parts-per-million (ppm) level, including Fe, Zn, Mg, Mn, K, Ca, Cu, and a small amount of Nickel (Ni). The distinct metal solutions of 1, 5, and 10 ppm are used as standards, and the counts in the sample solution were used for quantification.

2.4. Preparation of crude extract

Freshly harvested seeds were collected, cleaned, dehusked, and ground into flour using the mechanical grinder. Ten grams of dry powder was individually combined with 100 mL of aqueous and methanolic solvents. The mixture was gently agitated for 48 h to yield crude extracts. These extracts were subsequently concentrated using water bath maintained at 40-45°C and stored at 4°C for further analysis.

2.5. Qualitative estimation of carbohydrate and phytochemicals

To determine the presence of physiologically active chemicals, methanolic extracts (ME) and aqueous extracts (AE) of finger millet landraces underwent a preliminary qualitative phytochemical screening. The screening process for secondary metabolites, such as tannins, flavonoids, steroids, and saponins, was performed as described by Harborne (1998). Furthermore, the presence of carbohydrate, alkaloids, coumarins, terpenoids, glycosides, phenols, flavonoid, tannin, quinones, and saponin were analysed by following the procedures outlined by Roghini and Vijayalakshmi, 2018.

2.6. Quantitative estimation of phytochemical

2.6.1 Total phenol assay

The Folin-Ciocalteu colourimetric technique, established by Singleton et al. (1999), was used with minor modifications to assess the total phenolic content (TPC) of the plant extract. A standard gallic acid solution (1 mg/mL) was made by dissolving 5 mg of gallic acid in 5 mL of methanol. This solution produced methanol-based dilutions with different amounts of gallic acid (20, 40, 60, 80, and 100 µL/mL). To each concentration, 2 mL of 10% Folin-Ciocalteu reagent (FCR) and 2 mL of 7% Na_2CO_3 was added, totalling 4.5 mL. After thoroughly mixing, the blue solution was incubated for 30 minutes at room temperature (RT). Finally, after incubation, the absorbance was measured at λ_{max} = 750 nm compared to a blank. A UV-visible spectrophotometer was used to identify the dark blue colour developed by the FCR reagent's oxidation of phenols in the plant extract. All tests were carried out in triplicate, and the average absorbance values at various concentrations of gallic acid were used to create a calibration curve. The results were reported as milligrams of gallic acid equivalents per gram of dry weight (mg GAE/g dw).

2.6.2 Total flavonoid assay

The total flavonoid content (TFC) was estimated through aluminium chloride ($AlCl_3$) method, with some changes to meet the experiment's requirements (Kumar et al., 2013). To make a 1 mg/mL stock solution, 5 mg of quercetin was dissolved. The standard solution was then serially diluted to produce four distinct concentrations: 0.20 mg/mL, 0.40 mg/mL, 0.60 mg/mL, and 1.0 mg/mL. For each concentration, 0.5 mL of the quercetin solution was added to a test tube containing 1.4 mL of distilled water and 0.1 mL of 10% $AlCl_3$, making the total amount 2 mL. After 15 minutes of dark incubation, absorbance was measured through spectrophotometer at λ_{max}= 415 nm. This procedure was repeated three times, and the average absorbance value was determined. The TFC was estimated using the calibration curve's linear equation and expressed in milligrams quercetin equivalent per gram dry weight (mg QE/g dw).

2.6.3 Total tannin content assay

Tannic acid was used to generate a standard curve at 20-100 µg/mL concentrations from a 1 mg/mL stock solution. The absorbance of both the test samples and the reference solutions was measured at λ_{max} = 725 nm using a UV/visible spectrophotometer. A blank was made by replacing the sample with distilled water and repeating the process. Tannin content was calculated using the Folin-Ciocalteu technique with slight modifications (Ci and Indira, 2016). In this procedure, 0.1 mL of plant extract was mixed with 0.5 mL of Folin-Ciocalteu reagent (10%) and 1 mL Na_2CO_3 (20%). The mixture was thoroughly agitated and incubated for 30 minutes at RT. Three independent measurements were used to determine the total tannin content (TTC), and the results were reported as milligrams of tannic acid equivalents per gram of dry weight (TAE/g dw).

2.7. Antioxidant activity

2.7.1 Total antioxidant capacity (Phosphomolybdenum assay)

The sample's total antioxidant capacity was assessed using the phosphomolybdate technique with ascorbic acid as the standard (Prieto et al., 1999). Following the same technique, the reagent concentration was adjusted. Specifically, 0.3 mL of the sample solution was combined with 2.7 mL of phosphomolybdate reagent mixture (1.6 mL of H_2SO_4, 193 mg of Na_3PO_4, and 247.17 mg of $(NH_4)6Mi_7O_{24}$) to make a total volume of 50 mL). The mixture was sealed in a water bath and heated to 95°C for 90 minutes. After cooling to RT, absorbance was measured at λ_{max} = 695 nm with a UV spectrophotometer and compared to a blank. Using 0.3 mL of reagent solution and the proper solvent, a blank was prepared in the same manner. The estimation of total antioxidant capacity (TAC) results was expressed in ascorbic acid equivalents per gram of dry weight (mg AAE/g dw). These values were determined from a triplicate analysis (n = 3).

2.7.2 DPPH radical scavenging assay

DPPH activity was determined using a previously established approach with slight modification (Bursal & Gulcin, 2011). An absorbance of 0.98 ± 0.02 at λ_{max}= 515 nm was obtained by dissolving 4 mg of DPPH in 100 mL of methanol. The finger millet extracts and standard at different concentrations (20-220 µg/mL) were combined with 3 mL of the DPPH solution. The mixture was incubated in the dark at RT for 15 minutes. A blank was made by replacing the sample with 300 µL of methanol. The antioxidant potential of finger millet seed extracts was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay (RSA), with calculations performed according to the standard Equation (1). Ascorbic acid was used as a standard. The results are expressed as the concentration required to inhibit 50% of the DPPH radicals (IC_{50}) in µg/mL, and were determined by plotting % RSA against different concentrations of the sample.

$$\% \text{ RSA} = \frac{\text{Absorbance of control} - \text{Absorbance of the sample}}{\text{Absorbance of control}} \times 100 \quad \text{Eq (1)}$$

2.7.3 ABTS radical scavenging assay

The ABTS activity was assessed using the standard approach published by (Re et al., 1999). This assay is based on antioxidant ability to quench the blue-green ABTS radical cation, causing decolourization. Free radicals were produced using 2.45 mM $K_2S_2O_8$ and 7 mM ABTS in double-distilled water (DDW). These solutions were mixed in a 1:1 ratio and incubated in the dark for 24 hours. After incubation, the absorbance was measured at λ_{max} = 745 nm (0.700 ± 0.02). In the test reaction, 0.3 µL of the plant sample was mixed with 2.5 mL of ABTS solution, resulting in a total volume of 2.8 mL. The mixture was incubated in the dark for 15 minutes. The ABTS radical-scavenging activity (RSA) of the extract was first calculated using Equation (2). The IC_{50} value, or the concentration of extract required to scavenge 50% of free radicals, was determined by plotting the % RSA against different extract concentrations and using linear regression for interpolation. The IC_{50} value was provided as mean ± SD (n = 3).

