

## AN OVERVIEW: IDENTIFICATIONS AND ACTIVITIES OF CALOTROPIS PROCERA LEAVES

Md. Semimul Akhtar\* and Soeb Hussain

Shri Ram Murti Smarak College of Engineering and Technology (Pharmacy), Bareilly (U.P.),  
India.

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### \*Corresponding Author

Md. Semimul Akhtar

Shri Ram Murti Smarak  
College of Engineering and  
Technology (Pharmacy),  
Bareilly (U.P.), India.

### ABSTRACT

*Calotropis procera* is also known as the Crown Tree, Swallow Tree, Apple of Sodam, giant calotrope belonging to Apocynaceae family and subfamily is Asclepiadoideae that is native to North Africa, tropical Africa, Western Asia, South Asia, and Indochina.<sup>[1]</sup> It is having a number of therapeutic uses like it act as an anti-inflammatory activity, as antipyretic, in elephantiasis, as anticholinergic, in treatment of peptic ulcer, antiobestic, as antidiabetic. It also work as antidote in snake bite and it also shows anticancer activity. It is useful in treatment of disorders related to respiratory, circulatory and neurological disorders, effective in skin treatment, vomiting, nausea, diarrhea, latex

possesses wormicidal activity and thus may be useful as an anthelmintic.<sup>[2][3]</sup> A study is perform on *Calotropis procera* leaf powder and leaf extracted in different solvent systems such as petroleum-ether (60-80°C), benzene, chloroform, ethanol, and sterile water. Preliminary phytochemical identification was done along with measurement of the leaf constants, fluorescence characteristics, and extractive values. Quantitative estimation of total ash value, acid insoluble ash, and water- soluble ash may serve as useful indices for identification of the powdered drug. Histochemical studies which give information about the rows of cylindrical, palisade cells and vascular bundles may also help for identification of the tissues. These preliminary identification great value in quality control and formulation development of *Calotropis procera*.<sup>[4]</sup>

**KEYWORDS:** *Calotropis procera* leaves, macroscopy, microscopy, ash values, extractive value, extractions, preliminary phytochemical identification.

## INTRODUCTION

*Calotropis* was formerly placed in the family Asclepiadaceae (the milkweed family), which is now considered a subfamily of the Apocynaceae. *Calotropis procera* (giant milkweed) is a perennial, greyish-green, woody shrub with broad fleshy leaves.<sup>[6]</sup> The Apocynaceae is a large family of plants including 415 genera and about 4555 species distributed largely throughout the tropics but also in warm temperate climates. It is very common throughout the drier parts of West and East Africa, but much less common towards southern Africa. It also occurs naturally from northern Africa east throughout continental Asia to South-East Asia. It is sometimes planted outside its natural area of distribution area; e.g. in subtropical America and Australia where it was introduced possibly as an ornamental but it has since escaped and naturalized. It is considered as toxic plant in countries like South Florida and USA.<sup>[24]</sup>

- 1. Synonyms:** - Madar Tree, Crown Tree, Swallow Tree, Apple of Sodom auricula tree, cabbage tree, calotrope, calotropis, Dead Sea apple, giant milkweed, Indian milkweed, kapok tree, King Edward's crown, king's crown, king's crown kapok, Prince of Wales' crown, rubber bush, rubber plant, rubber tree, rubberbush, small crown flower, small crownflower, Sodom apple, Sodom's milkweed, swallowwort.<sup>[4]</sup>
- 2. Biological name:** - *Calotropis procera*.
- 3. Kingdom:** - Plantae.
- 4. Order:** - Gentianales.
- 5. Family:** - Apocynaceae (Milkweed).
- 6. Subfamily:** - Asclepiadoideae.
- 7. Genus:** - *Calotropis*.
- 8. Species:** - *C. Procera*.
- 9. Height:** - 4 meter long (13 feet).

It has been used widely in India in traditional medicine to treat many ailments and has been recommended for the treatment of rheumatism, lupus, eczema, asthma, leprosy and syphilis.<sup>[7]</sup> It is used in the treatment of pain as well as inflammation and to aid digestion. The latex of the plant is used as an antidysenteric, antirheumatic, a diaphoretic, an expectorant and for the treatment of bronchial asthma and skin conditions.<sup>[26]</sup> In African and Asian countries, the latex of *C. procera* is utilized as an arrow poison, molluscicide, a fungicide, an anti-syphilitic, an anti-inflammatory, antipyretic, a purgative, as well as in the treatment of leprosy, diabetes and bronchial asthma.<sup>[18]</sup>

