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### Shikha Sharma

Ph.D. Scholar, Department of  
Pharmaceutical Chemistry, IPGT & RA,  
Gujarat Ayurved University, Gujarat,  
India

### Vinay J Shukla

HOD, Department of Pharmaceutical  
Chemistry, IPGT & RA, Gujarat  
Ayurved University, Gujarat, India

### Chanappa R Harisha

HOD, Department of Pharmacognosy,  
IPGT & RA, Gujarat Ayurved  
University, Gujarat, India

### Bhupesh R Patel

Associate Professor, Department of  
Dravyguna, IPGT & RA, Gujarat  
Ayurved University, Gujarat, India

### Correspondence:

#### Shikha Sharma

Ph.D. Scholar, Department of  
Pharmaceutical Chemistry, IPGT & RA,  
Gujarat Ayurved University, Gujarat,  
India  
Email: shikhavishishth@gmail.com

## Pharmacognostic and Preliminary Phytochemical studies on leaf and fruit of *Adansonia digitata* L.

Shikha Sharma\*, Vinay J Shukla, Chanappa R Harisha, Bhupesh R Patel

### ABSTRACT

**Background:** *Adansonia digitata*, the *baobab*, is the well-known tree species of the genus *Adansonia*. **Aim and Objectives:** Review reveals that the leaves and fruit of the plant are used traditionally in the treatment of fever, diarrhea, dysentery, ophthalmic, insect bites, inflammation, and pain. Although it is used traditionally, scientifically the plant is yet to be evaluated for its pharmacognostical characters. Hence, the plant parts were subjected to macro-microscopic, photomicrographic, physicochemical, and preliminary phytochemical tests to fix the quality standards for this drug. **Method:** Plant authentication, pharmacognostical study, physicochemical and phytochemical study was performed by following standard procedures as per Ayurvedic Pharmacopeia of India. **Results:** Microscopy of T. S. of rachis shows circular to orbicular in shape, in the cortical region parenchyma cells heavily loaded by tannin content, cluster and rosette crystals. Schematic outline of T. S. of the leaflet is bulged in the center and consist of the outer epidermis, mesophyll cells differentiated into upper palisade and lower spongy parenchyma while lower epidermis interrupted by anisocytic type of stomata. T.S. of fruit shows the presence of epidermis followed by layers of stone cells and sclerids and T.S. of seed shows outer hard testa, spool cells layer, inner large endosperm. Both samples show Loss on drying less than 6% and very few amounts of acid insoluble ash which directly indicate the amount of inorganic residue. Fruits are slightly acidic in nature. Qualitative tests showed the presence of carbohydrate, tannin, flavonoid, and steroids in leaves and fruits. **Conclusion:** This study would be useful in the identification and authentication of the raw drug.

**Keywords:** Baobab, Bombacaceae, Microscopy, Phytochemistry.

### INTRODUCTION

The Baobab tree *Adansonia digitata* L. is a member of the family Bombacaceae. Bombacaceae (Bombax, Baobab or Kapok family) is a small family of flowering plants which contains about 28 genera and 200 species. Plants of this family are perennial, deciduous and woody trees [1]. The origin of the vernacular name "*Baobab*" is uncertain. However, most scientists believe it is derived from the Arabic name buhibab meaning fruit with many seeds. The genus name *Adansonia* is used in honor of *Michel Adanson* (1727–1806) who brought seeds to Paris in 1754 and who was the first person to provide a comprehensive description accompanied by a drawing of the plant after a trip to West Africa (Senegal). The species name *digitata* (hand-like) was selected in reference to the shape of the leaves [2]. It is medium-sized deciduous tree having smooth light brownish ash coloured bark. Leaves are pubescent beneath when young, glabrous *digitate* [3] leaflets three in young plant, five or seven in older plants [4], flowers soft yellowish- white, fruit is farinaceous from outside, whitish or yellowish sometimes pink-tinged pulp; seed reniform, shining brown or blackish with thick testa [5].