$$\%\text{RSA} = \frac{\text{Absorbance of control} - \text{Absorbance of the sample}}{\text{Absorbance of control}} \times 100 \quad Eq\ (2)$$

2.7.4 Ferric reducing antioxidant power (FRAP) assay

The Fe^{3+} reducing power was measured using a traditional FRAP assay, as described elsewhere (Viswanath et al., 2009), with modifications made to accommodate experimental conditions. To perform this experiment, 2.5 mL of phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of 1% potassium ferricyanide ($K_3[Fe(CN)_6]$) were added to the sample extracts and a standard solution of 0.5 mL ascorbic acid. The reaction mixture was then incubated at 50°C for 30 minutes. After incubation, 2.5 mL of 10% trichloroacetic acid ($C_2HCl_3O_2$) was added, and the mixture was centrifuged at 13,000 rpm for 10 min. The

supernatant was combined with 2.5 mL of DDW and 0.5 mL of 0.1% $FeCl_3$. The absorbance was measured at λ_{max} = 700 nm with a UV-VIS spectrophotometer. The extract's reducing power was assessed by measuring optical density. An increase in absorbance implies a higher reducing power. Results are expressed in milligrams of ascorbic acid equivalents (AAE) per gram of dry weight (dw) of extract (mg AAE / g dw), provided as mean ± SD (n=3).

2.7.5 Nitric oxide radical scavenging assay

The nitric oxide (NO) free radical scavenging activity was determined using the standard method (Sreejayan & Rao, 1997; Dubey et al., 2023). In separate test tubes, 80 µL of $Na_2[Fe(CN)5NO]$ and 200 µL of plant crude extract were mixed at different doses (20-220 µg/mL). The reaction was allowed to continue for 15 minutes with light exposure. After 15 minutes, 1.0 mL of Griess reagent (1% sulphanilamide in 2% H_3PO_4) was added to each tube, which was then incubated for 45 minutes at 30 °C in the presence of light. To stop the reaction, 40 µL of 0.1% naphthylamine dihydrochloride in 2.5% H_3PO_4 was added. The final volume was adjusted to 2 mL with DDW, and the absorbance was measured at λ_{max} = 540 nm. The nitric oxide radical scavenging activity was calculated as % inhibition using Equation (3). The IC_{50} value, defined as the concentration of extract required to scavenge 50% of the free radical, was subsequently determined. The IC_{50} value was expressed as mean ± SD (n = 3).

$$\% \text{ RSA } = \frac{\text{Absorbance of Control } - \text{ Absorbance of the Sample}}{\text{Absorbance of Control}} \times 100 \quad Eq\ (3)$$

2.8. coli Statistical analysis

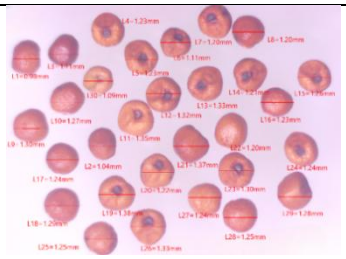
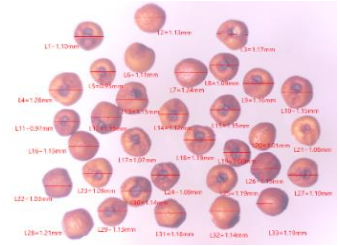
The values shown in the results are the mean ± standard deviation (SD) of three replicates. The Graphs are prepared using GraphPad Prism 9.0.1.

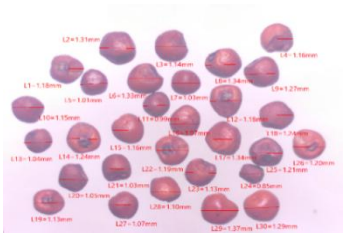
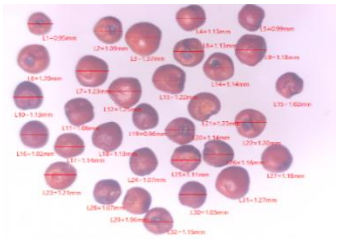
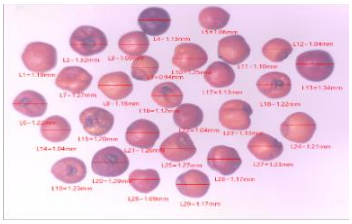
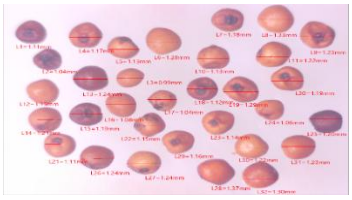
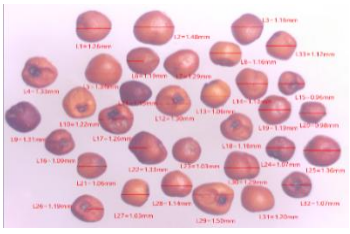
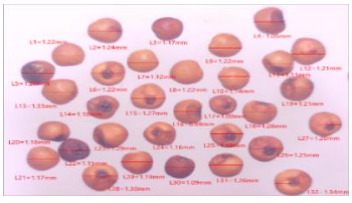
3. Results and discussion

3.1. Morphology and seed size of different finger millet landraces

Morphological analysis revealed that the grains are generally spherical, ranging in size from 1.11 ± 0.07 to 1.23 ± 0.09 mm, and colour varies from light brown to dark brown (Table 2). The present study supports earlier reports that finger millet displays substantial morphological diversity, particularly in grain colour and seed shape, previously identified from India, Choudhary et al. (2025) and Nepal (Thapa et al., 2025), similarly documents 66 and 72 finger millet shows wide variation in seed pigmentation, including light brown, white, black so on and morphology across landraces. These patterns, together with the significant genotype-environmental interaction reported by Bandyopadhyay et al. (2023), highlight the combined influence of genetic background and environmental conditions on phenotypic expression. Consistent with these studies, our research also finds clear differences in seed colour and size across the finger millet landraces. Such variability likely reflects variance in genetic composition and local cultivation environment, which influence pigment deposition, grain filling, and seed coat development, as noted by Xiang et al. (2019). These morphological traits are not only important for distinguishing varieties but also related to underlying biochemical and nutritional characteristics. However, the diversity observed in this study underscores the adaptive potential of finger millet and supports the value of conserving landraces diversity for future crop improvement.

Table 2: Morphology and seed size of different finger millet landraces.

