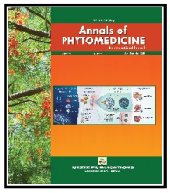
Annals of Phytomedicine 14(2): 466-475, 2025

DOI: http://dx.doi.org/10.54085/ap.2025.14.2.45

Annals of Phytomedicine: An International Journal
http://www.ukaazpublications.com/publications/index.php

Print ISSN : 2278-9839 Online ISSN : 2393-9885



Original Article : Open Access

Variability in secondary metabolites of vegetable cowpea genotypes (*Vigna unguiculata* L.) and their association with nutritional and functional quality

K. Sundharaiya, K. Bharathi Kumar*◆ and M. Kabilan**

Department of Fruit Science, Horticultural College and Research Institute, Periyakulam-625604, Theni, Tamil Nadu, India

** Cotton Research Station,Veppanthattai-621 116, Perambalur, Tamil Nadu, India*

*** Division of Agricultural Chemistry, Regional Coffee Research Station, Coffee Board of India, Thandigudi-624216, Dindigul, Tamil Nadu, India*

Article Info

Article history

Received 2 November 2025

Revised 6 December 2025

Accepted 7 December 2025

Published Online 30 December 2025

Keywords

Vigna unguiculata L.

Secondary metabolites

Phytochemicals

Antioxidant activity

Proximate composition

Mineral content

Functional foods

Abstract

Vegetable cowpea (*Vigna unguiculata* L.) is an important legume vegetable valued for its nutritional richness and functional bioactive compounds. The present study evaluated two prominent vegetable cowpea genotypes, PKMVU 02 and PKMVU 03, to determine genotypic variability in secondary metabolites, proximate composition, carbohydrate profile, mineral concentration and antioxidant activities. Significant differences were observed across all phytochemical parameters, with PKMVU 03 exhibiting notably higher levels of total phenolics (41.86 mg GAE g^{-1} FW), flavonoids (17.45 mg QE g^{-1} FW), tannins, glycosides, β-carotene, lutein and chlorophylls compared to PKMVU 02. Antinutritional factors such as phytic acid and oxalates were considerably lower in PKMVU 03, enhancing mineral bioavailability. Proximate analysis revealed that PKMVU 03 possessed higher protein (23.16%), fibre (4.86%), ash, carbohydrates and energy value (338.80 kcal 100 g^{-1}), indicating superior nutritional quality. Carbohydrate profiling also demonstrated higher soluble sugars, reducing sugars and starch in PKMVU 03, contributing to improved palatability. Mineral analysis showed consistently greater concentrations of Ca, Mg, K, P, Fe, Zn, Cu and Mn in PKMVU 03, reinforcing its nutrient-rich profile. Antioxidant assays including DPPH, ABTS, FRAP and ORAC confirmed significantly stronger antioxidant potential in PKMVU 03 due to its elevated phytochemical content. Overall, the findings identify PKMVU 03 as a superior genotype with enhanced nutritional, functional and antioxidant attributes, making it an ideal candidate for varietal recommendation, dietary improvement and use in crop improvement programs aimed at developing health-promoting vegetable cowpea cultivars.

1. Introduction

Vegetable cowpea (*Vigna unguiculata* L.), commonly known as yardlong bean, asparagus bean, or snake bean, is one of the most important legume vegetables cultivated across tropical and subtropical regions of Asia, Africa and parts of South America (Nwosu *et al*., 2019). It is an agriculturally significant crop due to its remarkable adaptability to diverse climatic conditions, short growth duration, tolerance to heat and drought and its ability to thrive under low-input production systems. The tender, elongated pods are widely consumed as a nutritious vegetable, contributing substantially to dietary protein, essential minerals, vitamins and beneficial bioactive compounds. As global interest shifts toward sustainable, nutrient-dense foods and plant-based diets, vegetable cowpea has gained attention as a promising crop for enhancing food and nutritional security.The tender pods of vegetable cowpea are valued not only for their palatability and culinary versatility but also for their considerable nutritional richness (Bpateng *et al*., 2022). They serve as an excellent source of dietary protein, complex carbohydrates, dietary fiber, vitamins such as vitamin C and folate and minerals including iron, calcium, magnesium, potassium and zinc. In addition, vegetable cowpea contains significant amounts of essential amino acids, making it a vital component of plant-based diets, especially in regions where animal protein is limited or expensive. The crop's nitrogen-fixing ability also contributes to soil fertility, making it an integral part of diversified, climate-resilient agricultural systems. Beyond its basic nutritional profile, vegetable cowpea is increasingly recognized for its rich array of phytochemicals and secondary metabolites (Adeyeye *et al*., 2024). These compounds, which include phenolic acids, flavonoids, tannins, carotenoids, anthocyanins, saponins, alkaloids and various glycosides, are known to contribute substantially to the crop's functional and therapeutic properties. Phenolic compounds such as gallic acid, caffeic acid, ferulic acid and chlorogenic acidare especially abundant in legumes and are well documented for their strong antioxidant potential. Flavonoids like quercetin, kaempferol and catechin are similarly important for regulating oxidative processes and supporting human health. These metabolites play a protective role by scavenging free radicals, reducing cellular oxidative stress and preventing lipid peroxidation (Amoah *et al*., 2023).

Recent studies have highlighted the substantial pharmacological potential of secondary metabolites present in cowpea pods. Phenolics and flavonoids exhibit potent antioxidant, anti-inflammatory and

Corresponding author: Dr. K. Bharathi Kumar
Associate Professor, Cotton Research Station, Veppanthattai-621 116, Perambalur, Tamil Nadu, India
E-mail: bharathisolanum@gmail.com
Tel.: +91-9443534330

antidiabetic activities, making them beneficial for reducing the risk of chronic diseases such as cardiovascular disorders, diabetes, obesity and certain cancers. Carotenoids contribute not only to attractive pod pigmentation but also to vitamin A activity, immune modulation and eye health (Affrifah *et al*., 2025). Anthocyanins, though typically present in lower concentrations in vegetable cowpea compared to pigmented grain legumes, possess notable antioxidant and anti-inflammatory effects. Saponins and tannins, though sometimes considered antinutritional factors, offer potential therapeutic benefits such as cholesterol reduction, immune modulation and antimicrobial activity when present in moderate amounts (Salisu *et al*., 2020). Secondary metabolites also perform indispensable physiological functions in plants. They help regulate plant-environment interactions, contributing to defense against pests, pathogens and abiotic stresses such as drought, heat and salinity. By acting as antioxidants, osmoprotectants, signaling molecules and structural components, phytochemicals improve plant resilience and productivity. Thus, understanding the biochemical composition of vegetable cowpea can provide valuable insights into both plant physiology and the crop's nutritional and functional attributes (Tofade *et al*., 2021).While grain-type cowpea has received considerable attention in phytochemical and nutritional research, studies focusing specifically on vegetable cowpea remain relatively limited. Most available studies have concentrated on general agronomic performance, yield traits, pod morphology and pest resistance, whereas comprehensive investigations into genotypic variability in secondary metabolites and their association with nutritional and functional quality are scarce. Given the increasing global interest in functional foods, nutraceutical crops and plant-based therapeutics, there is a pressing need to explore and characterize the phytochemical diversity present in vegetable cowpea genotypes. Genetic diversity within vegetable cowpea offers an untapped reservoir for crop improvement (Kamara *et al*., 2022). Different genotypes exhibit substantial variation in pod color, pod length, biochemical composition, seed characteristics and stress tolerance. Such diversity provides opportunities to identify superior genotypes that possess high levels of health-promoting phytochemicals and desirable nutritional traits. These elite genotypes can be utilized in breeding programs aimed at enhancing functional quality while maintaining or improving agronomic performance (Rubel *et al*., 2023). Furthermore, understanding the biochemical basis of variability can support the development of improved varieties with enhanced antioxidant capacity, better consumer appeal and increased market value.