It is native to Africa but also found in India in Gujarat, Utter Pradesh Bihar, Bombay, and Madras [6]. *Adansonia* has been used in traditional medicine since ancient times. Ethno-medicinally the leaves were used to treat a wide variety of conditions including fatigue, as an anti-asthmatic, as a tonic and for insect bites, Guinea worm, and internal pains, diseases of the urinary tract, ophthalmia, and otitis. Internally, they are given as an astringent, sudorific, tonic and febrifuge. As a lotion, they are employed in earache and ophthalmia. An infusion of both the leaves and flower is given in respiratory and digestive disorder [5]. The leaves are also used as fomentation and poultices for rheumatic affection of the limbs and irritable inflammatory ulcers. Fruit pulp has been traditionally used as an immunostimulant, anti-inflammatory, analgesic, pesticide, antipyretic, febrifuge, and astringent in the treatment of diarrhea [5] and used for treatment of smallpox and measles [7]. A refrigerant drink is made from pulp is soluble in water, given as diaphoretic in fever, in diarrhea and dysentery, and in hemoptysis. The dry pulp administered with water, gives considerable relief from chronic bronchial asthma, severe itching in the case of allergic dermatitis,

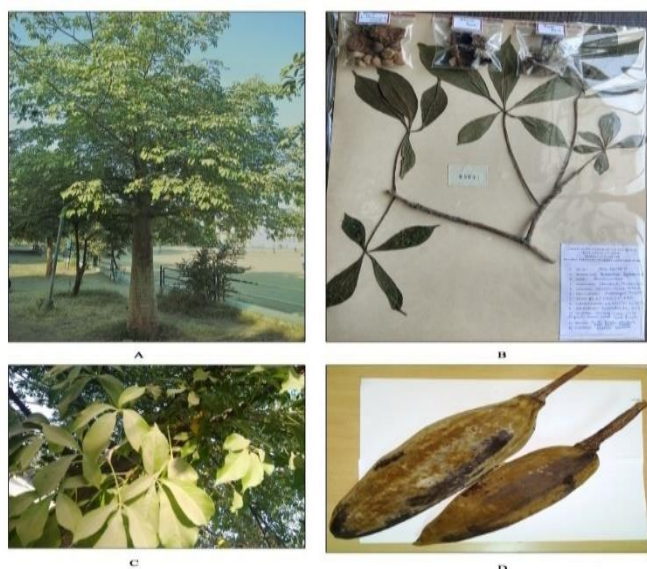
and urticaria [5]. Seeds were used in folk medicine to treat diarrhea, and hiccough while their oil extract mainly used to treat skin complaints and for cosmetic applications [8]. The seeds and oil are used as food, fuel, cosmetics and medicines in the tropical treatment of muscle wounds, dandruff, and other skin ailments [9].

The review reveals that the very few works have been done on the fruit of *Adansonia digitata* [10] whereas leaves are not explored pharmacognostically. Hence, this research article focuses on the pharmacognosy and preliminary phytochemical analysis of leaf and fruit of the *Adansonia*.

## MATERIALS AND METHODS

### Collection and Authentication

Leaves of *Adansonia digitata* L. was collected from its natural habitat Jamnagar, Gujarat in the month of July 2018 while fruits from Bhavnagar, Gujarat during the months of March 2018 with the help of the local taxonomist. Authentication of the plant was done in the Pharmacognosy Laboratory, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University and herbarium of the plant was submitted to the laboratory provided with herbarium reference number no. Ph.M: 6162/18-19 [Fig. 1].



**Figure 1:** (A) Plant in natural habitat; (B) Herbarium Ph.M: 6162/18-19; (C) Leaves; (D) Fruits

### Macroscopic evaluation

Morphological characters of leaves and fruits of *Adansonia digitata* L. were studied as per visual observation, following the standard procedure of taxonomy and verified with existing floras for authentication [11-13].

### Microscopic evaluation

The microscopic evaluation includes the thin free hand transverse sections of rachis and leaflets passing through midrib, fruit pericarp and seed were taken and observed under the microscope. After that sections were stained with phloroglucinol along with hydrochloric acid and iodine solution, respectively and again examined to assess different cellular structure and content. The samples were observed under the compound microscope (Quasma, India) and photomicrography was done.

### Surface study and micrometric evaluation

The surface study was done to determine the type of stomata and stomatal index. For the determination of stomatal index, the upper and

lower surface/epidermal layers of clear leaf piece were peeled out separately by means of forceps and kept on the slide to mount in glycerin water. The epidermal layer was observed under the microscope for the determination of the stomatal type and index [14-16].

Measurement of the length, breadth of stomata, crystal, and trichome was also taken into consideration for micrometric evaluation [11]. The samples were observed under the compound microscope (Quasma, India) and photomicrography was done.

### Preparation of leaf and fruit powder

The leaves and fruits with seeds were shade dried then powdered individually by using the mechanical grinder and sieved through 80# for further powder microscopy, physicochemical parameters, and qualitative tests. The powder was stored in the airtight glass container.

