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BIOCHEMICAL AND MINERAL EVALUATION OF NIGERIAN SPINACH (*AMARANTHUS HYBRIDUS* L.) AS INFLUENCED BY DOMESTIC PROCESSING PROCEDURES

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Abstract: This study evaluated the effects of selected domestic blanching procedures on the biochemical and mineral composition of Nigerian spinach (*Amaranthus hybridus* L.) to identify processing methods that maximize nutrient retention. Fresh spinach leaves were subjected to four blanching treatments: steam blanching without cutting (BAL), hot-water blanching without cutting (CAL), cutting before steam blanching (DAL), and cutting before hot-water blanching (EAL), while untreated leaves (FAL) served as the control. Biochemical and mineral analyses were conducted using standard methods, and data were analyzed using ANOVA and principal component analysis (PCA). Significant differences ($p < 0.05$) were observed among treatments. Fresh leaves contained the highest concentrations of phenolics, flavonoids, dietary fibre, and minerals. Steam blanching, particularly BAL, retained nutrients more effectively than hot-water blanching, whereas EAL resulted in the greatest nutrient losses. PCA explained 94.0% of the total variation and clearly distinguished treatments based on nutrient retention. The findings indicate that BAL is the most effective domestic processing method for preserving the nutritional quality of Nigerian spinach.

Keywords: *Amaranthus hybridus*, blanching, nutrient retention, phytochemicals, minerals, nigerian spinach.

INTRODUCTION

The increasing demand for nutrient-dense foods and sustainable dietary options has heightened interest in indigenous leafy vegetables because of their important role in food and nutrition security, particularly in developing countries (Zhu, 2024). These vegetables are valued for their affordability, accessibility, and richness in essential nutrients and bioactive compounds. In many African countries, indigenous vegetables constitute an important component of traditional diets and serve as valuable sources of vitamins, minerals, dietary fibre, and phytochemicals required for healthy growth and development (Mungofa *et al*., 2022). In Nigeria, they contribute significantly to household nutrition, especially among populations that depend largely on locally available food resources. Among these vegetables, Nigerian spinach (*Amaranthus hybridus* L.), commonly known as "Sokoyokoto," is widely consumed because of its nutritional value, ease of cultivation, and versatility in food preparation (Ogwu, 2020). Belonging to the family Amaranthaceae, *A. hybridus* is widely cultivated across tropical and subtropical regions due to its adaptability to diverse environmental conditions, rapid growth, and high biomass production (Macías-Naranjo *et al*., 2026). Its leaves and tender stems are commonly used in soups, stews, sauces, and other traditional dishes. Beyond its culinary importance, the vegetable provides income for smallholder farmers and vegetable traders and is also utilized in traditional medicine for the management of ailments such as gastrointestinal disorders, inflammation, and general body weakness (Ruth *et al*., 2021). The nutritional importance of Nigerian spinach is attributed to its rich biochemical and mineral composition. Previous studies have reported appreciable levels of vitamins A, C, and K, as well as essential minerals including calcium, iron, magnesium, potassium, phosphorus, and zinc (Olatoberu *et al*., 2019). The vegetable is also a good source of dietary fibre and bioactive compounds such as phenolics and flavonoids, which possess strong antioxidant properties. These compounds help combat oxidative stress and have been associated with reduced risks of cardiovascular diseases, diabetes, and certain cancers. Consequently, regular consumption of Nigerian spinach can contribute to improved nutritional status and overall health, particularly in regions where micronutrient deficiencies remain prevalent (Ukom and Obi, 2018).

Despite its nutritional benefits, Nigerian spinach is rarely consumed raw. Before consumption, the leaves are commonly subjected to domestic processing methods such as washing, cutting, blanching, boiling, drying, and cooking (Frances *et al*., 2022). These processes improve palatability, reduce microbial contamination, soften plant tissues, and enhance digestibility. Among them, blanching is widely employed as a pre-cooking treatment because it helps preserve colour, texture, and sensory quality while reducing enzymatic deterioration. However, processing can also alter the nutritional composition of vegetables through thermal degradation, oxidation, enzymatic reactions, and the leaching of water-soluble nutrients into the processing medium (Verma *et al*., 2018). As a result, the nutritional quality of vegetables may differ substantially between their fresh and processed forms. The effects of domestic processing on nutrient retention have received considerable attention because household preparation methods can significantly influence both nutrient concentration and bioavailability. Nutrient losses are often associated with prolonged heating,