Name of landraces	Finger millet grains	Picture showing diameter (mm) of individual seed	Average seed diameter (mm). Mean ± SD (n=29)
F1			1.23 ± 0.09
F2			1.11 ± 0.07

F3			1.16 ± 0.12
F4			1.12±0.09
F5			1.16 ± 0.09
F6			1.17 ± 0.08
F7			1.12 ± 0.13
F8			1.18 ± 0.07

3.2. Proximate nutritional composition of finger millet landraces

The nutritional composition of eight finger millet landraces was analysed for dietary fibre, moisture, protein, fat, ash content and carbohydrate, and the results were expressed in percentages (%). We observed notable variation in nutritional composition among the finger millet landraces, including dietary fibre content (Figure 1). The F4 landrace exhibited the highest dietary fibre content (14.88 ± 0.61%), while the F8 landrace exhibited the least (11.17 ± 1.25%). However, Protein content in landrace F4 exhibited a greater amount, 10.13 ± 0.12%, and F6 landrace exhibited the least amount, 5.59 ± 0.08%. Fat content in landrace F1 exhibited a higher amount, 1.22 ± 0.29%, and the least amount was exhibited in landrace F2, 0.81 ± 0.03%. Moisture and ash contents were maximum in F7 (15.60 ± 0.13%) and F6 (3.44 ± 0.06%), respectively, whereas F1 exhibited the least for both

moisture (11.12 ± 0.06%) and ash content (2.47 ± 0.03%). Carbohydrate content in landrace F1 exhibited a higher amount (64.84 ± 1.41%) and F4 landrace exhibited the least amount (56.77 ± 0.86%).

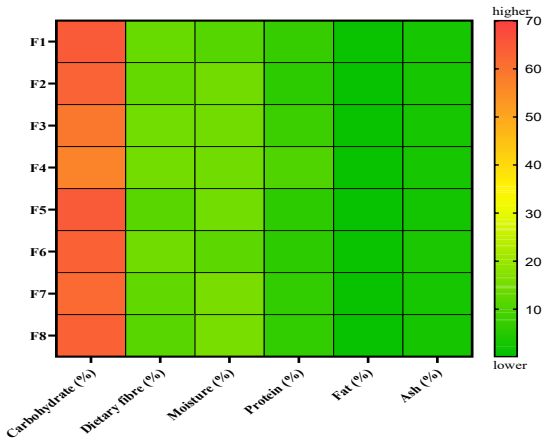


Figure 1: Heatmap showing proximal nutritional composition across landraces. Nutritional content (%) includes carbohydrate, dietary fibre, moisture, protein, fat, and ash. Dark green indicates lower levels, yellow represents moderate levels, and reddish colour represents higher levels of these values.

Our findings revealed considerable variability in the nutritional composition of the evaluated finger millet landraces, with F4 having higher dietary fibre and protein content compared to the other landraces. Such differences are consistent with earlier reports (Kumari et al., 2017; Gebreyohannes et al., 2025) and may be due to genetic composition, geographic effects, and environmental interactions. Genetic factors play a particularly important role, as demonstrated by Babu et al. (2014), who identified several quantitative trait loci (QTLs) associated with agronomic traits in finger millet. Specifically, two QTLs, OM5 and FM8, have been linked to tryptophan content, while another QTL, FMO2EST1, was associated with protein content. These findings highlight how specific allelic variations can influence amino acid composition and protein synthesis, possibly explaining the superior protein content observed in F4. Furthermore, Kumari et al. (2017) observed significant variation in the nutritional content among 11 finger millet varieties. Specifically, the protein content ranged from 6.92 to 9.14 g/100g, crude fat ranged from 0.10 to 0.84 g/100g, and moisture content ranged from 9.7% to 12.4%. Similar variations were also recorded by others (Onipe & Ramashia, 2022; Jorapur et al., 2024; Panwar et al., 2024). The relatively low carbohydrate content and high protein and fibre content of F4 landrace (Figure 1) emphasize the need to identify and promote nutritionally superior landraces for human consumption.

3.3. Mineral content analysis

We observed variation in mineral composition in all eight finger millet landraces (Figure 2). Potassium (K) content in landrace F1 exhibited a relatively higher amount (1776.22 ± 0.57 mg/kg), and F5 landrace exhibited the least amount (1061.15 ± 0.4mg/kg). Similarly, Ca, Mg, Zn, Fe, Mn, Cu and Ni content were recorded high in landraces F5 (1477.73 ± 0.37 mg/kg), F4 (480.83 ± 0.68 mg/kg), F4 (7.41 ± 0.20 mg/kg), F8 (121.38 ± 0.23 mg/kg), F2 (137.59 ± 0.57 mg/kg), F7 (2.19 ± 0.006 mg/kg), and F6 (1.59 ± 0.05 mg/kg), respectively (Figure 2).

This study highlights the detection of essential minerals via ICP-MS, particularly K, Ca, Mg, Zn, Fe, Cu, and Ni. The mineral content identified in this research is consistent with earlier findings, although the levels differ among landraces (Backiyalakshmi et al., 2024; Jayawardana et al., 2019; Onipe & Ramashia, 2022). In general, the recurring evidence of finger millet's mineral richness across various studies underscores its importance as a nutrient-rich cereal.

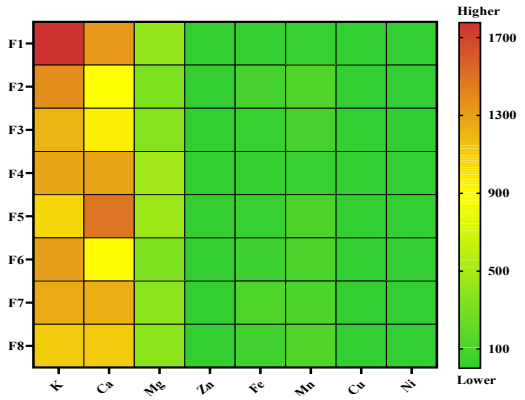


Figure 2: Heatmap showing variations in mineral content (mg/kg). Green indicates lower levels, yellow represents moderate levels, and reddish signifies higher values. The values are represented as the mean ± SD (n = 2).

The noticeable variation in mineral composition among the evaluated finger millet landraces and the higher Ca and K content, indicate a strong influence of both genetic and environmental factors. Finger millet is renowned for its substantial genotypic diversity, a fact supported by previous studies. For instance, Mirza et al. (2014) identified crucial Ca transport genes such as CAX1 and TPC1, which may partially account for the increased Ca accumulation observed in F5 landrace. Additionally, variations in soil characteristics and agronomic practices likely contributed to the observed mineral variability by affecting nutrient availability and uptake efficiency. Results

The current research focused mainly on antimicrobial resistance, the virulence genes of *E. coli* isolates from dead chickens in Dinajpur district, Bangladesh, and the significant public health hazard posed by resistance-carrying E. coli strains. Our study's central hypothesis was to identify the prevalence of MDR E. coli in different organs of poultry, with implications for public health.