Dried latex and root are used as a snake poisoning antidote. It is also used as an abortifacient, and for the treatment of piles and intestinal worms. It also shows anthelmintic activity. Dried leaf powder is sprinkled on wounds to improve healing and burnt ground leaves effectively reduce pain and swellings in rheumatic joints.<sup>[8][11]</sup>



**Figure 1: *Calotropis procera* plant and leaves.**

*Calotropis* is a spreading shrub or medium-sized tree having 2.5 to 6 m in height. It has a deep taproot and a secondary root system with woody lateral roots that can regenerate rapidly when the plant gets injured. The stems are crooked and covered with a fissured corky bark. The grey-green leaves are 15-30 cm long and 2.5-10 cm broad and have a succulent and waxy appearance, hence the name *procera*, which means wax in latin.<sup>[9][10]</sup> The flowers at the base are pentamerous, thin, translucent, or greenish white and purple violet at the lobe end. The fruit is a fleshy and inflated, up to 10 cm or more in diameter.<sup>[10]</sup>

## **METHODS AND MATERIALS**

### **Plant materials**

Fresh leaves were collected from growing plant of the *Calotropis procera* plant in the medicinal garden of Shri Ram Murti Smarak College of Engg. And Tech., Bareilly, U.P., India. The plant specimens was authenticate and the leaves was subjected to wash under tap water and then air-dried. The fresh leaf was subjected for the microscopic analysis, macroscopic analysis, for extraction preliminary phytochemical identification.<sup>[17]</sup> Some leaves were converted in to the powdered form for microscopic characteristics. The leaves

which were subjected for extraction they are converted into the coarse powdered form by crushing them.

### Instrumentation and techniques

Leaf specimens were cut into the rectangular pieces which should have midrib and a portion of the lamina. Specimens measuring 0.05 cm<sup>2</sup> were cut from the midrib portion of the lamina for paradermal parts. The leaf specimens were fixed and embedded in paraffin blocks<sup>[12]</sup>, followed by dehydration, infiltration, and sectioning<sup>[13]</sup> and finally staining and photographing of the sections.<sup>[14]</sup>

### Morphology of *Calotropis procera* leaf

The leaves have a waxy appearance and contain a milky white sap (i.e. latex). Branching occurs from the base of the plant upwards. Younger stems are greyish-green in colour, smooth in texture, and have a covering of whitish coloured hairs (i.e. they are hoary). Mature stems have a deeply fissured, cork-like, bark that is light brown in colour.<sup>[16]</sup>

The broad, relatively thick, leaves (5-30 cm long and 4-15 cm wide) are also coloured grayish green with maximum margins. These leaves are rounded (i.e. orbicular) or egg-shaped in outline (i.e. ovate) with shortly-pointed tips (i.e. acute apices). Their upper surfaces are mostly hairless (i.e. glabrous), while their undersides may be densely covered in tiny white hairs or have a tuft of stiff hairs at the base of the central vein (i.e. midrib).<sup>[4]</sup>

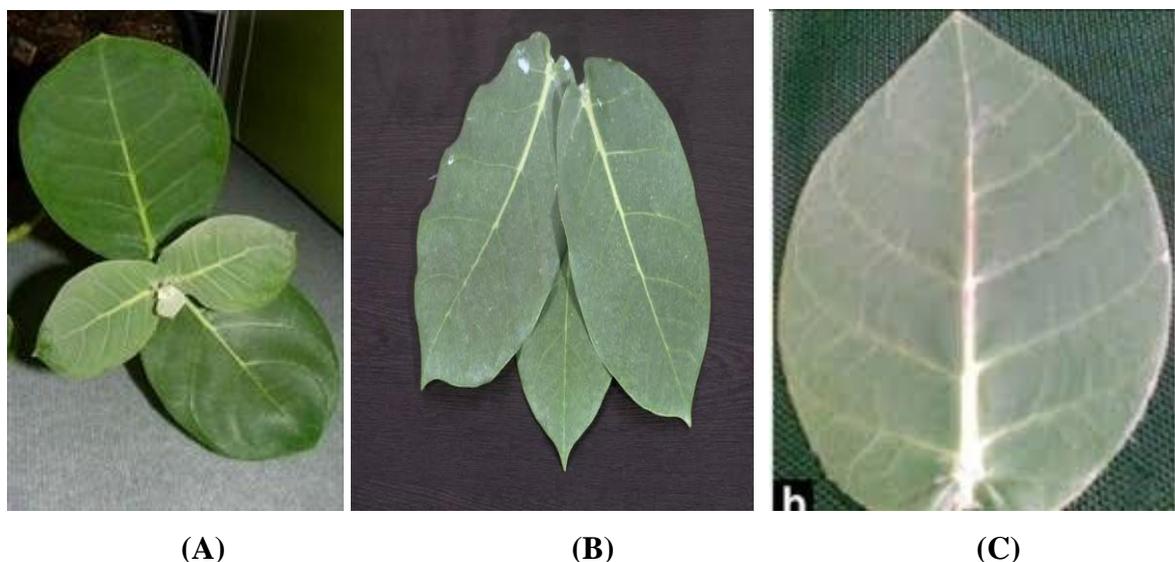


Figure 2: Morphology of *Calotropis procera*.

