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PROXIMATE AND MINERAL COMPOSITIONS OF LEAVES AND SEEDS OF BANGLADESHI BOMBAX CEIBA LINN

Shahin Aziz^{1*}, Husna Parvin Nur², Shahal Ahmed³, Aminul Ahsan², Abu Bakar Siddique² and Koushik Saha⁴

¹Chemical Research Division, BCSIR, Dhaka, Bangladesh.

²Bangladesh Council of Scientific and Industrial Research (BCSIR) Laboratories, Dhaka, Bangladesh.

³Department of Applied Chemistry and Chemical Technology, Islamic University, Kushtia, Bangladesh.

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*Corresponding Author Dr. Shahin Aziz

Chemical Research Division, BCSIR, Dhaka, Bangladesh.

ABSTRACT

Bangladesh is a rich store house of medicinal plants. Among them Bombax ceiba Linn. is an important one. In Bangladesh, Bombax ceiba is locally known as "Shimul tree". The whole part of the plant used as traditional folk medicine and has antidysentric, antidiahorreal and antipyretic effects. Taking these things into consideration it creates sufficient interest to carry out analysis for proximate and mineral compositions of leaf and seed of Bombax ceiba grown up in Bangladesh. Proximate analysis of the plant was carried out using

standard analytical procedures. The results of the proximate analysis showed that the leaves of Bombax ceiba have higher contents of crude fiber (28.44%), ash on drying (8.14%), acid insoluble ash (9.30%) and water soluble ash (13.84%) than that of seeds. Whereas seeds have higher carbohydrate contents (40.82%) than leaves of this plant. The atomic absorption spectrophotometer (AAS) was used for quantitative analysis of various elements. Total 13 important elements were analyzed in leaves and seeds of Bombax ceiba. Results indicated the presence of Na, K, Ca, Mg, Cr, Fe, Zn, Al, Cu, Ni, Pb, Cd and Mn in both leaves and seeds. But among the elements, concentration of Ca was found higher in leaves and concentration of K was found higher in seeds. The most important finding of the work is that leaves of Bombax ceiba showed high concentration of all elements than seedsexcept Na, Cu, Cd and Ni. The elemental composition in both leaves and seeds of the plantwere found to be different

⁴Department of Chemistry, Jahangirnagar University, Savar, Dhaka-1342, Bangladesh.

except in case of Ni. Therefore, leaves and seeds of this medicinal plant are enriched in some micro and macro nutrients like Fe, Ca, Na, K, Zn, which are very important for biological metabolic system as well as human health.

KEYWORDS: Bombax ceiba, Proximate analysis, Atomic Absorption Spectrophotometer, elemental compositions.

INTRODUCTION

Herbal remedies are gaining their revival as many sufferers shifting from modern drugs and embracing complementary medicine. World wide most clinical useful prescription drugs are of plant origin. [1] Medicinal plants are a source of great economic value all over the world. Nature has bestowed on us a very rich botanical wealth and a large number of diverse types of plants grown in different parts of the country. [2] In recent years, there has been a gradual revival of interest in the uses of medicinal plants in developing countries because herbal medicines have been reported safe and without any adverse side effect especially when compared with synthetic drugs. Thus a search for new drugs with better and cheaper substitutes from plant origin are a nature choice. [3]

Bombax ceiba is commonly known as silk cotton tree which belongs to the family of Bombacaeae. It is one of the important medicinal plants in tropical and subtropical region in Asia especially in India, Sri Lanka, Pakistan, Malaysia, Myanmar and in Bangladesh. It has number of traditional and medicinaluses in the traditional system of medicine such as Avrveda, Siddha and Unani. [4] The plant is known by different names such as red Cotton tree, Indian kapok tree (English), shalmali (Sanskrit), semal (Hindi), Shimul (Bengali), mullilavu (Malyalam) in different languages. [5] According to Ayurveda, the plant has stimulant, astringent, haemostatic, aphrodisiac, diuretic, antidiarrhoeal, cardiotonic, emtic, demulcent, antidysentric, alterative and antipyretic properties. [6,7] The different parts of the plant, BombaxceibaLinn are used for medicinal purposes for thousands of years in India or subcontinent. A paste of leaves and flowers of this tree is employed as external application for skin trouble. [8] Seed oil is used for the manufacture of soaps and lubrication substances. [9] Seeds are applied on the skin in small pox and chicken pox. [10] Leaves are used as laxative, haematinic.^[11] The tree is a strong light-demander and fast growing. It grows best on deep sandy loams or other well-drained soils, particularly in valleys, in regions receiving 50 to 460 cm annual rainfall well distributed throughout the year. [12] This plant has the compound leaves which are palmate, digitate, large, spreading, glabrous which has common petiole and the size of leaf is 15-30cm long. Five leaflets are common in one leaf but sometimes up to the seven leaflets could be found. Within the capsule the plant has many seeds which are obovoid, smooth, 6-9 mm long in size. The plant has bright red flowers which appear in January to March. The seeds are oily and surrounded by a thick mass of long silky hairs or floss, hence easily blown about by wind. Floss isolated from its fruits is an excellent material for making padded surgical dressings, insulating material for refrigerators, soundproof covers and walls and as being vermin proof: it is the most suitable for making cushions, pillows and upholstery. The plant is best matchwood resource and useful for reclamation of wastelands and mine spoils. So it can also be utilized to improve the barren soil and gain the economic benefits simultaneously. [14,15]