### Powder organoleptic and microscopic evaluation

The colour, odour and taste and texture of the leaves, fruit and seed powder were recorded separately for organoleptic evaluation. Powder microscopy was done by keeping the leaf powder, fruit powder and seed powder (80#) on the individual slide and studied under the microscope. The samples were also examined after staining with different suitable reagents i.e. phloroglucinol along with hydrochloric acid and iodine solution under the compound microscope and photomicrography was done [15].

### Physicochemical and Phytochemical evaluation

Assessment of the various physicochemical parameters such as foreign matter, moisture content, ash value, acid insoluble ash, pH, water-soluble extractive, and alcohol-soluble extractive was carried out by following standard procedures recommended by Ayurvedic Pharmacopoeia of India. Phytochemical evaluation for qualitative analysis of the presence of various secondary metabolites was done as per reference. All determinations were performed in triplicate and the results are presented as mean $\pm$ SEM [14, 17].

## RESULTS

### Morphological study

**Leaf:** leaves are alternate compound, petiolate, petiole 13-19cm long, slightly angular and stipulated. Length of the leaf is 27-35 cm and width is 18-20 cm. Leaflets are tetra to pentafoolate, obovate to lanceolate, 3-17 cm long having an acuminate apex and reticulate venation.

**Fruit:** Matured dried fruit of *Adansonia* is an indehiscent capsule with a long peduncle, shape somewhat conical to lanceolate. The outer surface is hard stony along with slight ridges and covered with a brown velvety surface. Internally chocolate brown contains numerous seeds embedded within the yellowish pulp. Fruits are 45-50 cm long, breadth is 11-12 cm with circumference 37-40cm and weighs up to 450-700gm.

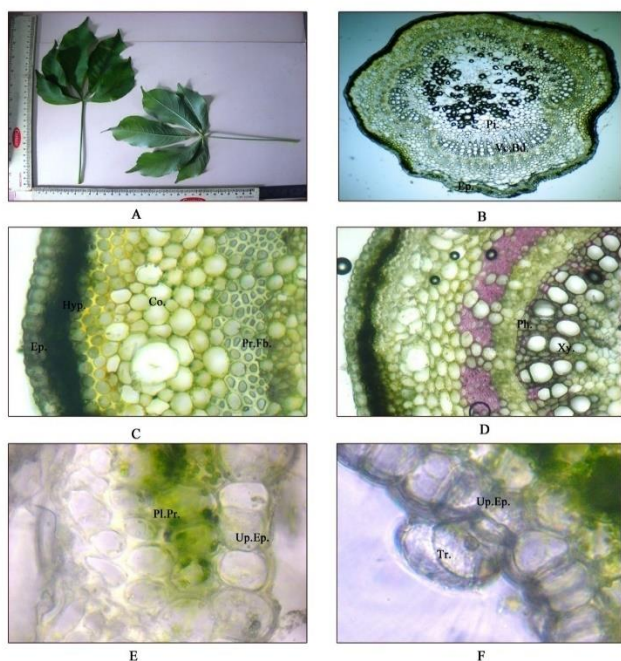
**Seed:** Seeds are Kidney-shaped, the outer seed coat is hard and testa consistent, brownish in colour, rough in touch, internally yellowish white measures about length 0.9-1.1 cm and breadth 0.6-0.8cm. Endosperm is large hook or U shaped with peppery perisperm. Cotyledons are white in colour kidney-shaped measures about length 1.2-1.3cm and breadth 0.4-0.7cm.

### Microscopy study

#### T.S. of Leaf

**Rachis:** The diagrammatic section of rachis is circular with a wavy margin. It consists of the outer epidermis, middle cortex followed by vascular bundle and central pith.

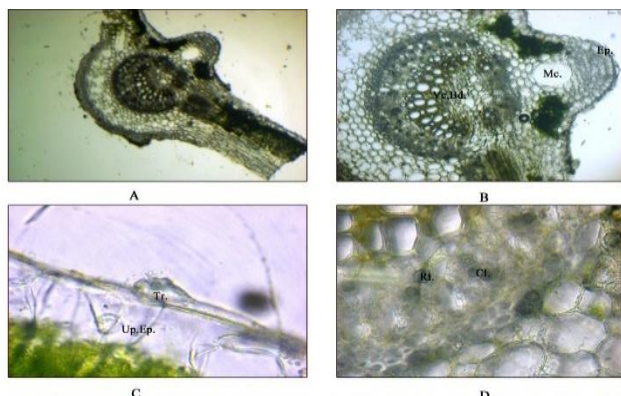
The detail T.S. shows outer single layered epidermis covered with cuticle consist of rounded square shaped compactly arranged epidermal cells intercepted by sessile glandular trichome (1µm × 0.4µm). Cortex immediately after epidermis consist of 3 -4 layered thick walled cells forming hypodermis followed by rounded oval shaped parenchyma cells forming the wide cortex. Some of the cells forming the cavity, later on give mucilaginous latex. Beneath the endodermis, 6-7 layer pericyclic fibers are circularly arranged all over the cortical region. Some part of the pericyclic fiber zone is intercepted by the lignified pitted parenchyma cells. Vascular bundles are the open and collateral type (radially arranged phloem present above the xylem). Phloem made up of phloem fibers and sieve elements and xylem consist of xylem parenchyma and its fibers and centered towards the pith. Pith occupies the larger area of T.S. made up of the parenchymatous cells whereas adjacent to the xylem some of the pith cells are pitted in nature (Fig.2).