Phytochemical variability also influences sensory qualities such as pod color, taste, texture and overall acceptability. For example, higher phenolic content may impart a slightly astringent or bitter taste, while carotenoids contribute to yellowish or green pigmentation. Balancing nutritional and sensory attributes is therefore important for both breeders and the food industry. In addition to secondary metabolites, vegetable cowpea pods contain various macro- and micronutrients essential for overall human health. Proteins and amino acids support growth and tissue repair; dietary fiber aids digestion and helps regulate blood glucose levels; minerals such as iron and calcium contribute to hematological and skeletal functions (Bello *et al*., 2019). Vitamins, particularly vitamin C, enhance immunity and antioxidant status. The combined presence of nutrients and bioactive compounds makes vegetable cowpea a highly valuable functional vegetable capable of supporting public health and combating micronutrient deficiencies in low-income regions. Antioxidant activity, one of the key functional attributes associated with phytochemicals, plays a vital role in mitigating oxidative stress-related diseases. Free radicals generated from metabolic processes or environmental exposures can damage proteins, lipids and DNA, leading to inflammation, early aging and disease development (Ikuerowo *et al*., 2021). Antioxidants such as phenolics, flavonoids and carotenoids neutralize these free radicals, thereby protecting tissues and enhancing physiological resilience. Evaluating antioxidant capacity through multiple assays, such as DPPH, ABTS, FRAP and ORAC, provides comprehensive insights into the biochemical potential of vegetable cowpea pods.

Antinutritional factors, including phytic acid, oxalates, tannins, trypsin inhibitors and lectins, are natural plant compounds that may reduce nutrient bioavailability or alter digestive processes. However, in vegetable cowpea, these compounds are generally present in moderate quantities and often exhibit dual functionalityacting as both antinutrients and health-promoting agents. For instance, phytic acid has antioxidant and anticancer properties, while tannins demonstrate antimicrobial activity. Assessing antinutritional factors is therefore essential for evaluating the overall functional quality and safety of vegetable cowpea as a dietary component (Dzotsi *et al*., 2023). In this context, the present study aims to provide a detailed evaluation of secondary metabolites, nutritional composition, antioxidant activity and antinutritional components across a diverse set of vegetable cowpea genotypes. By integrating phytochemical, nutritional and functional assessments, this research seeks to identify genotypes with superior health-promoting properties and to generate foundational knowledge that can support breeding and food innovation initiatives. The results of this study will contribute to improved utilization of vegetable cowpea in human diets, enhanced crop value chains and the advancement of sustainable agricultural systems that promote both nutritional security and public health.

2. Materials and Methods

2.1 Plant material and experiment site

PKMVU 02: It is a promising vegetable cowpea genotype collected from the Virudhachalam local region of Tamil Nadu. The pods are bold, attractive and visually appealing, making the genotype suitable for fresh market consumption. Its pod characteristics including enhanced pod girth, length and firmness contribute to its desirability as a high-quality vegetable type cowpea. Owing to its superior pod morphology and consumer-preferred appearance, PKMVU 02 holds significant potential for cultivation and varietal development programs aimed at improving vegetable cowpea productivity and market value.

PKMVU 03: It is a vegetable cowpea genotype collected from the Virudhachalam local region of Tamil Nadu. This accession is characterized by its slender, uniformly green pods with a smooth surface and tender texture, making it highly suitable for culinary use. The pods exhibit moderate length with a straight growth habit, ensuring ease of harvesting and reduced post-harvest damage. PKMVU 03 is also noted for its early pod set, consistent flowering pattern and desirable pod tenderness, which contribute to enhanced consumer acceptance. Its fine pod morphology, excellent cooking quality and stable performance under field conditions make PKMVU 03 a valuable genotype for vegetable cowpea improvement programs.

The present study was conducted using two high-performing vegetable cowpea genotypes, PKMVU 02 and PKMVU 03, which are

widely recognized for their superior pod quality, yield performance and adaptability to the agroclimatic conditions of Southern Tamil Nadu. The experiment was carried out at the Department of Vegetable Science, Horticultural College and Research Institute, Periyakulam, Tamil Nadu Agricultural University. Both genotypes were cultivated under field conditions following a randomized block design (RBD) with three replications to ensure experimental precision and minimize environmental variability. Recommended agronomic practices including optimum spacing, nutrient management, irrigation scheduling and plant protection measures were uniformly applied throughout the cropping period. Pods were harvested at commercial maturity, typically 10-12 days after pod set, when they attained ideal tenderness and marketable quality. Fresh pod samples were immediately transported to the laboratory for subsequent phytochemical, nutritional, antioxidant and antinutritional analyses.

2.2 Phytochemical analysis

Fresh pod samples of PKMVU 02 and PKMVU 03 were homogenized using chilled 80% methanol and filtered to obtain clear extracts for phytochemical quantification. All analyses were performed in triplicate and results were expressed on a fresh weight (FW) or dry weight (DW) basis (Bationo *et al*., 2021; Nyathi *et al*., 2020).

2.2.1 Total phenolics

Total phenolic content was estimated using the Folin-Ciocalteu method. 1 g of fresh vegetable cowpea pod tissue was extracted using 10 ml of 80% methanol and centrifuged at 10,000 rpm for 10 min. From the supernatant, 0.5 ml of extract was mixed with 2.5 ml of 10% Folin-Ciocalteu reagent and allowed to react for 5 min. Subsequently, 2.0 ml of 7.5% sodium carbonate solution was added and the mixture was incubated for 30 min at room temperature in the dark. Absorbance was recorded at 765 nm using a UV-Vis spectrophotometer. Total phenolics were quantified using a gallic acid standard curve and calculated using the formula:

$$\text{Total phenolic content} = \frac{C \times V}{W}$$

where, C is the concentration derived from the standard curve (mg ml^{-1}), V is the volume of extract (ml) and W is the sample weight (g FW). Results were expressed as mg gallic acid equivalents per gram fresh weight (mg GAE g^{-1} FW).