Several studies have been carried out on the isolation of pharmacologically active compounds on leaves and seeds of Bombax ceiba^[5] but no work has been reported about proximate analysis of the leaves and seeds of the plant and elemental compositions of leaves and seeds so far. Keeping in mind the wide application of different plant parts of the plant Bombax ceiba in traditional medicine and ayurvedic preparation, proximate analysis and mineral compositions of leaves and seedsof this important medicinal plant was carried out.

MATERIALS AND METHODS

Collection of plant material

Fully matured fresh leaves and seeds of Bombax ceiba were collected from chamta village of Natore district, Bangladesh in the month of June 2015 and identified by the taxonomist of Bangladesh national Herbarium, Dhaka where a voucher specimen (No.41877) has been deposited. The leaves and seeds of Bombax ceiba were separately air dried. These dried samples of leaves and seeds were powdered using 20 mesh screen in Willey mill and then used for subsequent analyses.

Proximate Composition

Moisture and Dry matter contents

The Moisture content was determined by heating the samples in an electric oven at 105-110°C until constant weight (6-10 hours). [16] The percentage was calculated by

Moisture content (%) =
$$\frac{\text{Weight of moisture}}{\text{Weight of sample taken}} \times 100$$

Dry matter (%) = 100-moisture%

Ash Contents

Ash was determined by incineration of the moisture free samples at about 600°C (about 6-12 hours) in a temperature controlled Muffle furnace until ash becomes almost white or grayish white in color.^[16] The percentage of ash was calculated by

Ash content on dry weight basis(%) =
$$\frac{\text{Weight of ash}}{\text{Weight of sample taken}} \times 100$$

Acid insoluble Ash

Acid insoluble ash was determined by boiling the ash sample of leaves and seeds of Bombax ceiba with 25 ml 3N HCl for 5 minutes and collecting the insoluble matter.^[17] Then they were dried, ignited and weighed. The percentage from the ash taken was calculated by

$$Acid \ insoluble \ ash\% \\ = Ash \ content\% \ (ODB) \\ - \frac{ \ (\text{Weight of ash taken} - \text{Weight of acid insoluble ash}) \times \text{ ash content (\%)} }{ \text{Weight of ash taken} }$$

Water Soluble Ash

Water soluble ash was determined by boiling the ash sample of leaves and seeds of Bombax ceiba with 25ml distilled water for 5 minutes and the insoluble matter was thus collected. Then they were dried, ignited at 450°C and weighed. The percentage from the ash taken was calculated by

$$Water soluble ash\% = \frac{\text{(Weight of ash taken - Wight of water in soluble ash)} \times \text{Ash Content(\%)}}{\text{Weight of ash taken}}$$

Crude Fiber

The moisture and fat free sample (2-5 g, after hot extraction with 40-60°C petroleum ether) of leaves and seeds of Bombax ceiba was digested with 200ml boiling 0.255N H_2SO_4 (1.25% w/v) and 0.313N NaOH (1.25%w/v) solution. After digestion, it was dried at 110°C for 12 hours, ignite at 550°C in a Muffle furnace for 8 hours and recoded the loss of weight^[19]. The percentage of crude fiber was calculated by

$$\frac{\textit{Crude Fibre}(\%)}{\textit{Weight of sample after digesting with acid and alkali - Weight of ash}}{\textit{Weight of dry sample taken}}$$

Carbohydrate Content

Carbohydrate content of leaves and seeds of Bombax ceiba was estimated by subtracting the sum of the protein, fat, ash and crude fiber from the dry sample.