**Figure 2:** Microscopic characters of Rachis of *Adansonia digitata* Linn. (A) Natural leaves with measurement (B) Transverse section of the rachis (C) Epidermis, Hypodermis, Cortex, and pericyclic fiber, (D) Phloem and Xylem (E) Upper epidermis and Palisade parenchyma (F) Sessile glandular trichome Pi.-Pith; Vc.Bd.- Vascular bundle; Ep.- Epidermis; Co.-Cortex; Pr.Fb.- Pericyclic fiber; Ph.- Phloem; Xy.- Xylem; Pl.Pr.- Palisade parenchyma; Up.Ep.- Upper epidermis; Tr.- Trichome

**T.S. of leaflet**

Leaflets are dorsiventral. Schematic T.S. of leaflet shows outer epidermis, mesophyll cells differentiated into upper palisade and lower spongy parenchyma. T.S. through midrib shows large vascular bundle present at the center (Fig.3).



**Figure 3:** Microscopic characters of Midrib and surface study of *Adansonia digitata* Linn. (A) Diagrammatic section of T.S. through midrib (B) Through midrib Epidermis, Mucilage cavity, Vascular bundle (C) Upper epidermis along with trichome, (D) Rossete and Cluster crystals (E) T.S. of midrib (stained) (F) Phloem and Xylem (stained), (G) Pitted parenchyma with microcrystal and mucilaginous cavity, (H) Epidermal cells with Anisocytic stomata

Ep.- Epidermis; Mc.- Mucilaginous cavity; Vc.Bd.- Vascular bundle; Tr.-Trichome; Up.Ep.- Upper epidermis; Ph.- Phloem; Xy.- Xylem; Pt. Pr. -Pitted parenchyma; Ani.- Anisocytic stomata; Epi.- Epidermal cells Through midrib

**Epidermis**

Upper epidermis consists of single-layered barreled shape compactly arranged cells. Some of the cells lead to form multicellular glandular sessile trichome. The lower epidermis also consists of epidermal cells as in upper epidermis having trichomes and some of the epidermal cells leads to giving the stomatal opening. Stomata are mainly Anisocytic in nature. Both the epidermis covered with a thin cuticle.

**Mesophyll**

Mesophyll is differentiated into upper palisade and lower spongy parenchyma. Palisade cells consist of 1-2 layer of elongated compactly arranged cells, below the upper epidermis. The lower 2-3 layer spongy parenchyma filled with chloroplast and oil globules. Some of the parenchyma cells loaded with rosette and cluster crystal. T.S. passing through midrib shows outer epidermis followed by 3-4 layers of collenchymatous cells forming hypodermis. Some of the mucilaginous cavities are also observed in the parenchymatous cell. Centrally located readily arranged arc-shaped vascular bundle surrounded by a bundle sheath. 2-3 layered pericyclic fibers are also present around the vascular bundle. Phloem present towards the lower epidermis made up of phloem fibers and sieve elements. Some of the parenchyma cells in the phloem region are loaded with rosette and cluster crystals of calcium oxalate. Xylem present towards the upper epidermis made up of xylem parenchyma and its fibers. Metaxylem is situated towards the lower epidermis whereas protoxylem towards upper epidermis. Medullary rays are uniseriate to triseriate present between the xylem vessels.