2.2.2 Total flavonoids

Total flavonoid content was determined using the aluminium chloride colorimetric assay. To 0.5 ml of the methanolic extract, 1.5 ml of methanol, 0.1 ml of 10% aluminium chloride, 0.1 ml of 1 M potassium acetate and 2.8 ml of distilled water were added sequentially. The mixture was incubated at room temperature for 30 min, after which absorbance was measured at 415 nm. Flavonoid concentration was calculated from a quercetin calibration curve and expressed as mg quercetin equivalents per gram fresh weight (mg QE g^{-1} FW) using the formula:

$$\text{Total flavonoids content} = \frac{C \times V}{W}$$

2.2.3 Tannins

Tannin content was estimated following the vanillin-HCl method. 1 ml of the methanolic extract was mixed with 5 ml of freshly prepared vanillin reagent (4% vanillin in methanol combined with 8% hydrochloric acid). The mixture was incubated for 20 min at room temperature and absorbance was measured at 500 nm. Tannin concentration was computed using a catechin standard curve and expressed as mg catechin equivalents per gram fresh weight (mg CE g^{-1} FW), calculated as:

$$\text{Tannins} = \frac{C \times V}{W}$$

2.2.4 Saponins

Saponin content was determined gravimetrically. 2 g of dried sample were extracted with 20 ml of 20% aqueous ethanol in a water bath maintained at 55°C for 4 h. After filtration, the residue was re-extracted twice under identical conditions and all extracts were combined and concentrated to 40 ml. The concentrate was partitioned using 20 ml diethyl ether to remove lipids and the aqueous phase was further extracted using 60 ml n-butanol. The butanol layer was washed with 5% NaCl solution, evaporated to dryness and the residue was dried at 60°C to constant weight. Saponin percentage was calculated as:

$$\text{Saponins (\%)} = \frac{\text{Weight of residue}}{\text{Weight of sample}} \times 100$$

2.2.5 Alkaloids

Alkaloid content was quantified *via* acid-base precipitation. Two grams of dried sample were treated with 10% acetic acid in ethanol and allowed to stand for 4 h. The mixture was filtered and concentrated to one-fourth of the original volume. Concentrated ammonium hydroxide was added dropwise until complete precipitation occurred and the mixture was allowed to stand for 2 h. The precipitate was filtered, dried at 60°C and weighed. Alkaloid percentage was calculated using:

$$\text{Alkaloids (\%)} = \frac{\text{Weight of alkaloid residue}}{\text{Weight of sample}} \times 100$$

2.2.6 Total glycosides

Total glycoside content was determined using Baljet's method. One millilitre of extract was mixed with an equal volume of Baljet's reagent (picric acid in sodium hydroxide) and kept in the dark for 1 h. Absorbance was recorded at 495 nm and glycoside concentration was estimated using a standard curve. The final value was computed using following formula and expressed as mg g^{-1} DW:

$$\text{Total glycosides content} = \frac{C \times V}{W}$$

2.2.7 Total anthocyanins

Anthocyanin concentration was measured using the pH differential method. Methanolic extracts were diluted separately in potassium chloride buffer (pH 1.0) and sodium acetate buffer (pH 4.5). Absorbance values were recorded at 510 nm and 700 nm for both pH solutions. The pigment concentration was determined using the equation:

$$\text{Total anthocyanin content} = \frac{A \times MW \times DF \times 1000}{D \times 1}$$

$$A = (A_{510} - A_{700})_{pH1.0} - (A_{510} - A_{700})_{pH4.5}$$

where, MW = 449.2 g mol^{-1} (cyanidin-3-glucoside), DF = dilution factor, ε = 26,900 l mol^{-1} cm^{-1}, l = path length (1 cm). Results were expressed as mg C^3G g^{-1} FW.

2.2.8 Total carotenoids (β-carotene and lutein)

Carotenoid fractions were quantified using HPLC. Pigments from 1 g of fresh sample were extracted using acetone:hexane (1:1), filtered through a 0.45 µm membrane and injected into an HPLC system equipped with a C18 column. The mobile phase consisted of acetonitrile:methanol:dichloromethane (70:20:10) and detection was performed at 450 nm. The carotenoid content was calculated as following formula and expressed as mg 100 g^{-1} FW:

$$\text{Total carotenoids} = \frac{C \times V}{W}$$

2.2.9 Chlorophylls (Chl-a and Chl-b)

Chlorophyll pigments were extracted from 0.5 g fresh tissue using 10 ml of 80% acetone. After centrifugation, absorbance of the supernatant was measured at 663 nm and 645 nm. Chlorophyll a, chlorophyll b and total chlorophyll contents were calculated using the following equations and the values were expressed as mg g^{-1} FW:

$$\text{Total chlorophyll} = \text{Chl-a+Chl-b}$$

$$\text{Chl-a} = 12.7\ (A_{663}) - 2.69\ (A_{645})$$

$$\text{Chl-b} = 22.9\ (A_{645}) - 4.68\ (A_{663})$$

2.2.10 Phytic acid

Phytic acid content was estimated using Wade reagent. 1 ml of extract was mixed with 1 ml of Wade reagent (0.03% $FeCl_3$) and 0.3% sulfosalicylic acid). Absorbance was measured at 500 nm and phytic acid concentration was calculated using a standard curve. The final value was computed as following formula and expressed as mg g^{-1}:

$$\text{Phytic acid} = \frac{C \times V}{W}$$

2.2.11 Oxalates

Oxalate content was quantified *via,* $KMnO_4$, titration. One gram of dried sample was ashed at 550°C, extracted with 2N HCl, filtered and diluted to 100 ml. The extract was titrated against 0.05 N potassium permanganate at boiling temperature until a faint pink color persisted for 30 sec. Oxalate content was calculated using:

$$\text{Oxalate (mg / 100 g)} = \frac{T \times N \times 45 \times 100}{W}$$

where, T = titration volume (ml), N = normality of $KMnO_4$, 45 = molecular weight of oxalate, W = sample weight (g).

2.3 Proximate and nutritional composition

Proximate composition of the vegetable cowpea genotypes PKMVU 02 and PKMVU 03 was determined following AOAC (2016) standard methods.

2.3.1 Moisture content

Moisture content was estimated using the oven-drying method, wherein approximately 5 g of fresh pod sample was weighed and placed in a hot-air oven at 105°C until a constant weight was achieved. The moisture percentage was calculated based on weight loss during drying.

2.3.2 Protein content

Protein content was determined using the Kjeldahl method by digesting 1 g of dried sample with concentrated sulfuric acid in the presence of a catalyst mixture (K_2SO_4:$CuSO_4$). The digested samples were distilled using 40% NaOH and titrated with 0.1 N HCl. Nitrogen percentage obtained was converted to crude protein using the conversion factor 6.25 according to the formula:

$$\text{Protein (\%)} = \text{Nitrogen (\%)} \times 6.24$$

2.3.3 Crude fat

Crude fat was measured using Soxhlet extraction. 2 g of dried sample were placed in a thimble and extracted with petroleum ether (60-80°C) for 6 h. After evaporation of the solvent, the residue was weighed and crude fat percentage was calculated.

2.3.4 Crude fibre

Crude fibre was estimated using the AOAC enzymatic-gravimetric method, wherein the sample was sequentially digested with dilute sulfuric acid and sodium hydroxide, filtered, dried and incinerated at 550°C. Fibre content was expressed as the difference between dried residue and ash weight.

2.3.5 Ash content

Ash content was determined by incinerating 2 g of dried sample in a muffle furnace at 550°C for 6 h until a white or grey residue remained. The ash percentage was computed as the ratio of ash weight to the original sample weight.

