

Comparative Study of the Proximate, Mineral and Amino Acid Compositions of *Boerhavia Diffusa* and *Boerhavia Erecta*

Akinsola Abiodun Folasade*¹, Oluwafemi Grace Adekemi², Osasona Ilesanmi², Omolayo Akinola³,
Oluwasola Esther Adeola⁴, Awolumate Kehinde Abigeal⁴

¹Department of Industrial Chemistry, Ekiti State University, P. M. B. 5363, Ado-Ekiti, Ekiti State, Nigeria

²Department of Chemical Sciences, Bamidele Olumilua University of Education, Science and Technology, P.M.B. 250, Ikere-Ekiti, Ekiti State, Nigeria

³Department of Chemistry, The Federal University of Technology, P.M.B. 704, Akure, Ondo State, Nigeria

⁴Department of Chemistry, Ekiti State University, P. M. B. 5363, Ado-Ekiti, Ekiti State, Nigeria

DOI: <https://dx.doi.org/10.51244/IJRSI.2026.1305000129>

Received: 06 May 2026; Accepted: 11 May 2026; Published: 03 June 2026

ABSTRACT

This study comparatively evaluated the proximate composition, mineral content, anti-nutrient levels, and amino acid profiles of the edible portions of *Boerhavia diffusa* and *Boerhavia erecta*, two wild edible vegetables commonly consumed in parts of Africa and Asia. Fresh plant samples were collected from Ado Ekiti, Nigeria, air-dried and analyzed using standard analytical procedures. The results showed that carbohydrate was the most abundant proximate component in both species, ranging from 48.49% in *Boerhavia diffusa* to 50.50% in *Boerhavia erecta*. Crude protein contents were relatively high and comparable, with values of 16.13% and 15.79%, respectively. *Boerhavia diffusa* contained higher ash content, whereas *Boerhavia erecta* had higher crude fat and fibre contents. Mineral analysis revealed appreciable levels of essential macro- and microelements, particularly calcium, potassium, phosphorus, iron, and magnesium. Lead was not detected in either sample. The Na/K ratios of both vegetables were below one, suggesting possible dietary benefits for cardiovascular health. Anti-nutrient analysis indicated low concentrations of saponin and alkaloid, while the level of cyanide and tannin in both samples could be reduced by processes such as cooking or boiling. Amino acid profiling showed the presence of both essential and non-essential amino acids in considerable amounts. Glutamic acid was the predominant amino acid in both samples, while leucine was the most abundant essential amino acid. *Boerhavia diffusa* generally recorded higher total amino acid content, essential amino acid index, and predicted protein efficiency ratio than *Boerhavia erecta*. Amino acid scoring patterns further demonstrated that both vegetables could contribute significantly to essential amino acid requirements, especially for children. Overall, the findings indicate that *Boerhavia diffusa* and *Boerhavia erecta* are valuable nutrient-rich leafy vegetables with promising potential for improving dietary quality and supporting food and nutrition security.

Keywords: *Boerhavia diffusa*, *Boerhavia erecta*, proximate, amino acid, mineral

INTRODUCTION

Wild edible plants (WEPs) are uncultivated plant species with edible parts that grow naturally in their native habitat without human intervention. These plants are well adapted to local agro-ecological conditions and often require minimal inputs such as irrigation and synthetic fertilizers (Heywood, 2011), making them suitable for cultivation in marginal environments where other crops may fail.

They are vital sources of food and medicinal remedies that have long been a part of human survival and cultural tradition. They also provide essential nutrients for animal growth, health and productivity (Rumicha *et al.*, 2025). Millions of individuals regularly include WEPs in their diets (Carvalho & Barata, 2016), and more than 7000 species of WEPs have been utilized as food at some point in human history (Agarwal *et al.*, 2025). The exploration of WEPs has gained momentum recently, primarily due to their potential to combat food insecurity and malnutrition in developing regions.

Two *Boerhavia* species, namely *Boerhavia diffusa* Linn. (*B. diffusa*) and *Boerhavia erecta* L. (*B. erecta*), are valuable WEPs found across tropical and subtropical regions of Africa and Asia. Both plants are species of flowering plants which belong to the family Nyctaginaceae (Four o'clock family). Historically, the *B. diffusa* plant has been used both as a food and as an Ayurvedic intervention among Asian and African ethnic populations (Das *et al.*, 2023).

African and Chinese traditional medicinal systems have used *Boerhavia* species for the treatment of different ailments because of their medicinal properties (Patil *et al.*, 2016). *B. erecta* has similar properties to *B. diffusa*, and both are used as diuretics, as well as for the treatment of asthma in moderate doses. Their leaves are prepared in a sauce as vegetables in West and East Africa (Schmelzer & Gurib-Fakim, 2008).

The medicinal properties of these plants have been studied extensively by many scientists (Kumar & Singh, 2026; Chaudhary & Dantu, 2011; Das *et al.*, 2022; Nisha *et al.*, 2018; Nugraha *et al.*, 2015). Furthermore, the nutritional evaluation of *B. diffusa* and *B. erecta* leaves has also been documented by some researchers (Thakur & Pathak, 2016; Ujowundu *et al.*, 2008; Ezeabara & Nwiyi, 2017). Juna Beegum *et al.* (2017) also gave a report on the nutraceutical evaluation of the *B. diffusa* whole plant.

However, a literature search has shown that nutritional information on the comparative analysis of the edible portion of *B. diffusa* and *B. erecta*, commonly used as vegetables, is lacking. This study aims to evaluate the comparative analysis of the proximate composition, mineral, and amino acid contents of *B. diffusa* and *B. erecta* plants. Findings from this research will reveal more about the nutritional value of these vegetables and their potential contribution to dietary needs, especially in regions where plant-based nutrition is predominant.

METHODOLOGY

Sample Collection and Preparation

Fresh *B. diffusa* and *B. erecta* plants were collected from farmland in Ado Ekiti. The plants were authenticated at the herbarium of the Plant Science and Biotechnology Department, Ekiti State University, Ado Ekiti, Nigeria. The assigned voucher numbers of *B. diffusa* and *B. erecta* were UHAE2024089 and UHAE2024092 respectively. The edible parts of the samples were removed, washed with distilled water, and air-dried under the shade for two weeks at room temperature. They were then ground to powder by using a VTCL Excella blender. All the chemicals used were of analytical grade.

Proximate Analysis

The proximate composition of the samples (moisture, ash, crude protein, crude fat, and crude fibre) was determined using standard methods of AOAC International (2012). The carbohydrate content was estimated by subtracting the sum of the percentages of moisture, fat, protein, ash and crude fibre contents from 100.

Mineral Analysis

For mineral analysis, the sample was initially dry-ashed at 550°C to a constant weight. The ash residue of each sample was dissolved in 10 ml of 50% nitric acid solution and diluted with distilled water, making the final volume of 25 ml. The aliquot was used to separately determine the following mineral contents: calcium, magnesium, iron, manganese, zinc, copper and lead using an atomic absorption spectrometer (Buck Scientific 210 VGP) (Kanu *et al.*, 2009). The concentrations of sodium and potassium were determined using a flame

emission photometer (Model 405, Corning, UK), while phosphorus was determined by using the vanadomolybdate spectrophotometric method (Gemede *et al.*, 2016).

Anti-nutrient Determination

Alkaloid Determination

Alkaloid concentration was determined by using the procedure described by Peduruhewa *et al.* (2021). 5 g of the sample was weighed into a 250 ml beaker, and 200 ml of 20% acetic acid in ethanol was added and covered to stand for 4 h. This was filtered, and the extract was concentrated using a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle, and the precipitate was collected by filtration and weighed.

Saponin Determination

Saponin value was estimated by following the report of Okwu & Josiah (2006). 20 g of the sample was dispersed in 200 ml of 20% ethanol. The suspension was heated over a hot water bath for 4 h with continuous stirring at about 55°C. The mixture was filtered, and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over a water bath at about 90 °C. The concentrate was transferred into a 250 ml separatory funnel, and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 ml of n-butanol was added. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation, the samples were dried in the oven to a constant weight, and the saponin content was calculated.

Determination of Tannin Content

500 mg of the sample was taken in a plastic bottle, and 50 ml of distilled water was added. Then it was shaken in a mechanical shaker for 1 h and filtered in a 50 ml volumetric flask made up to the mark. 5 ml of the filtrate was pipetted out into the test tube and mixed with 2 ml of 0.1 M FeCl₃ in 0.1 N HCl and 0.008 M K₄Fe(CN)₆ (potassium ferrocyanide). The absorbance was measured at 120 nm within 10 min (Khan *et al.*, 2011).

