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Research Article

Pharmacognostical Studies on Stems of *Clerodendrum infortunatum* L.

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Abstract

Clerodendrum infortunatum L. also known as *Clerodendrum viscosum* (Family Verbenaceae) is a perennial soft tomentose shrub that attains about 1m height consisting of light purple coloured or pinkish white flowers. It is widely used in traditional medicinal systems such as Ayurveda, Homeopathy and Unani due to its significant medicinal properties. It is commonly found in waste places of India, Burma and Bangladesh.

Aim: The present study was undertaken to lay down the pharmacognostical standards of stems of *Clerodendrum infortunatum* L. in order to validate / document its therapeutic benefits.

Materials and Methods: Stems of *Clerodendrum infortunatum* L. were subjected to pharmacognostical studies which include evaluation parameters such as histological study, microscopical study, fluorescence study, physicochemical analysis and phytochemical screening. Ethanolic and aqueous extracts were used for the physicochemical analysis and preliminary phytochemical screening

Results: Histological study revealed the presence and arrangement of various cells in the stem. Microscopical study revealed the presence of important characters such as covering trichomes, glandular trichomes, lignified fibres, wood elements, stone cells, medullary rays and calcium oxalate crystals. Preliminary phytochemical screening demonstrated the presence of phenolics, tannins, flavonoids, alkaloids, carbohydrates and glycosides.

Conclusion: The generated data could be significantly used to establish the pharmacognostic profile for the correct authentication, standardization and quality control of stems of *Clerodendrum infortunatum* L.

Keywords: *Clerodendrum infortunatum*, Verbenaceae, Bhandirah, Pharmacognostic evaluation, microscopy, macroscopy

INTRODUCTION

Clerodendrum infortunatum L., also known as *Clerodendrum viscosum* (Family Verbenaceae) is a salient medicinal plant in traditional medicinal systems such as Ayurveda, Homeopathy and Unani¹. It is also known as Hill Glorybower (Bhandirah in Sanskrit) and is widely distributed in Asia, Africa, Australia, Indo-malaysia, America and Western Ghats^{2,3}. It is a terrestrial perennial slightly woody flowering shrub found throughout Indian plains that has blunt quadrangular stems and branches. Leaves are oblong or elliptic, with serrate margin which are usually arranged three at a node or sometimes oppositely arranged⁴.

In Ayurveda, it is used in the treatment of tumors, cirrhosis, in scorpion sting, snake bite, malaria and jaundice. Indian folk medicine use various parts of the plant for the treatment of asthma, fever, bronchitis, diseases of the blood, burning sensation, inflammation, epilepsy sores, ulcers^{5,6}. Various indigenous communities utilize different parts of the plant as an ingredient in local wine, green salad and to treat various

ailments. Its stem is used in toothache and decoction of tender twigs is consumed orally to alleviate the menstrual complications. *Clerodendrum infortunatum* L. stem also has antibacterial and antifungal activity⁷. In spite of plenty of medicinal activity attributed to the stem of *Clerodendrum infortunatum* L., no pharmacognostical report is available in order to determine the anatomical standards necessary for the quality control of crude drugs.

The aim of the present investigation was to demonstrate the macroscopic, microscopic, physicochemical standards and conduct the preliminary phytochemical screening of *Clerodendrum infortunatum* L. stem, which could be utilized in preparation of plant monograph.

MATERIALS AND METHODS

Plant material collection

Clerodendrum infortunatum L. plant was collected from Thiruvalla, Pathanamthitta District in the month of September 2024 and authenticated by botanist Dr. Jacob

Thomas, Herbarium Curator, PG and Research Department of Botany, Mar Thoma College, Tiruvalla.

Organoleptic and macroscopic studies

Organoleptic characters of fresh stem like colour, taste, odour and shape were analyzed. Macroscopic characters of fresh stem were analyzed with the aid of simple microscope and the characters were observed⁸.

Histological studies

Histological studies of the stem were performed by taking free hand transverse section. Thin sections of *Clerodendrum infortunatum* L. stem were selected and cleared utilizing chloral hydrate solution. Thereafter they were stained with phloroglucinol and HCl, mounted in glycerin on a glass slide which was utilized for cellular and anatomical studies⁹⁻¹⁴.

Powder microscopic studies

Dried stem powder was utilized for investigating powder microscopic characters. Small amount of the powder sample was cleared using chloral hydrate solution and thereafter stained with 1 to 2 drops of phloroglucinol and HCl. Sample was spread evenly on a glass slide, thereafter mounted in glycerin and observed under the microscope to study the microscopic characters⁹⁻¹⁴.

