IKR Publishers [International Knowledge and Research Publishers]

IKR Journal of Agriculture and Biosciences (IKRJAB)

Journal homepage: https://ikrpublishers.com/ikrjab/

Volume-1, Issue-1 (May-June) 2025

ISSN: XXXX-XXXX (Online)

Nutritional, Antioxidant, and PhytochemicalCharacterization Leaves of Hibiscus sabdariffa L. Grown in SHESTCO, Abuja, Nigeria

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ARTICLE INFO

Article history:

Received : 15-05-2025

Accepted : 21-05-2025

Available online : 27-05-2025

Keywords:

Antioxidant, Hibiscus Sabdariffa L, Mineral, Phytochemical, Proximate, Vitamins.

ABSTRACT *Original research paper*

This paper examined the nutritional composition, phytoconstituents, mineral content, and antioxidant effect of Hibiscus sabdariffa L. (Roselle) leaves cultivated at the SHESTCO. The leaves were subjected to qualitative and quantitative phytochemical screening, proximate analysis, mineral profiling, and the DPPH scavenging radicalassaywas used to assesantioxidantactivity. Screening for Phytochemicals confirmed the existence of phenols, saponins, flavonoids, alkaloids, glycosides, and tannins, with phenols and flavonoids being abundant (374.71 mg/100g and 173.83 mg/100g, respectively). The leaves showed high crude protein (17.41%), dietary fiber (21.87%), and appreciable levels of ash (12.18%), carbohydrates (28.37%), and energy (211.11 kcal/100g). Mineral analysis indicated substantial amounts of calcium (546.75 mg/100g), magnesium (270.7 mg/100g), and phosphorus (63.3 mg/100g), supporting the potential of Roselle leaves as a mineral-rich vegetable. The extract also exhibited concentration-dependent of antioxidant activity. These findings highlight the nutritional and functional potential of H. sabdariffa leaves and support their incorporation into nutraceuticals and functional food formulations.

Introduction

Rosellecommonly known as, *Hibiscus sabdariffa* L. belongs to the Malvaceaefamily and is cultivated in tropical and subtropical regions worldwide for its calyces, leaves, seeds, and stems, all of which have various food and medicinal uses [1][2]. In Malaysia, Roselle is locally referred to as *asampaya*, *asamsusur*, or *ribena Malaysia* [3]. In several regions of Africa, India, and Bangladesh, the leaves are widely consumed as vegetables and used in soups, salads, and condiments [4].Despite its wide usage, scientific studies on Roselle have predominantly focused on the calyces, while the leaves remain underutilized and overlooked as a potential source of food and therapeutic agents in many countries[5][6].Recent reports, however, suggest that Roselle leaves possess considerable medicinal and nutritional value, including antioxidant, antitumor, anti-atherosclerotic, and antihyperlipidemic activities [7][8].These biological activities have been linked to the occurrence of flavonoids and other polyphenolcomposites in the leaves, which may contribute to their significant antioxidant potential[3][6][9].Given the limited scientific exploration of Roselle leaves compared to the calyces, there is a pressing need to investigate their phytochemical composition and nutraceutical potential. Therefore, the research was aimed at evaluate the nutritional composition and antioxidant properties of the leaves of Hibiscus sabdariffa using spectrophotometric methods, with a view to providing scientific justification for their inclusion in functional foods and natural health products.

Resources and Approaches

Collection of the Plant leaves and Extraction

Roselle red leaves was collected from the back of Physic Advanced Research Centre (PARC) SHESTCO. The leaves under tap water was washed and dried in the shade for 14 days before groundedinto fine powder form using a blender.

Quantitative and Qualitative Phytochemical Screening

Phytochemical screening of Roselle red leaves was carried out using water as the extraction solvent. Alkaloids were identified using Mayer's reagent, while saponins were detected through the foam test. The presence of tannins and phenols was assessed using the ferric chloride test. Flavonoids were screened with the sodium hydroxide test, and glycosides were evaluated using Legal's test. [10][11].The Quantitative analysis was conducted base on the procedure reported by I.M. Ibrahim et al.[12].

Proximate Composition Study

The Proximate analysis was performed using the standard methods prescribed by the Association of Official Analytical Chemists (AOAC) reported by Ibrahim et al. [11], with minor modifications. Each measurement was carried out in triplicate, and the results were reported as percentages (%). Furthermore, the total caloric content was calculated by aggregating the percentage of crude protein, Fat and carbohydrate content. As follow:

Calorific energy (kcal) = (Fat × 9)+ (Carbohydrate × 4) + (Protein ×4)

Minerals Composition Analysis

The mineral compositions of the sample were analysed by measuring 1g of dried sample digested in a micro-Kjeldahlflask (100 mL) with HNO_3/$HClO_4$and wait for a colourlesssolution. Then, the obtained sample was well-ventilated and concentrated to 50 mL in a volumetric flask with 0.1 M HCl. The elemental composition namely potassium (K), calcium (Ca), sodium (Na), magnesium (Mg), manganese (Mn), copper (Cu) iron (Fe), Lead (Pb), and zinc (Zn) was evaluated by using graphite furnace absorption atomic spectrophotometer (AAs). While phosphorus (P) content was determined by a colorimetric method following the AOAC Standard method [11].