Determination of Cyanide Content

The cyanide level was determined as described by Eleazu and Eleazu (2012). 5 g of the sample was dissolved in 50 ml of distilled water and allowed to stay overnight. The sample was filtered, and the filtrate was used for the cyanide determination. To 1 ml of the aqueous extract was added 4 ml of alkaline picrate (obtained by dissolving 1 g of picrate and 5 g of Na₂CO₃ in 200 ml of distilled water), and the whole setup was incubated in a water bath at a temperature of 50 °C for 5 min. The formation of a dark red colour was read spectrophotometrically at 490 nm against a reagent blank which contained 1 ml of distilled water and 4 ml of alkaline picrate solution. The cyanide content of the sample was extrapolated from a standard curve that was prepared by diluting potassium cyanide (KCN) standard (in water, acidified with HCl) to varying concentrations of 0.01 to 0.05 µg ml⁻¹ in 0.01 increments. The cyanide concentration was calculated from the equation of the calibration curve.

Amino Acid Determination

Following the AOAC (2006) method, sample was defatted by weighing about 2 g of sample into the extraction thimble of a Soxhlet apparatus. The fat was extracted with chloroform/methanol mixture (2:1v/v) and the extraction lasted for about 5-6 h.

According to the procedure described by Danko *et al.* (2012), 10 ml 6 N HCl, containing 4% thioglycolic acid was added to an accurately weighed 50 mg of the defatted sample. The solution was incubated for 72 h at 110°C in hermetically closed glass container. An aliquot part of hydrolysate was neutralized by adding of Na₂CO₃ to pH: 2.5-5.0. To 100 µl of the neutralized hydrolyzed sample was added 100 µl of solution of internal standard

Norvaline. The sample was purified by cation-exchange solid-phase extraction. The amino acids in purified solutions were derivatized with ethyl chloroformate. The derivatizing reagent was removed by scavenge with nitrogen. The derivatives of amino acids were dissolved in aliquot part of isooctane and were analyzed by gas chromatography with flame- ionization detector.

For the determination of tryptophan, 10 ml 1M KOH was added to an accurately weighed quantity (50 mg) of the defatted sample. The solution was incubated for 48 h at 110°C in hermetically closed glass container. After the alkaline hydrolysis, the hydrolysate was neutralized to get the pH in the range of 2.5-5.0. 100 µl of solution of internal standard Norvaline was added to 100 µl of the neutralized hydrolysate. The sample was purified by cation-exchange solid-phase extraction. The amino acids in purified solutions was derivatized with ethyl chloroformate. The derivatizing reagent was removed by scavenge with nitrogen. The derivatives of amino acids were dissolved in aliquot part of isooctane and analyzed by gas chromatography with flame-ionization detector

Assessment of Quality Parameters

The following quality parameters were evaluated as stated below:

Amino Acid Scores

The amino acid scores were computed using three different scoring patterns:

- Provisional amino acid scoring pattern: based on the recommendations from FAO/WHO (1973).
- Whole hen's egg pattern: determined for both essential and non-essential amino acids, following the scoring scheme proposed by Paul et al. (1976).
- Pre-school child EAA requirement: derived from the recommended amino acid requirement pattern for pre-school children reported by FAO/WHO/UNU (1985).

Essential Amino Acid Index

The Essential Amino Acid Index (EAAI) was determined using a method previously described in the literature (Nielsen, 2002)

Predicted Protein Efficiency Ratio

The Predicted Protein Efficiency Ratio (P-PER) was estimated using the equation derived by Alsmeyer et al. (1974).

$$\text{P-PER} = - 0.468 + 0.454 (\text{Leu}) - 0.105 (\text{Tyr})$$

where (Leu) and (Tyr) represent the g/100 g protein concentrations of Leucine and Tyrosine, respectively.

Isoelectric Point

The isoelectric point (pI) was calculated as stated by Olaofe and Akintayo (2000).

Other Determinations

Additional parameters evaluated included the concentrations and percentage values of Total Amino Acid (TAA), Total Essential Amino Acid (TEAA), Total Non-Essential Amino Acid (TNEAA), Total Acidic Amino Acid (TAAA), Total Basic Amino Acid (TBAA), Total Neutral Amino Acid (TNAA), Total Sulphur Amino Acid (TSAA), and Total Aromatic Amino Acid (TArAA). The percentage cystine in TSAA (% Cys/TSAA) and the leucine-to-isoleucine (Leu/Ile) ratio were also estimated.

Statistical Analysis

Statistical analysis was performed using the Statistical Package for the Social Sciences (SPSS) Version 25.0 software. Data were presented as means $\pm$ standard deviations. Differences between *B. diffusa* and *B. erecta* were examined using independent samples t-tests. Statistical significance was evaluated using two-tailed tests with an alpha level of $p < 0.05$.

RESULTS

The proximate composition of *B. diffusa* and *B. erecta* is presented in Table 1. Carbohydrate was the most abundant component in both samples, accounting for $48.49 \pm 0.13\%$ in *B. diffusa* and $50.50 \pm 0.17\%$ in *B. erecta*. Crude protein content was relatively high and comparable between the two species, with *B. diffusa* having a value of $16.13 \pm 0.04\%$ while *B. erecta* had a value of $15.79 \pm 0.06\%$. The crude fat and crude fibre contents were higher in *B. erecta* ($5.19 \pm 0.04\%$ and $6.30 \pm 0.01\%$), compared to *B. diffusa* ($3.70 \pm 0.03\%$ and $4.64 \pm 0.06\%$). In contrast, ash content was higher in *B. diffusa* ($15.74 \pm 0.05\%$) than in *B. erecta* ($11.49 \pm 0.04\%$). Moisture content was low and similar in both samples, with values of $11.30 \pm 0.14\%$ and $10.73 \pm 0.05\%$ for *B. diffusa* and *B. erecta*, respectively. The percentage composition for carbohydrate, crude fat, ash, crude fibre and moisture contents recorded for the two samples were all statistically different at $p < 0.05$. However, there was no significant difference in the value of protein contents of the two samples.

Table 1: Proximate composition of *B. diffusa* and *B. erecta*

Parameters (%)	<i>B. diffusa</i>	<i>B. erecta</i>
Moisture	11.30 ± 0.14^a	10.73 ± 0.05^b
Crude Protein	16.13 ± 0.04^a	15.79 ± 0.06^a
Crude Fat	3.70 ± 0.03^b	5.19 ± 0.04^a
Ash	15.74 ± 0.05^a	11.49 ± 0.04^b
Crude Fibre	4.64 ± 0.06^b	6.30 ± 0.01^a
Carbohydrate	48.49 ± 0.13^b	50.50 ± 0.17^a

Values are means of duplicate determination $\pm$ standard deviation.

Means bearing different superscripts within the same row for a given parameter are significantly different ($p < 0.05$).

Mineral Composition of *B. diffusa* and *B. erecta*

The mineral composition of *B. diffusa* and *B. erecta* (mg/100 g) is presented in Table 2. Among the macro-minerals, calcium was the most abundant in *B. diffusa* (821 ± 3 mg/100 g), while potassium had the highest concentration in *B. erecta* (725 ± 0 mg/100 g). Potassium levels were high in both samples, with values of 667 ± 2 mg/100 g in *B. diffusa* and 725 ± 0 mg/100 g in *B. erecta*. Phosphorus was the third most abundant macro mineral, with concentrations of 278 ± 0 mg/100 g and 286 ± 0 mg/100 g in *B. diffusa* and *B. erecta*, respectively.

Magnesium occurred at moderate levels in both samples, ranging from 77.05 ± 0.21 mg/100 g in *B. diffusa* to 98.33 ± 0.18 mg/100 g in *B. erecta*. Sodium was present at relatively low concentrations in both vegetables. For the micro minerals, iron and manganese were the most abundant. *B. erecta* recorded higher values of iron (17.13 ± 0.10 mg/100 g) and manganese (12.66 ± 0.08 mg/100 g) compared to *B. diffusa*, which had 10.63 ± 0.04

mg/100 g and 6.06 ± 0.06 mg/100 g, respectively. Zinc concentrations were similar in both samples, with values of 5.94 ± 0.02 mg/100 g for *B. diffusa* and 5.06 ± 0.02 mg/100 g for *B. erecta*.

Copper had the lowest concentration among the measured minerals in both samples. Lead was not detected in either sample. The sodium-to-potassium (Na/K) ratio was 0.05 for both vegetables, while the calcium-to-phosphorus (Ca/P) ratios were 2.95 for *B. diffusa* and 2.28 for *B. erecta*. All the percentage mineral compositions of the elements analyzed for were found to be statistically different ($p < 0.05$) except lead, which was not detected in both samples.

Table 2: Mineral composition of *B. diffusa* and *B. erecta* (mg/100 g)

Minerals	<i>B. diffusa</i>	<i>B. erecta</i>
Sodium	33.83 ± 0.07^b	38.61 ± 0.01^a
Potassium	667 ± 2^b	725 ± 0^a
Calcium	821 ± 3^a	653 ± 4^b
Magnesium	77.05 ± 0.21^b	98.33 ± 0.18^a
Phosphorus	278 ± 0^b	286 ± 0^a
Iron	10.63 ± 0.04^b	17.13 ± 0.10^a
Manganese	6.06 ± 0.06^b	12.66 ± 0.08^a
Zinc	5.94 ± 0.02^a	5.06 ± 0.02^b
Copper	1.71 ± 0.04^b	2.53 ± 0.11^a
Lead	ND	ND
Na/K ratio	0.05	0.05
Ca/P ratio	2.95	2.28

Values are means of duplicate determination $\pm$ standard deviation.