3.4. Qualitative screening of carbohydrate and phytochemicals

The presence of carbohydrates and phytochemicals in finger millet landraces, such as phenolics, flavonoids, tannins, alkaloids, coumarins, terpenoids, glycosides, quinones, and saponins, plays an essential role in human health. Therefore, qualitative screening of these phytochemicals was performed in all eight landraces (Figure 3 and Table 3). The carbohydrate content was found to be high across the landraces. Several of these phytochemicals were present in both AE and ME, and a few phytochemicals were absent either in both the solvents or in one of the solvents. Detection of several of these phytochemicals suggests the potential health benefits of these landraces. Phenolic compounds, primarily found in fruit and vegetables, exhibit anti-inflammatory, antioxidant, and anti-proliferative properties that can help inhibit disease progression (Obaid et al., 2024). Tannins enhance glucose absorption and may aid in treating non-insulin-dependent diabetes while also possessing antioxidant, antimicrobial, and anticancer activities (Kumari & Jain, 2015). Alkaloids exhibit a range of therapeutic properties, including anti-inflammatory, anti-cancer, antigenotoxic, antidiabetic, antinociceptive, and antiparasitic effects (Borsoi et al., 2024). Coumarin has anti-inflammatory, antinociceptive, antidiabetic, antioxidant, and anti-fibrotic qualities (Aydın et al., 2024). Terpenes have antibacterial properties against both antibiotic-sensitive and

antibiotic-resistant bacteria, affecting cell activity and reducing protein and DNA synthesis (Siddiqui et al., 2024). Glycosidic compounds offer numerous benefits, including antidiabetic, anti-inflammatory, anti-fatigue, anti-allergic, anti-ageing, anti-skin glycation, analgesic, wound-healing, and aphrodisiac effects (Saha et al., 2024).

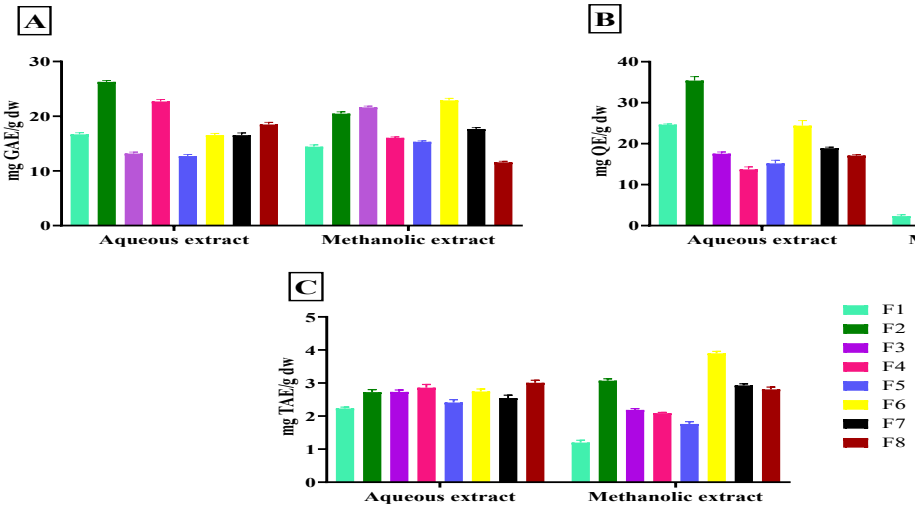


Figure 3: Bar diagrams showing quantitative estimation of phytochemicals in aqueous extract (AE) and methanolic extract (ME). (A) Total phenolic content (TPC), (B) Total flavonoid content (TFC), and (C) Total tannin content (TTC). The values are presented as mean ± SD (n = 3).

Table 3: Qualitative screening of carbohydrate and phytochemicals of finger millet landraces.

Phytochemical name	Solvent used for extraction	F1	F2	F3	F4	F5	F6	F7	F8
Carbohydrate	AE	***	***	***	***	***	***	***	***
	ME	***	***	***	***	***	***	***	***
Phenol	AE	**	**	**	*	**	***	**	**
	ME	-	-	-	-	-	-	-	-
Flavonoid	AE	**	**	**	**	**	***	**	***
	ME	*	*	*	*	*	**	*	*
Tannin	AE	**	**	**	**	**	***	**	**
	ME	-	-	-	-	-	-	-	-
Alkaloid	AE	*	-	***	***	*	*	***	***
	ME	-	-	-	-	-	-	-	-
Coumarin	AE	**	***	***	***	**	***	**	***
	ME	**	**	**	**	**	***	***	**
Terpenoid	AE	***	*	**	**	*	**	***	*
	ME	***	***	**	***	***	**	**	***
Glycoside	AE	-	-	-	-	-	-	-	-
	ME	-	-	-	-	-	-	-	-
Quinones	AE	**	**	***	**	**	***	***	**
	ME	**	***	***	**	**	***	***	***
Saponin	AE	-	**	-	**	**	*	**	**
	ME	-	-	-	-	-	-	-	-

Note: Heavily present, ***; Moderately present, **; Present, *; Absent, -

3.5. Quantitative estimation of phytochemicals

3.5.1 Phenolic estimation

This study investigated total phenol content (TPC) in all 8 finger millet landraces. Our observation revealed that the AE of landrace F2 exhibited the highest phenol concentration of 26.03 ± 0.19 mg GAE/g dw, whereas the landrace F5 had the lowest concentration of 12.72 ± 0.22 mg GAE/g dw (Figure 3A). In the ME, F6 showed the highest TPC level (22.89 ± 0.19 mg GAE/g dw), while landrace F8 exhibited the lowest (11.56 ± 0.17 mg GAE/g dw). All TPC results were presented as milligrams of gallic acid equivalent per gram dry weight (mg GAE/g dw).

Considerable variability in TPC was observed across all finger millet landraces, with F2 having the highest levels. In addition to sample composition and extraction efficiency, phenolic accumulation is influenced by genetic variation and environmental factors, including geography, soil composition, and rainfall (Kumari et al., 2017). The phenolic levels identified across the landraces are consistent with previous findings, which also report significant variability in free total phenolic content (TPC), ranging from 1.14 to 1.79 mg FAE/g dw (ferulic acid equivalent per gram dry weight) and bound TPC values between 0.58 to 1.23 mg FAE/g dw. Free and bound TPC were found to be highest in finger millet's brown seed coat (Xiang et al., 2019). Our findings along with other earlier studies (Hithamani and Srinivasan, 2014; Lansakara et al., 2016) have documented wide ranges of TPC among cultivated and local varieties suggesting finger millet's value as a functional food.