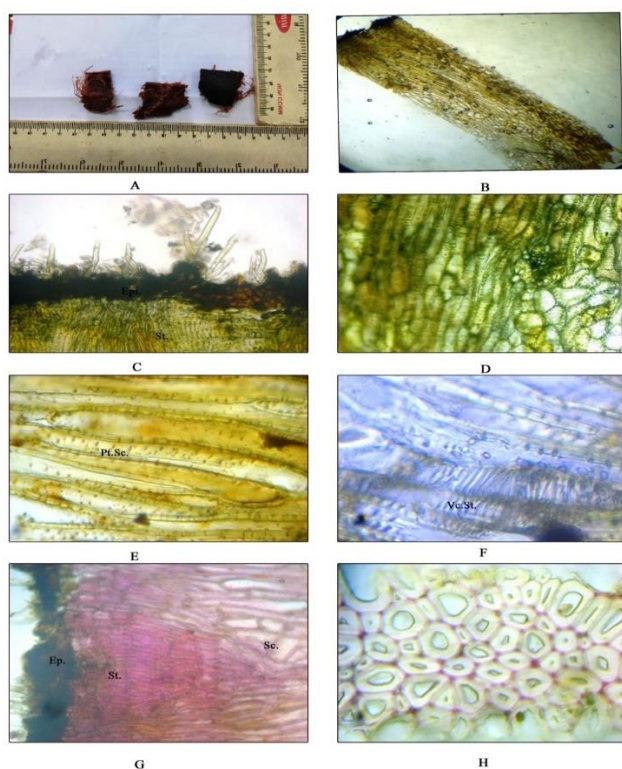
**Surface and micrometric evaluation of Leaf**

Surface study of lower epidermis shows presence of anisocytic type of stomata. Length and breadth of stomata measures about 0.9-1.2 µm × 0.5-0.6 µm. Stomatal index is 22.67 whereas palisade ratio of the leaflet is 4.

**T.S. of Fruit**

Diagrammatic section of the fruit shows outer epicarp, mesocarp, and endocarp.

Epicarp made up of several layered regular shaped compactly arranged cells which greatly consist of brown content (tannin content). Epicarp is covered with epidermis having numerous unicellular and bilobed trichomes. Inner to the epicarp a stony mesocarp layer shows two distinct regions. Below the epicarp, there are several layered different shaped rounds to rectangular with narrow lumen stone cells along with thick lignified pericyclic fibers are present. Stone cells layers are followed by longitudinally elongated thick pitted sclereids with narrow lumen present all over the remaining mesocarp area. The other parenchyma cells of mesocarp area consist of largely brown content and rarely isolated simple starch grains. The vascular bundle which present in mesocarp mainly consists of annular to spiral vessels. Pulpary endocarp consists of numerous seeds distributed all over the fruit. Endocarp mainly consists of parenchyma cells composed of brown content and rarely simple starch grains (Fig.4).



**Figure 4:** Microscopic characters of Fruit of *Adansonia digitata* Linn.

(A) Fruit cut pieces (B) Diagrammatic section of Fruit (C) Epicarp with tannin content and stone cells, (D) Mesocarp with stone cells and sclereids (E) Pitted sclereids (F) Mesocarp with vascular strands (G) Epicarp, Mesocarp with lignified stone cells and sclereids (H) Pericyclic fibers

Ep.- epidermis; St.- stone cells; Pt.Sc.- Pitted sclereids; Vc. St.- Vascular strands

### Seeds

Diagrammatic section of the seed shows outer hard testa, spool cells layer, inner large endosperm. (Fig.5)

### Testa

The outermost part of the seed made up of single layer barrel to uneven shaped compactly arranged epidermal cells forming epidermis embedded by brown content. The epidermis consists of thin cuticle along with papillose like structure all over the epidermis. Epidermis followed by 2-3 layers uneven shaped parenchyma cells forming hypodermis. Hypodermis contains some of the isolated osteosclerides with tannin content. Some of the vascular strands found all over the hypodermis. Beneath the hypodermis, longitudinal elongated compactly arranged single layered spool like cells are arranged

followed by single layered longitudinal elongated compactly arranged lignified palisade-like cells occupied the larger area of testa. Below this aleurone layer is present embedded with aleurone grains on oil globules. Beneath the aleurone layer 2-3 layered collapsed parenchyma cells longitudinal elongated embedded by dark brown content is present followed by a single layer perisperm made up of parenchyma cells.