Means with different superscript for a given parameter in the same row are significantly different ($p < 0.05$).

Anti-nutrients Composition of *B. diffusa* and *B. erecta*

Table 3 shows the anti-nutrient contents of *B. diffusa* and *B. erecta*. Saponin content was identical in *B. diffusa* and *B. erecta* (0.10 mg/g). Tannin was higher in *B. diffusa* (10.36 ± 0.11 mg/g) than in *B. erecta* (8.32 ± 0.02 mg/g). Alkaloid was slightly higher in *B. erecta* (0.14 ± 0.01 mg/g) compared to *B. diffusa* (0.11 ± 0.00 mg/g). Cyanide levels were 7.10 ± 0.42 mg HCN/kg in *B. diffusa* and 6.92 ± 0.74 mg HCN/kg in *B. erecta*. Statistically, there was no significant difference at $p < 0.05$ among the saponin contents of the two samples; however, tannin, alkaloid and cyanide values were all significantly different at $p < 0.05$.

Table 3: Anti-nutrients composition of *B. diffusa* and *B. erecta*

Parameters	<i>B. diffusa</i>	<i>B. erecta</i>
Saponin (mg/g)	0.10 ± 0.00^a	0.10 ± 0.00^a

Tannin (mg/g)	10.36±0.11 ^a	8.32±0.02 ^b
Alkaloid (mg/g)	0.11±0.00 ^b	0.14±0.01 ^a
Cyanide (mg HCN/kg)	7.10±0.42 ^a	6.92±0.74 ^b

Values are means of duplicate determination ± standard deviation.

Means with different superscripts for a given parameter within the same row differ significantly at $p < 0.05$.

Amino Acid Concentration and Quality Parameters of *B. diffusa* and *B. erecta*

The results of the amino acid concentration of *B. diffusa* and *B. erecta* and the quality parameters of the amino acid profiles are shown in Tables 4 and 5, respectively. Glutamic acid was the most abundant amino acid in *B. diffusa* (16.30±0.12 g/100 g cp) and *B. erecta* (12.65±0.17 g/100 g cp), followed by aspartic acid (9.97±0.09 and 10.50±0.10 g/100 g cp, respectively). Among the essential amino acids, leucine had the highest value in both samples (7.26±0.26 g/100 g cp in *B. diffusa* and 6.39±0.11 g/100 g cp in *B. erecta*), while methionine had the lowest (1.14±0.13 and 0.79±0.02 g/100 g cp, respectively). Lysine, phenylalanine, histidine, arginine, isoleucine and tryptophan were higher in *B. diffusa*, whereas threonine was slightly higher in *B. erecta*. For the two vegetable plants, all the amino acids obtained in the present study were significantly different at $p < 0.05$ except glycine, serine, proline, valine, methionine and tyrosine, which are not significantly different.

Table 4: Amino acid composition of *B. diffusa* and *B. erecta* (g/100g crude protein)

Amino acid Profile	<i>B. diffusa</i> (g/100g cp)	<i>B. erecta</i> (g/100g cp)
Glycine (Gly)	5.37±0.07 ^a	4.09±0.19 ^a
Alanine (Ala)	6.17±0.17 ^a	4.46±0.16 ^b
Serine (Ser)	5.16±1.02 ^a	6.27±0.27 ^a
Proline (Pro)	4.86±0.71 ^a	5.03±0.01 ^a
Valine (Val)*	4.55±0.16 ^a	4.77±0.09 ^a
Threonine (Thr)*	4.36±0.22 ^b	5.28±0.08 ^a
Isoleucine (Ile)*	4.49±0.18 ^a	3.38±0.04 ^b
Leucine (Leu)*	7.26±0.26 ^a	6.39±0.11 ^b
Aspartic acid (Asp)	9.97±0.09 ^b	10.50±0.10 ^a
Lysine (Lys)*	5.88±0.15 ^a	4.27±0.18 ^b
Methionine (Met)*	1.14±0.13 ^a	0.79±0.02 ^a
Glutamic acid (Glu)	16.30±0.12 ^a	12.65±0.17 ^b
Phenylalanine (Phe)*	6.88±0.18 ^a	4.14±0.12 ^b
Histidine (His)*	3.94±0.09 ^a	2.44±0.06 ^b

Arginine (Arg)*	6.21±0.39 ^a	4.75±0.17 ^b
Tyrosine (Tyr)	2.34±0.07 ^a	2.41±0.12 ^a
Tryptophan (Trp)*	1.94±0.04 ^a	1.22±0.08 ^b
Cystine (Cys)	1.06±0.01 ^b	2.03±0.18 ^a

Values are means of duplicate determination ± standard deviation.

Means with different superscript for a given parameter in the same row are significantly different ($p < 0.05$).

For the quality parameters of the amino acid profiles (Table 5), the TAA values were 97.88 g/100 g cp for *B. diffusa* and 84.87 g/100 g cp for *B. erecta*. *B. diffusa* recorded higher values than *B. erecta* in TEAA with histidine (46.65 and 37.43); without histidine (42.71 and 34.99); TNEAA (51.23 and 47.44); TNAA (55.58 and 50.26); TAAA (26.27 and 23.15); TBAA (16.03 and 11.46); and TArAA (11.16 and 7.77). *B. erecta* showed higher values for %TNEAA (55.90), %TNAA (59.22), %TAAA (27.28), %TSAA (3.32), %cystine in TSAA (71.99%), and Leu/Ile ratio (1.89) compared to *B. diffusa*. However, *B. diffusa* had higher values than *B. erecta* for P-PER (2.58 and 2.18), EAAI (1.34 and 1.13), and pI (5.63 and 4.77).

Table 5: Quality parameters of the amino acid profiles of *B. diffusa* and *B. erecta*

Parameter	<i>B. diffusa</i>	<i>B. erecta</i>
Total Amino Acid (TAA)	97.88	84.87
Percent total amino acid (%TAA)	100	100
Total non-essential amino acid (TNEAA)	51.23	47.44
Percent total non-essential amino acid (% TNEAA)	52.34	55.90
Total essential amino acid (TEAA) with Histidine	46.65	37.43
Percent total essential amino acid (% TEAA) with Histidine	47.66	44.10
Total essential amino acid (TEAA) without Histidine	42.71	34.99
Percent total essential amino acid (% TEAA) without Histidine	43.64	41.23
Total neutral amino acid (TNAA)	55.58	50.26
Percent total neutral amino acid (% TNAA)	56.79	59.22
Total acidic amino acid (TAAA)	26.27	23.15
Percent total acidic amino acid (% TAAA)	26.84	27.28
Total basic amino acid (TBAA)	16.03	11.46
Percent total basic amino acid (% TBAA)	16.38	13.50
Total sulphur amino acid (TSAA)	2.20	2.82

Percent total sulphur amino acid (% TSAA)	2.25	3.32
Percent cystine in TSAA	48.20	71.99
Total aromatic amino acid (TArAA)	11.16	7.77
Percent total aromatic amino acid (% TArAA)	11.40	9.16
Leu/Ile	1.62	1.89
Calculated isoelectric point (pI)	5.63	4.77
Predicted protein efficiency ratio (P-PER)	2.58	2.18
Essential amino acid index (EAAI)	1.34	1.13

Essential Amino Acid Scores of *B. diffusa* and *B. erecta*

Table 6: Essential amino acid scores of *B. diffusa* and *B. erecta* based on FAO/WHO [1973] standard

Amino acid	Suggested value (mg/g)	Sample score (<i>B. diffusa</i>)	Sample score (<i>B. erecta</i>)
Ile	40	1.12	0.85
Leu	70	1.04	0.91
Lys	55	1.07	0.78
Met+Cys	35	0.63	0.81
Phe+Tyr	60	1.54	1.09
Thr	40	1.09	1.32
Trp	10	1.94	1.22
Val	50	0.91	0.95

Results of the essential amino acid score of *B. diffusa* and *B. erecta* based on FAO/WHO [1973] and the requirements of pre-school children based on FAO/WHO/UNU (1985) scoring patterns are shown in Tables 6 and 7. Based on the FAO/WHO (1973) standard, *B. diffusa* recorded higher scores in Ile (1.12), Leu (1.04), Lys (1.07), Phe+Tyr (1.54), and Trp (1.94), while *B. erecta* showed higher values in Met+Cys (0.81), Thr (1.32), and Val (0.95). Both samples had scores above 1.0 for several amino acids, with *B. diffusa* exceeding the standard in Ile, Leu, Lys, Phe+Tyr, Thr, and Trp, while *B. erecta* exceeded in Phe+Tyr, Thr, and Trp.

