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PHARMACOGNOSTIC AND PHYTOCHEMICAL EVALUATION OF LEAVES OF *GARDENIA RESINIFERA* ROTH.

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ABSTRACT: *Gardenia resinifera* Roth. is a medicinal plant belonging to family Rubiaceae. It is an important traditional medicinal plant employed in various indigenous system of medicine against several diseases. The current communication provides pharmacognostic, physicochemical and phytochemical investigation carried out on the leaves of *Gardenia resinifera* Roth, which are useful in setting some diagnostic indices for the identification and preparation of a monograph of the plant.

INTRODUCTION: Gardenias are members of the madder or Rubiaceae family. Gardenias are most prevalent in China, Japan, tropical regions of Southeast Asia and the Pacific islands, South Africa and India. Today there are over 200 different species of *Gardenia* mostly hybrid in existence throughout the world. *Gardenia resinifera* Roth. is commonly known as dikamali which belongs to the family Rubiaceae. It is a large glabrous shrub or small tree attaining a height up to 6-7.5 m high and found in different Hills and Ghati areas of India.

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Different parts of this plant contain different phytoconstituents which are used to increase appetite, used as astringent, used to relieve the pain of bronchitis, in vomiting, liver disorders [1] constipation, and cutaneous diseases [2, 3, 4, 5, 6].

MATERIALS AND METHODS:

Collection and Authentication: The Fresh leaves of *Gardenia resinifera* Roth. were collected from the local areas of Hubli-Dharwad district, Karnataka, India in June 2010 and authenticated by Dr. B. D. Huddar, Head of the Department of Botany, Shri Kadasiddheshwar Arts College, and H. S. Kotambari Science Institute, Vidyanagar, Hubli, and Karnataka, India.

Extraction: The leaves of *Gardenia resinifera* Roth. were washed under tap water and dried in the air under the shade at room temperature and converted to a coarse powder and stored in an

airtight container. For the physicochemical investigation, 50 g of dried powder was successively extracted with different solvents such as petroleum ether, chloroform, ethyl acetate, and ethanol in increasing order of polarity using Soxhlet apparatus. The extracts were concentrated under reduced pressure using rotary flash evaporator, and the residues were dried in a desiccator over sodium sulfite. After drying, the respective extracts were weighed, and the percentage yield was determined and stored in airtight container [9, 14, 15, 16].

Pharmacognostic Studies:

Macroscopic Characteristics: For morphological observations, fresh leaves (approx. 6-20 cm in length) were used. And organoleptic features *viz.* color, odor, taste, shape, sizes were observed and evaluated botanically [7].

Microscopic Characteristics: The freehand section of the leaf was taken and stained by safranine for lignified cells and Sudan red for oil cells and then mounted in glycerine and observed under low power [8, 9] and powder microscopy was also carried out and their specific diagnostic characters were recorded [10].

Physico-Chemical Parameters: The physico-chemical parameters were like total ash value, loss on drying, water soluble ash, insoluble acid ash, petroleum ether, alcohol, and water-soluble extractive value, etc were determined as per who guidelines [11, 12].

Phytochemical Analysis: The qualitative phytochemical tests were carried out on all the extracts of *Gardenia resinifera* Roth. leaves to detect the presence of various Phytoconstituents[7, 8, 13, 14].

RESULTS:

Macroscopic Characteristics: Macroscopically, the leaf was simple in composition, opposite, elliptic-oblong, entire, midrib distinct on both surfaces, leathery texture, wavy margin, narrowed leaf base, with acuminate apex, innately reticulate venation, the average leaf size was 06 to 20 cm (length)and 2.5 to 7.5 cm (width). The fresh leaf was green in color **Fig. 1**.

Morphological Evaluation of *Gardenia resinifera* Roth:

FIG. 1: *GARDENIA RESINIFERA* ROTH. LEAVES

TABLE 1: MORPHOLOGY OF THE LEAVES OF *GARDENIA RESINIFERA* ROTH.