Carbohydrate (%)= 100- (Protein+Fat+Ash+CrudeFibre)

Food Energy

Food Energy of leaves and seeds of Bombax ceiba was estimated according to the equation. [20,21]

Food Energy (FE) = (% of crude protein \times 4) + (% of lipids \times 9) + (% of Carbohydrate \times 4) cal/g

Mineral Compositions

For the determination of mineral compositions in dried leaves and seeds of the plant, we used Nitric acid (69%, Merck India) and Percholoric acid (70%, Merck India) for digestion of our experimental sample without further purification. For standard calibration of respective elements we purchased Na, K, Ca, Mg, Cr, Fe, Zn, Al, Ni, Cd, Mn standard solution (100mg/ml) from Hach (Germany). We prepared the respective desired standard from the stock solution using lab made double distilled water.

Ashing and digestion of plant parts

Accurately weight 2.0g of leaves of Bombax ceiba (previously cleaned) was taken separately in a porcelain crucibleand was heated to about 650°C and cooled and then it was weighed. The crucible with sample was placed in the Bunsen burner (at low flow rate gas) until the smoke ceased. Then the crucible was placed in a temperature controlled muffle furnace at 525°C for about 8-10 hours to obtain carbon free white ash. It was then cooled in desiccators and weighed. This procedure was repeated till a constant weight was obtained and the color of the ash was changed to almost white. The same was done for 2.0 g of seeds of the plant. About 1.0g ash sample for both leaves and seeds of Bombax ceiba were taken separately in 50ml volumetric flask and then 15ml 1M HNO₃ acid was added. Then the flask was placed on magnetic stirrer heater in fume hood for four hours at 250°C. When the color of the solution was changed to milky solutions, it was cooled for 10 minutes and then 7.5ml concentrated perchloric acid (HClO₄) was added. Then it was heated until colorless solution was obtained. For the determination of dissolved elements, the sample was filtered through 0.45 micron filter paper. In all the cases, the pH of the sample was maintained and verified to be less than 2.0 prior to analysis. The standard working solution of interest was prepared to make the

standard calibration curve. Absorption for a sample solution used the calibration curve to determine the concentration of particular element in that sample.

Analytical procedure

Among all elements only Sodium (Na) and Potassium (K) were estimated by using flame photometer (Model AnA-135, OSK, Japan). Most of the elements like Calcium (Ca),Magnesium (Mg), Chromium (Cr), Iron(Fe), Zinc (Zn), Aluminum (Al),Copper (Cu), Nickel (Ni), Lead (Pb), Cadmium (Cd) and Manganese (Mn) in leaves and seeds of our plant samples were analyzed by usingAtomic Absorption Spectrophotometer (Varian, AA 240FS, Australia) which was equipped with flame and graphite furnace. For our experiment, we choose air acetylene flame mode. Thecondition fixedacetylene 1.8 l/min and air 15 l/min, argon gas flow for inert atmosphere. The instrumental default temperature parameters were automatically fixed for each element analysis. For quantitative measurement of each element with its linear working range, its respective wavelength and statistical calibration graph of correlation coefficient are listed in Table 1. Data recorded of respective elements in triplicate measurements for its authentication which was used for standard deviation calculation.

Table1: Operating parameter for working element

Element	Wavelength	Lamp intensity	Slit width
	(nm)	(mA)	(nm)
Na	589.0	12	0.2
K	766.49	10	0.2
Ca	422.7	10	0.5
Mg	285.2	4	0.5
Cr	357.9	7	0.2
Fe	248.3	5	0.2
Zn	213.9	5	1.0
Al	396.2	10	0.5
Cu	324.8	4	0.5
Pb	217.3	10	1.0
Cd	228.8	4	0.5
Mn	279.5	5	0.2
Ni	232.0	4	0.2

RESULTS AND DISCUSSION

Proximate composition of leaves and seeds of the plant Bombax ceiba were recorded and the results are presented in Table 2.