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direct contact with water, and tissue disruption caused by cutting prior to processing (Ogwu, 2020). These losses are particularly important in developing countries where leafy vegetables serve as major dietary sources of essential micronutrients (Gelaye, 2023). Although previous studies have reported the nutritional composition of *A. hybridus*, information on the comparative effects of different blanching methods on its biochemical and mineral constituents remains limited. In particular, the influence of cutting before blanching and the relative effectiveness of steam and hot-water blanching in preserving nutrients have not been adequately investigated. Therefore, this study evaluated the effects of selected blanching treatments on the biochemical and mineral composition of Nigerian spinach with the aim of identifying processing methods that maximize nutrient retention and preserve nutritional quality.

MATERIALS AND METHODS

Sample Collection

Fresh Nigerian spinach leaves (2 kg) were obtained from an accredited farm in Ikole Ekiti, Ekiti State, Nigeria. Leaves were harvested in the morning to ensure freshness and immediately transported to the laboratory for processing.

Sample Preparation and Pretreatments

The leaves were sorted and washed thoroughly before being subjected to different blanching treatments following the method of Amit *et al.* (2023). Four pretreatment methods were employed, as illustrated in Figures 1–4:

Cutting before hot-water blanching (EAL): Leaves were chopped into small pieces and blanched in hot water (leaf-to-water ratio of 1:5, w/w) for 2 min. The blanched leaves were cooled in cold water and drained.

Cutting before steam blanching (DAL): Leaves were chopped into small pieces and steam-blanched for 5 min.

Hot-water blanching without cutting (CAL): The whole leaves were blanched in hot water (1:5, w/w) for 2 min, cooled in cold water, and drained.

Steam blanching without cutting (BAL): The whole leaves were steam-blanched for 5 min.

Following pretreatment, all samples were dried at 60°C for 4 h, ground, sieved through a mesh sieve, packaged, and stored for subsequent analyses.

Untreated spinach leaf flour served as the control sample (FAL).

Experimental Design

Five samples were prepared:

FAL: Untreated spinach leaf flour (control)

BAL: Steam-blanched spinach leaves without cutting

CAL: Hot-water-blanched spinach leaves without cutting

DAL: Cut spinach leaves subjected to steam blanching

EAL: Cut spinach leaves subjected to hot-water blanching

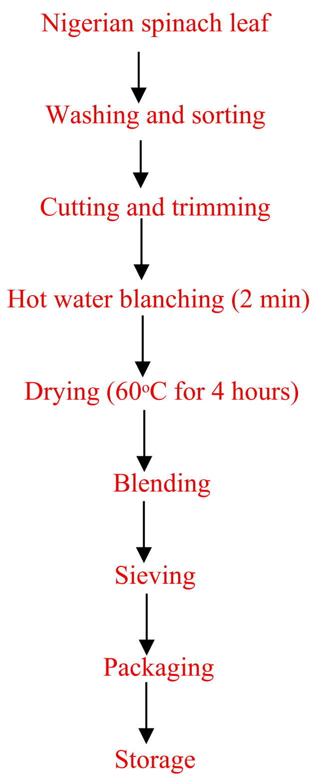

Fig. 1. Cutting before Hot water blanching pretreatment of Nigerian spinach leaf.

Nigerian spinach leaf
↓
Washing and sorting
↓
Cutting and trimming
↓
Steam Blanching (2 min)
↓
Drying (60°C for 4 hours)
↓
Blending
↓
Sieving
↓
Packaging
↓
Storage

Fig. 2. Cutting before Steam blanching pretreatment of Nigerian spinach leaf.

Nigerian spinach leaf
↓
Washing and sorting
↓
Draining
↓
Hot water blanching (2 min)
↓
Drying (60°C for 4 hours)
↓
Grinding
↓
Sieving
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Packaging
↓
Storage

Fig. 3. Hot water blanching without cutting pretreatment of Nigerian spinach leaf.