2.3.6 Carbohydrate profile

Total soluble sugars were determined using the phenol–sulphuric acid method. 1 g of fresh sample was homogenized in 80% ethanol, heated at 80°C for 30 min, filtered and the filtrate was collected. To 1 ml of extract, 1 ml of 5% phenol and 5 ml of concentrated sulfuric acid were added rapidly. After cooling for 20 min, absorbance was measured at 490 nm. A glucose standard curve was used to quantify soluble sugars. Reducing sugars were estimated using the dinitrosalicylic acid (DNS) method. 1 ml of alcoholic extract was mixed with 1 ml of DNS reagent and heated in a boiling water bath for 10 min. After cooling, absorbance was read at 540 nm. Concentration was determined using a glucose standard curve and expressed as mg g^{-1} fresh weight. Starch content was determined using the anthrone method. After ethanol extraction of sugars, the pellet was hydrolysed with 52% perchloric acid for 1 h. The hydrolysate was diluted and 1 ml of the solution was mixed with 4 ml of anthrone reagent (0.2% anthrone in concentrated sulfuric acid). The mixture was heated for 10 min in a boiling water bath, cooled and absorbance was measured at 620 nm. Starch content was calculated using a glucose standard curve and expressed as mg g^{-1} fresh weight.

2.3.7 Energy value

Energy value was calculated using Atwater physiological fuel factors, assigning 4 kcal g^{-1} to protein, 9 kcal g^{-1} to fat and 4 kcal g^{-1} to carbohydrates. The total energy was estimated as:

$$\text{Energy (kcal/100 g)} = (4 \times \text{Protein}) + (9 \times \text{Fat}) + (4 \times \text{Carbohydrates})$$

2.4 Mineral composition

Mineral elements including calcium (Ca), magnesium (Mg), potassium (K), phosphorus (P), iron (Fe), zinc (Zn), copper (Cu) and manganese (Mn) were quantified using Atomic Absorption Spectroscopy (AAS). Initially, 1 g of oven-dried sample was subjected to wet digestion using a mixture of concentrated nitric acid and perchloric acid in the ratio 9:4 (v/v). The digestion was carried out on a hot plate until a clear solution was obtained. The final volume was made up to 50 ml with deionised water and filtered. Samples were aspirated into an AAS instrument calibrated using analytical-grade mineral standards. Absorbance values were recorded at the characteristic wavelengths for each element (Ca - 422.7 nm, Mg - 285.2 nm, Fe - 248.3 nm, Zn - 213.9 nm, Cu - 324.8 nm, Mn - 279.5 nm). Phosphorus was estimated calorimetrically using ammonium molybdate reagent at 660 nm. Mineral concentration was expressed as mg kg^{-1} dry weight using:

$$\text{Mineral concentration (mg kg}^{-1}) = \frac{A \times V}{W}$$

where, A = concentration obtained from calibration curve (mg ml^{-1}), V = final volume (ml) and W = sample weight (kg).

2.5 Antioxidant activity

Antioxidant capacity of the vegetable cowpea extracts was evaluated using four complementary assays: DPPH, ABTS, FRAP and ORAC, providing a comprehensive assessment of free radical scavenging and reducing potential.

2.5.1 DPPH radical scavenging activity

DPPH assay was performed by mixing 1 ml of 0.1 mM DPPH solution in methanol with 1 ml of plant extract at various concentrations. The reaction mixture was incubated in the dark for 30 min and absorbance was measured at 517 nm. Percentage inhibition was calculated using:

$$\text{Inhibition (\%)} = \left(\frac{A_0 - A_1}{A_0} \right) \times 100$$

where, A_0 is absorbance of control and A_1 is absorbance of sample. IC_{50} values were obtained by plotting inhibition versus concentration.

2.5.2 ABTS radical cation decolorization assay

ABTS radicals were generated by reacting 7 mM ABTS solution with 2.45 mM potassium persulfate and incubating for 16 h in the dark. The ABTSz solution was diluted to an absorbance of 0.70 ± 0.02 at 734 nm. One millilitre of diluted ABTSz was mixed with 100 µl of extract, incubated for 6 min and absorbance was measured at 734 nm. Percentage scavenging activity was calculated similarly to DPPH.

2.5.3 Ferric reducing antioxidant power (FRAP)

FRAP reagent was prepared by mixing acetate buffer (pH 3.6), TPTZ (2,4,6-tripyridyl-s-triazine) solution and $FeCl_3$ in a 10:1:1 ratio. In the assay, 100 µl of extract was added to 3 ml of FRAP reagent and incubated for 30 min at 37°C. Absorbance was recorded at 593 nm. Antioxidant power was expressed as ìmol Fe^2z equivalents g^{-1} FW using a $FeSO_4$ standard curve.

2.5.4 Oxygen radical absorbance capacity (ORAC)

ORAC assay was carried out using fluorescein as the fluorescent probe. The reaction mixture contained 150 µl fluorescein, 25 µl plant extract and 25 µl of 153 mM AAPH (radical generator). The decay of fluorescence (excitation 485 nm, emission 535 nm) was monitored every minute for 60 min. Antioxidant capacity was calculated using area under the curve (AUC) and results were expressed as µmol Trolox equivalents g^{-1} FW:

$$\text{ORAC value} = (AUC_{sample} - AUC_{blank}) \times \text{Trolox slope}$$

2.6 Statistical analysis

All experimental data obtained from the phytochemical, nutritional, antioxidant and antinutritional assays were subjected to appropriate statistical analyses to determine the significance of variation between the vegetable cowpea genotypes PKMVU 02 and PKMVU 03. A one-way analysis of variance (ANOVA) was carried out using the statistical software SPSS version 25.0 to test the significance of mean differences at a 5% probability level ($p<0.05$).

3. Results

3.1 Phytochemical analysis

The phytochemical composition of the two vegetable cowpea genotypes exhibited considerable variation (Table 1). Total phenolic content ranged from 32.14 mg GAE g^{-1} FW in PKMVU 02 to 41.86 mg GAE g^{-1} FW in PKMVU 03, indicating significantly higher phenolic accumulation in PKMVU 03. A similar trend was observed for flavonoids, where PKMVU 03 (17.45 mg QE g^{-1} FW) recorded substantially higher levels than PKMVU 02 (11.23 mg QE g^{-1} FW). Tannin content also followed this pattern with PKMVU 03 (3.84 mg CE g^{-1} FW) surpassing PKMVU 02 (2.31 mg CE g^{-1} FW), signifying greater antioxidant potential (Table 1).