3.5.2 Total flavonoid content

The total flavonoid content (TFC) of all eight finger millet landraces was examined, which gave a variable composition of flavonoid content in each finger millet landrace. The highest flavonoid content was observed in AE of F2 landrace with a value of 35.39 ± 0.76 mg QE/g dw, and the lowest in F4 landrace with a value of 13.17 ± 0.34 mg QE/g dw (Figure 3B). In ME, F3 showed the highest value of 7.62 ± 0.30 mg QE/g dw, and F1 had the lowest content of flavonoid (2.31 ± 0.26 mg QE/g dw).. Considerable difference in flavonoid composition was observed among the landraces, with F2 having highest total flavonoid content. This variability aligns with earlier studies, which also reported notable TFC differences among finger millet varieties (Chandrasekara & Shahidi, 2010; Lansakara et al., 2016; Ajiboye et al., 2017; Xiang et al., 2019). Flavonoid levels are influenced by various factors, including genotype, environmental stress, seed coat properties, and phenylpropanoid metabolic pathways (Zhang et al., 2024). The consistently high flavonoid concentrations reported in multiple studies highlight finger millet's potential as a functional grain.

3.5.3 Total tannin content

The total tannin content (TTC) in both AE and ME of eight finger millet landraces was analyzed using the Folin-Ciocalteu method. TTC in the AE of F8 landrace exhibited a higher amount (3.00 ± 0.06 mg TAE/g dw), whereas the F1 landrace had the lowest value (2.23 ± 0.03 mg TAE/g dw) (Figure 3C). In contrast, ME of F6 landrace possessed the highest tannin content (3.89 ± 0.04 mg TAE/g dw), and F1 had the lowest (1.19 ± 0.05 mg TAE/g dw). Typically, tannin concentration varies significantly across extraction methods and landraces. In our study, we found variation in the tannin composition across finger millet landraces. A recent study in Maharashtra, India, found that the white landrace had a tannin content of 13.47 mg/g, although brown-pigmented millet types have been shown to have higher tannin content (Xiang et al., 2019). Research on various finger millet cultivars has demonstrated variations in tannin levels. (Panwar et al., 2016; Abioye et al., 2022). Another study yields similar findings to ours, suggesting that the tannin content in finger millet is highly cultivar-dependent, with condensed tannins in free fractions typically exhibiting variable levels (Xiang et al., 2019).

3.6. Result of *E. coli* serogrouping

Secondary metabolites exhibit antioxidant activities, known for their roles as lipid stabilizers and inhibitors of oxidative processes that contribute to ageing and cancer (Namiki, 1990). Their protective effects manifest through several mechanisms: inhibiting oxidizing enzymes, chelating transition metals, donating hydrogen or electrons to neutralize radicals, deactivating singlet oxygen, and enzymatically detoxifying reactive oxygen species. Given the complexity of antioxidant mechanisms, multiple assays are essential to fully evaluate the antioxidant capacity of extracts (Dykes, L., 2006; Huang et al., 2005). Millets, particularly the pericarp layers of these grains, are rich in polyphenols, such as phenolic acids, flavonoids, and tannins that serve as reducing agents, effectively quenching free radicals, chelating metals, and neutralizing singlet oxygen (Kumar et al., 2021). These polyphenols enhance the nutritional and therapeutic value of millet, providing significant antioxidant benefits that can protect cells from oxidative damage associated with chronic diseases.

3.6.1 Total antioxidant activity (TAA)/ Phosphomolybdenum assay

The total antioxidant activity (TAA) of the eight finger millet landrace extracts was assessed using the phosphomolybdenum method. The analysis revealed significant differences in antioxidant capacity among the millet extracts. In the AE, landrace F1 showed the highest antioxidant activity of 9.85 ± 0.30 mg AAE/g dw, while landrace F3 exhibited the lowest value of 2.30 ± 0.33 mg AAE/g dw. In ME, landrace F6 demonstrated the highest antioxidant capacity of 11.68 ± 0.42 mg AAE/g dw, and landrace F8 had the lowest value of 0.99 ± 0.41 mg AAE/g dw. Additionally, other finger millet landraces displayed moderate levels of antioxidant activity. A summary of each extract's antioxidant capacity is presented in Figure 4.

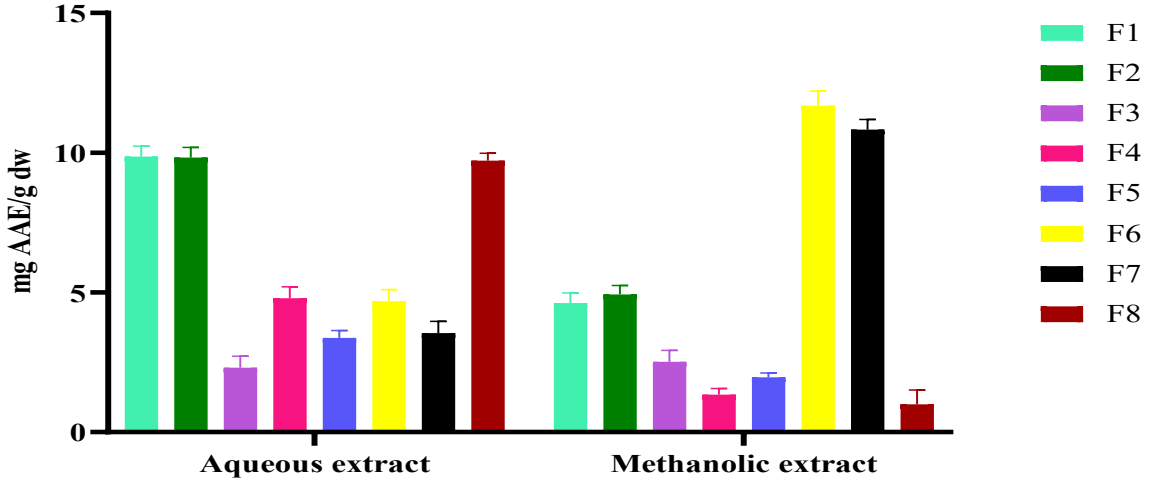


Figure 4: Total antioxidant activity (TAA) of AE and ME of finger millet landraces. The values of TAA are expressed in ascorbic acid equivalents per gram of dry weight (mg AAE/g dw).

The ME generally showed higher antioxidant levels than AE, likely due to methanol's ability to extract a broader range of antioxidant compounds, particularly the phenolics. Previous studies have highlighted the efficiency of methanol in extracting phenolic compounds, which contribute significantly to antioxidant activity (Kazi et al., 2024; Jaiswal et al., 2025). Jaiswal et al. (2025) conducted a comparative analysis of various millets, which reported that finger millet has higher antioxidant activity than other millets, including kodo, proso, barnyard, foxtail, and little millet. Variation in antioxidant levels among landraces can be attributed to differences in polyphenolic profiles, which are likely influenced by both genetic and environmental factors. Antioxidant-rich landraces, such as F1 and F6 (Figure 4), may have potential applications in the food and nutraceutical industries as natural sources of antioxidants. Overall, these findings support the relevance of finger millet as a source of dietary antioxidants. Further research into the specific phenolic compounds responsible for antioxidant activity could provide deeper insights into the health benefits associated with different millets.

