Quest Journals
Journal of Research in Agriculture and Animal Science
Volume 10 ~ Issue 5 (2023) pp: 01-06
ISSN(Online) : 2321-9459
www.questjournals.org

Research Paper

Evaluation of Proximate Composition, Mineral Nutrients, Antioxidant, Flavonoids and Phenolic Compounds of Barun (*Crataeva nurvala* Buch. Ham.) from Netrokona District of Bangladesh

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ABSTRACT: *The people of Netrokona district of Bangladesh have anethnobotanical belief that Barun Shak has nutritional and medicinal benefits without knowing its nutritional profiles. Therefore, the objective of the study was to determine the nutritional profile of this native plant's edible parts, such as leaf and stem bark by analyzing its proximate composition, mineral nutrients, and phytochemical compounds. The proximate analysis of leaf and stem bark revealed that leaf had higher moisture, crude protein, and crude fat contents than stem bark, whereas stem bark had higher ash, crude fiber, and carbohydrate contents. In terms of mineral nutrients, leaf had higher levels of potassium, magnesium and sodium, but stem bark had higher levels of calcium and sulfur. When taking phytochemicals into account, it was shown that stem bark contained higher levels of antioxidants, flavonoids and phenolic compounds than leaf did. However, vitamin C level was higher in leaf than in stem bark. The findings of the study indicated that the Barun Shak is an important source of nutrition and different phytochemicals. Thus, the ethnobotanical belief in the beneficial impact of Barun leaf and stem bark may be supported, and it will act both as a potent food and medicinal source. As a result, the generated nutritional profiles are being used in nutrition and health-related programs in Netrokona district of Bangladesh.*

Key words: ***P****roximate, Nutrition, Phytochemicals, Ethnobotanical, Barun*

R*eceived 07 May, 2023;* ***R****evised 16May, 2023;* ***A****ccepted 19 May, 2023 © The author(s) 2023.*
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I. INTRODUCTION

Bangladesh is a tropical and agrarian country with rich biodiversity that can be utilized in all aspects of community life. Different types of plants grow naturally around throughout the country. Among them some plants have been used by human since prehistoric times [1] for vegetables, fruits as well as medicinal purposes. Over centuries, cultures around the world have learned how to use plants to maintain health and fight against illness[2,3].These readily available and culturally important indigenous plant form an accessible and affordable health-care regime is an important source of livelihood for rural population of Bangladesh[4].Regarding this WHO has estimated that 80% of the population of developing countries being unable to afford pharmaceutical drugs, rely on traditional medicines mainly plant based to sustain primary health care needs [5].

Barun (*Crataeva nurvala* Buch. Ham.), belonging to family Cappridaceae is deciduous indigenous tree in Bangladesh [6]. These plants are naturally grown in forests, bushes and marginal land along the canal in swamp places. A long tradition of indigenous herbal medicinal systems, based on the rich local plant diversity is considered a very important component of the primary health-care system [7]. This plant has a special importance due to its effectiveness in a wide range of disease [8]. *C. nurvala* has been extensively used in traditional medicines as a blood purifier and in treating cardiac and lung weakness, fever, joint problems, blood flow, memory loss, respiratory complications, metabolic syndromes, wound healing and weak immune system. [9,10].

C. naruvala which is called Barun shak by Bangladeshi people. Rural people of Netrokona district of Bangladesh consume Barun shak as leafy vegetable based on their ethnobotanical beliefs and dietary diversity. They are also familiar to use the leaves, stem bark, root, flower and other parts of this plant for different physical

problems as herbal medicinal purposes in different form without knowing nutritional or medicinal value of this plant. Primary health care of the rural people is covered by Barun with their own choice. Therefore, the study has been undertaken to determine the proximate composition, mineral nutrients, vitamin C, antioxidant, phenolic and flavonoid compounds of Barun grown in Netrokona district of Bangladesh. Findings of this study will help the scientists and researchers to screen the compounds responsible for further evaluation.