Fluorescence analysis⁹⁻¹⁴

Fluorescence analysis is an important pharmacognostic parameter for the qualitative evaluation of crude drugs and was performed using standard methods. *Clerodendrum infortunatum* L. stem powder was treated with various neutral, acidic and basic solvents for 5 min and thereafter they were exposed to day light, short wavelength and long wavelength UV light.

Preparation of the extracts



Figure 1: *Clerodendrum infortunatum* L. plant

Histological studies

Transverse section of the *Clerodendrum infortunatum* L. stem is bluntly square shaped with four prominent convex protuberances. Outermost layer is single layered epidermis with numerous covering trichomes and glandular trichomes. Cortex is present below the

200g of *Clerodendrum infortunatum* L. stems was collected, air dried and pulverized to coarse powder. Then the powder was passed through sieve no:40 and extracted in Soxhlet apparatus by continuous hot percolation method using ethanol as solvent. Aqueous extract was prepared by hydro distillation method^{15,16}.

Physicochemical analysis

The shade dried stem powder was utilized for physicochemical analysis. Total ash value, water soluble ash value, acid insoluble ash value, loss on drying, foreign matter, swelling index and foaming index^{15,16} were determined.

Preliminary phytochemical screening

Ethanollic and aqueous extracts of *Clerodendrum infortunatum* L. stem were subjected to preliminary phytochemical screening. Qualitative chemical tests for the identification of various phytoconstituents such as phenolics, tannins, flavonoids, alkaloids, carbohydrates, proteins, steroids and glycosides were performed using standard methods^{15,16}.

RESULTS AND DISCUSSION

Organoleptic and macroscopic studies of *Clerodendrum infortunatum* L. stem

Colour: Dark green

Odour: Characteristic

Taste : Bitter

Shape : Bluntly quadrangular, slightly pubescent, annulate nodes

Size : 4 to 8 mm (w)

50 cm to 1m (h)

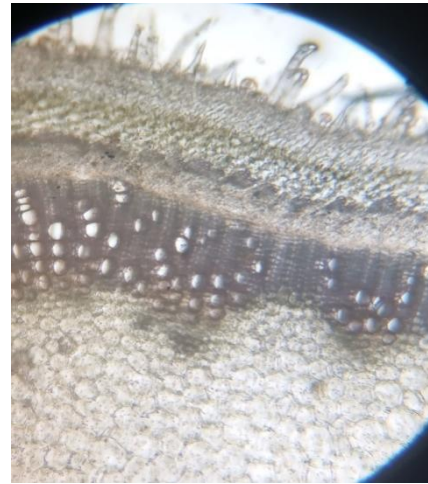


Figure 2: *Clerodendrum infortunatum* L. stem

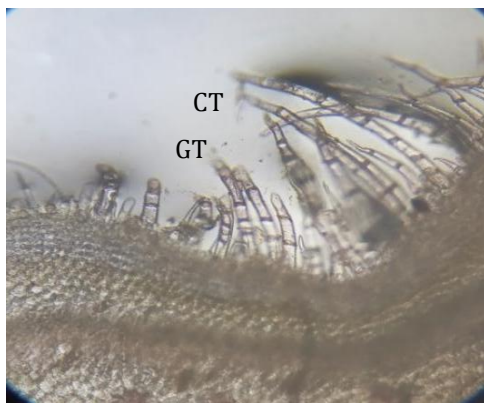
epidermis, which consist of several layers of collenchyma, followed by numerous layers of parenchyma and a single layer of stone cells. Secondary xylem region is lignified and contain numerous large xylem vessels, xylem fibres and medullary rays. The pith region consists of large parenchyma cells where a group of lignified cells are present at the centre.



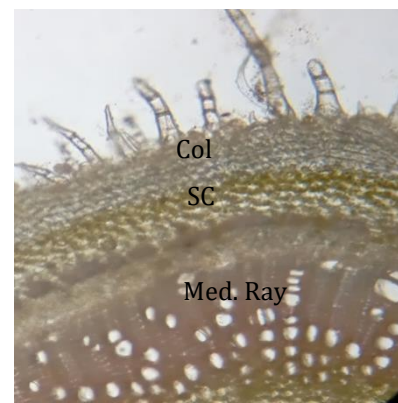
T.S of the *Clerodendrum infortunatum* L. stem



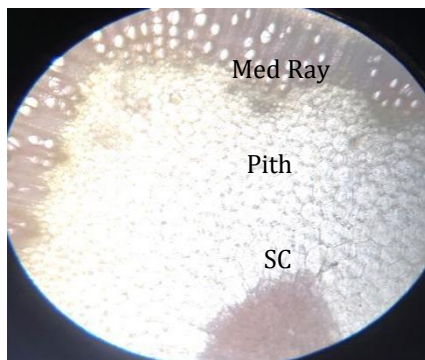
T.S showing trichomes, collenchyma, parenchyma, stone cells, medullary ray