(A) Showing fresh waxy leaves, (B) ovulated and hairless, (C) leaf showing pink vertex and venation pattern respectively.

**Table 1: Microscopic Characteristics of *Calotropis procera* leaf.**<sup>[21]</sup>

S.No	Microscopic Characteristics	Description
1.	Epidermis:	Epidermal cells are small in size, polyhedral. Shows presence of thick cuticle. Trichomes are less developed.
2.	Collenchyma:	3-5 layered collenchymatous cells present on both sides of upper and lower epidermis.
3.	Vascular bundles:	Stele crescent shaped consisting of two collateral and open vascular bundles. A strip of exchange present between xylem and phloem tissue.
4.	Parenchyma:	4-8 layered parenchymatous cells are present which are thin walled, isodiametric to circular with intercellular spaces present.
5.	Lamina:	Mesophyll is seen to be differentiated into palisade and spongy tissue. Below upper epidermis there are 4 rows of palisade mesophyll tissue.

#### Determination of Ash value of *Calotropis Procera*

Take and weight the silica crucible, weight a known amount of the *Calotropis procera* leaves powder. Heat it with the help of a Bunsen burner at low Flame, until the material gets charred. It transferred into the muffle furnace and heated strongly at a dull red heat (400-500<sup>0</sup>C) till a white ash was obtained. It was cooled in a desiccators for about 15 to 30 minutes, and weight by the help of electronic balance and the readings are noted down.<sup>[23]</sup>

$$\text{Total ash value} = Z - X/Y * 100$$

Where Z= Weight of crucible with ash.

X= Weight of ash.

Y= Weight of crude drug.

#### Determination of acid in soluble ash value of the *C. procera* leaves

Total ash was boiled with 25 ml of 25% hydrochloric acid for 15 minute. Then filter it and insoluble matter was collected to ash less Whatman filter paper, ignited in sintered crucible and then kept in a desiccators for 15 minute. The residue was weighed in electronic balance and the acid insoluble ash was calculated

$$\text{Acid insoluble ash value} = A/Y * 100$$

Where A= weight of residue obtained.

Y= weight of crude drug.

### **Florescent studies of powdered *Calotropis procera* leaves**

The dried powder of *Calotropis procera* leaves then passes through sieve plate number 120 and then goes for studies.

A pinch of powder was taken in a test tube with 10 ml of solvent likewise several test tubes were made by adding various solvent like alcohol, methanol, ethyl acetate, chloroform, benzene, hexane, water, hydrochloric acid and concentrate sulphuric acid.

All test tubes were shaken and allow for incubation for about 30 minutes. Color of drug solution thus obtained observed for their characteristic color reaction under the visible light (florescent tube) and the UV light (UV<sub>356</sub>) and were recorded by comparing with standard color chart.<sup>[15]</sup>

### **Methods of extraction and determination of extractive value**

#### **Extractive value**

Accurately weight 100gm of powdered leaves of the *Calotropis procera* were extracted successively in a Soxhlet with solvent like alcohol, methanol, chloroform, petroleum ether and distilled water separately. Extracts were allowed to dry at room temperature .After evaporation of solvent, weight, color and nature of the extracts were noted.

Extractive value (%) = weight of residue /weight of \*100

Extraction of leaf of *C.procera* was done with water, ethanol 60% and chloroform. The leaf powder (10g) were dissolved in 100 ml of each solvent. The suspended leaves solutions left to stand for 5 days for maceration process and labeled accordingly. The extracts were filtered and stored at 4<sup>0</sup>C for further use.<sup>[25]</sup>

### **Phytochemical Studies**

Firstly the leaves were washed by 5% mercuric chloride solution for about 5 minutes after that wash with sterile distilled water for purpose to remove the dirt or small dust particles, air dried and then converted into the fine powder after proper drying. Then powdered drug was extracted and the solvent were evaporated in rotary evaporater, the residue subjected to the phytochemical analysis.