For the EAA scores based on the requirements of preschool children's scoring pattern, *B. diffusa* showed higher scores in Ile (1.60), Leu (1.10), Lys (1.01), Phe+Tyr (1.46), Trp (1.76), and His (2.07), while *B. erecta* recorded higher values in Val (1.36), Thr (1.55), and Met+Cys (1.13). Scores above 1.0 were observed in *B. diffusa* for Val, Thr, Ile, Leu, Lys, Phe+Tyr, Trp, and His, and in *B. erecta* for Val, Thr, Ile, Met+Cys, Phe+Tyr, Trp, and His.

Table 7: Essential amino acid scores of *B. diffusa* and *B. erecta* based on requirements of pre-school children scoring pattern (FAO/WHO/UNU, 1985)

Amino acid	Preschool (g/100g)	Sample score (<i>B. diffusa</i>)	Sample score (<i>B. erecta</i>)
Val	3.50	1.30	1.36
Thr	3.40	1.28	1.55
Ile	2.80	1.60	1.21
Leu	6.60	1.10	0.97
Lys	5.80	1.01	0.74
Met + Cys	2.50	0.88	1.13
Phe + Tyr	6.30	1.46	1.04
Trp	1.10	1.76	1.11
His	1.90	2.07	1.28

Amino acid scores of *B. diffusa* and *B. erecta* based on the whole hen's egg scoring pattern (Paul *et al.*, 1976)

The outcome of the amino acid score of *B. diffusa* and *B. erecta* based on the whole hen's egg scoring pattern (Table 8) showed that *B. diffusa* recorded higher scores in Ile (0.80), Leu (0.87), Lys (0.95), Met (0.36), Phe (1.35), Trp (1.08), Gly (1.79), Ala (1.14), Glu (1.36), His (1.64), and Arg (1.02), while *B. erecta* showed higher values in Val (0.64), Thr (1.04), Cys (1.13), Tyr (0.60), Ser (0.79), Pro (1.32), and Asp (0.98). Scores above 1.0 were observed in *B. diffusa* for Phe, Trp, Gly, Ala, Pro, Glu, His, and Arg, and in *B. erecta* for Thr, Cys, Gly, Pro, Glu, and His.

Table 8: Amino acid score of *B. diffusa* and *B. erecta* based on whole hen's egg scoring pattern (Paul *et al.*, 1976)

Amino acid	Whole hen's egg (g/100g)	Sample score (<i>B. diffusa</i>)	Sample score (<i>B. erecta</i>)
Val	7.50	0.61	0.64
Thr	5.10	0.85	1.04
Ile	5.60	0.80	0.60
Leu	8.30	0.87	0.77
Lys	6.20	0.95	0.69
Met	3.20	0.36	0.25
Cys	1.80	0.59	1.13

Phe	5.10	1.35	0.81
Tyr	4.00	0.59	0.60
Trp	1.80	1.08	0.68
Gly	3.00	1.79	1.36
Ala	5.40	1.14	0.83
Ser	7.90	0.65	0.79
Pro	3.80	1.28	1.32
Asp	10.7	0.93	0.98
Glu	12.0	1.36	1.05
His	2.40	1.64	1.02
Arg	6.10	1.02	0.78

DISCUSSION

Proximate analysis is the method that determines the value of macronutrients in food samples (Echebiri *et al.*, 2022). The moisture levels of *B. diffusa* and *B. erecta* were within the range of moisture contents (10.00 and 12.08%) of some Nigerian leafy vegetables (Asaolu *et al.*, 2012) but slightly lower than 13.09% obtained by Juna Beegum *et al.* (2017) for *B. diffusa*. A low moisture level is crucial for storage and preservation because it lowers the chance of microbiological spoilage. Protein is necessary for building the structural components of the human body, such as muscles and organs (Tenyang *et al.*, 2016). It also plays an important role in the immune system (Insel and Turner, 2004). The crude protein values of *B. diffusa* (16.13%) and *B. erecta* (15.79%) recorded in this study were not significantly different. These results were consistent with the report of Ezeabara & Nwiyi (2017), where they obtained 16.41% and 15.89% for *B. diffusa* and *B. erecta* leaves. The high level of protein present in *B. diffusa* and *B. erecta* would contribute to the growth and repair of worn-out tissues and also improve human nutrition.

Fats are the primary structural components of cellular membranes and are also sources of energy (Cena & Calder, 2020). The level of fat present in *B. erecta* and *B. diffusa* was higher than those of *T. occidentalis* (2.12%), *A. hybridus* (2.67%), and *C. olitorius* (2.43%) (Obembe *et al.*, 2025), but lower than the obtainable fat contents of typical oil seeds such as soy beans (28.2%) (Etiosa *et al.*, 2017); groundnuts (46.10%) (Ayoola *et al.*, 2012), and calabash gourd seeds (46.2%) (Akinsola *et al.*, 2023). These vegetables may be beneficial for individuals with obesity and related health conditions because of their low fat contents. Ash content represents the level of inorganic matter and oxides in a sample, serving as a key indicator of its mineral composition. The percentage value of ash was higher in *B. diffusa* (15.74) than in *B. erecta* (11.49). Fibre aids in lowering blood cholesterol level and slows down the absorption of glucose, thereby keeping the blood glucose level in control (Anderson *et al.*, 2009). The crude fibre content of the two vegetables examined varied between 4.64% and 6.30%, with *B. erecta* having a higher significant value. The primary function of carbohydrates in the body is to supply energy (Mudambi & Rajagopal, 2007). Higher carbohydrate content was obtained in the present study for *B. diffusa* and *B. erecta* in comparison with the reported carbohydrate contents of *B. diffusa* (30.90%) (Juna Beegum *et al.*, 2017) and the leaves of *B. diffusa* and *B. erecta* (33.78% and 33.62%) (Ezeabara & Nwiyi, 2017). The amount of carbohydrate content in leafy vegetables can vary with prevailing environmental conditions.

The results of the mineral composition of *B. diffusa* and *B. erecta* (Table 2) revealed that both samples contained appreciable amounts of essential macro- and micro-minerals. The two vegetables contained high concentrations of calcium and potassium, which suggests that they can contribute significantly to meeting daily mineral requirements. Calcium is believed to regulate mitochondrial oxidative phosphorylation, thereby contributing to the maintenance of cellular energy homeostasis (Glancy *et al.*, 2013). Potassium performs a vital role in cellular functions, including maintaining fluid balance and osmolality in cells (McLean and Wang, 2021). In the level of concentration, phosphorus came third in both samples. Phosphorus is an essential mineral needed for cell structure, signalling, energy transfer, and other important functions (Sabuj *et al.*, 2021). Magnesium, an essential cofactor for several phosphorylation-related enzymes (Van Niekerk *et al.*, 2018), was present at moderate concentration in both samples. In the two samples, sodium concentrations were notably lower than those of potassium. Sodium is an essential nutrient involved in the maintenance of normal cellular homeostasis and in the regulation of fluid and electrolyte balance and blood pressure (Seldin & Giebisch, 1990). Iron and manganese had higher concentrations in *B. erecta* than in *B. diffusa*. Iron is crucial for red blood cell formation and numerous cellular reactions (Falodun *et al.*, 2010). Manganese is essential for numerous vital processes, including nerve and brain development and cognitive functioning (Balachandran *et al.*, 2020). The difference in the level of zinc present in the two samples was not high. Zinc participates in regulating numerous immune processes, including immune cell activation and differentiation, cytokine production, and the overall maintenance of immune cell function (Stefanache *et al.*, 2023). Zinc deficiency produces hair loss and hypochromic anaemia (Shenkin, 2008).

Copper, which had the least concentration in both samples, is an essential micronutrient required for proper metabolic function and normal tissue development (Petruzzielli *et al.*, 2026). The Na/K and the Ca/P ratios are nutritionally important parameters. The Na/K ratio obtained in the present study for *B. diffusa* and *B. erecta* was 0.05. A sodium-to-potassium (Na/K) ratio of less than one (< 1) is associated with a reduced risk of cardiovascular disease (Averill *et al.*, 2019). The lower sodium content in both samples would be beneficial, especially considering the established link between sodium intake and hypertension in humans. The calcium-to-phosphorus (Ca/P) ratios were 2.95 for *B. diffusa* and 2.28 for *B. erecta*, respectively. The Ca/P ratio exceeds 0.5, which is the minimum required for efficient calcium absorption in the intestine to support bone formation (Adeyeye *et al.*, 2014).