II. METHODS AND MATERIALS

Preparation of plant materials

The leaves and stem bark of Barun were collected from naturally growing populations, distributed along the of Netrokona district, Bangladesh. The samples were cleaned by hand properly and sun dried for few days till complete drying. After complete drying the samples were ground into powder using a grinder (IKA® MF 10 Basic Microfine Grinder Drive, Breisgau, Germany) at a dimension of 0.5 mm. Finally, the dried samples were stored in zipper bags and kept in cool and dry place until further analyses.

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Proximate Analysis

The proximate analysis of the samples for moisture, ash, protein, fat and carbohydrate were determined by the method of AOAC (2016) [11].

Determination of total phenolic compounds

Total phenolic compounds amount was measured using the Folin-Ciocalteu method [12]. Barun leaves and stem bark tissue (2 g) was homogenized with 2 mL of methanol and then centrifuged at 10,000 g for 20 min, and 1.5 mL of Folin-Ciocalteau's reagent (is a mixture of phosphomolybdate and phosphotungstate; 10-fold diluted) was added to 0.15 mL of the supernatant. The reaction was neutralized by adding 1.5 mL of 6% (w/v) sodium carbonate. The mixture was incubated at 75°C for 10 min and the absorbance at 760 nm was measured. Gallic acid was used as a standard, and results were expressed as mg of gallic acid equivalents (GAE)/100 g dry weight.

Determination of total flavonoid content

The total flavonoid content was estimated using aluminum chloride colorimetric assay [13,14]. The 0.5 mL of test samples' solution in methanol (5 mg/100 mL) were mixed with 2mL of distilled water and 150 µl of 5% sodium nitrate. After 6 min, 150 µl of 10% aluminum chloride and 2mL of 1 M sodium hydroxide were added and left at room temperature for 15 min. Absorbance of the mixtures was measured at 510 nm) and total flavonoid contents were calculated as rutin equivalents from a calibration curve of rutin. The calibration curve was prepared in the same manner using 0.01562-1 mg/mL of rutin solutions in methanol.

Determination of total antioxidant (phosphomolybdate assay)

The total antioxidant capacity assay of samples was carried out by the phosphomolybdenum assay. A 0.1-ml aliquot of the sample solution was shaken with 1 mL of reagent solution (0.6M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate). The test tubes were covered and incubated in a water bath at 95 °C for 90 min. After the samples were cooled, the absorbance of the mixture was measured at 695 nm [14]. Ascorbic acid was used as standard. The antioxidant capacity was estimated using the following formula:

Total antioxidant capacity (%) = [(Abs. of control − Abs. of sample)/ (Abs. of control] × 100

Determination of ascorbic acid content

Ascorbic acid was determined by the method of [15]. Fresh sample tissues (2 g) were homogenized with 6 mL of 2% oxalic acid and titrated against 2, 6-dichlorophenol-indophenol dye. The ascorbic acid content in samples was determined from the standard ascorbic acid and the results was expressed as mg/100 g FW.

Determination of K, Ca, Mg, S and Na contents

The oven-dried samples of both the leaves and stem bark were ground separately using a mortar and pestle and preserved for subsequent chemical analyses. A sub-sample weighing 1g was transferred into a clean, dry digestion flask. It was followed by adding a di-acid mixture (HNO_3: $HClO_4$ in the ratio of 2:1) of 10 mL to the flask. After leaving for overnight, the flask was heated with a sand bath at a temperature slowly elevated to 180 °C. The substances of the flask were heated to boiling point until wavy white precipitate became visible. The digested sample was allowed to cool, then it was diluted with distilled water and filtered using Whatman No. 42 filter paper. The volume was made up to 100 mL with distilled water and transferred into clean plastic bottle and stored in freezer for later use. From digested solution, Na and K contents were measured with the help of flame emission spectrophotometer (Jenway PEP-7) at 589 nm and 768 nm wavelength, respectively [16]. S content of the samples was quantified by the absorbance reading taken at 425 nm wavelength with a spectrophotometer. EDTA titrimetric method was used to determine Ca and Mg contents [17.18].

Statistical analysis

The data were analyzed and the figure were plotted using Microsoft Excel version 2016.Data were reported as mean ± standard SE (standard error) of four replicates.

III. RESULTS

Proximate composition

The proximate composition in leaves and stem bark of Barun is shown in Table 1. The moisture content in Barun leaves was 69.12% and 63.95% in stem bark. Higher amount of ash content (8.36%) was recorded in stem bark than leaves (5.84%).