Saponin and alkaloid levels showed moderate but meaningful differences. PKMVU 03 recorded 0.47% saponins and 0.34% alkaloids, while PKMVU 02 had lower values of 0.32% and 0.21%, respectively. Total glycoside content was more than 70% higher in PKMVU 03 (4.92 mg g^{-1} DW) compared to PKMVU 02 (2.86 mg g^{-1} DW), indicating enhanced bioactive composition. Among pigment compounds, PKMVU 03 demonstrated higher anthocyanin (1.42 mg C3G g^{-1} FW), β-carotene (4.63 mg 100 g^{-1} FW) and lutein (5.18 mg 100 g^{-1} FW) contents compared to PKMVU 02 (0.84, 2.91 and 3.12 mg, respectively). PKMVU 03 also showed substantially greater chlorophyll accumulation, with 3.12 mg g^{-1} chlorophyll-a, 1.18 mg g^{-1} chlorophyll-b and 4.30 mg g^{-1} total chlorophyll, while PKMVU 02 recorded 1.84, 0.64 and 2.48 mg g^{-1}, respectively. Antinutritional factors such as phytic acid (21.42 mg g^{-1} in PKMVU 02; 14.68 mg g^{-1} in PKMVU 03) and oxalates (6.72 vs. 4.28 mg 100 g^{-1}) were higher in PKMVU 02, suggesting better nutrient bioavailability in PKMVU 03. Overall, PKMVU 03 consistently exhibited higher levels of beneficial phytochemicals while maintaining lower antinutritional contents, indicating superior functional quality.

3.2 Proximate and nutritional composition

Moisture content was slightly higher in PKMVU 03 (87.15%) than PKMVU 02 (84.26%), reflecting its tender pod quality. Protein content varied significantly between genotypes; PKMVU 03 recorded 23.16%, considerably higher than PKMVU 02 (18.42%), making it a richer protein source (Table 2).

Table 1: Phytochemical composition of vegetable cowpea genotypes

Parameter	PKMVU 02	PKMVU 03	SE.d	CV%	ANOVA	CD (5%)
Total phenolics (mg GAE g^{-1} FW)	32.14 ± 1.84	41.86 ± 2.12	1.11	4.9	0.003**	3.62
Total flavonoids (mg QE g^{-1} FW)	11.23 ± 0.62	17.45 ± 0.84	0.39	5.1	0.001**	1.42
Tannins (mg CE g^{-1} FW)	2.31 ± 0.14	3.84 ± 0.21	0.09	6.3	0.002**	0.38
Saponins (% DW)	0.32 ± 0.03	0.47 ± 0.04	0.02	7.8	0.015*	0.08
Alkaloids (% DW)	0.21 ± 0.02	0.34 ± 0.03	0.01	7.9	0.009**	0.06
Total glycosides (mg g^{-1} DW)	2.86 ± 0.18	4.92 ± 0.22	0.12	6.2	0.001**	0.46
Anthocyanins (mg C3G g^{-1} FW)	0.84 ± 0.05	1.42 ± 0.08	0.03	6.7	0.002**	0.16
β-Carotene (mg 100 g^{-1} FW)	2.91 ± 0.18	4.63 ± 0.26	0.11	6.1	0.001**	0.48
Lutein (mg 100 g^{-1} FW)	3.12 ± 0.20	5.18 ± 0.27	0.12	5.8	0.001**	0.52
Chlorophyll-a (mg g^{-1} FW)	1.84 ± 0.12	3.12 ± 0.17	0.07	6.4	0.001**	0.31
Chlorophyll-b (mg g^{-1} FW)	0.64 ± 0.04	1.18 ± 0.07	0.02	6.9	0.003**	0.13
Total chlorophyll (mg g^{-1} FW)	2.48 ± 0.16	4.30 ± 0.22	0.10	6.2	0.001**	0.44
Phytic acid (mg g^{-1})	21.42 ± 1.11	14.68 ± 0.86	0.52	4.8	0.004**	2.12
Oxalates (mg 100 g^{-1})	6.72 ± 0.38	4.28 ± 0.26	0.16	5.6	0.006**	0.74

Table 2:Proximate composition of vegetable cowpea genotypes

Parameter	PKMVU 02	PKMVU 03	SE.d	CV%	ANOVA	CD (5%)
Moisture (%)	84.26 ± 2.84	87.15 ± 3.11	1.12	3.3	NS	4.92
Crude protein (% DW)	18.42 ± 0.92	23.16 ± 1.14	0.48	4.6	0.003**	2.12
Crude fat (% DW)	0.72 ± 0.05	1.11 ± 0.06	0.02	5.7	0.002**	0.14
Crude fibre (% DW)	3.24 ± 0.18	4.86 ± 0.22	0.11	5.3	0.001**	0.46
Ash (% DW)	0.84 ± 0.06	1.22 ± 0.08	0.03	6.4	0.003**	0.18
Total carbohydrates (% DW)	65.78 ± 3.14	69.42 ± 3.28	1.24	4.5	NS	6.82
Energy (kcal 100 g^{-1})	312.50 ± 10.74	338.80 ± 12.16	4.74	3.5	0.015*	21.64

Crude fat content was low in both genotypes, characteristic of vegetable legumes, but PKMVU 03 (1.11%) maintained a marginally higher value than PKMVU 02 (0.72%). Crude fibre content ranged from 3.24% in PKMVU 02 to 4.86% in PKMVU 03, indicating better dietary fibre composition in PKMVU 03. Similarly, ash content, reflecting overall mineral density, was higher in PKMVU 03 (1.22%) than PKMVU 02 (0.84%). Total carbohydrates were slightly higher in PKMVU 03 (69.42%) compared to PKMVU 02 (65.78%). In line with this, energy value derived from macronutrients was greater in PKMVU 03 (338.8 kcal 100 g^{-1}) compared to PKMVU 02 (312.5 kcal 100 g^{-1}). Collectively, PKMVU 03 demonstrated superior proximate composition, contributing to its enhanced nutritional quality.

3.3 Carbohydrate profile

Carbohydrate fractions differed significantly between the two genotypes. PKMVU 03 recorded higher total soluble sugars (5.32 mg g^{-1} FW) than PKMVU 02 (3.14 mg g^{-1} FW) and a similar trend was observed for reducing sugars, where PKMVU 03 exhibited 2.68 mg g^{-1} FW compared to 1.42 mg g^{-1} FW in PKMVU 02. This suggests a sweeter and more palatable pod in PKMVU 03 (Table 3).

Table 3: Carbohydrate profile of vegetable cowpea genotypes

Parameter	PKMVU 02	PKMVU 03	SE.d	CV%	ANOVA	CD (5%)
Total soluble sugars (mg g^{-1} FW)	3.14 ± 0.21	5.32 ± 0.32	0.11	5.8	0.001**	0.52
Reducing sugars (mg g^{-1} FW)	1.42 ± 0.09	2.68 ± 0.16	0.05	6.1	0.002**	0.28
Starch (mg g^{-1} FW)	5.82 ± 0.34	8.41 ± 0.45	0.17	5.5	0.001**	0.73

Starch concentration was also considerably greater in PKMVU 03 (8.41 mg g^{-1} FW) relative to PKMVU 02 (5.82 mg g^{-1} FW), indicating improved carbohydrate storage and higher energy contribution.The overall sugar-starch profile reinforces PKMVU 03 as a genotype with improved taste and nutritional attributes.

3.4 Mineral composition

Mineral analysis revealed that PKMVU 03 consistently outperformed PKMVU 02 across all macro- and micro-minerals. PKMVU 03 recorded higher levels of calcium (325.4 mg kg^{-1} DW), magnesium (244.8 mg kg^{-1}), potassium (3486.3 mg kg^{-1}) and phosphorus (435.8 mg kg^{-1}),

compared to PKMVU 02 (246.2, 182.6, 2845.6 and 412.4 mg kg^{-1}, respectively) (Table 4).