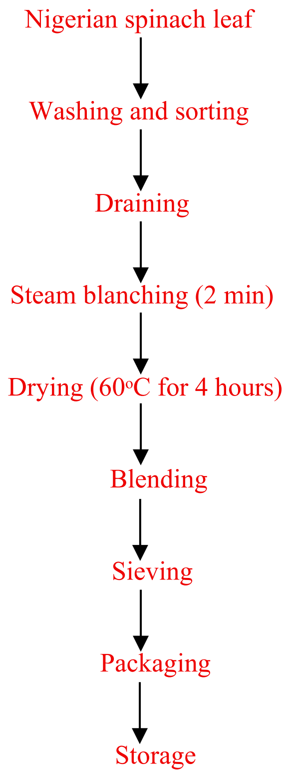

Fig. 4. Steam blanching without cutting pretreatment of Nigerian spinach leaf.

Biochemical Properties

pH Determination

The pH was determined using a Mettler Delta 350 pH meter according to AOAC (2010). Ten grams of each sample was homogenized in 100 mL of deionized water, filtered, and the pH of the filtrate was measured.

Amino Acid Determination

Essential and non-essential amino acids were determined according to AOAC (2010). Samples were hydrolysed with 6 M HCl containing phenol and 2-mercaptoethanol at 110°C for 22 h. Following hydrolysis, amino acids were quantified using reverse-phase high-performance liquid chromatography (RP-HPLC) equipped with an Agilent 1100 system and a Zorbax 80A C18 column. Cystine was determined after performic acid oxidation, while tryptophan was analyzed using the ninhydrin method. Amino acid concentrations were calculated from the resulting chromatograms.

Essential Amino Acid Index (EAAI)

The Essential Amino Acid Index was calculated according to Steinke *et al.* (1980):

$$EAAI = \sqrt[8]{\frac{(Phe \times Val \times Thr \times Ile \times Met \times His \times Lys \times Leu)_{sample}}{(Phe \times Val \times Thr \times Ile \times Met \times His \times Lys \times Leu)_{casein}}}$$

where the numerator and denominator represent amino acid concentrations in the test sample and standard protein (casein), respectively.

Predicted Protein Efficiency Ratio (P-PER)

The predicted protein efficiency ratio was estimated using the equation of Adeyeye (2017):

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

Predicted Biological Value (BV)

Biological value was calculated according to Okpala *et al.* (2016):

$$BV = 1.09(EAAI) - 11.7$$

Total Phenolic Content

Total phenolic content (TPC) was determined using the Folin–Ciocalteu method described by Singleton *et al.* (1999). Absorbance was measured at 725 nm using a UV–visible spectrophotometer, and results were expressed as mg gallic acid equivalents per gram dry weight (mg GAE/g).

Total Flavonoid Content

Total flavonoid content was determined according to Nabavi *et al.* (2013). Absorbance was measured at 500 nm and flavonoid concentration was calculated from a rutin standard curve. Results were expressed as mg rutin equivalents per gram sample (mg RE/g).

Mineral Determination

Sodium, potassium, calcium, magnesium, and iron contents were determined according to AOAC (2010). Samples were dry-ashed at 600°C, digested using hydrochloric and nitric acids, filtered, and analysed using Atomic Absorption Spectrophotometry (AAS).

Statistical Analysis

Data were expressed as mean ± standard deviation of triplicate determinations. One-way analysis of variance (ANOVA) was used to evaluate the effects of blanching treatments on the biochemical and mineral properties of Nigerian spinach leaves, while Duncan's Multiple Range Test (DMRT) separated significant means at $p < 0.05$. Pearson's correlation analysis was performed to determine relationships among variables. Multivariate analysis was further performed using principal component analysis (PCA) based on standardized data to identify major sources of variation among treatments. All analyses were performed using SPSS Statistics 20.0.

RESULTS AND DISCUSSION

Biochemical composition of Nigerian spinach leaves

The biochemical composition of fresh and processed Nigerian spinach leaves is presented in Table 1, while the corresponding retention percentages are shown in Table 3. Significant differences ($p < 0.05$) were observed among all treatments for the biochemical parameters evaluated. Generally, FAL recorded the highest values for total phenolics, flavonoids, pH, and dietary fibre, indicating that blanching treatments influenced the biochemical quality of the leaves.