#### **Test for alkaloids (Mayer's test)**

About 2-3 ml of leaf extract in a test tube, 3-4 drops of Mayer's reagent are added in the test tube containing extract. Orange color indicates presence of alkaloid.

- **Test for cardiac glycosides (Keller-Killiani test)**

2-3ml of leaf extract in a test tube, treated with 1ml of glacial acetic acid having one drop of ferric chloride solution. A deoxy sugar characteristic can be indicated by the presence of brown ring. Just below the brown ring a violet ring may appear, while in the acetic acid layer, a greenish ring may form slowly throughout thin layer, which indicates the positive test for cardiac glycosides.

- **Test for Tannins**

The small amount of leaf extract in a test tube, mixed with basic lead acetate solution. White colour precipitate shows the presence of Tannins.

- **Test for Saponins**

2g of the powdered leaf weigh and transferred into a conical flask, 20ml of sterile distilled water was added and boiled the mixture on water bath for 5 min. Then filter the mixture by the help of filter media and then take 2.5ml of the filtrate which was added to 10ml of sterile distilled water in a test tube. The test tube was closed and shaken for 30 seconds vigorously. Test tube having mixture stand for half an hour at room temperature. Honeycomb froth indicated the presence of saponins.

- **Test for Flavonoids**

2.5 ml of dilute ammonia solution was added to the aqueous filtrate of plant extract followed by addition of concentrated  $H_2SO_4$ . Yellow colour shows the presence of flavonoids.

- **Test for Steroids**

2 gram of plant extracts dissolved in small amount of acetic acid in a test tube. Simultaneously test tube gently warmed and cooled under the running tap water; add a drop of concentrated sulphuric acid in the test tube along with the sides. Green colour indicates the presence of Steroids.

- **Test for Terpenoids (Salkowski test)**

5ml of each plant part extract was mixed in 2 ml of chloroform, and concentrated  $H_2SO_4$  (3ml) was carefully added to form a layer. Formation of reddish brown color shows presence of terpenoids.

## RESULTS

The various studies which are performed on the leaves of the *Calotropis procera* includes the microscopically evaluations has also made which tells about the anatomical studies like presence of epidermis, xylem, inner and outer phloem and presence of trichomes. Epidermis cells are small in size, chlorenchyma (3-5) layered present on the both upper and lower epidermis, parenchyma present (4-8) layered, parenchymatous cells and mesophyll tissue present in leaves of *Calotropis procera*.