Crude protein content was ranged from 7.21% to 4.17% in leaves and stem bark on dry weight basis. The green leafy vegetables are rich sources of crude fibre, an important dietary component for bowl movement, Barun stem bark content higher amount 9.72% of crude fibre than leaves (6.32%). Though vegetables are poor sources of fat. However, significant amount of crude fat was recorded in leaves (0.16%) and in stem bark (0.12%) on dry weight basis. Considerable amount of carbohydrate was found in leaves (17.68%) and in stem bark (23.40%) of Barun.

Table 1. Proximate composition in leaves and stem bark of Barun.

Proximate composition	% of proximate composition in	
	Leaf	Stem bark
Moisture Content	69.12% ± 0.10	63.95% ± 0.16
Ash	5.84% ± 0.14	8.36% ± 0.41
Crude Protein	7.21% ± 0.3	4.17% ± 0.2
Crude fiber	6.32 %± 0.52	9.72% ± 0.27
Crude Fat	0.16% ± 0.08	0.12% ± 0.05
Carbohydrate	17.68% ± 0.54	23.40% ± 0.58

Values are ± standard error (SE); (n =4).

Mineral nutrient contents

Barun is an important traditional vegetable to the rural people of Netrokona district of Bangladesh, which showed a rich source of different mineral nutrients (Table 2). Higher amount of potassium content was recorded in leaves (299.70 mg/100 g) than in stem bark (117.80 mg/100 g) on dry weight basis. Comparatively, higher amount of calcium (276.60 mg/100 g) was found in stem bark than leaves (104.20 mg/100 g). Substantial amount of magnesium content was recorded in leaf (150.70 mg/100 g) and in bark (94.80 mg/100 g). Slightly higher quantity of sulphur was found in leaves (213.80 mg/100 g) than in stem bark (212.90 mg/100 g). Higher amount sodium content was recorded in leaves (33.70 mg/100 g) than in stem bark (20.40 mg/100g).

Table 2. Amount of some mineral nutrient contents in leaves and stem bark of Barun.

Mineral	Amount (mg/100 g DW)	
	Leaf	Stem bark
K	299.70 ± 2.50	117.80 ± 2.40
Ca	104.20 ± 1.80	276.60 ± 2.00
Mg	150.70 ± 1.20	94.80 ± 2.00
S	212.90 ± 2.00	213.80 ± 2.10
Na	33.70 ± 1.40	20.40 ± 1.10

Values are ± standard error (SE); (n =4).

Phytochemical compounds

The phytochemical compounds determined in this study refer to total antioxidants, phenolic and flavonoid; and vitamin C were found rich food source of these phytochemicals (Figure 1). Total antioxidant content was found to be 82.33 mg/100 g in leaves, whereas it was found 148.25 mg/100 g in stem bark. Higher amount of total flavonoid compound was recorded in stem bark (6.21 mg/100 g) than in leaves (2.59 mg/100 g). Total phenolic compounds in leaves and stem bark were recoded 10.04 mg/100 g and 24.00 g/100 mg, respectively. Ascorbic acid content was estimated to be higher in leaves48.74 mg/ 100 g than in stem bark 8.59 mg/100 g.