Vitamins Content Determination

The analysis of vitamin C was conducted using the Barakat titrimetric method in accordance with [12], in addition to vitamin E, B1. B2, and niacin were all asses [13].

DPPH Antioxidant property

The Roselle leaf extract was evaluated for antioxidant activity using the radical-scavenging assay DPPH (1, 1-diphenyl-2-picrylhydrazyl), as refer byAbubakar et al. [13], with slight amendments. The method is based on the antioxidants abilityto scavenge the stable DPPH free radical, which fallouts in a reduction in absorbance at 517 nm. To prepare the extract, 2 g of the dried leaf sample was soaked in 10 mL of methanol, and left to stay for proper extraction. A stock solution was prepared in 1 mg/mLby dissolving the dried methanolic extract in methanol. Methanol was used to prepare freshly 0.01 mM DPPH solution. For the analysis, DPPH solution 3 mL was mixed with extract solution 2 mL in a test tube. Serial dilutions of the extract were made to obtain varying concentrations for dose-response analysis. For 30 minutes the mixture was incubated in the dark at room temperature to avoid light-induced degradation of DPPH. At 517 nm the absorbance was then measured using a UV-Vis spectrophotometer. Ascorbic acidas a positive control was used as a standard antioxidant, at the same stock concentration (1 mg/mL in methanol). A control solution was prepared by replacing the extract with methanol, while the blank contained only methanol to zero the spectrophotometer. The DPPH radical percentage of inhibition was calculated using the following formula:

Inhibition % = [(controlstandard absorbance – sampleabsorbance)/controlstandard absorbance] X 100

Results and Discussions

Phytochemicals analysis

Table 1: Quantitativeand Qualitative Phytochemical analysis of dried leavesofHibiscus sabdariffa

Phytochemicals		Composition (mg/100g)
Alkaloids	+	8.62± 0.42
Saponins	+	36.07±0.52
Flavonoids	+	173.83±0.62
Glycosides	+	31.83±0.13
Phenols	+	374.71±0.42
Tannins	+	53.93±0.62

The results data are articulated in mean ± SD (n=3).

Phytochemical screening is a vital step in the identification of new sources of bioactive compounds with therapeutic and industrial value, enabling the optimal use of natural resources. Numerous medicinal plants have undergone chemical investigation by researchers [14]. In this study, leaf

samples were selected for analysis, as leaves are often rich in secondary metabolites, making them ideal candidates for phytochemical evaluation.Various phytochemicals were extracted from the Roselle (Hibiscus sabdariffa) leaves using appropriate methods, and their presence (+) or absence (−) is summarized in Table 1. The results revealed the presence of key bioactive compounds including alkaloids, flavonoids, saponins, glycosides, phenols, and tannins—major classes of phytochemicals known for their biological activities. The phytochemical composition of Roselle leaves aligned with previously reported data, although slight variations were observed, potentially due to factors such as genetic differences, soil composition, and the nature of the solvent used [15].Alkaloids, a diverse group of nitrogen-containing compounds, are well-known for their anticancer properties and their ability to disrupt cell division, making them potential candidates for cancer therapy [16]. The presence of alkaloids in Roselle leaves supports its possible application in cancer treatment. Flavonoids are recognized for a wide range of biological activities, including antiviral, anti-inflammatory, antioxidant, and cytotoxic effects. They are also employed in managing hypertension, diabetes, and rheumatic conditions [17]. The detection of flavonoids in Roselle leaves suggests their potential in treating these disorders and as a natural antioxidant source. Polyphenols were also detected in the current study. These compounds have attracted considerable attention due to their health-promoting effects. Extensive research has demonstrated the pharmacological and biological significance of polyphenols, particularly their antioxidant, anti-inflammatory, antiviral, and cytotoxic properties [17, 18]. Polyphenols are known to support kidney and gastrointestinal health and are associated with protection against degenerative conditions such as cardiovascular disease, Alzheimer's disease, neurodegenerative disorders, cataracts, and atherosclerosis [19]. Saponins were found in the Roselle leaves as well. These compounds exhibit hypotensive and cardiodepressant effects and are recognized for their chemopreventive potential against cancer [20].Tannins, also present in the leaves, are known to inhibit bacterial growth by disrupting microbial enzymes. Tannins are potent antioxidants and possess astringent properties, making them useful in the treatment of gastrointestinal disorders such as diarrhea and dysentery [21].Overall, the presence of these phytoconstituents in Roselle leaves highlights their potential as a valuable natural resource for the development of future therapeutics targeting a range of diseases.