Total phenolic content ranged from 121.53 mg/100 g in EAL to 169.10 mg/100 g in FAL. Fresh leaves had the highest phenolic concentration, while EAL showed the lowest value. The reduction in phenolics after blanching is mainly due to thermal degradation and leaching of water-soluble compounds into the blanching water. Similar trends were reported by Mathivanan *et al*. (2022), who observed substantial losses of phenolics in vegetables subjected to heat and water-based processing. Among processed samples, steam blanching retained phenolics more effectively than hot-water blanching. DAL recorded 155.74 mg/100 g with 92.11% retention, whereas EAL retained only 71.86% (Table 3). These findings align with Narra *et al*. (2024) and Sharma *et al*. (2024), confirming reduced nutrient losses under steam blanching. Phenolic compounds are important antioxidants that help reduce oxidative stress and may lower the risk of chronic diseases such as cardiovascular disorders and cancer (Luna-Guevara *et al*., 2020).

A similar pattern was observed for total flavonoids, which ranged from 91.15 mg/100 g in EAL to 142.75 mg/100 g in FAL. The higher flavonoid content in FAL reflects the absence of heat treatment, which prevents degradation of heat-sensitive compounds. Lower values in blanched samples are attributed to thermal breakdown and leaching during processing, as also reported by Ahmed and Eun (2018) and Chen *et al*. (2022). Among the treatments, BAL showed the highest flavonoid retention (93.53%), followed by DAL (90.35%), while EAL recorded the lowest (63.87%). This confirms that steam blanching is more effective than hot-water blanching in preserving flavonoids. Since flavonoids play key roles in antioxidant defence and in preventing oxidative stress-related conditions such as type 2 diabetes and cardiovascular diseases, their retention enhances the nutritional quality of processed spinach (Batool *et al*., 2025).

The pH values of the samples ranged from 6.06 to 6.30, indicating that all treatments remained slightly acidic. Fresh leaves recorded the highest pH value (6.30), while CAL and EAL had the lowest values (6.06). Although the differences were relatively small, can cause minor reductions in vegetable pH due to changes in organic acid composition. Retention values further showed that pH was only minimally affected by processing, with all treatments retaining over 96% of the pH value of the fresh leaves. Total dietary fibre content ranged from 8.52% in EAL to 9.38% in FAL. Although blanching caused a reduction in fibre content, the losses were relatively small compared with those observed for phenolics and flavonoids. Retention values ranged from 90.83% in EAL to 95.09% in BAL, indicating that dietary fibre remained largely stable during processing. The slight reduction observed in EAL may be associated with cell wall softening and structural modifications caused by combined cutting and hot-water treatment. Similar findings have been reported by Dong *et al*. (2019) and Coe and Spiro (2022), who concluded that blanching generally has minimal effects on dietary fibre content in vegetables. As dietary fibre plays an important role in digestive health, glycaemic regulation, and the prevention of metabolic disorders, its high retention across treatments is nutritionally advantageous.

Table 1.

Biochemical compositions of Nigerian spinach leaves

Treatments	Total phenolic (mg/100g)	Total flavonoids (mg/100g)	pH	Total dietry fibre (%)
BAL	134.40±0.69^{d}	133.51±0.64^{b}	6.27±0.02^{b}	8.92±0.13^{b}
CAL	147.52±0.77^{c}	108.05±2.29^{d}	6.06±0.07^{d}	8.81±0.08^{c}
DAL	155.74±1.31^{b}	128.93±0.39^{c}	6.20±0.02^{c}	8.80±0.02^{c}
EAL	121.53±3.06^{e}	91.15±0.24^{e}	6.06±0.02^{d}	8.52±0.03^{d}
FAL	169.10±0.16^{a}	142.75±0.43^{a}	6.30±0.01^{a}	9.38±0.12^{a}

Values are mean ± S.D. for triplicate determinations. Means with the same superscript across a column are significantly different (p<0.05).
BAL = Steam blanching without cutting; CAL = Hot-water blanching without cutting; DAL = Cutting before steam blanching; EAL = Cutting before hot-water blanching;
FAL = Fresh Nigerian spinach leaf.