Anti-nutrients are natural or synthetic compounds that interfere with the absorption of nutrients (Egielewa *et al.*, 2021). Some of these compounds act as anti-nutrients for human consumption because of their adverse effects on the palatability, digestibility, and bioavailability of nutrients (Kumar *et al.*, 2022). Tannin was present at a concentration of 10.36 mg/g and 8.32 mg/g in *B. diffusa* and *B. erecta*. Daily intake of tannin below the range of 1.5–2.5 g is safe for consumption but the consumption beyond this range is responsible for low absorption of iron from diet (Rao & Prabhavathi, 1982). Tannins can also cause growth depression by decreasing the digestibility of protein and carbohydrates (Vadivel & Janardhanan 2005). These vegetables should be subjected to processing such as cooking or boiling before consumption to reduce the level of tannin. Alkaloid was detected at a low level of 0.11 mg/g in *B. diffusa* and 0.14 mg/g in *B. erecta*. Alkaloids are premeditated to be anti-nutrients due to their effect on the nervous system, hindering or erroneously enhancing electrochemical transmission (Salim *et al.*, 2023). Saponin content was relatively low in both samples. Saponins have been reported to inhibit proteolytic enzymes, such as trypsin and chymotrypsin, which play a crucial role in breaking down dietary proteins into absorbable amino acids (da Silva Magedans *et al.*, 2021). The cyanide content of *B. diffusa* and *B. erecta* was far lesser than the concentration range of 1480 and 2179 mg HCN/kg obtained by Umuhozariho *et al.* (2014) for the fresh leaves of three cassava species. Moreover, our result was below the WHO recommended safe level of 10 mg of HCN/kg body weight (FAO/WHO, 1991). Consumption of high dietary cyanide has been linked with a number of chronic health disorders (Nhassico *et al.*, 2008; CCDN, 2007). However, Ezeabara & Nwiyi (2017) reported lower cyanide values for *B. diffusa* (2.24 mg/kg) and *B. erecta* (2.17 mg/kg) leaves. The variation in these results may be as result of environmental factors, location, season, and soil types (Adejoh *et al.*, 2020). Research has shown that effective food processing techniques such as soaking, fermentation, cooking, and autoclaving can significantly reduce the concentrations of these anti-nutrients (Okaiyeto *et al.*, 2025).

Amino acids are the major nitrogen-containing compounds of plants and are also the building blocks of proteins (Lea & Azevedo, 2003). They are key precursors for syntheses of hormones and low-molecular weight

nitrogenous substances (Takahashi *et al.*, 2011). The amino acid composition and the quality parameters of *B. diffusa* and *B. erecta* (Tables 4 and 5) revealed the presence of both essential and non-essential amino acids, although their concentrations varied between the two samples. Among the non-essential amino acids, glutamic acid was the most abundant in both *B. diffusa* (16.30 g/100 g cp) and *B. erecta* (12.65 g/100 g cp), followed by aspartic acid with values of 9.97 and 10.50 g/100 g cp, respectively. The high levels of glutamic and aspartic acids suggest their importance in nitrogen metabolism and protein synthesis. Glycine, alanine, serine, and proline were also present in appreciable amounts.

Essential amino acids (EAAs) are amino acids that must be obtained from the diet or supplements (Tsomele *et al.*, 2023). Among the EAAs, leucine (7.26 g/100 g cp), phenylalanine (6.88 g/100 g cp), and arginine (6.21 g/100 g cp) were present in appreciable amounts in *B. diffusa*, while leucine (6.39 g/100 g cp), threonine (5.28 g/100 g cp), and arginine (4.75 g/100 g cp) were detected in *B. erecta*. Leucine, the dominant EAA in this study, also dominates the EAAs in pumpkin leaf (5.95 g/100 g), spinach (3.57 g/100 g), bitter leaf (5.11 g/100 g), and water leaf (2.97 g/100 g) (Arowora *et al.*, 2017). However, it is worthy of note that our samples had a higher concentration of this EAA than in the aforementioned vegetable leaves. In *B. diffusa*, methionine (1.14 g/100 g cp) and cystine (1.06 g/100 g cp) were present in lower quantities, indicating they may be limiting amino acids, while tryptophan (1.22 g/100 g cp) and methionine (0.79 g/100 g cp) had the least concentration in *B. erecta*.

From the summary of the quality parameters of the amino acid profiles, it was observed that *B. diffusa* had a higher TAA (97.88 g/100 g cp) when compared with *B. erecta* (84.87 g/100 g cp), suggesting a richer protein quality. The TAA of *B. diffusa* and *B. erecta* were better than those found in *S. oleracea* (75.56 g/100 g cp), *G. latifolium* (73.93 g/100 g cp), and *P. guineense* (73.93 g/100 g cp) (Aremu *et al.*, 2024). The concentration of the total non-essential amino acid (TNEAA) was higher than the total essential amino acid (TEAA) in both samples. The TEAA with histidine of *B. diffusa* (46.65 g/100 g cp) was of higher value than the obtainable value (37.43 g/100 g cp) for *B. erecta* in this study and those of *G. latifolium* (40.53 g/100 g cp), *S. oleracea* (39.86 g/100 g cp) and *P. guineense* (38.67 g/100 g cp) (Aremu *et al.*, 2024), an implication that *B. diffusa* would be a better source of essential amino acids. The TSAA, which comprises the sum of the concentrations of methionine and cystine, was 2.20 in *B. diffusa* and 2.82 in *B. erecta*, respectively. A similar result was recorded for *P. guineense* (2.05), *S. oleracea* (2.38) and *G. latifolium* (1.98) (Aremu *et al.*, 2024). The recommended range of aromatic amino acids for infant protein (6.8–11.8 g/100 g crude protein) (FAO/WHO/UNU, 1985) was met by the total aromatic amino acid (TArAA) content of *B. diffusa* (11.16 g/100 g cp) and *B. erecta* (7.77 g/100 g cp).

In *B. diffusa* and *B. erecta*, % TEAA with histidine were 47.66 and 44.10, while % TEAA without histidine were 43.64 and 41.23, respectively. The %TNAA varied from 56.79 (*B. diffusa*) to 59.22 (*B. erecta*), suggesting that these made up the majority of the amino acids in the plant samples. The %TAAA which ranged from 26.84 to 27.28 in *B. diffusa* and *B. erecta*, was lower than the %TNAA but greater than the %TBAA, which ranged from 16.38 to 13.50 in the samples. The %cystine in TSAA were 48.20 and 71.99 for *B. diffusa* and *B. erecta*, respectively, implying a higher percentage of cystine in *B. erecta*. Cystine has positive effects on mineral absorption, especially zinc (Mendoza, 2002).

B. diffusa and *B. erecta* had Leu/Ile ratios of 1.62 and 1.89. These values were higher than 1.49 obtained by Akinsola *et al.* (2021) for *B. patula* leaves. The calculated isoelectric point (pI) obtained in this study for *B. diffusa* and *B. erecta* showed that the samples were in the acidic medium of the pH range. The pI of any organic matter is important when the protein isolate is to be prepared (Adeyeye, 2010). With the predicted protein efficiency ratio (P-PER) values of 2.58 and 2.18 for *B. diffusa* and *B. erecta*, it can be inferred that the proteins from these vegetables would be of good quality because a protein whose protein efficiency ratio (PER) is below 1.5 is considered of poor quality; a PER between 1.5 and 2.0 indicates intermediate quality; and a PER above 2.0 signifies a good-quality protein (Benjamin *et al.* (2011); Friedman (1996)). However, the result obtained in the present study was based on the equation derived by Alsmeyer *et al.* (1974) using only leucine and tryrosine and not on rat bioassay. The EAAI of the two samples was better than the recorded EAAI of *C. adansonii* leaves, 1.09 (Akinsola *et al.*, 2022), and the seeds of *B. eurycoma* and *P. guineense*, with 0.664 and 0.571, respectively (Ajayi *et al.*, 2014).

The EAA scores based on FAO/WHO [1973] scoring pattern revealed that EAAs were present in appreciable amounts in *B. diffusa* when compared with *B. erecta*. Sample scores of Trp, Phe + Tyr, Ile, Thr, Lys and Leu in *B. diffusa* all exceeded 100%, while Val and Met + Cys scores were below 100%. However, *B. erecta* recorded values exceeding 100% only for Thr, Trp, and Phe + Tyr, while Val, Leu, Ile, Met + Cys, and Lys scored above 50% but less than 100% when compared with the FAO/WHO standard. Furthermore, the EAA with the highest score in *B. diffusa* was Trp (1.94), while Thr (1.32) was highest in *B. erecta*. Tryptophan is the precursor to serotonin, a brain neurotransmitter, platelet-clotting factor and neurohormone found in organs throughout the body (Braverman, 2003). Threonine is an important bioactive molecule that has vital mediation effects on protein synthesis, energy metabolism, and nutrient absorption (Chen *et al.*, 2017; Estalkhizir *et al.*, 2013). The limiting amino acid in *B. diffusa* was Met + Cys, which had the least score. However, Lys was the limiting amino acid recorded in *B. erecta*.

The EAA scores of *B. diffusa* and *B. erecta* based on the amino acid requirement recommended for preschool children (Table 7) revealed that His (2.07) and Trp (1.76) had the highest scores in *B. diffusa*, while Thr (1.55) and Val (1.36) had the highest scores in *B. erecta*. Histidine is a vital amino acid associated with various metabolic functions, such as histamine production, which is associated with allergic and inflammatory reactions (Salman *et al.*, 2021). Infants require tryptophan for growth as well as for the production and maintenance of the body's proteins, muscles, enzymes, and neurotransmitters (Dinesh *et al.*, 2015). Valine promotes mental vigour, muscle coordination and calm emotions (Zarkadas *et al.*, 1997). Met+Cys (0.88) was the limiting amino acid in *B. diffusa*, while Lys (0.74) was the limiting amino acids in *B. erecta*. *B. diffusa* would be able to supply more than the required EAA for the pre-school child as shown by EAA scores of His, Trp, Ile, Phe+Tyr, Val, Thr, Leu, and Lys, while Met+Cys would supply less than 100% of the requirement. Moreover, for *B. erecta*, the EAA scores for the preschool child indicated that Thr, Val, His, Ile, Met+Cys, Trp and Phe+Tyr would be able to provide more than the necessary amount for the preschool child, while Leu and Lys would both provide less than 100%.