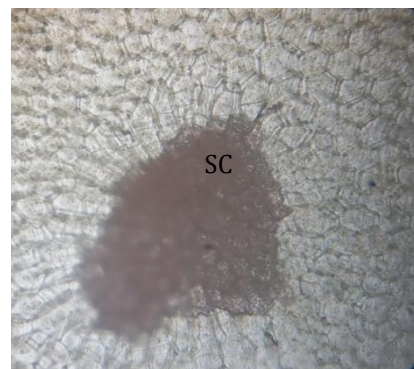
CT-Covering trichomes Col-Collenchyma, Sc-stone cells



GT-Glandular trichomes Med. Ray- medullary ray



Med Ray-medullary ray,



Sc-Stone cells

Figure 3: T.S of *Clerodendrum infortunatum* L. stem

Powder microscopy of *Clerodendrum infortunatum* L. stem

Clerodendrum infortunatum L. stem powder consists of important diagnostic characters such as multicellular uniseriate covering trichomes and multicellular

uniseriate glandular trichomes. It also consists of thin elongated cylindrical bundle of fibres, wood elements, needle shaped (acicular) calcium oxalate crystals, medullary rays, pitted xylem vessels and lignified stone cells.

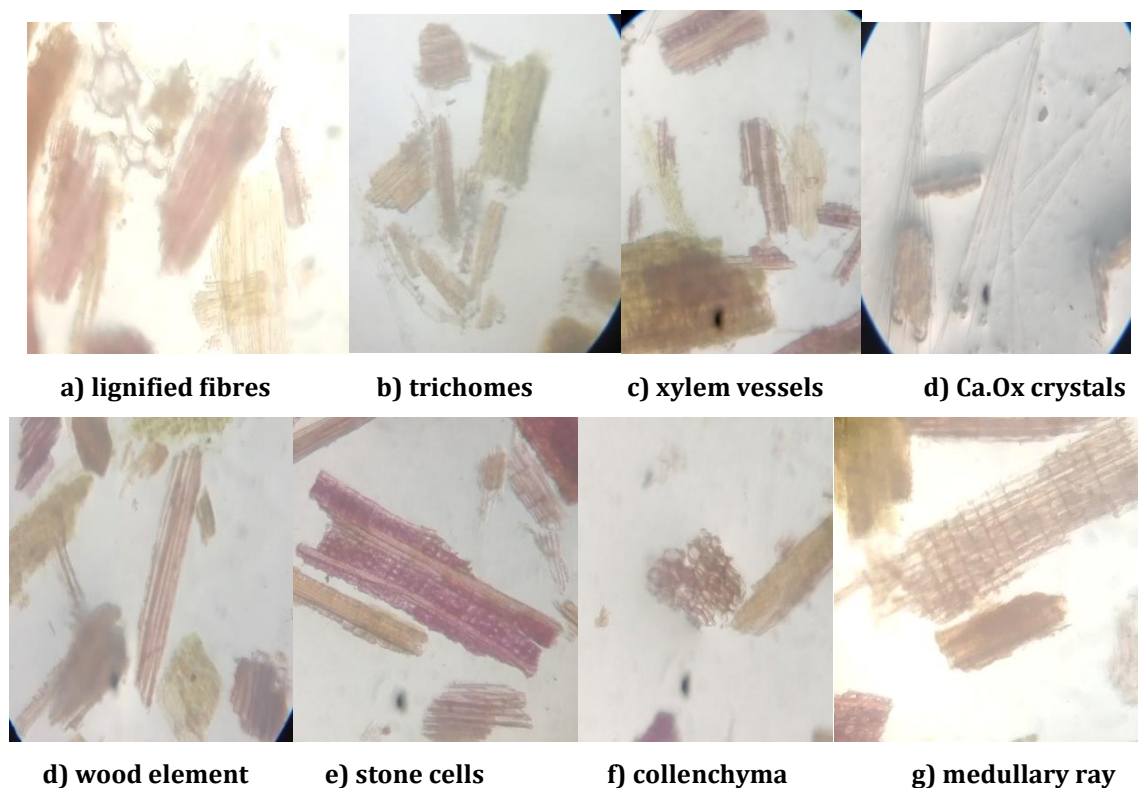


Figure 4: Powder microscopy of *Clerodendrum infortunatum* L. stem

Fluorescence analysis of *Clerodendrum infortunatum* L. stem

Fluorescence analysis of the stem powder was conducted using standard methods. *Clerodendrum infortunatum* L. stem powder was treated with various

acidic, neutral and basic solvents after which they were exposed to day light, short wavelength and long wavelength UV light. Many constituents of crude drugs manifested fluorescence in visible range of day light or short/long range of wavelengths. The results are depicted in table 1.