Table 2.

Mineral compositions of Nigerian spinach leaves (mg/100g)

Treatments	Ca	Fe	Mg	Mn	P	Na	K	Zn
BAL	94.92 ± 0.15^{b}	21.17 ± 0.28^{b}	48.75 ± 0.25^{b}	0.78 ± 0.02^{b}	34.82 ± 0.29^{b}	195.29 ± 0.39^{b}	284.93 ± 1.12^{b}	3.63 ± 0.12^{b}
CAL	87.98 ± 0.22^{d}	16.92 ± 0.22^{d}	43.82 ± 0.29^{c}	0.56 ± 0.03^{d}	26.08 ± 0.19^{d}	181.93 ± 1.15^{d}	268.96 ± 0.45^{c}	3.42 ± 0.03^{c}
DAL	89.78 ± 0.36^{c}	21.94 ± 0.09^{b}	42.34 ± 0.27^{d}	0.71 ± 0.03^{c}	30.02 ± 0.42^{c}	189.29 ± 1.12^{c}	265.35 ± 0.49^{d}	3.31 ± 0.09^{d}
EAL	73.76 ± 0.27^{e}	10.97 ± 0.13^{d}	33.73 ± 0.35^{e}	0.49 ± 0.03^{e}	19.97 ± 0.12^{e}	148.35 ± 0.98^{e}	253.45 ± 0.98^{e}	3.07 ± 0.05^{e}
FAL	107.75 ± 0.83^{a}	26.29 ± 0.45^{a}	51.85 ± 0.57^{a}	0.85 ± 0.03^{a}	35.75 ± 0.35^{a}	207.67 ± 1.97^{a}	305.34 ± 1.10^{a}	4.08 ± 0.05^{a}

Values are mean ± S.D. for triplicate determinations. Means with the same superscript across a column are significantly different ($p<0.05$).

Table 3.

Percentage Retention of Biochemical Properties in Processed Nigerian Spinach Leaves (FAL = 100%)

Treatment	Total Phenolics (%)	Flavonoids (%)	pH (%)	Dietary Fibre (%)
BAL	79.47	93.53	99.52	95.09
CAL	87.21	75.73	96.19	93.92
DAL	92.11	90.35	98.41	93.60
EAL	71.86	63.87	96.19	90.83
FAL	100.00	100.00	100.00	100.00

Table 4.

Percentage retention of mineral contents in processed Nigerian spinach leaves (FAL = 100%)

Treatment	Ca (%)	Fe (%)	Mg (%)	Mn (%)	P (%)	Na (%)	K (%)	Zn (%)
BAL	88.10	80.50	94.01	91.76	97.39	94.06	93.30	88.97
CAL	81.65	64.38	84.58	65.88	72.98	87.60	88.13	83.82
DAL	83.33	83.44	81.69	83.53	83.96	91.04	86.88	81.13
EAL	68.44	41.74	65.07	57.65	55.86	71.45	83.02	75.24
FAL	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

