Original Article

Evaluation of nutritional value and antioxidant activity of root and leaf of *Samarakhai* (*Byttneria herbacea* Roxb.): An extra pharmacopoeial herb

Tarun Sharma, Rabinarayan Acharya[1]

PG Department of Dravyaguna Vigyana, J S Ayurved Mahavidyalaya, Nadiad, [1]Department of Dravyaguna Vigyana, Institute of Teaching and Research in Ayurveda, Jamnagar, Gujarat, India

Abstract

Background: *Samarakhai* (*Byttneria herbacea* Roxb.), family Sterculiaceae, is one of the reputed folklore medicinal herbs, found in many parts of India. Although consumed as a vegetable since long time, its root and leaves are not yet reported for its nutritive value and antioxidant activities. **Aim:** The aim of this study was to evaluate nutritional value and antioxidant potential of root and leaf of *B. herbacea* Roxb. **Materials and methods:** Nutritional parameters such as carbohydrate, fat, protein, energy value, calcium, iron, zinc, manganese, phosphorus and vitamin C were evaluated. Antioxidant activity was evaluated through three test methods, i.e., 1,1-diphenyl-2-picrylhydrazyl, ferric-reducing antioxidant power (FRAP) and phosphomolybdenum assay. **Results:** *B. herbacea* roots and leaves showed the presence of total carbohydrate 46.39 g/100 g and 40.12 g/100 g, total fat 0.63 g/100 g and 1.20 g/100 g, true protein 11.46 g/100 g and 10.49 g/100 g, energy content 237.07 kcal/100 g and 213.24 kcal/100 g, iron 821.10 ppm and 889.64 ppm, zinc 9.2 ppm and 47.98 ppm, manganese 329.86 ppm and 474.59 ppm, phosphorus 0.40 ppm and 0.10 ppm and calcium 4856.84 ppm and 14964.49 ppm, respectively. The half-maximal inhibitory concentration values of the methanol extract of root, leaf and ascorbic acid were found to be 217.25 µg/ml, 131.42 µg/ml and 178.88 µg/ml, respectively. In FRAP assay, antioxidant activity of methanol extract of leaf (129.15 µM) was found to be more than root (73.13 µM). **Conclusion:** *B. herbacea* root contains high amount of true protein, carbohydrate and energy value, while micronutrients such as iron, zinc, manganese and calcium are more in its leaf. Both roots and leaves exhibited potent antioxidant activity where the leaves possess more values than the roots.

Keywords: Antioxidant, *Byttneria*, Gandhamardan hills, nutritive value, Samarakhadyam

Introduction

Importance of herbs in the management of human ailments cannot be over emphasized as approximately 30%–40% of today's conventional system been depending on the herbal source for their nutritional and curative purposes.[1] Plants not only provide us phytochemicals of medicinal value but also provide many nutrients and minerals. Leafy vegetables are known as potential sources of minerals and vitamins.[2] Among the different food articles, vegetables are the cheapest available sources of carbohydrates, proteins, vitamins and minerals.[3,4] Minerals are inorganic substances, present in all body tissues and fluids, and their presence is necessary for the maintenance of certain physicochemical processes. Minerals are chemical constituents used by the body in many ways. Although they yield no energy, they have important roles to play in many activities in the body.[5]

Plants, especially fruits and vegetables, are known to possess phytochemicals such as flavonoids and vitamins that exhibit significant amounts of antioxidant activity and that can be utilized to scavenge the excess free radicals from human body.[6]

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Address for correspondence: Dr. Tarun Sharma, PG Department of Dravyaguna Vigyana, J S Ayurved Mahavidyalaya, Nadiad - 387 001, Gujarat, India.
E-mail: tarunsharma1286@gmail.com

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Plants are good sources of natural antioxidants which provide protection against harmful free radicals and have been strongly associated with reduced risk of chronic diseases.[7] Natural antioxidants exhibit many biologically important functions which include protection against oxidative stress and degenerative diseases and are reported to possess antibacterial, antiallergic, antiviral, anti-inflammatory, anticancer, antiaging activity and hepatoprotective properties.[8,9]