Proximate analysis

Table 2: Proximate composition of the leavesof Hibiscus sabdariffa

Parameters	Composition (%)
Moisture	7.60±0.53
Crude fat	3.11±0.51
Ash	12.18±0.42
Carbohydrate	28.37±0.32
Crude fibre	21.87±0.51
Crude protein	17.41±0.31
Energy (Kcal)	211.11±0.41

The results data are expressed in mean ± SD (n=3).

The major nutrient composition of Hibiscus sabdariffa leaves collected from PARC, SHESTCO, FCT Abuja, was analyzed and is presented in Table 2. The moisture content of the sample was approximately 7.60%, indicating relatively low humidity. This low moisture level is advantageous for prolonged storage and reduced susceptibility to microbial contamination. These findings are consistent with those of Ellis and Roberts. [22] and Nyam et al. [23], who noted that seed and leaf samples with moisture contents within the range of 4–8% are optimal for long-term storage and microbial safety. The crude protein content was found to be 17.41%, which is significantly greater than the values reported in earlier studies by El-Sayed et al.[24], Tounkara et al. [18], Nyam et al. [23], and El-Deab and Ghamry [25]. This suggests that the Roselle leaves sourced from this particular region may serve as a richer dietary protein source compared to those from other agro-ecological zones. Regarding crude fat, the sample contained 3.11%, aligning with the findings of El-Deab and Ghamry.[25], but notably lower than the 20.83% and 27.83% fat content reported by El-Sayed et al.[24] and Tounkara et al.[18], respectively. These discrepancies may be attributed to genetic differences, agronomic practices, and environmental conditions, including soil type and climate. The ash content, representing the total mineral content, was 12.18%, closely matching values reported by El-Sayed et al.[24] and Tounkara et al. [18], indicating a comparably high mineral composition. Notably, the crude fiber content was high, at 21.87%, approximately twice the values previously reported by El-Sayed et al. [24] and Nyam et al. [23], suggesting the prospective of these leaves as a significant source of dietary fiber. The total carbohydrate content, estimated by summing the crude fiber and nitrogen-free

extract, was 28.37%, aligning with values reported by El-Sayed et al. [24], [18][23][25]. Additionally, the energy value was calculated at 799.94 kcal/100g, higher than the energy values reported by [26], indicating the potential of Roselle leaves as a high-energy leafy vegetable. Overall, the proximate composition analysis demonstrates that Roselle leaves are a valuable source of essential nutrients including carbohydrates, fat, fiber, and protein. These findings support their potential application as an affordable, nutrient-dense ingredient in food fortification and functional food product development [27][28]. However, it is important to note that the nutritional quality of Roselle leaves can vary significantly based on environmental and agro ecological factors. Variations in soil composition, water availability, temperature, sunlight intensity, and other climatic conditions can influence the biosynthesis and accumulation of nutrients in plant tissues [29][30].

Mineral analysis

Table 3: Mineral composition of the Hibiscus sabdariffa leaves

Elements	Composition (mg/100g)
Mg	270.7±1.02
Ca	546.75±1.23
Na	27.85±1.36
Mn	1.68±1.64
Zn	1.08±0.81
P	63.3±1.23
Cu	0.42±1.22
Fe	33.45±1.73
K	1207.6±1.44
Pb	0.03±0.43

The results Values are expressed in mean ± SD (n=3).

Mineral nutrients are essential for maintaining human health and are required in small quantities to support a range of physiological functions. These include both macronutrients such as magnesium (Mg), potassium (K), calcium (Ca),sodium (Na), and phosphorus (P),and micronutrients such as chromium (Cr), zinc (Zn), copper (Cu), iron (Fe),selenium (Se), andmanganese (Mn). The mineral composition of the Roselle leaves, comprising both macro- and micronutrients, is presented in Table 3.Among the macronutrients, potassium (K) was found in the highest concentration, followed by calcium (Ca), magnesium (Mg), phosphorus (P), and sodium (Na) in descending order. For the micronutrients, iron (Fe) showed the highest concentration, followed by manganese (Mn), zinc (Zn), copper (Cu), and lead (Pb), respectively, in decreasing quantity. These findings are consistent with previous reports that emphasize the high mineral density of Roselle leaves [18][23].Variations in mineral composition between studies may be attributed to differences in soil mineral content, environmental factors such as rainfall and sunlight, and agricultural practices across different cultivation regions. Each of the minerals identified plays a critical role in human health: for example, calcium and phosphorus contribute to strong bones and teeth; sodium and potassium are essential for fluid balance and nerve transmission; magnesium supports muscle contraction and enzymatic reactions; while iron and zinc are vital for oxygen transport, immune function, and cellular metabolism. In summary, Roselle leaves can be considered a nutrient-rich botanical resource, particularly valued for their diverse mineral content. They hold promise as a functional food or nutritional supplement for improving human health and combating micronutrient deficiencies.