The amino acid score (AAS) based on the whole hen's egg scoring pattern showed that eight amino acids, namely Gly, His, Glu, Phe, Pro, Ala, Trp and Arg, had better scores in *B. diffusa* when compared with the whole hen's egg score. However, only the scores obtained for six amino acids (Gly, Pro, Cys, Glu, Thr, and His) in *B. erecta* exceeded the reference values established for whole hen's egg. Gly showed the highest scores in the two samples, specifically 1.79 in *B. diffusa* and 1.36 in *B. erecta*. Glycine plays a role in diabetes. It is a secretagogue of glucagon-like peptide-1 (GLP-1) (Gameiro *et al.*, 2005), insulin, and glucagon (Gonzalez-Ortiz *et al.*, 2001). The amino acid with the least score in both samples was Met with 0.36 and 0.25 in *B. diffusa* and *B. erecta* respectively. Methionine is crucial for several biological functions, including protein synthesis, cell proliferation, and protection against oxidative stress (Lv *et al.*, 2025)

Limitation of the Study

A key limitation of this study is that the vegetable samples were collected from a single location within the country. This restricts the ability to generalize the proximate, mineral, and amino acid composition results across different regions, as environmental factors such as soil type, climate, and seasonal variation can significantly influence nutrient composition. A broader sampling approach involving multiple geographic locations and seasons would provide a more comprehensive and reliable assessment of the nutritional profile of these vegetables.

CONCLUSION

The present study demonstrated that both *B. diffusa* and *B. erecta* possess appreciable nutritional value and can serve as important dietary sources of nutrients. The two vegetables contained substantial amounts of carbohydrates, proteins, essential minerals, and amino acids. Anti-nutrient analysis indicated low concentrations of saponin and alkaloid. However, the level of cyanide and tannin obtained in both samples could be reduced by processes such as cooking or boiling. *B. diffusa* showed comparatively higher ash content, total amino acids, essential amino acid index, and predicted protein efficiency ratio, suggesting superior protein quality. On the other hand, *B. erecta* contained higher crude fibre, fat, iron, manganese, and sulphur-containing amino acids.

The favourable Na/K and Ca/P ratios observed in both vegetables further support their nutritional importance, particularly in relation to cardiovascular and bone health. The amino acid score evaluations also revealed that both species can contribute meaningfully to essential amino acid requirements, especially among growing children and populations relying heavily on plant-based diets. These findings highlight the nutritional potential of these underutilized wild edible plants and support their wider dietary inclusion and possible domestication as affordable nutrient sources for improving food and nutrition security.

REFERENCES

1. Adejoh, J., Idoko, F. E., Ujah, E. A., & Okoh, M. P. (2020). Cyanide content analysis in selected food crops (soya beans, maize, white beans), nuts (ground nut, tiger nut) cultivated and consumed in Nigeria and, espouse of environmental toxicants as epigene (s) modifier. *Chemical Science & Engineering Research*, 2(4), 26-33. <https://doi.org/10.36686/Ariviyal.CSER.2020.02.04.017>.
2. Adeyeye, E. I., Oyarekua, M. A., & Adesina, A. J. (2014). Proximate, mineral, amino acid composition and mineral safety index of *Callinectes latimanus*. *International Journal of Development Research*, 4(12), 2641-2649.
3. Adeyeye, E. I. (2010). Effect of cooking and roasting on the amino acid composition of raw groundnut (*Arachis hypogaea*) seeds. *Acta Scientiarum Polonorum Technologia Alimentaria*, 9(2), 201-216.
4. Agarwal, V., Singh, S., & Datta, R. (Eds.). (2025). Exploring Traditional Wild Edible Plants. CRC Press, Taylor & Francis Group. Boca Raton.
5. Ajayi O. B., Akomolafe S. F & Adefioye A. (2014). Proximate analysis, mineral contents, amino acid composition, anti-nutrients and phytochemical screening of *Brachystegia eurycoma* Harms and *Pipper guineense* Schum and Thonn. *American Journal of Food and Nutrition*, 2(1), 11-17. DOI: [10.12691/ajfn-2-1-3](https://doi.org/10.12691/ajfn-2-1-3)
6. Akinsola, A. F., Olatunde, O. C., Osasona, I., Sekayo, O. F., & Omotayo, F. O. (2021). Nutritional evaluation of *Brillantaisia patula* leaves. *Asian Plant Research Journal*, 8(4), 63-73. DOI: 10.9734/aprj/2021/v8i430186.
7. Akinsola, A. F., Osasona, I., Akintayo, E. T., Siyanbola, T. O., & Omosebi, S. O. (2023). Nutritional evaluation of calabash gourd (*Lagenaria siceraria*) seeds and oil. *Journal of Culinary Science & Technology*, 21(5), 737-758. DOI: [10.1080/15428052.2021.2016527](https://doi.org/10.1080/15428052.2021.2016527)
8. Akinsola, A. F., Osasona, I., Ojo, C. O., Awe, A., & Omolayo, P. E. (2022). Phytochemical screening, proximate and amino acid compositions of two edible indigenous leafy vegetables from Ekiti State, Nigeria. *International Journal of New Technology and Research*, 8(7), 5-13. <https://doi.org/10.31871/IJNTR.8.7.21>
9. Alsmeyer, R. H., Cunningham, A. E., & Happich, M. L. (1974). Equations to predict PER from amino acid analysis. *Food Technology*, 2, 24-38
10. Anderson, J. W., Baird, P., Davis Jr, R. H., Ferreri, S., Knudtson, M., Koraym, A., ... & Williams, C. L. (2009). Health benefits of dietary fiber. *Nutrition reviews*, 67(4), 188-205. DOI: [10.1111/j.1753-4887.2009.00189.x](https://doi.org/10.1111/j.1753-4887.2009.00189.x)
11. AOAC (2006). Official Methods of Analysis, 18th edition. Association of Official Analytical Chemists, Washington DC.
12. AOAC (2012). Official methods of analysis of AOAC International (19th ed.), Gaithersburg, M.D. USA
13. Aremu, M. O., Osonwa, N. V., Omotehinwa, F., Aremu, S. O., Ortutu, S. C., Jibrin, M., & Muhammad, H. I. (2024). Comparative studies on the nutritional composition of three commonly sold leafy vegetables in Lafia modern market, Nasarawa State, Nigeria. *Lafia Journal of Scientific and Industrial Research*, 2 (1), 40-46.
14. Arowora, K. A., Ezeonu, C. S., Imo, C., & Nkaa, C. G. (2017). Protein levels and amino acids composition in some leaf vegetables sold at Wukari in Taraba State, Nigeria. *International Journal of Biological Sciences and Applications*, 4(2), 19-24.
15. Asaolu, S. S., Adefemi, O. S., Oyakilome, I. G., Ajibulu, K. E., & Asaolu, M. F. (2012). Proximate and mineral composition of Nigerian leafy vegetables. *Journal of Food Research*, 1(3), 214.