Samarakhai (*Byttneria herbacea* Roxb.), family Sterculiaceae, is a branched, unarmed herb with a perennial woody stock, glabrous ovate-acuminate toothed leaves, small pale purple flowers, has been reported for its traditional use as food and medicinal purpose. It is commonly found in peninsular India from Gujarat southwards to Tamil Nadu and in Odisha and Bihar.[10-12] It is also known by *Samarkhoi, Samarkhai, Sambarkai* and *Samar Kayee* by the tribes of Odisha.[13] The vernacular names denote its uses as a favorite odder (khai) of deer (sambar/samar). Different parts of the plant are used to treat different disease conditions such as wounds, fractures, cholera, dysentery, leukorrhea, and swelling.[11,13] The paste of roots is being used topically for wound healing[14] and orally for body pain.[15] Leaves of this plant are used for dysentery, impaction and leukorrhea.[16,17] The whole plant is used in cholera, diarrhea and gynecological disorders.[10,11] Its roots and leaves are being cooked and eaten as vegetable in Odisha.[18]

Recent literature review shows that only few works have been carried out on this herb regarding antioxidant activity. Antioxidant activities of aqueous extract from the leaves, stem, and root of *B. herbacea*, through three favorable *in vitro* test methods, including nitric oxide, catalase, and superoxide dismutase, have been reported. The results indicated that *B. herbacea* exhibits a good antioxidant activity.[19] In spite of its importance as a food source, there are no published reports on the nutritional composition of herb. Hence, the present study reports the nutritional value including antioxidant potential of methanolic extract of root and leaf of *B. herbacea.*

Materials and methods

Collection, authentication and preservation of the sample

Samarakhai, growing in Gandhamardan hill ranges of Bargarh district of Odisha, India, was identified as *B. herbacea* Roxb., belonging to Sterculiaceae family, on the basis of its morphological characters, comparing with the reported characters mentioned in flora of Odisha[12] and with the help of local taxonomist [Figure 1]. The fresh plant samples were collected in the month of September 2017 from its natural habitat. Plant specimen is authenticated by BSI Kolkata with letter no. CNH/2016/Tech. II/68. Herbarium was prepared and submitted to museum of Pharmacognosy Laboratory, I.P.G.T. and R.A., Jamnagar, vide herbarium no. Phm. 6200/16-17, for future reference [Figure 2]. The collected plant samples were shaken to remove adherent soil and dirt. The roots and leaves were separated from the stem and washed under running fresh water [Figures 3 and 4]. Then, they were shade dried, powdered, passed through mesh no. 80 and preserved in an airtight glass container.

Nutritional evaluation

The sample energy value was estimated (in kcal) by multiplying the percentage crude protein, crude lipid and carbohydrate by the recommended factor (2.44, 8.37 and 3.57, respectively) used in the analysis. The caloric value was determined based on the Atwater factor,[20] total carbohydrates content were estimated by phenol-sulfuric acid method[21] and total soluble protein content was estimated by the Folin–Lowry method.[22] Total fat was estimated using the Soxhlet extraction method,[23] total protein content was determined by Micro-Kjeldahl method,[24] Vitamin C by 2,4-dinitrophenylhydrazine method[25] and Vitamin A by official methods of food analysis.[26] Microwave plasma-atomic emission spectrometry was used to determine iron, zinc, manganese and calcium.[27] The vanadomolybdophosphoric acid colorimetric method[28] was used to determine phosphorus content of acid extractions.

Antioxidant assay

1,1-diphenyl-2-picrylhydrazyl radical scavenging

The 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay was performed by the method of Blois.[29] Antioxidant activities of the methanolic extracts of the different samples were expressed as half-maximal inhibitory concentration (IC_{50}), defined as the concentration of the test material required to cause a 50% decrease in initial DPPH concentration. Ascorbic acid was used as standard.

FRAP assay

The ferric-reducing antioxidant power (FRAP) assay was assessed according to Benzie and Strain[30] using a Hewlett-Packard 8453 diode array spectrophotometer.

Phosphomolybdenum assay

The total antioxidant capacity of the methanolic extracts of root and leaf samples was measured spectrophotometrically using the phosphomolybdenum (PM) method.[31]

Results and Discussion

Nutritional evaluation

The results of nutritional analysis of dried root and leaf of *B. herbacea* are presented in Table 1.

The energy value was found 237.07 kcal/100 g in root and 213.24 kcal/100 g in leaf. Lower calorific value of leaf makes it good in the diet of the obese. Carbohydrate, proteins, and fats are the main sources of energy. Fats have the greatest amount of food energy per mass 9 cal/g. Proteins and most carbohydrates have about 4 cal/g.[32] The root was found to be rich in carbohydrate (46.39 g/100 g) and true protein content (11.46 g/100 g) which is comparatively little higher than the leaf, where carbohydrate and true protein are 40.12 g/100 g and 10.49 g/100 g, respectively. Intake of rich carbohydrate diet regularly prevents the formation of ketone bodies and also