S. no.	Features	Observation
1	Color	Green
2	Taste	Pungent
3	Odor	Aromatic
4	Size	6.3-20 / 2.5-7.5 cm
5	Shape	Elliptic-oblong
6	Texture	Leathery
7	Margin	Wavy
8	Leaf base	Narrowed
9	Apex	Acuminate
10	Venation	Pinnately reticulate
11	Midrib	Distinct on both surfaces

Microscopic Characteristics: The transverse section of *Gardenia resinifera* Roth. leaf showed the presence of upper and lower epidermis. The epidermis consists of single-layered polygonal cells covered with a thick, warty cuticle; some of the lower and upper epidermal cells are interrupted with the paracytic type of stomata and unicellular covering trichomes. Mesophyll region is differentiated into palisade cells and spongy parenchyma which contain prism type of calcium oxalate crystals. The midrib region consists of a collateral type of vascular bundles, and on either

side of vascular bundle, collenchymatous cells are present below the upper and above the lower epidermis. Xylem was lignified, phloem was non-lignified, and it also contains mucilage with resin ducts. The salient diagnostic characteristics of leaf were the collateral type of vascular bundle, paracytic stomata, xylem vessels, mucilage with resin ducts and prismatic type of calcium oxalate crystals. These characters can be used for standardization of drugs and also used for the preparation of plant monographs **Fig. 2**.

Microscopical Evaluation of T. S. of *Gardenia resinifera* Roth. Leaves:

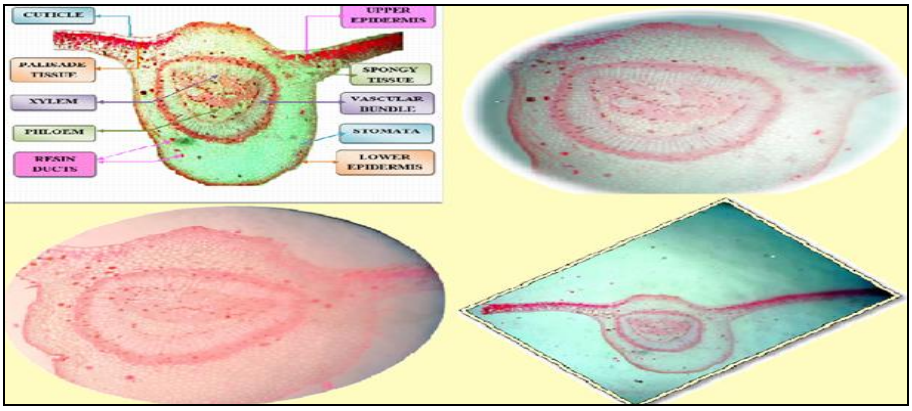

FIG. 2: T. S. OF *GARDENIA RESINIFERA* ROTH. LEAVES

Powder Study: The crude powder of *Gardenia resinifera* Roth. Leaf was dark green with characteristic odor and pungent taste. The diagnostic features of powder were prism type of calcium oxalate crystals present on the surface of epithelial cells. In surface view, fragments of epidermis were embedded with paracytic stomata, xylem vessels with spiral thickening and mucilage were observed **Fig. 3**.

Diagnostic Characters of Powdered Leaves of *Gardenia resinifera* Roth:

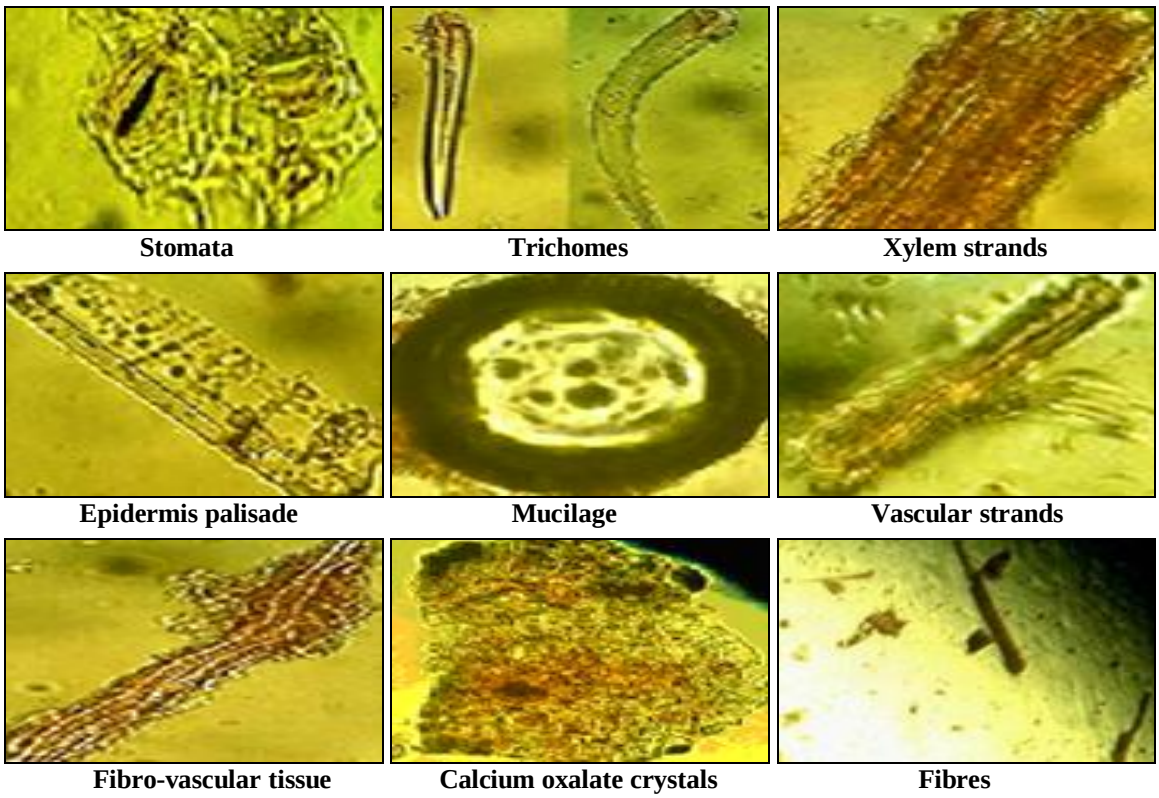

FIG. 3: POWDER MICROSCOPY

Physiochemical Investigations: The constant physical evaluation of the drugs is an important parameter in detecting adulteration or improper handling of drugs. The moisture content of dry powder of *Gardenia resinifera* Roth. leaves were 7.28% which is not very high; hence it would discourage bacteria, fungi or yeast growth. The ash value was determined by three different forms *viz*., total ash, water-soluble ash and acid insoluble ash. The total ash is particularly important in the evaluation of purity of drugs, *i.e*. the presence or absence of foreign inorganic matter such as metallic salts or silica. Acid-insoluble ash measures the amount of silica present, especially sand. Water soluble ash is the water-soluble portion of the total ash [24]. The total ash of crude powder of *Gardenia resinifera* Roth. leaf was 8%; water-soluble ash was 3%, and acid insoluble ash was 1%. Less amount of these three parameters indicate that the inorganic matter and silica was less in *Gardenia resinifera* Roth. leaves. The extractive value of crude powder was maximum in water (18.4%) followed by alcohol (16%) and the minimum was in petroleum ether (3.2%) **Table 2**.

TABLE 2: PHYSIOCHEMICAL INVESTIGATIONS

S. no.	Parameter	Determined value (% w/w)
	(A) Extractive value	
1	Petroleum ether soluble Extractive value	3.2%
2	Alcohol soluble extractive value	16%
3	Water soluble extractive value	18.4%
	(B) Moisture content	
1	Total Moisture content	7.28%
	(C) Ash values	
1	Total ash	8%
2	Acid-insoluble ash	1%
3	Water soluble ash	3%

Phytochemical Analysis: The results of the qualitative phytochemical analysis of the crude powder extract of *Gardenia resinifera* Roth. leaves are shown in **Table 3**. Qualitative chemical examinations of various extracts revealed the presence of steroids in pet ether extract, carbohydrates in chloroform and ethanolic extracts, phenolic compounds, tannins and flavonoids in ethyl acetate and ethanolic extracts, glycosides in ethanolic extract respectively **Table 3**.

TABLE 3: QUALITATIVE CHEMICAL ANALYSIS PERFORMED ON SUCCESSIVE EXTRACTS OF *GARDENIA RESINIFERA* ROTH. DRIED LEAVES

Phyto-constituents	Pet. ether extract	$CHCl_3$ extract	Ethyl acetate extract	EtOH extract
Carbohydrates	-	+	-	+
Glycosides	-	-	-	+
Phytosterol steroids	+	-	_	-
Triterpenoids	-	-	-	-
Tannins & Phenolic group	-	-	+	+
Alkaloids	-	-	-	-
Flavonoids	-	-	+	+

ETOH = Ethanolic, P. E. = Petroleum Ether, $CHCl_3$ = Chloroform, E. A. = Ethyl acetate. + = Present, - = Absent.

CONCLUSION: As there is no pharmacognostic work on a record of this traditionally much-valued drug, the present work was taken up to lay down standards, which could be useful to establish the authenticity of this medicinally useful plant. Macro and microscopically standards discussed here can be considered as identifying parameters to authenticate the drug. In the present study, we have found that most of the biologically active phytochemical were present in the ethanolic extract and crude powder of *Gardenia resinifera* Roth. leaves. The medicinal properties of *Gardenia resinifera* Roth. leaves may be due to the presence of the above mentioned phytochemical. Further studies are in progress about this drug in our laboratory.

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CONFLICT OF INTEREST: Nil

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