Table 1: Fluorescence analysis of *Clerodendrum infortunatum* L. stem

Sl. No	Reagents	Visible light	UV 254nm	UV 366 nm
1.	Powder drug	Dull green	Greenish brown	Dark brown
2.	Powder drug+ ethanol	Dark green	Dark green	Fluorescent buff
3.	Powder drug+ lead sub acetate	Brown	Dark green	Black
4.	Powder drug+ diethyl ether	Brown	Dark green	Black
5.	Powder drug + 20% NaOH	Dark brown	Dark green	Fluorescent light green
6.	Powder drug + 0.5N KOH	Brown	Dark green	Fluorescent buff
7.	Powder drug+ HCl	Light brown	Dark green	Black
8.	Powder drug+ 50% H ₂ SO ₄	Light brown	Dark green	Black
9.	Powder drug+ FeCl ₃	Brown	Dark green	Black
10.	Powder drug+ CHCl ₃	Dark brown	Light green	Black
11.	Powder drug+ picric acid	Brown	Fluorescent green	Black
12.	Powder drug+ ethyl acetate	Brown	Dark green	Fluorescent light brown
13.	Powder drug+ petroleum ether	Pale brown	Dark green	Black
14.	Powder drug+ ammonia	Brown	Dark green	Fluorescent buff

Physicochemical analysis of *Clerodendrum infortunatum* L. stem

Physicochemical parameters like total ash value, water soluble ash value and acid insoluble ash value of *Clerodendrum infortunatum* L. stem were found to be 8.96 ± 0.27 , 7.25 ± 0.24 and 1.8 ± 0.12 respectively. Loss on drying was found to be 7.01 ± 0.22 whereas foreign organic matter was found to be nil. Swelling Index was found to be 0.80 ± 0.05 cm and Foaming Index was found to be less than 100 (Table 2.0).

Preliminary phytochemical screening

Ethanollic and aqueous extracts of *Clerodendrum infortunatum* L. stem were subjected to qualitative chemical tests for the identification of various phytoconstituents. The results of the chemical tests were recorded and tabulated in table 3.

Table 2: Physicochemical analysis of *Clerodendrum infortunatum* L. stem

Physicochemical Parameters	%w/w
Ash Values	
Total ash value	08.96 ± 0.27
Water soluble ash value	7.25 ± 0.24
Acid insoluble ash value	1.80 ± 0.12
Loss on drying	7.01 ± 0.22
Foreign organic matter	Nil
Swelling Index	0.80 ± 0.05 cm
Foaming Index	Less than 100

Values are expressed as mean \pm SEM; n=3

Table 3: Preliminary phytochemical screening of *Clerodendrum infortunatum* Linn.

S.N.	Phytoconstituents	CE	CA
1.	Tests for Alkaloids Mayer's test Wagner's test Hager's test Dragendorff's test	++ ++ ++ ++	+ + + +
2.	Tests for Terpenoids	+	-
3.	Tests for Sterols Liebermann burchard test Salkowski test	- -	- -
4.	Tests for Flavonoids Aqueous sodium hydroxide test Shinoda test	++ ++	++ ++
5.	Tests for Phenolics and Tannins Ferric chloride test Lead acetate test	+ +	++ ++
6.	Tests for Carbohydrates Molisch's test Barfoed's test Fehling's test	++ ++ ++	++ ++ ++
7.	Tests for Cardiac Glycosides Legal test Baljet test Raymond's test	- - -	- - -
8.	Tests for Proteins and Aminoacids Millon's test Biuret test Ninhydrin test	- - -	- - -
9.	Tests for Saponins Foam test/froth test	-	-

(++) High active constituents (+) Presence of active constituents (-) Absence of active constituents

CE- Ethanollic extract of *Clerodendrum infortunatum* L. stem

CA- Aqueous extract of *Clerodendrum infortunatum* L. stem

Ethanol and aqueous extracts of *Clerodendrum infortunatum* L. stem revealed the presence of important phytoconstituents such as phenolics, tannins, flavonoids, alkaloids, carbohydrates and glycosides.

CONCLUSION

Current study was focused to set up the pharmacognostical standards for the identification and standardization of stems of *Clerodendrum infortunatum* L. The established pharmacognostical parameters can be considered as distinguishable characters that facilitate the authentication of plant material, which in turn will aid in preventing the adulteration of raw materials in the herbal industries and facilitate the assurance in quality of raw materials as well as formulations. Preliminary phytochemical screening revealed the presence of important phytoconstituents such as phenolics, tannins, flavonoids, alkaloids, carbohydrates and glycosides.

The outcome of the present findings can be served as excellent tools to formulate the pharmacopoeial standards for the upcoming studies and research which in turn will result in the quality control and availability of potent marketed formulations.

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