Figure 1: *Byttneria herbacea* Roxb. in its natural habitat

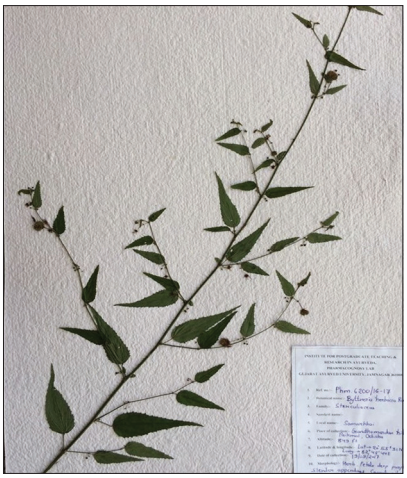


Figure 2: Herbarium specimen of *Byttneria herbacea* Roxb.

Figure 3: Roots of *B. herbacea*

Figure 4: Leaves of *B. herbacea*

Table 1: Nutritional values of root and leaf of *Byttneria herbacea* Roxb.

Parameters	Results	
	Root	Leaf
Energy (kcal/g)	237.07/100	213.24/100
Carbohydrate (g)	46.39/100	40.12/100
True protein	11.46/100	10.49/100
Fat	0.63/100	1.20/100
Protein (%)	8.17	11.29
Vitamin C (mg/g)	23.21/100	23.35/100
Vitamin A (mg/g)	Not detected	1.110
Iron (ppm)	821.10	889.64
Zinc (ppm)	9.2	47.98
Manganese (ppm)	329.86	474.59
Calcium (ppm)	4856.84	14,964.49
Phosphorus (ppm)	0.40	0.10

helps in breaking down of fatty acids.[33] Food proteins have been marketed as functional food ingredients for prevention of various lifestyle diseases and improving human well-being.[34]

Fat was found more in leaf (1.20 g/100 g) than the root (0.63 g/100 g). Lipids help the body to absorb fat-soluble vitamins such as vitamins A and E.[35] Hence, it can be depicted that a diet including leaf of *B. herbacea* should be more palatable than that with root because dietary fats function to increase food palatability by absorbing and retaining flavors.[36] A diet providing 1%–2% of its caloric energy as fat is said to be sufficient to human beings, as excess fat consumption yields to certain cardiovascular disorders such as atherosclerosis, cancer, and aging.[37] Crude protein was found more in leaf (11.29%) than the root (8.17%). According to the Food and Nutrition Board, food plants that provide more than 12% of their calorific value of protein are a good source of protein.[38]

Vitamin A was found only in leaf and not detected in the root of *B. herbacea.* Vitamin A (retinol) is an essential nutrient needed in small amounts by humans for the normal functioning of the visual system, growth and development, immune function, and reproduction.[39] Vitamin C was found slightly more in leaf (23.35 mg/100 g) compared to root (23.21 mg/100 g). The current recommended dietary allowance for Vitamin C

for adult men and women is set at 75 mg/day for women and 90 mg/day for men.[40] Vitamin C is potentially involved in cancer and cardiovascular disease prevention.[41]

Minerals are important in the diet because they serve as cofactors for many physiologic and metabolic functions. The biological effects of the trace elements in living system strongly depend on their concentration and thus should be carefully controlled, especially when herbs and drugs are used in human.[42] The leaf was found to be rich in iron (889.64 ppm), zinc (47.98 ppm) and manganese (474.59 which is comparatively higher than the root where iron, zinc and manganese are 821.10 ppm, 9.2 ppm and 329.86 ppm, respectively. In cellular respiration, iron functions as essential components of enzymes involved in biological oxidation such as cytochromes c, c1 and a1.[43] Iron is involved in synthesis and packaging of neurotransmitters and their uptake and degradation into other iron-containing proteins which may directly or indirectly alter brain function.[44] Zinc-dependent enzymes are involved in macronutrient metabolism and cell replication.[45] Vitamin A and E metabolism and bioavailability are dependent on zinc status.[46] It is needed for tissue repair and wound healing, plays a vital role in protein synthesis and digestion, and is necessary for optimum insulin action as zinc is an integral constituent of insulin.[43] Manganese is a part of enzymes involved in pyruvate metabolism, urea formation, and galactosyltransferase of connective tissue biosynthesis.[47]