3.6.2 Ferric reducing antioxidant power (FRAP) assay

The FRAP examination of finger millet landraces yielded values for both AE and ME, demonstrating considerable variability. In the AE, the highest FRAP activity was observed in landrace F4, with a value of 11.39 ± 0.19 mg AAE/g dw, while the lowest was in landrace F8, with a value of 9.55 ± 0.14 mg AAE/g dw. In contrast, the ME exhibited even more variation, with landrace F5 showing the highest FRAP activity of 18.70 ± 0.35 mg AAE/g dw, and landrace F1 having the lowest value of 8.61 ± 0.18 mg AAE/g dw. Other samples demonstrated intermediate FRAP activities, as shown in Figure 5.

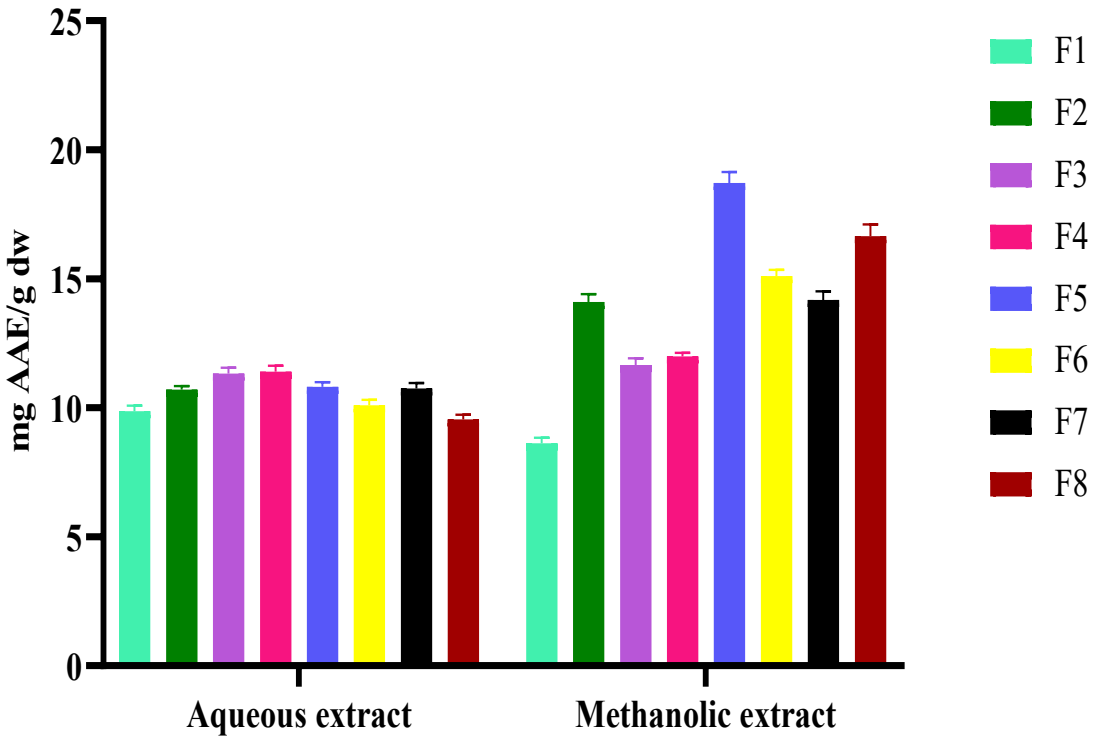


Figure 5: Bar diagram showing ferric reducing antioxidant power (FRAP) of AE and ME of finger millet landraces. The values are represented as mean ± SD (n = 3).

The landraces F4 and F5 exhibited higher FRAP activity, signifying a greater reducing capacity compared to other finger millet landraces. Previous studies have reported a wide range of FRAP values for finger millet. For instance, the whole grain reducing power of finger millet was found to range from 11.1 ± 0.1 to 23.9 ± 0.2 µM AAE/g dw (Kumari et al., 2017). Jayawardana et al. (2018) observed that finger millet exhibited a higher FRAP activity as compared to foxtail millet. Similarly, Semere et al. (2024) reported variable levels of FRAP activity in 12 finger millet landraces. The variability is linked to genotype, seed composition, phenolic and flavonoid content, and environmental growing conditions (Kumari et al., 2017). Studies by Abeysekera et al. (2022) and others also confirm notable differences in FRAP activity among millet and sorghum varieties, emphasizing the influence of specific landraces' traits on antioxidant strength. The pronounced variation observed in our extracts indicates that antioxidant activity is dependent on landraces and chemical nature of bioactive compounds in the grain.

3.6.3 DPPH radical scavenging activity

Our analysis revealed significant differences in DPPH scavenging activity among the millet extracts. The standard drug, ascorbic acid, showed the strongest antioxidant activity, with an IC_{50} value of 76.24 ± 0.44 µg/mL (Figure 6). IC_{50} value represents the concentration of an extract required to inhibit 50% of free radicals, and it is inversely related to antioxidant strength. The lower the IC50 value, the higher the potency; a higher IC50 value indicates weaker radical scavenging activity. The AE of landrace F1 exhibited the most potent scavenging activity with an IC_{50} value of 1058.11 ± 10.5 µg/mL, while landrace F7 showed the weakest activity with an IC_{50} of 2119.49 ± 433.2 µg/mL. Similarly, the ME showed variability in its antioxidant potential. The most potent scavenging activity was recorded for ME of landrace F2, with an IC_{50} value of 655.19 ± 19.85 µg/mL, whereas landrace F1 showed the least potent activity with an IC_{50} of 1820.41 ± 101.2 µg/mL. Other extracts demonstrated intermediate DPPH scavenging activity (Figure 6).

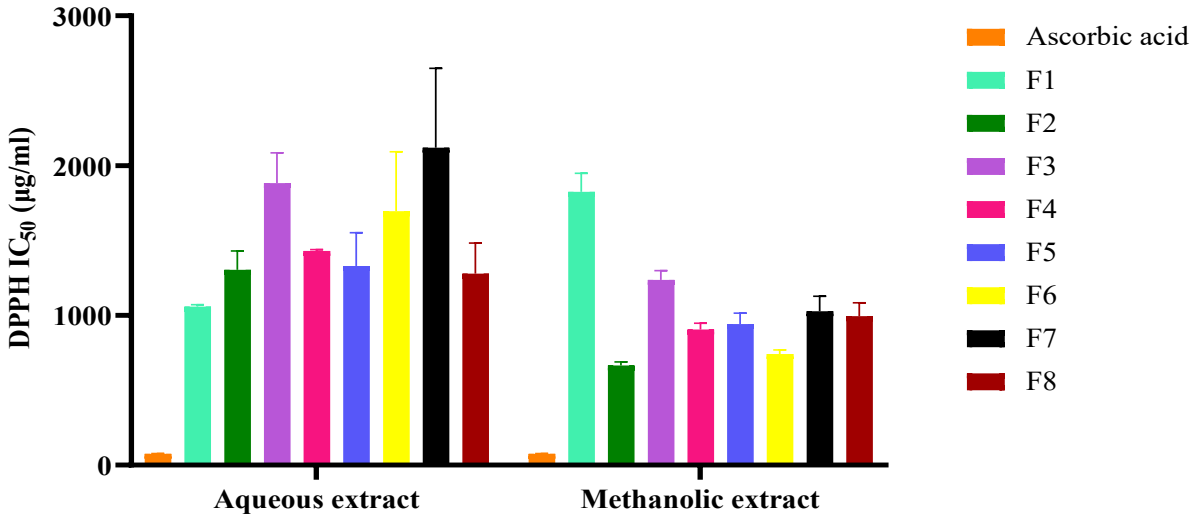


Figure 6: Bar graph showing DPPH radical scavenging activity (IC_{50} µg/mL) of AE and ME of finger millet landraces. The values are represented as mean ± SD (n = 3).