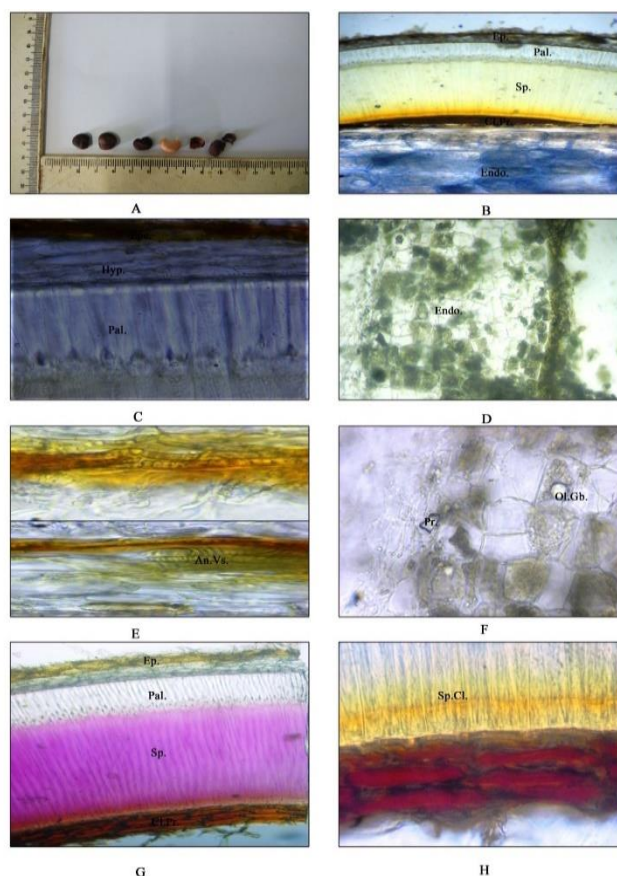
Endosperm consists of single-layered compactly arranged endosperm cells. Endosperm cells are squarous to unevenly shaped parenchyma cells embedded with starch grains, aleurone grains, oil globules, and prismatic crystals.

### Powder microscopy

The organoleptic characters of leaf powder is dark greenish in colour with the faint aroma, salty, Mucilaginous taste and rough in touch while fruit powder is light brownish in colour with slightly fragrant odour, astringent and sour taste and rough in touch. Seed powder is creamish brown in colour with astringent and sour taste; odour is light sour and rough in touch.

### Powder microscopy of Leaf powder

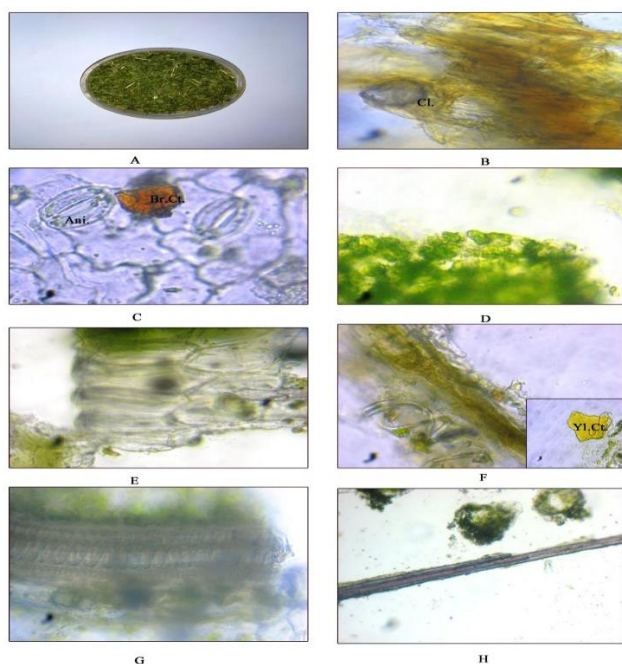
The diagnostic characters of leaf powder show the presence of fragments of anisocytic stomata, fragments of simple fiber, fragments of spongy parenchyma cells, fragments of palisade parenchymatous cells, yellowish brown content, spiral vessels, oil content and lignified fibers (Fig.6).



**Figure 5:** Microscopic characters of the seed of *Adansonia digitata* Linn.

(A) Seed macroscopy (B) T. S. of seed (C) Epidermis and Hypodermis, Palisade like cells, spool cells (D) Endosperm cells with crystals and oil globules (E) Collapsed parenchyma cells with tannin and annular vessels (F) Crystal, oil globules, and aleurone grains (G) Lignified spool cells (H)

Ep.- Epidermis; Pal.-palisade like cells; Endo.- Endosperm; An.Vs.- Annular vessels; Pr.- Prismatic crystal; Ol.Gb.- Oil globule; Sp.- Spool cells