Calcium content was found more in leaf (14964.49 ppm) compared to root (4856.84 ppm). According to Mitchell and Curzon, the mean requirement of calcium is 9.8 mg/kg/day or about 640 mg (16 mmol) at a mean body weight of 65 kg.[48] Calcium functions as a constituent of bones and teeth, regulation of nerve and muscle function. A reduced extracellular blood calcium increases the irritability of nerve tissue, and very low levels may cause spontaneous discharges of nerve impulses leading to tetany and convulsions.[43,49] Phosphorus content is found more in root (0.40 ppm) in comparison to leaf (0.10 ppm). Phosphorus is located in every cell of the body and is vitally concerned with many metabolic processes, including those involving the buffers in body fluids.[45]

Antioxidant activity

1,1-diphenyl-2-picrylhydrazyl radical scavenging activity

The DPPH radical has been used widely to test the antioxidant activities of plant extracts. This method is based on the reduction of DPPH in methanol solution in the presence of a hydrogen-donating antioxidant, bringing about a color change from purple to yellow, which is measured at 517 nm.[29] The IC_{50} values of the methanol extract of root, leaf, and ascorbic acid were found to be 217.25 µg/ml, 131.42 µg/ml and 178.88 µg/ml, respectively. Percentage scavenging of DPPH radical was found to rise with increasing concentration of the methanolic extract of root and leaf [Graph 1a and b]. DPPH method allows testing of both lipophilic and hydrophilic compounds[50,51] in comparison to other methods that are restricted in the nature of antioxidants that they can be used to quantify. Based on these facts, DPPH assay is one of the most widely employed methods for screening antioxidant activities of plant extracts.[52]

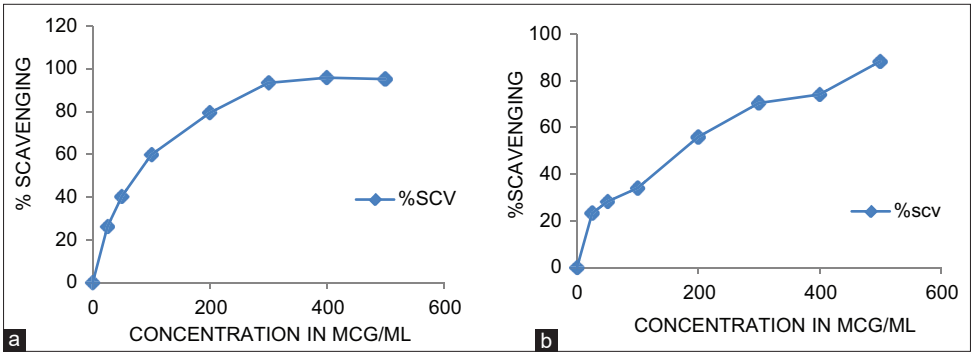
Ferric-reducing antioxidant power (FRAP) assay

In this assay, reduction of the ferric-TPTZ to the ferrous complex forms an intense blue color which can be measured at a wavelength of 593 nm. The intensity of the color is related to the amount of antioxidant reductants in the samples. The ferric-reducing activities of *B. herbacea* root and leaf are 73.130 µM and 129.15 µM, respectively. The absorbance of test drugs clearly increased, due to the formation of the Fe^{2+}-TPTZ complex with increasing concentration [Graph 2].

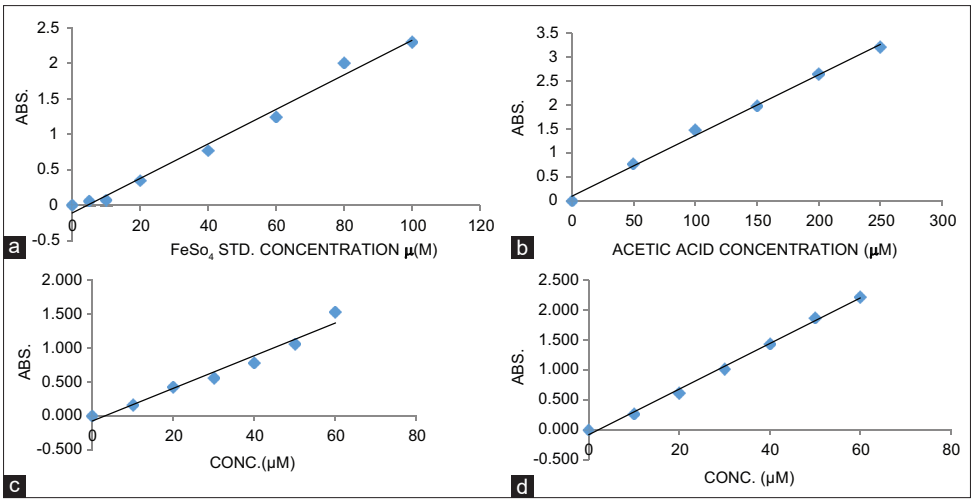
FRAP is an advanced method to assess the reduced concentration of ferric ions. FRAP assay estimates the reducing potential of an antioxidant reacting with a ferric-tripyridyltriazine (Fe^{3+}-TPTZ) complex and generating a colored ferrous tripyridyltriazine (Fe^{2+}-TPTZ).[53] It incorporates the simultaneous use of ferricyanide and ferric ions as chromogenic oxidants supplied more favorable redox conditions for a greater variety of antioxidants. FRAP assay possesses an immediate result of a large range of individual antioxidants in dose–response manner. FRAP presents the antioxidants in the sample as a reductant in a redox-linked colorimetric reaction.[54]