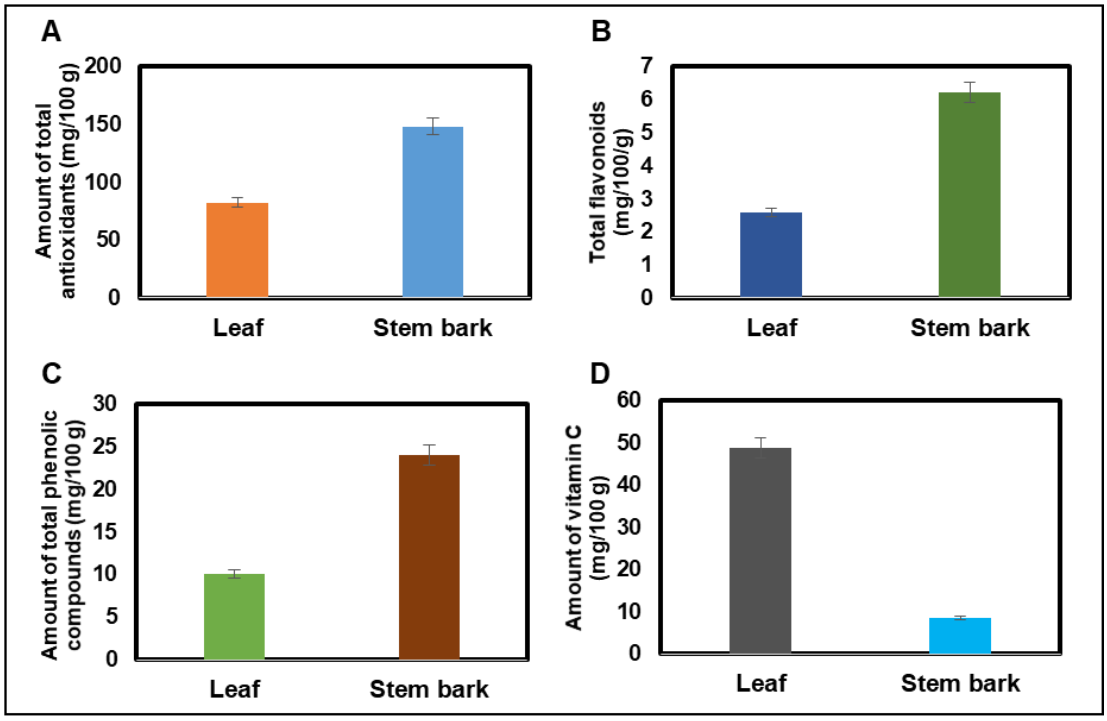

Figure 1. Amount of phytochemical compounds in leaf and stem bark of barun (mg/100 g DW). (**A**) total antioxidants, (**B**) total flavonoids, (**C**) total phenolic compounds, and **(D**) vitamin C. Data are means ± standard error (SE) of four replicates. The SE in each case is represented by the vartical bar in each graph. DW, dry weight basis.

IV. DISCUSSION

The result of proximate analysis (in %) of leaves and stem bark of Barun shown in table 1. In this study, the moisture content of leaves and stem bark was 69.12% and 63.95% respectively. According to [19, 20] moisture content more than 75% in the leaves is mainly accountable for higher degree of food spoilage. In case of Barun less than 75% moisture in leaves and stem bark were observed. In addition, it was lower from the resulting moisture contents of some other leafy vegetables in Bangladesh ranged from 81.06% to 85.49% which indicate Barun leaves and stem bark aid to prolong their shelf life [21].

C. nurvala leaves contain lower amount of carbohydrates (17.68%) compared to stem bark (23.40%). It is however, higher than leafy vegetable like indian spinach and red amaranth, contained only 1.38% and 2.23% of carbohydrates respectively. Carbohydrate constitute a major class of naturally occurring organic compounds that are essential for the maintenance and sustenance of life in plants and animals and also provide raw materials for many industries [22]. Leafy vegetables are good source of carbohydrate because it meets the Recommended Dietary Allowance (RDA) values [23]. That grades the Barun leaves a good source of vegetable.

The crude protein content of leaves and stem bark was 7.21% and 4.17% respectively showed in Table 1. Barun comprises higher protein content in comparison with other leafy vegetables like *Amaranthus* viridus (2.11%),*Chenopodium murale*leaves (2.98%) and *Nasturitium officinale*(2.76 %) [24]. The protein content of some other medicinal plants like *Ranunculus arvensis*, *Equisetum ravens*, *Carathamus lanatus* and *Fagoniacritica*, cannot exceed the protein content of Barun leaves and stem bark [25]. Moreover, red amaranth and Indian spinach contained 4.32% and 2.21% of protein respectively, also lower than the protein content of Barun leaves and stem bark.