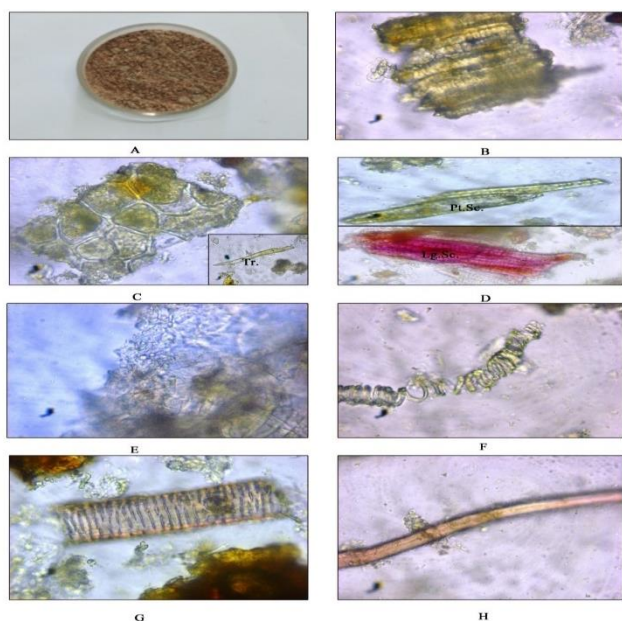


**Figure 6:** Microscopic characters of Leaf Powder of *Adansonia digitata* Linn. (A) Dry leaf powder (B) Cluster crystal (C) Upper Epidermis in surface view with Anisocytic stomata (D) Upper Epidermal cell with chlorophyll, Brown content with Cluster crystal (E) Upper Epidermal cells in tangential view (F) Spiral vessels and yellow content (G) Lignified spiral vessels (H) Lignified fiber

Cl.- Cluster crystal; Ani.- Anisocytic stomata; Br.Ct.- Brown content; Yl.Ct.- yellow content

### Powder microscopy of Fruit powder

The diagnostic characters of fruit powder shows presence of unicellular trichome, oil content, fragments of epicarp cells containing tannin, group of stone cells, group of sclerids, fiber, spiral vessels, simple starch grains with hilum, fragments of endosperm cells, fragments of stone cells, fragments of spool cells, fragments of mesocarp cells, lignified fibers and scalariform vessels (Fig7).

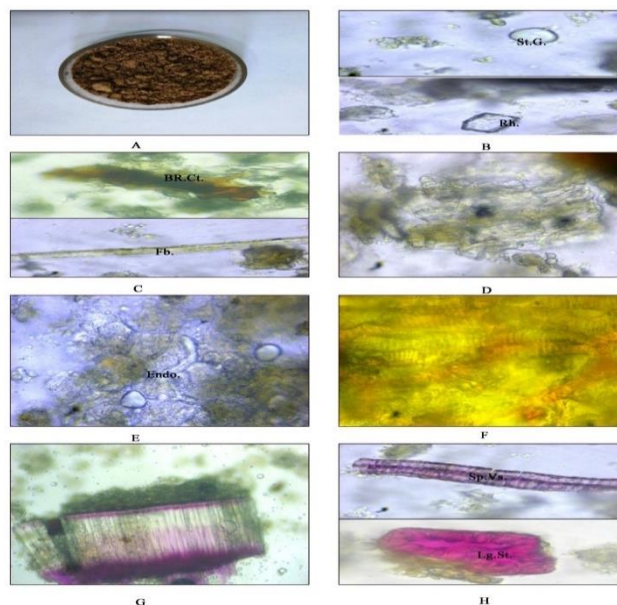


**Figure 7:** Microscopic characters of Fruit Powder of *Adansonia digitata* Linn. (A) Fruit powder (B) Group of Pitted Sclerids (C) Epicarp cells in surface view and Trichome (D) Pitted sclereids and Lignified stone cells (E) Mesocarp cell with oil globules (F) Spiral vessels (G) Scalariform vessels (H) Lignified fiber

Pt.Sc.- Pitted Sclerids; Lg.Sc.- Lignified sclereids; Tr.- Trichome

### Powder microscopy of Seed powder

The diagnostic characters of seed powder show the presence of oil globule, osteosclerids with tannin content and oil globules, stone cells, prismatic crystal, simple fiber, lignified annular vessel, fragments of endosperm cells and the fragment of palisade cells (Fig.8).



**Figure 8:** Microscopic characters of Seed Powder of *Adansonia digitata* Linn. (A) Seed powder (B) Starch grain, Rhombohedral crystal (C) Brown content and Fiber (D) Parenchymatous cells with oil globules (E) Endosperm cells (F) Osteosclerids (G) Fragments of Spool cells (H) Lignified Spiral vessel and stone cell

Rh. – Rhombohedral crystals; St.G.- Starch grains; Sp.vs.- Spiral vessels; Lg.St.- Lignified Stone cell; Endo.- Endosperm cell

### Physico-chemical study

Leaves and Fruit powder of *Adansonia digitata* were subjected to physicochemical parameters like the loss on drying, total ash, acid insoluble ash, alcohol soluble extractive value, pH value, etc. Results are depicted in Table no. 1