Iron content, a crucial micronutrient for combating anemia, was significantly higher in PKMVU 03 (52.3 mg kg^{-1}) than in PKMVU 02 (36.8 mg kg^{-1}). Zinc and copper levels were also notably higher in PKMVU 03 (24.1 mg kg^{-1} Zn; 8.72 mg kg^{-1} Cu) compared to PKMVU 02 (18.4 mg kg^{-1} Zn; 6.14 mg kg^{-1} Cu). Manganese content followed the same pattern, with PKMVU 03 (17.52 mg kg^{-1}) surpassing PKMVU 02 (12.36 mg kg^{-1}).These results clearly demonstrate the mineral superiority of PKMVU 03, making it a more nutrient-rich option for consumers.

3.5 Antioxidant activity

The antioxidant assays showed significant differences between genotypes, with PKMVU 03 consistently outperforming PKMVU 02 (Table 5).

Table 4: Mineral composition of vegetable cowpea genotypes

Mineral (mg kg^{-1} DW)	PKMVU 02	PKMVU 03	SE.d	CV%	ANOVA	CD (5%)
Calcium	246.2 ± 12.4	325.4 ± 14.2	6.32	4.2	0.002**	28.6
Magnesium	182.6 ± 9.84	244.8 ± 11.2	4.54	4.8	0.003**	23.4
Potassium	2845.6 ± 132.6	3486.3 ± 142.4	58.4	4.6	0.001**	302.5
Phosphorus	412.4 ± 21.2	435.8 ± 22.6	9.34	5.1	NS	48.2
Iron	36.8 ± 2.14	52.3 ± 2.86	1.14	5.3	0.001**	4.98
Zinc	18.4 ± 1.08	24.1 ± 1.42	0.58	5.6	0.003**	2.62
Copper	6.14 ± 0.38	8.72 ± 0.46	0.18	5.2	0.002**	0.84
Manganese	12.36 ± 0.72	17.52 ± 0.84	0.32	5.4	0.001**	1.68

Table 5: Antioxidant activities of vegetable cowpea genotypes

Antioxidant parameter	PKMVU 02	PKMVU 03	SE.d	CV%	ANOVA	CD (5%)
DPPH IC_{50} (µg ml^{-1})	72.42 ± 3.26	49.28 ± 2.42	1.34	4.8	0.001**	6.12
ABTS IC_{50} (µg ml^{-1})	81.16 ± 3.84	57.84 ± 2.86	1.52	4.9	0.001**	7.42
FRAP (µmol $Fe^{2}z$ g^{-1} FW)	6.12 ± 0.32	9.82 ± 0.42	0.14	4.6	0.001**	0.82
ORAC (µmol TE g^{-1} FW)	12.46 ± 0.68	18.42 ± 0.8	0.45	4.3	0.001**	0.78

PKMVU 03 exhibited a lower IC_{50} value (49.28 µg ml^{-1}) compared to PKMVU 02 (72.42 µg ml^{-1}), indicating stronger free radical scavenging efficiency. Similarly, PKMVU 03 recorded a superior ABTS radical scavenging ability with an IC_{50} of 57.84 µg ml^{-1}, compared to 81.16 µg ml^{-1} in PKMVU 02.FRAP values demonstrated a remarkable difference, with PKMVU 03 achieving 9.82 µmol $Fe^{2}z$ g^{-1} FW, while PKMVU 02 recorded only 6.12 µmol $Fe^{2}z$ g^{-1} FW. This signifies better reducing potential in PKMVU 03.ORAC values further confirmed the antioxidant superiority of PKMVU 03 (18.42 µmol TE g^{-1} FW) over PKMVU 02 (12.46 µmol TE g^{-1} FW). The combined results of DPPH, ABTS, FRAP and ORAC show a strong positive relationship between antioxidant activity and the higher phenolic, flavonoid and carotenoid contents of PKMVU 03.

4. Discussion

The present investigation evaluated two vegetable cowpea genotypes, PKMVU 02 and PKMVU 03, for a wide spectrum of secondary metabolites, proximate components, carbohydrate fractions, mineral composition and antioxidant activities. The results revealed clear and consistent genotypic variations, with PKMVU 03 outperforming PKMVU 02 in most biochemical and nutritional parameters. These findings underscore the importance of genotype selection in improving the nutritional and functional quality of vegetable cowpea, a widely consumed legume vegetable in Asia and Africa. The substantial genotypic differences in secondary metabolites observed in the present study confirm the strong influence of genetic makeup on phytochemical accumulation in legumes. PKMVU 03 recorded significantly higher total phenolics (41.86 mg GAE g^{-1} FW) and flavonoids (17.45 mg QE g^{-1} FW) than PKMVU 02 (32.14 and 11.23 mg, respectively). Phenolic compounds play central roles in plant defense, oxidative stress regulation and human health by functioning as antioxidants, anti-inflammatory agents and modulators of metabolic pathways (Abebe *et al.*, 2022). Therefore, higher phenolic and flavonoid levels in PKMVU 03 suggest superior antioxidant potential and greater resilience to abiotic and biotic stress. Tannins, which contribute to both antioxidative and antimicrobial functions, were also more abundant in PKMVU 03 (3.84 mg CE g^{-1} FW) than PKMVU 02 (2.31 mg CE g^{-1} FW). Moderate tannin levels are desirable, as excessive amounts may reduce palatability, but the observed values fall within acceptable consumption limits (Shevkani *et al.*, 2025). Similarly, saponins and alkaloids, which are known for their cholesterol-lowering, anti-inflammatory and cytoprotective effects, were higher in PKMVU 03, indicating greater therapeutic value.

Glycosides showed a pronounced difference between the genotypes, with PKMVU 03 accumulating nearly double the amount found in PKMVU 02. Glycosides in legumes contribute to cardioprotective, antioxidant and antimicrobial properties, suggesting that PKMVU 03 may possess broader biological activities. The carotenoid pigments β-carotene and lutein, essential for vision and immune health, were also higher in PKMVU 03, indicating greater provitamin A potential. Higher β-carotene is particularly desirable for addressing vitamin A

deficiency in populations relying heavily on plant-based diets. Pigment analyses revealed that PKMVU 03 also possessed significantly greater chlorophyll-a and chlorophyll-b, explaining its deep green pod color. Chlorophylls not only influence visual appeal but also possess antioxidant and anti-mutagenic properties. The increased chlorophyll content implies enhanced photosynthetic capacity and potentially higher biomass accumulation (Imade *et al*., 2025). On the contrary, anti-nutritional factors such as phytic acid and oxalates were found in higher amounts in PKMVU 02 than in PKMVU 03. Reductions in these compounds are desirable because phytic acid chelates essential minerals (like iron and zinc) and oxalates interfere with calcium absorption. Therefore, the comparatively lower phytic acid and oxalate concentrations in PKMVU 03 indicate improved mineral bioavailability. Overall, differences in secondary metabolite accumulation between the two genotypes highlight PKMVU 03 as a plant with superior functional food potential.