16. Ayoola, P. B., Adeyeye, A., & Onawumi, O. O. (2012). Chemical evaluation of food value of groundnut (Arachi hypogaea) seeds. *American Journal of Food and Nutrition*, 2(3), 55-57. doi:10.5251/ajfn.2012.2.3.55.57
17. Balachandran, R.C., Mukhopadhyay, S., McBride, D., Veevers, J., Harrison, F.E...& Bowman, A. B. (2020). Brain manganese and the balance between essential roles and neurotoxicity. *Journal of Biological Chemistry*, 295, 6312-6329. <https://doi.org/10.1074/jbc.rev119.009453>
18. Benjamin, A. A., Ayalogu, O. E. & Onyeike, E.N. (2011). Performance characteristics and organ weight of rats fed “onunu” and “mgbam” traditional diets of the Ikwerre people of Niger Delta, Nigeria”. *Continental J. Food Science and Techn.* 6(1), 12-19. <http://dx.doi.org/10.5707/cjfst.2012.6.1.13.20>
19. Braverman, E. R. (2003). The Healing Nutrients Within: Facts, findings, and new research on amino acids. Basic Health Publications Inc., Laguna Beach, CA.
20. Cassava Cyanide Diseases and Neurolathyrism Network (CCDN) (2007). Working together to eliminate cyanide poisoning, konzo and Tropical Ataxic Neuropathy (TAN). CCDN News. Issue No. 9.
21. Cena, H., & Calder, P. C. (2020). Defining a healthy diet: evidence for the role of contemporary dietary patterns in health and disease. *Nutrients*, 12(2), 334. DOI: 10.3390/nu12020334
22. Chaudhary, G., & Dantu, P. K. (2011). Morphological, phytochemical and pharmacological, studies on Boerhaavia diffusa L. *Journal of Medicinal Plants Research*, 5(11), 2125-2130.
23. Chen, Y.P., Cheng, Y.F., Li, X.H., Yang, W.L., Wen, C., Zhuang, S. & Zhou, Y. M. (2017). Effects of threonine supplementation on the growth performance, immunity, oxidative status, intestinal integrity, and barrier function of broilers at the early age. *Poultry Science*, 96, 405–413. <https://doi.org/10.3382/ps/pew240>
24. da Silva Magedans, Y. V., Phillips, M. A., & Fett-Neto, A. G. (2021). Production of plant bioactive triterpenoid saponins: from metabolites to genes and back. *Phytochemistry Reviews*, 20(2), 461-482.
25. Danka, P. O., Dobrina, D. T. & Kalin, V. I. (2012). Simultaneous identification and determination of total content of amino acids in food supplements-tablets by gas chromatography. *Asian Journal of Pharmaceutical Clinical Research*, 5(2), 57 – 68.
26. Das, S., Sahoo, B. M., & Bhattamisra, S. K. (2022). Multifunctional role of phytochemicals derived from Boerhaavia diffusa L. in human health, ailments, and therapy. *Current Nutrition & Food Science*, 18(6), 574-588. DOI: 10.2174/1573401318666220308141939
27. Das, S., Singh, P. K., Ameeruddin, S., Kumar Bindhani, B., Obaidullah, W. J., Obaidullah, A. J., ... & Mohapatra, R. K. (2023). Ethnomedicinal values of Boerhaavia diffusa L. as a panacea against multiple human ailments: a state of art review. *Frontiers in Chemistry*, 11, 1297300. <https://doi.org/10.3389/fchem.2023.1297300>
28. Dinesh, B., Yadav, B., Reddy, R. D., Padma, A. S., & Sukumaran, M. K. (2015). Determination of ascorbic acid content in some Indian spices. *International Journal of Current Microbiology and Applied Sciences*, 4(8), 864-868.
29. Echebiri U. V., Ugoh C. I., Eze, F. C., Omeje, I. L. & Momoh, B. (2022) Analytical methods in proximate composition of ten commonly used seasonings in Nigeria. *European Journal of Food Science and Technology*, 10(2), 24-34
30. Egielewa, S. J., Odiachi, P. C., & Okodugha, G. O. (2021). Evaluation of nutrient and anti-nutrient of some traditional green leafy vegetables in Edo North, Nigeria. *Applied Science & Technology*, 56, 12 (1), 56-64.
31. Eleazu, C. O. & Eleazu, K. C. (2012). Determination of the proximate composition, total carotenoid, reducing sugars and residual cyanide levels of flours of 6 new yellow and white cassava (Manihot esculenta Crantz) varieties. *American Journal of Food Technology*, 7, 642-649. DOI: 10.3923/ajft.2012.642.649.
32. Estalkhzir, F.M., Khojasteh, S. & Jafari, M. (2013). The effect of different levels of threonine on performance and carcass characteristics of broiler chickens. *Journal of Novel Applied Sciences*, 9, 382–386.
33. Etiosa, O. R., Chika, N. B., & Benedicta, A. (2017). Mineral and proximate composition of soya bean. *Asian Journal of Physical and Chemical Sciences*, 4(3), 1-6. DOI: 10.9734/AJOPACS/2017/38530
34. Ezeabara, C. A., & Nwiyi, U. C. (2017). Investigation of phytochemical and proximate components in

- different parts of *Boerhavia diffusa* L. and *B. erecta* L. *Advances in Applied Sciences*, 2(5), 60-3. doi: [10.11648/j.aas.20170205.11](https://doi.org/10.11648/j.aas.20170205.11)
35. Falodun, A., Uzoekwe, A. S., & Shengxiang, Q. (2010). Phytochemical, anticancer and antioxidant evaluation of potential chemical constituents of *Calliandria Surinamensis*. *Nigerian Journal of Biotechnology*, 21, 55-59.
 36. FAO/WHO (1973). Energy and protein requirements, Technical Report Series No 522, WHO, Geneva.
 37. FAO/WHO/UNU (1985). Energy and protein requirements: Report of a Joint FAO/WHO/UNU Expert Consultation, WHO Technical Report Series, No. 724. Geneva: WHO.
 38. FAO/WHO (1991). Food Standard programmes. Cordex Alimentaris Commission XII supplement 4 FAO Rome Italy.
 39. Friedman, M. (1996). Nutritional value of proteins from different foods sources. A review. *Journal of Agricultural and Food Chemistry*, 44, 6-29. <http://dx.doi.org/10.1021/jf9400167>
 40. Gameiro, A., Reimann, F., Habib, A. M., O'malley, D., Williams, L., Simpson, A. K., & Gribble, F. M. (2005). The neurotransmitters glycine and GABA stimulate glucagon-like peptide-1 release from the GLUtag cell line. *The Journal of Physiology*, 569(3), 761-772. doi: [10.1113/jphysiol.2005.098962](https://doi.org/10.1113/jphysiol.2005.098962)
 41. Gemedede, H. F., Haki, G. D., Beyene, F., Woldegiorgis, A. Z., & Rakshit, S. K. (2016). Proximate, mineral, and antinutrient compositions of indigenous Okra (*Abelmoschus esculentus*) pod accessions: implications for mineral bioavailability. *Food Science & Nutrition*, 4(2), 223-233. <https://doi.org/10.1002/fsn3.282>
 42. Glancy, B., Willis, W. T., Chess, D. J., & Balaban, R. S. (2013). Effect of calcium on the oxidative phosphorylation cascade in skeletal muscle mitochondria. *Biochemistry*, 52(16), 2793–2809. <https://doi.org/10.1021/bi3015983>
 43. González-Ortiz, M., Medina-Santillan, R., Martinez-Abundis, E., & von Drateln, C. R. (2001). Effect of glycine on insulin secretion and action in healthy first-degree relatives of type 2 diabetes mellitus patients. *Hormone and Metabolic Research*, 33(06), 358-360. doi: [10.1055/s-2001-15421](https://doi.org/10.1055/s-2001-15421).
 44. Heywood, V. H. (2011). Ethnopharmacology, food production, nutrition, and biodiversity conservation: towards a sustainable future for indigenous peoples. *Journal of Ethnopharmacology*, 137, 1–15. doi: [10.1016/j.jep.2011.05.027](https://doi.org/10.1016/j.jep.2011.05.027)
 45. Insel, P. M., Turner, R. E., & Ross, D. (2004). Nutrition. 2nd Pkg Edition.
 46. Juna Beegum, G.R., Sugunan, V. S., & Beevy, S. S. (2017). Neutraceutical evaluation of *Boerhavia diffusa* L. *International Journal of Current Pharmaceutical Research*, 9(1), 101-104. DOI: <http://dx.doi.org/10.22159/ijcpr.2017v9i1.16626>
 47. Khan, A. M., Qureshi, R. A., Ullah, F., Gilani, S. A., Nosheen, A., Sahreen, S., ... & Murad, W. (2011). Phytochemical analysis of selected medicinal plants of Margalla Hills and surroundings. *Journal of Medicinal Plants Research*, 5(25), 6017-6023.
 48. Kumar, S., & Singh, B. (2026). *Boerhavia diffusa* L.: Integrative traditional uses, phytochemistry, pharmacological properties, and quality control/quality assurance. *Chemistry & Biodiversity*, 23(2), e01724. DOI: [10.1002/cbdv.202501724](https://doi.org/10.1002/cbdv.202501724)
 49. Kumar, Y., Basu, S., Goswami, D., Devi, M., Shivhare, U. S., & Vishwakarma, R. K. (2022). Anti-nutritional compounds in pulses: Implications and alleviation methods. *Legume Science*, 4(2), e111. <https://doi.org/10.1002/leg3.111>
 50. Lea, P. J. & Azevedo, R.A. (2003). Primary Products: Amino Acids, Editor (s): Brian Thomas, Encyclopedia of Applied Plant Sciences, Elsevier, 871-883, <https://doi.org/10.1016/B0-12-227050-9/00241-6>.
 51. Lv, Y., Feng, Y., Lu, J., Zhang, S., Wei, Y., & Gao, C. (2025). New insights into methionine's role in maintaining intestinal stem cell regeneration via autophagy regulation. *Animal Nutrition*. DOI: [10.1016/j.aninu.2025.04.008](https://doi.org/10.1016/j.aninu.2025.04.008)
 52. McLean, R. M., & Wang, N. X. (2021). Potassium. *Advances in Food and Nutrition Research*, 96, 89–121. <https://doi.org/10.1016/bs.afnr.2021.02.013>.
 53. Mendoza, C. (2002). Effect of genetically modified low phytic acid plants on mineral absorption. *International Journal of Food Science and Technology*, 37(7), 759-767.
 54. Mudambi, S. R. & Rajagopal, M.V. (2007). Fundamentals of Foods, Nutrition and Diet Therapy. New Age International.