Leaves seeds **Test parameters** Percent (%) **Percent (%) Composition Composition** Moisture 11.87 ± 0.23 9.46 ± 0.02 90.54±0.20 Dry matter 88.13 ± 0.23 8.14 ± 0.10 6.50 ± 0.30 Ash on drying 9.30±0.02 Acid Insoluble ash 7.10 ± 0.05 Water soluble ash 13.84 ± 0.01 10.95 ± 0.02 Crude fiber 28.44 ± 0.50 25.17±0.01 Carbohydrate 32.47±0.30 40.82 ± 0.05 Food energy 290.29±.02cal/g 320.08±.03cal/g

Table2: Proximate composition of leaves and seeds of Bombax ceiba.

Data are expressed as Mean \pm SD (n=3)

The leaves of Bombax ceiba have high crude fiber (28.44%) and high ash content (8.14%) than seeds. The ash content for leaves is 8.14% and forseeds 6.50%. The present result found that the high value of ash in the case of leaves indicateshigh quality of mineral contents. The moisture content in both the case was determined on the fresh weight basis whereas the organic content was calculated on the dry weight basis. Acid insoluble ash is an indication of silicate impurity and water soluble ash indicates the content of soluble minerals. In both cases leaves of the plant showed the higher values than seeds. Also in the current studies, the amount of acid insoluble ash was found low compare to water soluble ash in both cases.

Crude fiber refers to the indigestible carbohydrate component that is present in plants. Fiber is characterized by low or no nutritional value but because of its effect on the digestive system, it is thought to help with such problems in diabetes and high levels of blood cholesterol. [22] In the present study the crude fiber content for leaves (28.44%) and for seeds (25.17%) showed good values.

Like all living organisms, plants require energy in chemical form so they can grow and carry out basic life functions. Plants produce, store and burn carbohydrates in the form of sugar to provide themselves with energy.^[23] The present study showed the presence of carbohydrate content in the case of leaves is 32.47% and in the case of flowers is 40.82%. The Table 2 also presented the food energy in the case of leaves and seeds of this plant are 290.23 cal/g and 320.08 cal/g, respectively.

The elemental compositions of leaves and seeds of Bombax ceiba were determined by using Atomic Absorption Spectrophotometer. A total of 13 elements i.e. Na, K, Ca, Mg, Cr, Fe, Zn, Al, Cu, Pb, Cd, Mn and Niwere analyzed from both leaves and seeds of the plant which are accountable for curing various diseases. The result of the analyses is presented in the Table 3. It may be noted each result is an average of at least three independent measurements. These elements play a vital role in the formation of secondary metabolites which are responsible for pharmacological actions of these elements in both leaves and seeds of the plant.

Table: 3 Mineral compositions (mg/g) of leaves and seeds of Bombax ceiba.

Elements	Leaves (dry weight basis, mg/g)	Seeds (dry weight basis, mg/g)
Na	19.07±0.26	23.77±0.53
K	153.66±5.05	144.44±4.98
Ca	177±5.05	31.05±3.04
Mg	48.15±0.09	41.33±0.26
Cr	0.005 ± 0.0004	$0.001 \pm .001$
Fe	1.54±0.044	0.617±0.083
Zn	27.09±<0.005	17.12±0.002
Al	3.320±0.005	0.831±0.004
Cu	$0.059 \pm < 0.001$	$0.128 \pm < 0.001$
Pb	$0.020 \pm < 0.001$	$0.003 \pm < 0.001$
Cd	$0.0004 \pm < 0.001$	$0.0006 \pm < 0.001$
Mn	0.177 ± 0.001	$0.104 \pm .009$
Ni	$0.012 \pm < 0.001$	0.012±0.001

Measured values are mean \pm Standard Deviation (SD) of three replicate analyses

In the present study it was observed that leaves showed higher concentration of Ca, Mg, Cr, Fe, Al, Pb and Mn compared to seeds of Bombax ceiba. But seeds showed higher concentration of Sodium (Na), Cupper (Cu) and Cadmium (Cd).

Sodium (Na) maintains the osmotic equilibrium between the extra cellular fluid and the tissue cells and maintains the pH of blood within normal limit. It is also concerned with the conduction of nervous impulses, muscle contractility and control of heart muscle conduction.^[25] The average concentration of Na was 19.07 mg/g for leaves and 23.77 mg/g for seedsin the present study.

Potassium (K) is helpful in reducing hypertension and maintaining cardiac rhythm. In the human body, Potassium plays vital role in many physiological reactions and its deficiency or excess can affect human health.^[26] The average concentration of K was 153.66 mg/g for leaves and 144.44 mg/g for seeds in the present study.