Proximate composition forms the basis for evaluating the overall nutritional adequacy of vegetable crops. PKMVU 03 recorded higher levels of crude protein, crude fibre, crude fat and ash compared to PKMVU 02, suggesting that it is both nutritionally richer and functionally superior. Protein content is one of the most desirable attributes of legumes and PKMVU 03 contained considerably more protein (23.16%) than PKMVU 02 (18.42%). This difference is nutritionally significant because cowpea serves as a major protein source in many developing regions where animal protein consumption is limited. Higher protein levels in PKMVU 03 thus make it a valuable cultivar for addressing protein-energy malnutrition (Owade*et al*., 2019). Crude fibre content was also markedly greater in PKMVU 03 (4.86%) than in PKMVU 02 (3.24%). Dietary fibre is essential for digestive health, maintenance of gut microbiota and reduction of chronic diseases such as diabetes, obesity and cardiovascular disorders. Fibre-rich vegetables like PKMVU 03 can thus contribute to improved health outcomes. The higher ash content observed in PKMVU 03 (1.22% vs. 0.84%) further supports its richer mineral density, as confirmed by mineral analysis. Total carbohydrate content was slightly, but consistently, higher in PKMVU 03, contributing to its higher calculated energy value (338.8 kcal 100 g^{-1}) compared to PKMVU 02 (312.5 kcal). Moisture content, although not a direct indicator of nutrient quality, affects shelf-life and texture. PKMVU 03 exhibited slightly higher moisture content (87.15%), consistent with its tender pod nature. Collectively, these proximate values indicate that PKMVU 03 possesses a more balanced nutritional profile, making it a better candidate for breeding programs aimed at improving cowpea quality traits.

Carbohydrate analysis revealed that PKMVU 03 recorded higher levels of total soluble sugars (5.32 mg g^{-1} FW), reducing sugars (2.68 mg g^{-} FW) and starch (8.41 mg g^{-1} FW) than PKMVU 02 (3.14, 1.42 and 5.82 mg g^{-1} FW, respectively). Soluble sugars contribute directly to pod sweetness and consumer acceptability. Therefore, the higher sugar levels in PKMVU 03 may impart better sensory qualities, making it more preferable in fresh markets. Reducing sugars and starch content also influence both nutritional and functional qualities (Sombie *et al*., 2018). Higher starch indicates more effective carbohydrate storage, while reducing sugars determine flavor and digestibility. The superior carbohydrate profile of PKMVU 03 suggests that it may be suitable not only for fresh consumption but also for value-added processing industries such as dehydrated vegetables and ready-to-eat food products (Affrifah *et al*., 2022).

Mineral composition is a critical determinant of the micronutrient value of vegetables. PKMVU 03 consistently displayed superior mineral concentrations compared to PKMVU 02, confirming its higher potential for improving micronutrient intake in human diets. The enhanced levels of calcium (325.4 mg kg^{-1}), magnesium (244.8 mg kg^{-1}), potassium (3486.3 mg kg^{-1}) and phosphorus (435.8 mg kg^{-1}) in PKMVU 03 are nutritionally significant. Calcium and magnesium support bone health and enzymatic activities, while potassium plays a vital role in regulating blood pressure and fluid balance. The slightly higher phosphorus concentration further strengthens PKMVU 03's nutritional value. Micronutrient deficiencies, particularly of iron and zinc, are widespread in developing countries. PKMVU 03 exhibited notably higher levels of iron (52.3 mg kg^{-1}) and zinc (24.1 mg kg^{-1}) compared to PKMVU 02 (36.8 and 18.4 mg kg^{-1}, respectively). These differences are nutritionally meaningful because iron is essential for hemoglobin formation and zinc is required for immune function and cell division. Copper and manganese were also present in higher quantities in PKMVU 03. The superior mineral profile of PKMVU 03, combined with its lower antinutritional factors, indicates greater mineral bioavailability and fortifies its status as a nutrient-dense genotype (Murga-Orrillo *et al*., 2024).

Antioxidant capacity reflects the ability of plant extracts to neutralize free radicals and prevent oxidative damage associated with chronic diseases. PKMVU 03 demonstrated significantly higher antioxidant activity across all assays. The DPPH and ABTS radical scavenging assays, both of which measure free radical quenching capacity, showed lower IC_{50} values in PKMVU 03 (49.28 and 57.84 µg ml^{-1}, respectively) compared to PKMVU 02 (72.42 and 81.16 µg ml^{-1}). Lower IC_{50} values denote higher antioxidant potency, suggesting that PKMVU 03 is more effective at neutralizing radicals. FRAP values (9.82 µmol $Fe^{2}z$ g^{-1} FW for PKMVU 03 vs. 6.12 µmol for PKMVU 02) further support this finding by indicating superior ferric-reducing ability. Similarly, the ORAC assay, which measures peroxyl radical scavenging capacity, demonstrated higher antioxidant capacity in PKMVU 03 (18.42 µmol TE g^{-1} FW) compared to PKMVU 02 (12.46 µmol TE g^{-1} FW). These results correlate strongly with the heightened levels of phenolics, flavonoids, tannins, carotenoids and chlorophylls in PKMVU 03. Since antioxidant activity is largely driven by phytochemical composition, the superiority of PKMVU 03 in both biochemical and functional assays is expected. This genotype may therefore provide enhanced protective effects against oxidative stress-related diseases such as diabetes, cardiovascular disorders, obesity and certain cancers (Choi *et al*., 2024).

The combined analysis of all parameters clearly demonstrates that PKMVU 03 is a superior genotype with respect to nutritional quality, functional metabolites and antioxidant potential. Its higher concentrations of dietary protein, fibre, minerals, sugars, pigments and bioactive compounds make it a versatile and health-promoting vegetable. These integrated characteristics suggest the suitability of PKMVU 03 for inclusion in breeding programs targeting high-nutrition

and functional vegetable cowpea varieties. The present study clearly establishes PKMVU 03 as the superior genotype in almost all evaluated parameters. Its higher biochemical, nutritional and antioxidant properties, combined with lower antinutritional factors, make it an excellent candidate for genetic improvement, dietary inclusion and functional food development. PKMVU 02, while nutritionally moderate, may still serve as a suitable cultivar but lacks the enhanced health-promoting attributes of PKMVU 03.

5. Conclusion

The present investigation demonstrated substantial genotypic variability in the phytochemical, nutritional, mineral and antioxidant attributes of the two vegetable cowpea genotypes, PKMVU 02 and PKMVU 03. Across all evaluated parameters, PKMVU 03 consistently exhibited superior performance, characterized by significantly higher levels of total phenolics, flavonoids, tannins, carotenoids, chlorophyll pigments, soluble sugars and essential minerals such as calcium, magnesium, potassium, iron, zinc, copper and manganese. These enhanced biochemical traits directly contributed to its stronger antioxidant capacity, as reflected in the lower IC_{50} values for DPPH and ABTS assays and higher FRAP and ORAC activities. In addition to its rich phytochemical profile, PKMVU 03 also recorded higher protein, fibre, carbohydrate and energy content, alongside lower concentrations of antinutritional factors such as phytic acid and oxalates, thereby improving nutrient bioavailability. Collectively, these findings establish PKMVU 03 as a nutritionally dense and functionally superior genotype with considerable potential for addressing dietary deficiencies and promoting human health. The integrative superiority of PKMVU 03 suggests its value as a promising candidate for varietal recommendation, consumer nutrition, functional food development and as a strong parental line in breeding programs aimed at enhancing nutritional quality in vegetable cowpea. Overall, the study highlights the significance of genotype selection in improving the health-promoting properties of vegetable cowpea and emphasizes the potential of PKMVU 03 as a high-value cultivar for future cultivation and crop improvement initiatives.