### Microscopic and morphological characteristics of *Calotropis procera* leaf

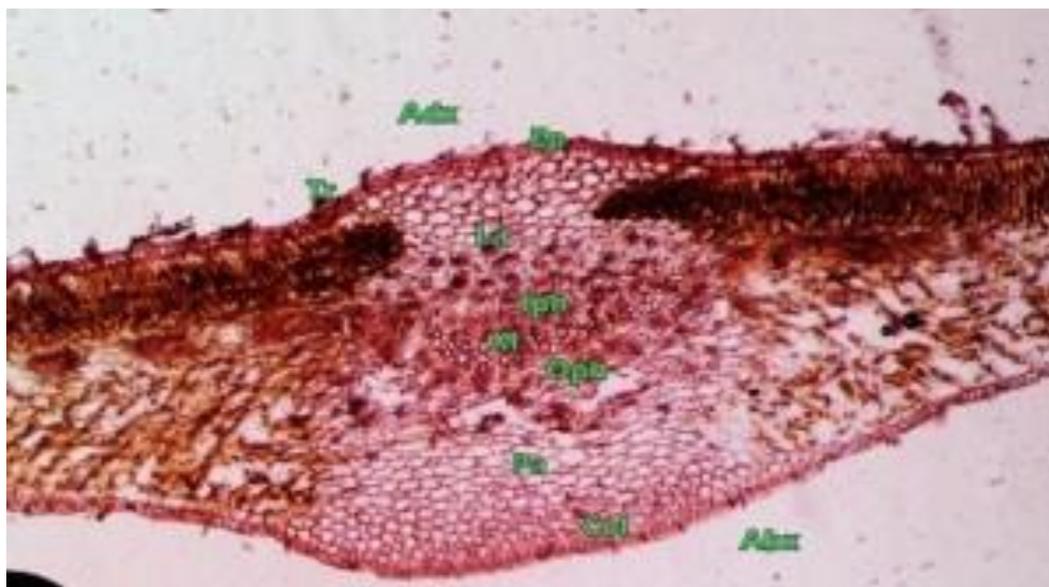


Figure 3: T.S of *Calotropis procera* leaf midrib.<sup>[21]</sup>

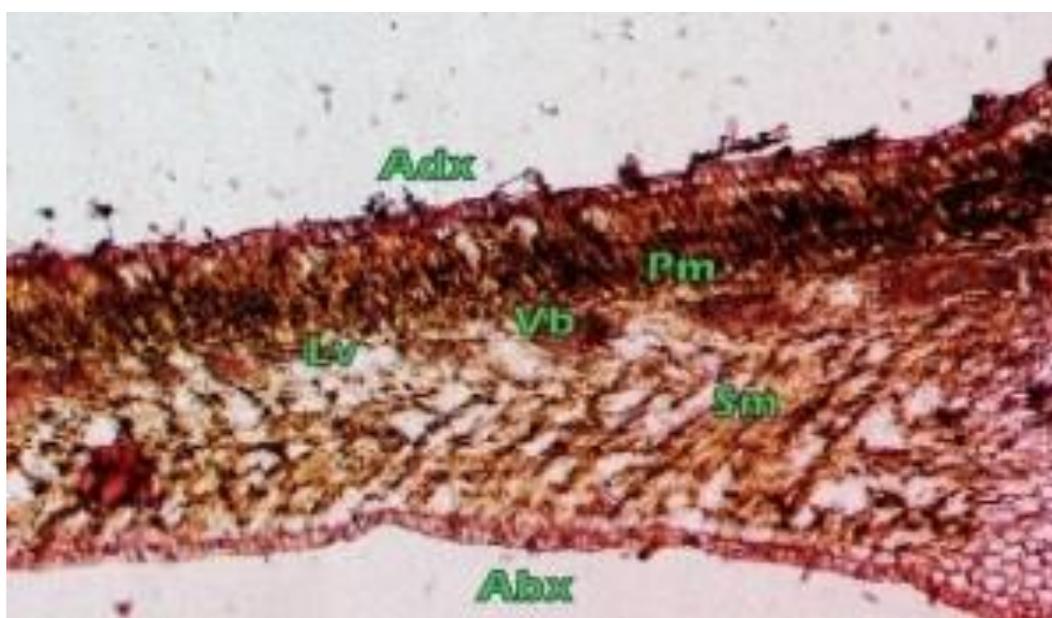


Figure 4: T.S OF *Calotropis procera* leaf lamina.<sup>[21]</sup>

**Table 2: Leaf constant *Calotropis procera* leaf.**

Leaf constant	Value
Stomatal index	44
Stomatal number	11
Palisade ratio	14.8
Vein islet number	25.2
Vein islet termination number	19.7

**Table 3: Ash value of leaf of *Calotropis procera*.**

Type of ash	Ash value (%)
Total ash	18.3
Acid insoluble ash	1.6
Water soluble ash	1.9

**Table 4: Fluorescence of *Calotropis procera* leaf in different solvent systems.**

Treatment	Under visible light	U.V.light(short wavelength;254)
Powder as such	Green	No change
Powder + 1N NaOH (Aqueous)	Light green	Green
Powder + 1N NaOH (ethanolic)	Pale green	Light green
Powder + 1N HCl	Green	Green
Powder + 50% HNO <sub>3</sub>	Brown	Green

**Table 5: Phytochemical studies perform on leaf extract of *Calotropis procera*.**

Phytochemicals	Petroleum ether extract	Distilled water extract	Ethanol extract	Chloroform extract
Alkaloids	–	–	+	–
Glycosides	+	+	+	+
Saponins	–	+	+	–
Tannins	+	+	+	+
Flavonoids	–	+	+	–
Steroids	+	–	+	–
Terpenoids	+	+	+	+

Where (+): Positive, (-): Negative test respectively.

## CONCLUSION

The leaves are the important source of primary metabolites, and secondary metabolites like alkaloids, glycosides, saponins, tannins, flavonoids, steroids, terpenoids has been found to be present in different solvent system extract. The anatomical studies like leaf constants, stomata index and quantitative estimation like extractive value, ash value, fluorescence, and preliminary phytochemical identification help in the of this drug. These studies have been found to be very much important part in the discovery of new herbal drugs and also have a

vital role in primary health care system of medicines. These studies also suggested that observed pharmacognostic and physiochemical parameters having great value in quality control and formulation development. Phytochemical studies will help in the identification of chemical constituents. The above studies may be prove helpful in the with regards to its identification, standardization and also having a great value in the further research on this plant and plant parts and also use in the Ayurvedic System of medicine. These above studies are also helpful in development of the dosage form.

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