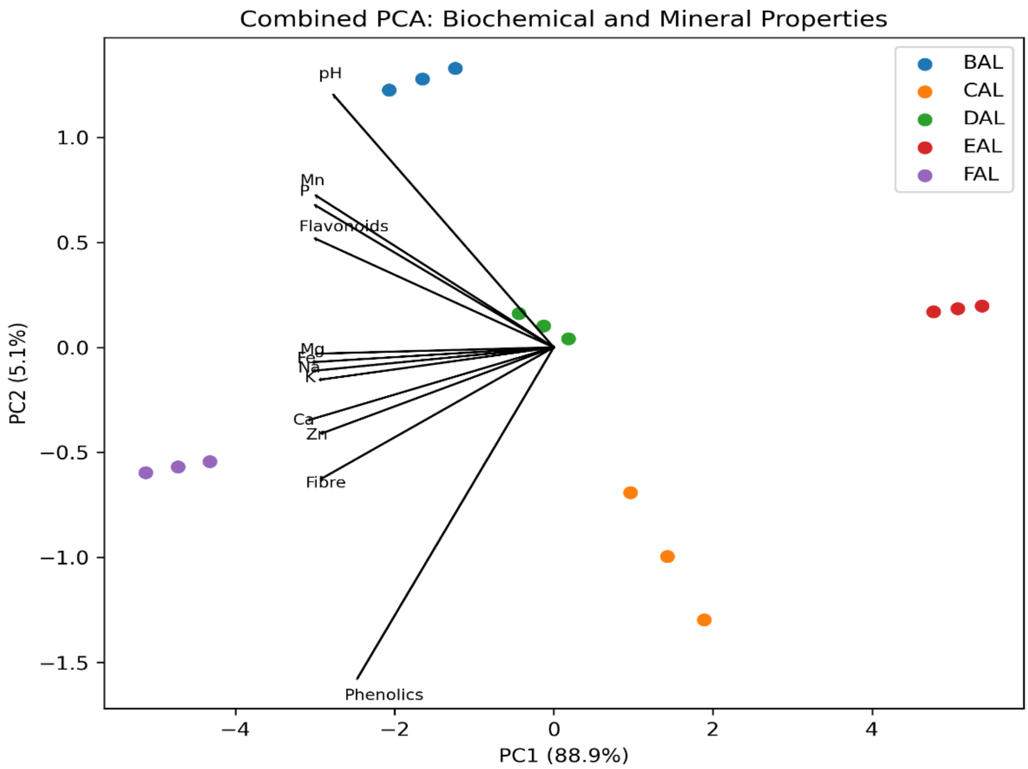

Fig. 5. Principal component analysis biplot.

Table 5.

PCA loadings for biochemical and mineral properties

Variables	PC1 loading	PC2 loading
Phenolics	-0.82108	-0.52511
Flavonoids	-0.99969	0.172898
pH	-0.92162	0.400646
Fibre	-0.97458	-0.20905
Ca	-1.02456	-0.11538
Fe	-1.00729	-0.02349
Mg	-0.99545	-0.01014
Mn	-0.99603	0.241112
P	-0.99882	0.225679
Na	-1.00417	-0.03723
K	-0.97906	-0.05145
Zn	-0.97211	-0.13686

Mineral composition of Nigerian spinach leaves

The mineral composition of Nigerian spinach leaves subjected to different domestic processing procedures is presented in Table 2. The results revealed significant ($p \leq 0.05$) differences among the treatments for all mineral elements evaluated. FAL consistently recorded the highest concentrations of all the minerals, whereas the lowest values were generally observed in EAL. Calcium content decreased significantly from 107.75 mg/100 g in fresh leaves to 73.76 mg/100 g in EAL, with retention values ranging from 68.44% (EAL) to 88.10% (BAL) (Table 4). Similarly, iron declined from 26.29 to 10.97 mg/100 g, with retention values of 80.50% and 83.44% in BAL and DAL, respectively, compared with only 41.74% in EAL. Magnesium also decreased from 51.85 to 33.73 mg/100 g, while retention ranged from 65.07% (EAL) to 94.01% (BAL). These results indicate that steam blanching preserved minerals more effectively than hot-water blanching, likely because the absence of direct water contact reduced mineral leaching. Similar findings have been reported by Verma *et al*. (2018), Rana *et al*. (2020), and Putriani *et al*. (2022). The retention of calcium, iron, and magnesium is nutritionally important due to their roles in bone development, oxygen transport, enzyme activation, and energy metabolism. Manganese content decreased from 0.85 mg/100 g in FAL to 0.49 mg/100 g in EAL, with retention values ranging from 57.65% to 91.76%. Likewise, phosphorus declined from 35.75 to 19.97 mg/100 g, while retention ranged from 55.86% (EAL) to 97.39% (BAL), the highest among all minerals evaluated. These reductions are consistent with reports that mineral losses during blanching are primarily caused by diffusion into the blanching medium (Nardis *et al*., 2021; Wang *et al*., 2021; Sarkar *et al*., 2023). Retaining these minerals is beneficial because manganese supports enzymatic and antioxidant functions, whereas phosphorus is essential for skeletal development and energy transfer.