Phosphomolybdenum assay

PM assay is based on the reduction of phosphate-Mo (VI) to phosphate-Mo (V) by the sample and subsequent formation



Graph 1: (a) 1,1-diphenyl-2-picrylhydrazyl radical scavenging activity of *Byttneria herbacea* leaf. (b) 1,1-diphenyl-2-picrylhydrazyl radical scavenging activity *Byttneria herbacea* root



Graph 2: (a) $FeSO_4$ Std. curve at 593 nm. (b) Acetic acid Std. curve at 593 nm. (c) Absorbance of *Byttneria herbacea* root extract at 593 nm. (d) Absorbance of *Byttneria herbacea* leaf extract at 593 nm

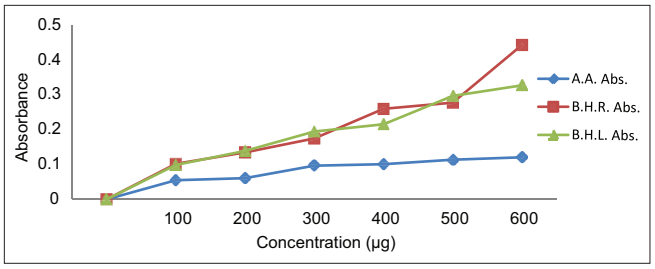
Table 2: Phosphomolybdenum assay of root and leaf of *Byttneria herbacea*

Concentration (µg)	Absorbance at 695 (nm)		
	Ascorbic acid	Root	Leaf
0	0	0	0
100	0.055	0.102	0.099
200	0.061	0.135	0.139
300	0.097	0.175	0.195
400	0.101	0.260	0.216
500	0.114	0.278	0.297
600	0.121	0.444	0.328

of a bluish green-colored phosphate/Mo (V) complex at acid pH. This method is routinely applied in the laboratory to evaluate the total antioxidant capacity of plant extracts.[31] Phosphomolybdenum assay of root and leaf of *B. herbacea* are presented in Table 2. Graph 3 represents the total antioxidant capacity which evaluates the increase in concentration of methanolic extract of root and leaf, reducing capacity of antioxidant.

FRAP and PM both methods are based on the redox antioxidant reaction which allow phyto products to act as reducing agents, hydrogen donators, singlet oxygen quenchers, and metal chelators.[55] PM assay is a quantitative method to explore the reduction reaction rate among antioxidant, oxidant and molybdenum ligand. It forms a green PM complex without induction of free metal ions solution. Hence, it shows uniqueness among *in vitro* antioxidant assays.[56]

In the present era, the use of natural antioxidants for scientific research purpose is increasing. Free radical-induced oxidation can result in cell membrane disintegration, membrane protein damage, and DNA mutation which can further proliferate many life-killing diseases such as cancer, cardiovascular diseases, and liver injury.[57] Hence, one should rely on nature



Graph 3: Total antioxidant capacity by phosphomolybdenum assay. B.H.L.: *Byttneria herbacea* leaf, B.H.R.: *Byttneria herbacea* root, A.A.: Acetic acid

as natural products exhibit unique mechanism of action on these therapeutic problems.

Conclusion

B. herbacea Roxb. is one of the important and widely used medicinal herbs. Root and leaf of the plant are good sources of energy and contain an appreciable amount of nutrients and minerals and can be used as nutritious food in daily life. Its root contains high amount of true protein, carbohydrate, and energy value, while micronutrients such as iron, zinc, manganese and calcium are more in its leaf. Root and leaf of the plant are rich source of Vitamin C and can be used in Vitamin C deficiency. Results indicates that *B. herbacea* have great potential for use as a source of natural antioxidants. Its leaf shows better antioxidant activity than root through DPPH and FRAP assay. Further investigations may be carried out to find active component of the extract and to confirm the mechanism of action.

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Conflicts of interest

There are no conflicts of interest.

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