Vatamins analysis

Table 4: Vitamin Composition of the Hibiscus sabdariffa leaves

Parameter	Composition (mg/100g)
Vitamin A	0.28±0.42
Vitamin C	300.10±1.31
Vitamin E	6.81±0.74
Vitamin-B1	0.32±0.13
Vitamin-B2	0.25±0.31
Vitamin-B3	2.72±0.23
Vitamin-B5	0.52±0.21
Vitamin-B6	0.47±0.14

Vitamin -K	0.47±0.32

The results Values are expressed in mean ± SD (n=3).

Generally, Roselle leaves (Hibiscus sabdariffa) are recognized as one of the essential leafy vegetables for human consumption owing to their high content of nutritional, medicinal, and bioactive substances[18][28]. Among these nutrients, vitamins play a crucial role in supporting normal cell function, growth, and development. Therefore, the determination of vitamin content in Roselle leaves is necessary, particularly to promote their use in the functional foods and nutraceutical products formulation[31].In the present study, the vitamin content of Roselle leaves was generally found to be low in most water- and vitamins fat-soluble, such as, B-complex vitamins (B5, B2, B1, andB6), vitamin A, and as well as vitamin K. These were present only in trace or minimal amounts. However, the analysis revealed a high concentration of ascorbic acid (vitamin C), that is consistent with earlier findings [32][33]. This indicates that leaves of Rosellecould serve as a significant dietary source of vitamin C, which is essential for antioxidant defense, immune support, and collagen formation. Thus, Roselle leaves may not only contribute to micronutrient intake but also offer potential for value-added food fortification, especially as a source of ascorbic acid and possibly vitamins (fat-soluble) such as vitamin A and K, depending on postharvest handling and processing methods.

Dpph antioxidant properties

Table 5: scavengingradical- activity of the leaves extract of Hibiscus sabdariffa using DPPH method

Concentration (mg/mL)	% inhibition	Vitamin C
0.5	91.43±1.02	96.4±0.2
0.25	87.76±1.04	87.2±0.5
0.625	69.21±1.12	74.05±0.3
0.6125	52.94±0.15	60.21±0.4
0.03125	35.83±0.82	49.54±0.1

The results Values are expressed in mean ± SD (n=3).

In this study, the dried leaves of Hibiscus sabdariffawas evaluated for antioxidant activity using the DPPH (2, 2-diphenyl-1-picrylhydrazyl) radical scavenging assay, by comparing the total antioxidant capacity of the extract to that of ascorbic acid, a known reference standard. The DPPH assay is based on the donation of hydrogen atoms or electrons to neutralizeantioxidantsability of the DPPH radical, which is purple in color and absorbs maximally at 517 nm. Upon interaction with antioxidants, the DPPH radical becomes decolorized, and this change in absorbance is used to quantify antioxidant activity [34].The results, presented in Table 5, showed that the scavenging activity of the Roselle leaf extract was a dependent of dosage, with all concentrations exhibiting good radical inhibition, comparable to ascorbic acid. The maximum inhibition ranged from 91% to 96% at a0.5 mg/mL concentration of extract, though complete inhibition was not achieved. These results are in agreement with one previously studies [35][28], which also reported strong antioxidant potential in Roselle leaf extracts. This antioxidant capacity is largely ascribed to the incidence of polyphenol compounds, including flavonoids, anthocyanin's, and phenolic acids, which are known for their redox properties and their capability to neutralize chelate metal ions, inhibit lipid peroxidation, and free radicals[36][37]. The data further confirm that H. sabdariffa leaves are rich in these bioactive compounds that can subsidize to the prevention of oxidative stress-induced damage, suggesting their potential use in food and pharmaceutical applicationsas a natural antioxidants. Therefore, the incorporation of Roselle leaf extract into functional foods, dietary supplements, or as natural food colorants and preservatives holds promising potential, especially in light of growing consumer interest in plant-based, antioxidant-rich ingredients.

Conclusion

The findings from this study demonstrate that ***Hibiscus sabdariffa*** leaves are rich in phytochemicals, essential nutrients, and minerals with significant health-promoting properties. The existence of active ingredients such as saponins, phenols, and flavonoids, as well as their potent antioxidant activity, supports their potential role in disease prevention and therapeutic applications. The leaves also show favorable proximate and mineral compositions, suggesting their utility as an affordable and accessible source of nutrients. These attributes make ***Hibiscus sabdariffa*** leaves a promising candidate for incorporation into functional foods and nutraceutical products. However, further studies, including toxicity profiling and medical trials, are recommended to fully establish their efficacyandsafety.

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