Acknowledgements

The authors express their sincere thanks to Horticultural College and Research Institute, TNAU, Periyakulam, for providing the necessary support to carry out this work.

Conflict of interest

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

Reference

Abebe, D. A.; Tolesa, T. B. and Daba, M. K. (2022). A review of the nutritional use of cowpea (*Vigna unguiculata*) and prospects for improvement. Journal of Agriculture and Food Research, **10**:100383.

Adeyeye, I. O.; Aluko, E. M. and Lawal, B. J. (2024). Phenolic composition and antioxidant activities of improved cowpea cultivars grown in contrasting ecologies. Journal of Food Composition and Analysis, **120**:105503.

Affrifah, N. S.; Anang, M. K. and Yeboah-Agyepong, E. (2022). Cowpea: Nutritional profile, processing methods, and potential health implications. Legume Science, **4(3)**:e131.

Affrifah, N. S.; Antwi, V. A. and Ofori-Boateng, M. A. (2025). Modern breeding strategies for improving nutritional quality and abiotic stress tolerance in cowpea. Plant Breeding, **144(1)**:1-18.

Affrifah, N. S.; Atiemo-Obeng, I. Y.; Asante, K. and Essumang, A. (2022). A review of the nutritional use of cowpea (*Vigna unguiculata*): Promises and challenges. Current Research in Food Science, **5**:103-118.

Amoah, H. K.; Nartey, L. B. and Adjei, E. T. (2023). Impact of soil type and cultivation environment on cowpea phytochemical accumulation. South African Journal of Plant and Soil, **40(3)**:249-257.

Bationo, F. G.; Compaoré, M.; Tignégré, J.B. and Kiendrébéogo, M. (2021). Variation in antioxidant capacity and phenolic compounds among cowpea landraces from West Africa. Food Chemistry, **348**:129088.

Bello, M. O.; Olajide, R. A. and Ojediran, J. A. (2019). Effect of boiling and fermentation on cowpea nutritional and antioxidant properties. Asian Journal of Dairy and Food Research, **38(1)**:40-47.

Boateng, E. F.; Asante, K. and Adu-Gyamfi, S. (2022). Impact of genotype and environment interaction on cowpea proximate and mineral composition. Food Research International, **157**:111504.

Choi, Y.-M.; Kim, H.-J.; Lee, M.-G.; Park, J.-H. and Kim, S.Y. (2024). Nutritional qualities, metabolite contents, and antioxidant activities of 14 field-grown yardlong bean accessions. Plants, **13(12)**:1564.

Dzotsi, M. V.; Nyadanu, D. A. and Kusi, F. (2023). Influence of genotype and environment on nutritional traits of cowpea in Ghana. Journal of Plant Nutrition, **46(2)**:250-262.

Ikuerowo, P. O. E.; Chukwuma, M.A. and Eze, C. (2021). Nutritional and bioactive diversity of underutilized cowpea landraces: Implications for breeding. Heliyon, **7(2)**:e06311.

Imade, E. M. and Usunomena, U. (2025). Comparative study of phytochemical composition and antioxidant properties of *Vigna unguiculata* cultivated on mining and non-mining soils in Edo North. Tropical Journal of Phytochemistry and Pharmaceutical Sciences, **4(2)**:73-78.

Kamara, J. M.; Elnasikh, A. M. and Adjei, R. K. (2022). Genotypic variation in cowpea for mineral composition and protein content. Plant Foods for Human Nutrition, **77(4)**:547-556.

Murga-Orrillo, H.; Vásquez-Torres, A.; Huamán-Gutiérrez, Y. and Zúñiga-Dávila, L. (2024). Physiological characterization and bioactive compounds of cowpea accessions under organic mulch conditions. Frontiers in Agronomy, **6**:1392068.

Nwosu, R. N.; Ogbulie, F. O. and Okafor, C. E. (2019). Phytochemical constituents and anti-nutritional factors of cowpea varieties available in Southeastern Nigeria. International Journal of Food Science and Nutrition Engineering, **9(3)**:47-52.

Nyathi, S.; Mlambo, B. and Gasura, E. (2020). Correlating seed coat colour with phenolic content and antioxidant activity in cowpea genotypes. Journal of Food Biochemistry, **44(9)**:e13376.

Olalekan, B. A.; Fadahunsi, O. M. and Okoro, N. E. (2024). Nutritional profiling and bioactive potential of cowpea flour blends for functional food development. Journal of Food Measurement and Characterization, **18(4)**:3012-3024.

Omodara, A. A.; Ogunwolu, K. A. and Adebowale, F. A. (2023). Phytochemical screening and antioxidant activity of improved cowpea varieties under drought stress. Agronomy, **13(7)**:1684.

Owade, J. O.; Abong', C. O. and Wandayi, M. (2019). Cowpea leaves: A review of its nutritive value, processing methods, and potential health

benefits. African Journal of Food, Agriculture, Nutrition and Development, **19(3)**:14772-14789.

Rubel, M.G; Morales, J. P. and Sanchez, L. L. (2023). Functional properties and nutraceutical potential of cowpea proteins and peptides. Food Hydrocolloids, **150**:108270.

Salisu, H. M.; Adeoti, O. A. and Musa, A. A. (2020). Evaluation of nutritional, phytochemical, and antioxidant properties of cowpea varieties consumed in northern Nigeria. Global Journal for Research Analysis, **9(6)**:1-6.

Shevkani, G; Mehra, N.; Singh, J. and Kaur, H. (2025). Cowpeas for sustainable agriculture and nutrition security. Discover Food, **5(1)**:374-382.

Sombié, P. A. E. D.; Compaoré, M.; Coulibaly, A. Y.; Sawadogo, A.; Tignégré, J.-B. D. L. S. and Kiendrébéogo, M. (2018). Antioxidant and phytochemical studies of 31 cowpeas (*Vigna unguiculata* (L. Walp.)) genotypes from Burkina Faso. Foods, **7(9)**:143.

Tofade, S. A.; Olawale, A. J. and Ojo, M. B. (2021). Antioxidant and anti-inflammatory activities of selected cowpea genotypes. Journal of Food Science and Technology, **58**(11):4230-4238.

Citation **K. Sundharaiya, K. Bharathi Kumar and M. Kabilan (2025). Variability in secondary metabolites of vegetable cowpea genotypes (*Vigna unguiculata* L.) and their association with nutritional and functional quality. Ann. Phytomed., 14(2):466-475. http://dx.doi.org/10.54085/ap.2025.14.2.45.**