DPPH is a stable synthetic free radical commonly used to assess the radical-scavenging potential of antioxidants in various dietary samples (Pyrzynska & Pekal, 2013). Our findings confirm that F1 and F2 landraces exhibit notable radical scavenging activity, although the scavenging capacity was lower than that of ascorbic acid (Figure 6). Differences in DPPH IC_{50} values across landraces and varieties have been reported in several previous studies (Lansakara et al., 2016; Abeysekera et al., 2017; Ofosu et al., 2020). Kumari et al. (2017) also observed varying DPPH radical scavenging activities depending on the type of extract (methanolic or ethanolic) and highlighted the influence of genotype, grain colour, and phenolic and flavonoid content on antioxidant responses.

3.6.4 ABTS radical scavenging activity

The current study revealed notable differences in ABTS scavenging activity across the millet extracts (Figure 7). The AE of landrace F8 demonstrated the strongest radical scavenging activity with an IC_{50} value of 190.73 ± 2.81 µg/mL, while landrace F3 showed the lowest activity at 380.55 ± 8.62 µg/mL. Among ME, landrace F2 exhibited the highest potency with an IC_{50} value of 251.93 ± 5.42 µg/mL, while landrace F1 had the least activity with an IC_{50} value of 580.93 ± 25.3 µg/mL. Other extracts exhibited intermediate radical-scavenging activity, as shown in Figure 7.

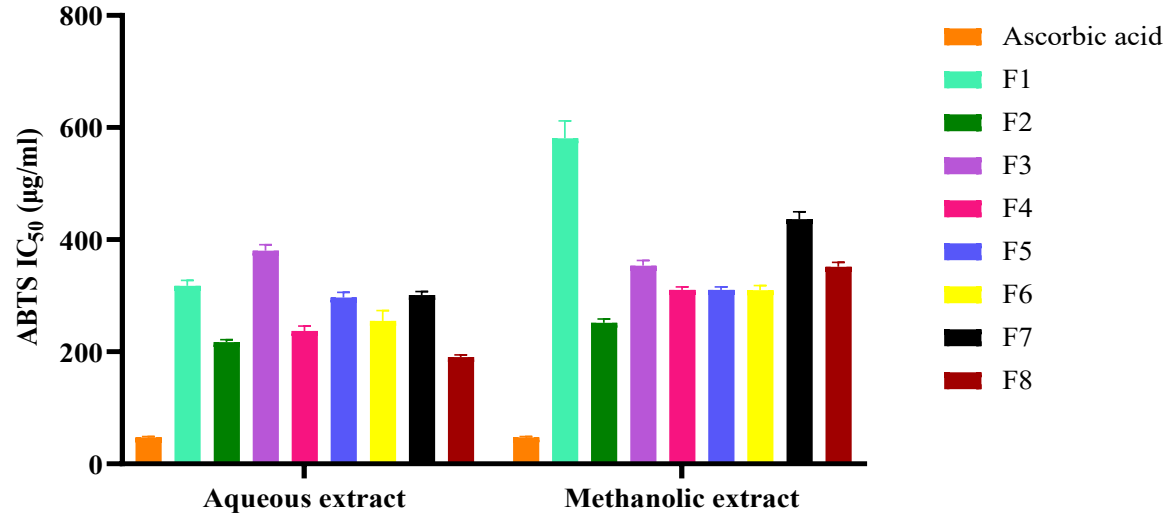


Figure 7: Bar graph showing ABTS radical scavenging activity (IC_{50} µg/mL) of AE and ME of finger millet landraces. The values are represented as mean ± SD (n = 3).

The variation in ABTS scavenging activity across the tested landraces reflects the differences in their phenolic, flavonoid, and tannin composition, which influence electron-transfer and radical-quenching efficiency. The strong ABTS activity in F8 and F2, therefore, suggests a richer presence of electron-donating bioactive compounds, reinforcing their potential as nutritionally valuable landraces with enhanced antioxidant functionality.

3.6.5 Nitric oxide (NO) radical scavenging activity

The antioxidant potential of finger millet seed extracts was also assessed using the NO radical scavenging assay, with ascorbic acid serving as the standard reference. Results were expressed as IC_{50}

values, indicating the concentration required to inhibit 50% of the NO radicals, measured in µg/mL. The standard drug, ascorbic acid, showed the strongest antioxidant activity, with an IC50 of 67.36 ± 0.95 µg/mL. The current analysis revealed considerable differences in NO scavenging activity among the millet extracts (Figure 8). In AE, F1 landrace exhibited the highest radical scavenging activity with an IC_{50} value of 86.03 ± 1.77 µg/mL, whereas F3 landrace showed the lowest activity of 227.07 ± 6.80 µg/mL. In ME, F4 landrace had the highest NO scavenging activity, 94.77 ± 3.71 µg/mL, while F6 landrace had the least potent activity, 172.04 ± 8.47 µg/mL. Other extracts demonstrated intermediate NO scavenging activity, as shown in Figure 8.