Ash content denote the accessible impurities, organic and inorganic matters that help to predict the soluble and insoluble minerals in the samples [26] indicate the mineral content of plant. Barun leaves and stem bark contained 5.84% and 8.36% of ash respectively which is higher than red amaranth 1.68% and Indian spinach 1.05%. These results indicate that Barun has higher mineral content than other common vegetables. Another proximate component, crude fiber of Barun leaves 6.32% and stem bark 9.72% was estimated. It has been reported that the crude fiber offers a variety of health benefits and is essential in reducing the risk of chronic disease such as diabetes, obesity, cardiovascular and other non-communicable diseases [26]. The aerial parts of Barun leaves and stem bark showed rich content of crude fiber and fell in the range of the standard recommendation for fiber in diet.

From the nutritional point of view fats are very important as it is a high energy supplier and help to absorb fat-soluble vitamins [27]. But leafy vegetables always carried low-fat content also supporting poor fat content of Barun leaves and stem bark. A diet providing 1.20% of its caloric of energy as fat is said to be deficient for human being whereas excess fat consumption is implicated in certain cardiovascular disorders. [10].

The result of the total antioxidants capacity, total phenolic compounds. flavonoids and vitamin C were given in Figure 1. In this study total antioxidants of Barun leaves 82.33% and stem bark 148.25% were estimated. Different literatures demonstrated that natural antioxidants collected from leafy vegetables create protection against the action of free radicals [28] also support the phytochemical results of this study. Total phenolic compound of Barun stem bark was higher 24.04% than leaves 10.02%. The presence of phenolic compounds might be responsible for the free radical quenching activity. On the other hand, flavonoids one of the most important classes of phenolic compounds have the antioxidant activity is associated with multiple features of chemical structures show maximum antiradical activity. Flavonoids suppress reactive oxygen formation, chelate trace elements involved in free-radical production, scavenge reactive species and up-regulate and protect antioxidant defenses [29]. Moreover, vitamin C content of Barun leaves 48.76% and stem bark 8.59% were determined. Vitamin C is one of the most powerful antioxidants and important for oxidation reduction processes which play an important role in reducing oxidation for human health [30].

Mineral determination of Barun leaves and stem bark also done and the elemental contents Ca, K, Mg, S and Na were found in appreciable quantities in Table 2. Almost all herbal plants provide health benefits that can be well correlated to their nutritional content. Calcium is an important mineral found 104.20 mg/100g in Barun leaves and 276.60 mg/100g in stem bark, which have many vital roles in human and animal life and the deficiency causes poor development, growth and different abnormalities [31] Magnesium has a great role in the activities of various enzymes that are involved in carbohydrate metabolism and glucose oxidation observed 150.70 mg/100g and 94.80 mg/100g in leaves and stem bark respectively. Barun leaves and stem bark are rich source of S performsvital role in the human body to make hair, nails, and skin cells rigid [32]. To maintain ionic stability of human body, sodium (Na) and potassium (K) act as essential tools. Na and K rich food have a strong relationship to prevent atherosclerosis and hypertension [32,33]. Due to the imbalance of calcium and phosphorus levels in blood vessels, kidney disease and bone disorders may happen. Thus. Barun might be a mineral enrich plant source for food and medicine.

Conclusions

Plants have contributed immensely for food and medicinal purposes. It can be summarized that *Crataevanurvala*contain the important constituent needed to combat variouskinds of infection in human body. The leaves and stem bark of Barunshowed very good potential source of antioxidant, proximate composition, mineral nutrients and ascorbic acid. Thus, the ethnobotanical believe on the beneficial effect of Barun leaves and stem may be evidenced and be act as an active source of food and medicine. Further research can be done to isolate novel bioactive compounds to produce new drugs in next generation against non-communicable diseases.

Funding:
This research has been conducted by the revenue fund of Peoples Republic of Bangladesh, Ministry of Agriculture.

Acknowledgement

The author is grateful to Bangladesh Institute of Research and Training on Applied Nutrition, Ministry of Agriculture for providing financial support to M.A.The authors are grateful appreciate the sincere help of Professor Dr. Mohammad Gulzarul Aziz, Department of Food Technology and Rural Industries, BangladeshAgricultural University (BAU), Mymensingh for assistance in proximate and phytochemical analyses. The authors also thank to Professor Dr. ShohidulAlam, Department of Agricultural Chemistry, BAU for providing facilities of mineral nutrient analysis.

Conflicts of interest: The author reports no conflict of interest.

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