55. Nhassico, D., Muquingue, H., Cliff, J., Cumbana, A. & Bradbury, J. H. (2008). Rising African cassava cyanide intake and control measures. *Journal of the Science of Food and Agriculture*, 88, 2043-2049.
56. Nielsen, S. S. (2002). Introduction to the chemical analysis of foods. CBS Publishers and Distributors, New Delhi.
57. Nisha, M., Vinod, B. N., & Sunil, C. (2018). Evaluation of *Boerhavia erecta* L. for potential antidiabetic and antihyperlipidemic activities in streptozotocin-induced diabetic Wistar rats. *Future Journal of Pharmaceutical Sciences*, 4(2), 150-155. <https://doi.org/10.1016/j.fjps.2017.12.001>
58. Nugraha, A. S., Hilou, A., Vandegraaff, N., Rhodes, D. I., Haritakun, R., & Keller, P. A. (2015). Bioactive glycosides from the African medicinal plant *Boerhavia erecta* L. *Natural Product Research*, 29(20), 1954-1958.
59. Obembe, O. M., Dada C. A. & Adanikin B. A. (2025). Evaluation of the nutritional and antinutritional composition of selected green leafy vegetables consumed in Ekiti State, Nigeria. *International Journal of Life Sciences Research*, 13(2), 32-40. DOI: <https://doi.org/10.5281/zenodo.15257599>
60. Okaiyeto, S. A., Liu, D., Zhang, C., Bai, J. W., Chen, C., Sharma, P., ... & Xiao, H. W. (2025). Anti-nutrients of plant-based food: physicochemical properties, effects on health and degradation techniques-a comprehensive review. *Journal of Future Foods*. DOI: [10.1016/j.jfutfo.2025.06.022](https://doi.org/10.1016/j.jfutfo.2025.06.022)
61. Okwu, D. E., & Josiah, C. (2006). Evaluation of the chemical composition of two Nigerian medicinal plants. *African Journal of Biotechnology*, 5, 357-336.
62. Olaofe, O. & Akintayo, E. T. (2000). Prediction of isoelectric points of legume and oilseed proteins from their amino acid compositions. *The Journal of Technoscience*, 4, 49-53.
63. Patil, K. S., & Bhalsing, S. R. (2016). Ethnomedicinal uses, phytochemistry and pharmacological properties of the genus *Boerhavia*. *Journal of Ethnopharmacology*, 182, 200-220. <https://doi.org/10.1016/j.jep.2016.01.042>
64. Paul, A. A., Southgate, D. A. & Russel, J. (1976). First supplement to McCance and Widdowson's. The composition of foods. HMSO, London; 1976
65. Peduruhewa, P. S., Jayathunge, K. G. L. R., & Liyanage, R. (2021). Nutritional evaluation and phytochemical screening of *Commelina diffusa*: An underutilized wild edible plant of Sri Lanka. *American Journal of Food and Nutrition*, 9(3), 106-111. DOI: [10.12691/ajfn-9-3-2](https://doi.org/10.12691/ajfn-9-3-2)
66. Petruzzelli, R., Polishchuk, E. V., & Polishchuk, R. S. (2026). Copper in Human Health and Disease: Insights from Inherited Disorders. *Physiology* (Bethesda, Md.), 41(3), 253-271. <https://doi.org/10.1152/physiol.00032.2025>
67. Rao, B. S. & Prabhavathi, T. (1982). Tannin content of foods commonly consumed in India and its influence on ionisable iron. *Journal of the Science of Food and Agriculture*, 33 (1), 89-96.
68. Rumicha, T. D., Belew, S., Hasen, G., Teka, T. A., & Forsido, S. F. (2025). Food, feed, and phytochemical uses of wild edible plants: a systematic review. *Food Science & Nutrition*, 13(6), e70454. doi: [10.1002/fsn3.70454](https://doi.org/10.1002/fsn3.70454)
69. Sabuj, S. S., Islam, M. S., Uddin, M. N., Islam, M. N., & Islam, M. S. (2021). Vitamin-C and Mineral Composition Analysis of Some Fresh Vegetables in Patuakhali, Bangladesh. *Asian Food Science Journal*, 20(9), 78-88. DOI: [10.9734/afsj/2021/v20i930344](https://doi.org/10.9734/afsj/2021/v20i930344)
70. Salim, R., Nehvi, I. B., Mir, R. A., Tyagi, A., Ali, S., & Bhat, O. M. (2023). A review on anti-nutritional factors: unraveling the natural gateways to human health. *Frontiers in Nutrition*, 10, 1215873. DOI [10.3389/fnut.2023.1215873](https://doi.org/10.3389/fnut.2023.1215873)
71. Salman, M., Suraiya, S., Das, P., Islam, M. A., & Haq, M. (2021). Variation in the proximate composition, amino acids content and fatty acids of Thai Pangus (*Pangasianodon hypophthalmus*) Fish Depending on Size. *Asian Food Science Journal*, 20(6), 35-49. DOI: [10.9734/afsj/2021/v20i630307](https://doi.org/10.9734/afsj/2021/v20i630307)
72. Schmelzer, G.H. and Gurib-Fakim, A., Eds. (2008) Plant Resource of Tropical Africa 11 (1) Medicinal Plants 1. PROTA Foundation, Wageningen, Netherlands/ Backhuys Publishers, Leiden, Netherlands/CTA, Wageningen, Netherlands, 117-121.
73. Seldin, D. W., & Giebisch, G. H. (1990). The Regulation of sodium and chloride balance. Raven Press.
74. Shenkin, A. (2008). Basics in clinical nutrition: Physiological function and deficiency states of trace elements. *European e-Journal of Clinical Nutrition and Metabolism*, 3(6), e255-e258.

75. Stefanache, A., Lungu, I. I., Butnariu, I. A., Calin, G., Gutu, C., Marcu, C., ... & Damir, D. (2023). Understanding how minerals contribute to optimal immune function. *Journal of Immunology Research*, 2023(1), 3355733. doi: [10.1155/2023/3355733](https://doi.org/10.1155/2023/3355733).
76. Takahashi, T., Toda, E., Singh, R. B., De Meester, F., Wilczynska, A., Wilson, D., & Juneja, L. R. (2011). Essential and non-essential amino acids in relation to glutamate. *The Open Nutraceuticals Journal*, 4(1), 205-212
77. Tenyang, N., Ponka, R., Tiencheu, B., Womeni, H. M., & Tonfack, F. (2016). Proximate composition, fatty acid and mineral contents of four freshwater fish from Maga Lake (far North region of Cameroun). *American Journal of Food Science and Technology*, 4(3), 64-69. doi: [10.12691/ajfst-4-3-2](https://doi.org/10.12691/ajfst-4-3-2).
78. Thakur, M. K., & Pathak, S. (2016). Phytochemical, Nutritional and Antimicrobial Properties of *Boerhaavia Diffusa*. *International Journal of Science and Research*, 5(9), 400-404.
79. Tsomele, G. F., Meiring, B., Anyasi, T. A., Mlambo, V., Amonsou, E., Lepule, S. P., ... & Wokadala, O. C. (2023). Influence of amino acid profile and secondary structure on nutritional and functional properties of *Trichilia emetica* and *Trichilia dregeana* protein concentrates. *International Journal of Food Science & Technology*, 58(10), 5489-5500. DOI:[10.1111/ijfs.16425](https://doi.org/10.1111/ijfs.16425)
80. Ujowundu, C. O., Igwe, C. U., Enemor, V. H. A., Nwaogu, L. A., & Okafor, O. E. (2008). Nutritive and anti-nutritive properties of *Boerhavia diffusa* and *Commelina nudiflora* leaves. *Pakistan Journal of Nutrition*, 7(1), 90-92.
81. Umuhozariho, M. G., Shayo, N. B., Msuya, J. M., & Sallah, P. Y. K. (2014). Cyanide and selected nutrients content of different preparations of leaves from three cassava species. *African Journal of Food Science*, 8(3), 122-129.
82. Vadivel, V., & Janardhanan, K. (2005). Nutritional and antinutritional characteristics of seven South Indian wild legumes. *Plant Foods for Human Nutrition*, 60(2), 69-75. DOI: [10.1007/s11130-005-5102-y](https://doi.org/10.1007/s11130-005-5102-y)
83. van Niekerk G, Mitchell M, Engelbrecht A-M (2018) Bone resorption: supporting immunometabolism. *Biol Lett* 14. [10.1098/rsbl.2017.0783](https://doi.org/10.1098/rsbl.2017.0783)
84. Zarkadas, C. G., Voldeng, H. D., Yu, Z. R., & Choi, V. K. (1997). Determination of the protein quality of three new northern adapted cultivars of common and miso type soybeans by amino acid analysis. *Journal of Agricultural and Food Chemistry*, 45(4), 1161-1168.