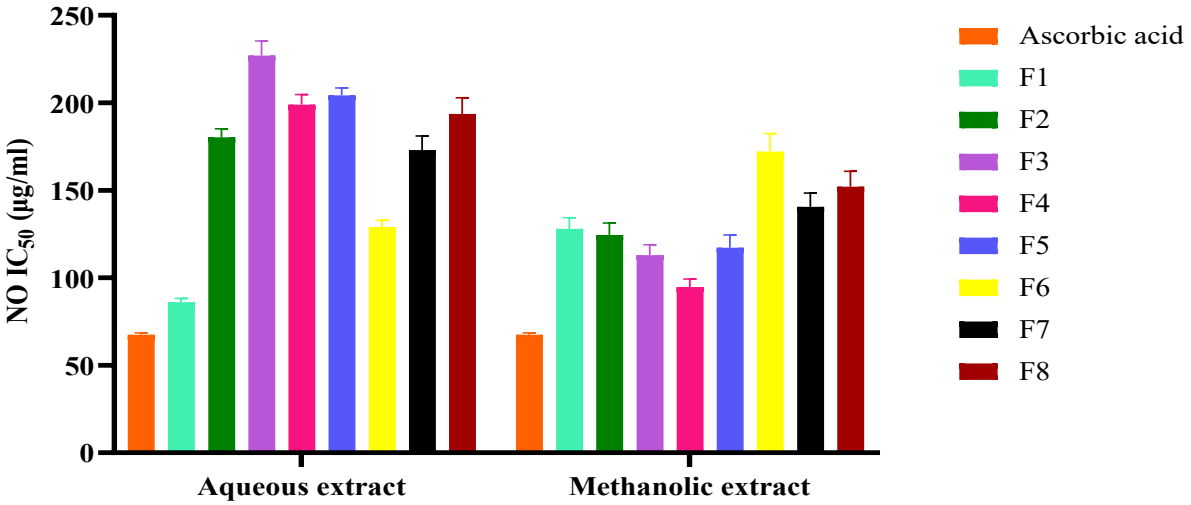


Figure 8: Bar graph showing Nitric oxide (NO) radical scavenging activity (IC_{50} µg/mL) of AE and ME of finger millet landraces. The values are represented as mean ± SD (n = 3).

The NO scavenging assay targets reactive nitrogen species and quantifies the inhibition of NO radical formation by sodium nitroprusside (Marcocci et al., 1994), revealing significant NO scavenging activity in the examined millet extract. The current analysis revealed noticeable differences in NO scavenging activity among the millet extracts (Figure 8). Notably, the F1 and F4 finger millet landraces demonstrated superior NO scavenging capabilities. This finding aligns with other research findings indicating that the use of plant extracts for treating inflammation and related disorders (Mueller et al., 2010). Further supporting our observations, Abeysekera et al. (2022) observed NO inhibitory activities in various millet and sorghum varieties, with the Oshadha variety of finger millet and sweet sorghum showing the highest inhibitory effects. Jayawardana et al. (2021) observed that ME and the ethanolic extract (EE) of the Oshadha and Rawana varieties of finger millet inhibited arachidonate 5-lipoxygenase, with a strong correlation with antioxidant activity, further emphasizing the potential therapeutic benefits of these millet varieties. Collectively, our results highlight the antioxidant properties of millet extracts, suggesting their utility in combating oxidative stress.

Given the different mechanisms of action of antioxidants, we evaluated the antioxidant capacity of finger millet landraces using multiple assays. While each method provides distinct insights, results may vary due to differences in radical type, radical conditions, and antioxidant solubility. Thus, combining these assays provides a more holistic understanding of antioxidant properties. The IC_{50} values for the DPPH, ABTS, and NO assays for finger millet extracts were relatively high compared with those of pure antioxidant standard drugs. This could be due to the complex nature of crude extracts; unlike a purified molecule, crude extracts comprise a mixture of phenolic acids, flavonoids, tannins, and other constituents that can interact and modulate radical-scavenging activity. Overall, the observed differences in IC_{50} values across samples suggest that factors such as the type of extract, the specific millet landrace, and the assay method significantly influence antioxidant activity. Further studies are needed to investigate the mechanisms underlying these variations and to determine the broader health benefits of incorporating finger millet into a balanced diet.

3.7. Conclusions

This study underscores the nutritional richness and functional potential of traditional finger millet landraces from the tribal regions of Chhattisgarh. The key findings regarding nutrients and phytochemicals are summarised in Table 4. The significant variability observed in seed morphology, phytochemical profiles, nutrient composition, and antioxidant activity indicates that these landraces are valuable agricultural resources. The findings show that the F2 landrace stands out for its high total phenolic and flavonoid content and exceptional antioxidant activity. Meanwhile, the F4 landrace, characterized by lower carbohydrate levels and higher protein and dietary fibre content, offers nutritional benefits crucial for managing metabolic disorders. These variations among landraces are likely attributed to environmental conditions, soil characteristics, and agroecological factors inherent to their native habitats. Promoting the consumption of these millet landraces, especially among rural and marginalized populations, can play a significant role in addressing malnutrition and mitigating the risk of non-communicable diseases like diabetes and cancer. Furthermore, the conservation and incorporation of these landraces into mainstream agriculture and food systems are essential for enhancing dietary diversity, supporting sustainable agricultural practices, and ensuring long-term food security. Their distinct nutrient and phytochemical profiles also make them strong candidates for breeding nutritionally superior and climate-resilient millet varieties. Conserving this diversity through in-situ and ex-situ strategies, including a community seed bank, is essential for ensuring its long-term preservation. Overall, this work emphasizes the importance of recognizing and utilizing traditional agricultural resources in the fight against malnutrition and health disparities.

Table 4: A summary of nutritional and phytochemical compositions among 8 finger millet landraces (F1-F8).

Name of the analysis		Performance of landraces (landraces showing highest and lowest contents)
Proximate (%)	Carbohydrate	F1 (64.84 ± 1.41) and F4 (56.77 ± 0.86)
	Dietary fibre	F4 (14.88 ± 0.61) and F8 (11.17 ± 1.25)
	Protein	F4 (10.13 ± 0.12) and F6 (5.59 ± 0.08)
	Fat	F6 (1.18 ± 0.02) and F2 (0.81 ± 0.03)
	Ash	F6 (3.44 ± 0.06), F5 (2.47 ± 0.03)
Mineral content (mg/kg)	Potassium (K)	F1 (1776.22 ± 0.57) and F5 (1061.15 ± 0.4)
	Calcium (Ca)	F5 (1477.73 ± 0.37) and F2 (888.13 ± 0.23)
	Magnesium (Mg)	F4 (480.83 ± 0.68) and F2 (320.06 ± 0.43)
	Zinc (Zn)	F4 (7.41 ± 0.20) and F8 (5.09 ± 0.05)
	Iron (Fe)	F7 (121.38 ± 0.23) and F1 (13.75 ± 0.1)
	Manganese (Mn)	F2 (137.59 ± 0.57) and F1 (23.88 ± 0.23)
	Copper (Cu)	F7 (2.19 ± 0.006) and F3 (1.22 ± 0.005)
	Nickel (Ni)	F6 (1.59 ± 0.05) and F3 (0.59 ± 0.05)
Phytochemicals	Phenol (mg GAE/g dw)	F2 (26.03 ± 0.19) and F5 (12.72 ± 0.22)
	Flavonoid (mg QE/g dw)	F2 (35.39 ± 0.76) and F4 (13.17 ± 0.34)
	Tannin (mg TAE/g dw)	F8 (3.00 ± 0.06) and F1 (2.23 ± 0.03)

Declarations

Prior informed consent was obtained from the seed-donor farmers who cultivated the landraces under study. The research was conducted in full adherence to the ethical standards and policies of the authors' institution.

Ethical consideration

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Author's Contribution

AK and SND designed the work. MM, MVM, AM and VR performed the experiments and prepared the draft of the article. SND, AK, and MVM corrected the article. Finally, all authors read and approved the final version of the article.

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Data Availability

All data generated or analysed during this study are included in this published article

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