Sodium and potassium concentrations also decreased following processing. Sodium declined from 207.67 mg/100 g in FAL to 148.35 mg/100 g in EAL, with retention values of 71.45–94.06%, while potassium, the most abundant mineral, decreased from 305.34 to 253.45 mg/100 g, with retention values ranging from 83.02% to 93.30%. The greater losses observed in hot-water-blanched samples reflect the high solubility of these minerals and their susceptibility to leaching, in agreement with the findings of Kachhadiya *et al*. (2018), Aadil *et al*. (2019), Deng *et al*. (2019). Both minerals are important for electrolyte balance, nerve transmission, and muscle function. Among the trace minerals, zinc showed the least reduction, decreasing from 4.08 mg/100 g in fresh leaves to 3.07 mg/100 g in EAL, with retention values ranging from 75.24% to 88.97%. Although relatively stable during thermal processing, some zinc losses occurred, particularly during hot-water blanching, due to leaching effects (Zhang *et al*., 2021; Reis, 2023). The higher retention of zinc in steam-blanched samples contributes to maintaining the nutritional quality of the processed spinach because of its role in immune function, protein synthesis, and cellular metabolism.

Principal Component Analysis (PCA)

The PCA of the biochemical and mineral properties of Nigerian spinach leaves revealed that PC1 and PC2 accounted for 88.9% and 5.1% of the total variation, respectively, explaining 94.0% of the overall variability (Figure 5). The high proportion of variance explained by PC1 indicates that it was the dominant factor influencing differences among treatments. All measured variables exhibited strong negative loadings on PC1, ranging from −0.821 for phenolics to −1.025 for calcium (Figure 5), indicating that this component represents an overall nutrient-quality gradient. Similar interpretations of PCA loadings have been reported in studies evaluating the effects of processing on vegetable quality (Durmuş, 2025). The PCA score plot showed a clear separation of treatments according to processing method. FAL clustered at the negative extreme of PC1 and were strongly associated with higher concentrations of phenolics, flavonoids, dietary fibre, and minerals. This finding agrees with previous reports that fresh vegetables generally retain higher levels of bioactive compounds and nutrients than processed samples (Ding *et al*., 2025). In contrast, CAL and EAL were positioned on the positive side of PC1, indicating greater nutrient losses, particularly in hot-water-blanched samples. The distinct separation of EAL from the other treatments suggests that cutting before hot-water blanching intensified nutrient degradation through increased leaching and thermal exposure (Ahmed and Eun, 2018; Sharma *et al*., 2024). PC2 contributed only 5.1% of the variation but further differentiated the treatments based on pH, Mn, P, and flavonoids, which showed positive loadings, whereas phenolics and fibre had negative loadings. The location of BAL and DAL closer to the origin and upper quadrants indicates better preservation of biochemical and mineral constituents, particularly under steam blanching.

CONCLUSION

The results of this study revealed that blanching significantly influenced the biochemical and mineral composition of Nigerian spinach leaves. FAL consistently recorded the highest concentrations of total phenolics, flavonoids, dietary fibre, and mineral elements, indicating that thermal processing resulted in varying degrees of nutrient loss. However, the magnitude of these losses depended largely on the blanching method employed. BAL and DAL generally retained higher levels of bioactive compounds and minerals than CAL and EAL, demonstrating the effectiveness of steam blanching in preserving nutritional quality. The superior retention observed in steam-blanched leaves can be attributed to reduced leaching of water-soluble nutrients and limited exposure to direct water contact. In contrast, hot-water blanching, particularly when combined with EAL, resulted in the greatest nutrient losses. This suggests that tissue disruption prior to blanching increases the surface area available for nutrient diffusion and accelerates the loss of valuable compounds during processing. The findings highlight the importance of selecting appropriate processing methods to maintain the nutritional and functional quality of Nigerian spinach leaves. Steam blanching emerged as the most effective technique for preserving both biochemical constituents and mineral nutrients. Therefore, its adoption in household and food-processing practices could help maximize nutrient retention and enhance the dietary value of this important leafy vegetable.

AUTHORS CONTRIBUTIONS

Conceptualization: E.B.F., and A.B.A.; Methodology: E.B.F., and A.B.A.; Data collection: E.B.F.; Data validation: A.B.A.; Data processing: E.B.F. and B.M.F.; Writing—original draft preparation: E.B.F., and B.M.F.; Writing—review and editing: E.B.F., A.B.A., and B.M.F.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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