Calcium (Ca) overcomes the problems of high blood pressure, heart attack, pre-menstrual syndrome, colon cancer and keeping the bones strong and reduces the risks of osteoporosis in old age. ^[24,26] The average concentration of Ca was 177.0 mg/g for leaves and 31.05mg/g for seeds in the present study.

Magnesium (Mg) improves insulin sensitivity, protects against diabetes and its complications and reduces blood pressure.^[27] Also Mg involves in many enzymatic reactions of oxidative metabolism of nutrients and cell constituents synthesis, transmission of nerve impulses, body temperature regulation, detoxification, energy production and the formation of health bones and teeth.^[28] The average concentration of Mg was 48.15mg/g for leaves and 41.33 mg/g for seeds. Zinc is an important constituent of viable sperm especially human sperm. It is necessary for the growth and multiplication of cells (enzymes responsible for DNA and RNA synthesis, for skin integrity, bone metabolism and functioning of taste and eyesight.^[26] Zinc (Zn) deficiency may contribute to arrested sexual maturation, growth retardation and hair loss, delayed wound healing and emotional disturbance.^[29] The average concentration of Zn was 27.09mg/g for leaves and 17.12mg/g forseeds in the present study.

For the formation of the oxygen carrying protein haemoglobin and myoglobin, the human body needs iron (Fe). It is an essential mineral to prevent anemia and cough associated with angiotensin-converting enzyme (ACE) inhibitors. Also in the synthesis of neurotransmitters such as dopamine or epinephrine and serotonin, Fe is an essential cofactor. In the present study the average concentration of Fe was found 1.54 mg/g for leaves and 0.617 mg/g for seeds.

Manganese (Mn) can help to assist the body in metabolizing protein, helps the diabetic to metabolize carbohydrates and in treating diabetes.^[29] The average concentration of Mn was 0.177 mg/g for leaves and 0.104mg/g for seeds in the present study.

Lead (Pb) is toxic metal and nonessential element for human body as it causes a rise in blood pressure, kidney damage, miscarriage and subtle abortion, brain damage, declined fertility of men through sperm damage, diminished learning abilities of children and disruption of nervous systems.^[29,30] The average concentration of Pb was 0.020 mg/g for leaves and 0.003 mg/g for seeds in our present study.

The value of toxic element Cd for the case of leaves was0.0004 mg/g and in the case of seed was0.0006 mg/g. The maximum limit for Cd is 0.3 mg/kg in herbal medicines and products while the dietary intake limit is 10.3 mg/kg which is prescribed by WHO.^[31]

Cr showed the value for leaves 0.005mg/g whereas in the case of seeds the value was 0.001 mg/g. In the present experiment the toxic elements found to be below the prescribed limits. Excessive intake can cause poisoning in human body. Pb and Cd cause acute and chronic poisoning, adverse effects on the kidney, liver, heart vascular and immune system, brain damage, subtle abortion, declined fertility of men through sperm damage, diminished learning abilities of children and disruption of nervous systems.^[32,33]

The environmental factors including atmosphere and pollution, season of collection samples, age of plant and soil condition in which plant grows. These factors may affect the concentration of elements in the plant region to region.^[34]

CONCLUSION

The proximate analysis of leaves and seeds of the plant Bombax ceiba showed that the leaves of the plant have higher content of crude fiber (28.44%), ash content (8.14%), acid insoluble ash (9.30%) and water soluble ash (13.84%) than that of seeds of the plant. The results found that the higher value of ash content in leaves indicates the presence of high quantity of minerals. These results suggested that the plant parts could be an excellent source of investigated minerals and thus could help in maintaining normal physiological functions of human body. Elemental uptake by a plant is influenced by various factors including types of plant, nature of soil, climate and agriculture practices. [33,34] The concentration of elements is not uniformly distributed throughout the plant. In the current study different elemental concentrations vary due to those factors. This is the first report of proximate analysis and mineral compositions of leaves and seeds of Bombax ceiba and will be helpful in the synthesis of new modern drugs with various combinations of plant parts which can be used in the cure of many diseases ethanomedicinally. Also the different concentration of elements in different parts of Bombax ceiba leads to the conclusion that the plant will have different specific roles in the treatment of different diseases. However more detailed analysis of chemical composition of different part of this important medicinal plant is required.

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