**Table 1:** Results of Physicochemical parameter of the Leaf and Fruit

S. No.	Parameter	Leaves	Fruit
1.	Loss on drying (%w/w)	5.92 ± 0.34	5.66 ± 0.55
2.	Ash value (% w/w)	7.97 ± 0.35	4.13 ± 0.22
3.	Acid insoluble ash (%w/w)	0.14 ± 0.01	0.52 ± 0.34
4.	Alcohol soluble extractive(%w/w)	12.03 ± 0.28	4.99 ± 0.16
5.	Water-soluble extractive (%w/w)	19.97 ± 0.17	23.79 ± 1.97
6.	pH (5% aq. Sol <sup>n</sup> )	6.4	5.9

### Preliminary phytochemical study

Preliminary phytochemical results showed the presence of tannin, flavonoids, steroids, and carbohydrate in the root bark and stem bark of *Adansonia digitata*. Results are depicted in Table no. 2

**Table 2:** Results of Phytochemical screening of the Leaf and Fruit

S. No.	Phyto-constituents	Tests	Leaf		Fruit	
			ME	WE	ME	WE
1.	Carbohydrates	Molisch's	+	-	+	-
2.	Proteins	Biuret	-	-	-	-
3.	Amino acids	Ninhydrin	-	-	-	-
4.	Alkaloids	Wagner	-	-	-	-
		Mayor	-	-	-	-
5.	Tannins	FeCl <sub>3</sub>	-	+	+	+
6.	Steroids	Salkowski	-	+	-	+
7.	Flavonoids	Lead Acetate	+	-	+	+
8.	Saponin glycoside	Foam	-	-	-	-

## DISCUSSION

*Adansonia digitata*. is a medium-sized deciduous tree. Leaves are alternate, digitate, so the name *Adansonia digitata* is the key character for identification. Diagrammatic section of the rachis is circular to orbicular in shape, in the cortical region parenchyma cells heavily loaded by cluster crystals, rosette crystals, tannin content are the key characters. Diagrammatic section of the leaf through midrib is circular to oval with two Lamina wings, Upper Epidermis made up of thick compactly barrel-shaped cells interrupted by glandular and simple trichomes and also some of the stomatal openings present in the lower epidermis exclusively anisocytic. Through the midrib beneath the epidermis 3-4 layers of collenchyma tissues present, largely deposited by the cluster crystals, rosette crystals followed by few layer parenchymatous cells are the key characters. The peeled lower epidermis of the leaf shows stomata of anisocytic type are the key characters. Leaf powder shows the presence of cluster crystals, rosette crystals, tannin content, glandular and simple trichomes anisocytic type epidermal cell with brown content, the fragment of the spiral vessel, are the key characters. Fruit of *Adansonia* is an indehiscent capsule. It consists of epicarp, mesocarp, and endocarp. Epicarp extremely hard and with several layers of stone cells and sclereids and pericyclic fibers which forms the fruit very hard. Mesocarp and fleshy endocarp consist of a large amount of tannin. vascular bundles are the distinguished characters of the fruit. Trigonous seeds with outer hard testa with papillose like outgrowth followed by hypodermis and large spool cells layer. Endosperm with squarish shaped endosperm cells with oil globules starch grains, prismatic crystals, and aleurone grains are the important characters. Fruit powder shows the presence of fragments of spool cells and sclerids while seed powder shows the presence of osteosclerids is the key character of identification.

Phytochemical analysis of whole plant powder exhibits various results viz., moisture content is less than 6%. Both samples show very few amounts of acid insoluble ash which directly indicate the amount of inorganic residue. Water-soluble extractive value has been found more in fruit sample whereas leaf shows higher alcohol soluble extractive value. Extractive value gives an idea about the type of chemical moieties present in the plant. Fruits are slightly acidic in nature. Qualitative tests showed the presence of carbohydrate, tannin, flavonoid, and steroids in leaves and fruits.

## CONCLUSION

*Adansonia digitata* is a medium-sized deciduous tree. Leaves are alternate, digitate are the key identification character of the plant. The typical characteristic of leaf, upper epidermis made up of glandular and simple trichomes, while lower epidermis interrupted by anisocytic type of stomata. Fruit is hard with stony endocarp covered with a velvety surface. The typical characteristic of fruit is the presence of layers of stone cells and sclerids and in seed presence of spool cells, palisade-like cells and osteosclerids in the hypodermis. The results obtained

from the physicochemical parameter, qualitative, and quantitative study will serve as standardization values providing information regarding authentication and act as standards for quality assurance.

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## Conflict of Interests

All the authors declared that there is no conflict of interest

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