

PHYTOCHEMICAL AND PHARMACOGNOSTICAL STUDIES ON *PEDALLIUM MUREX* LINN

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ABSTRACT

Pedallium Murex Linn (entire plant) (Pedaliaceae), a plant of immense medicinal values was taken for the present study. It is essential to ensure the quality and purity of the popular fragrance plant. The entire plant of *Pedallium Murex* Linn was subjected to various Pharmacognostical evaluations like Morphological, Microscopical and Powder analysis. Results have revealed clearly that the entire plant is genuine. The Phytochemical constituents of Leaves of *Pedallium Murex* Linn have been worked out. The dry powder of the entire plant was successfully extracted with Total Petroleum ether extract, Alcohol Extract, Chloroform extract, and Aqueous Extract. All the extracts were subjected to Preliminary Phytochemical screening. It showed the presence of Carbohydrates, Glycosides, Alkaloids, Steroids and Flavonoids. As per Materia medica (Krithikar and Basu), studies are required for the screening of various Pharmacological activities like plant pacifies vitiated vata, pitta, urinary retention, kidney stone, seminal weakness, amenorrhea, inflammation, flatulence and fever.

KEYWORDS: *Pedallium Murex* Linn, Phytochemical, Pharmacognosy

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INTRODUCTION

Pedallium murex is an important medicinal plant that contains several alkaloids like pedalitin, Diosmetin, Dinatin, Pedalin dinatin-7-glucuronide. The leaf decoction is used to control white discharge due to excessive body heat. Root decoction is used as an antibilious agent, while the juice of the fruit is used as an emmenagogue and to promote lochial discharge¹. The decoction of the seeds and glycosides obtained from it showed mild diuretic activity and the alcoholic extract of the fruits reduced blood pressure in dog and rat². It is reported that many Indian medicinal plants show beneficial effects against renal injury³. A good example is a succulent herb, *Pedallium murex* Linn, commonly called Gokhru a member of family Pedaliaceae. It is commonly found in Deccan and in some parts of Ceylon and Gujarat and in the coastal areas of southern India⁴. It is about 15 to 40 cm in height, having four angle spiny brownish colour fruits (1-2 cm). The fruits are rich in flavonoids, sapogenin (diosgenin-0.06%) and soluble proteins (20.14mg/yml). An infusion extract prepared using cold water from the leaves, stems and fruits of

P. murex are demulcent, diuretic and also found to be useful in the treatment of disorders of urinary systems such as gonorrhoea, dysuria, incontinence of urine, etc. Herbal medicine is a triumph of popular therapeutic diversity. Almost in all the traditional medicine, the medical plants play a major role and constitute the backbone for the same. In order to make sure the safe use of these medicines, a necessary first step is the establishment of standards of quality, safety and efficacy. Keeping this fact in to consideration, the attempts were made to establish pharmacognostical studies⁵⁻¹¹ of the plant *Pedallium murex* linn belonging to family Pedaliaceae.

The therapeutic potentials of plant and animal origin are being used from the ancient times by simple process without isolation of pure compounds that is in the form of crude drugs. The pharmacological action of crude drug is determined by the nature of its constituents.

Thus plant species may be considered as a biosynthetic laboratory not only for chemical compounds, e.g. Carbohydrates, proteins and fats that are utilized as a food by humans and animals, but also for a multitude of

compounds including alkaloids, flavonoids, glycosides etc. which exert definite pharmacological activity.

To obtain these pharmacological activities, plant materials were used as such in their crude form or may be extracted with suitable solvents to take out the desired components and the resulting principle being employed as therapeutic agents. The phytochemistry of herbal drug embraces a thorough consideration of these chemical entities that are termed as constituents. As the herbal drugs contain so many chemical compounds, it is essential to single out those responsible for therapeutic effect to be called as active constituents.

By considering the above facts, it is necessary to evaluate the nature of extract before evaluating the biological activity of same. We have been selected such extracts for pharmacological activity which contain large number of chemical constituents. Hence for this purpose, we have to go for preliminary tests to evaluate chemical nature of extracts qualitatively.

MATERIALS AND METHODS

The plant is an exact sub-succulent, annual herb. The stem is angular when young. The plant emits an unpleasant smell through mucilage. The leaves are alternate and sub opposite with unequal leaves; it is broadly ovate oblong, crenate, emarginated, base truncate and succulent. Flowers are yellow solitary and auxiliary with a pair of glands at the base.

Collection of specimens

The plant specimens for the proposed study were collected from Pattukottai district. Care was taken to select healthy plants and normal organs. The required samples of different organs were cut and removed from the plant and fixed in FAA (Formalin-5ml + Acetic acid-5ml + 70% ethyl alcohol-90ml). After 24hrs of fixing, the specimens were dehydrated with graded series of tertiary butyl alcohol as per the schedule given by Sass, 1940. Infiltration of the specimens was carried by gradual addition of paraffin wax (melting point 58-60 C) until TBA solution attained super saturation. The specimens were cast into paraffin blocks.

Sectioning

The paraffin embedded specimens were sectioned with the help of Rotary Microtome. The thickness of the sections was 10-12mcg. Dewaxing of the sections was by customary procedure¹². The sections were stained with toluidine blue as per the method published by O'Brien *et al*¹³. Since toluidine blue is a polychromatic stain. The staining results were remarkably good; and some cytochemical reactions were also obtained. The dye rendered pink colour to the cellulose walls, blue to the lignified cells, dark green to suberin, violet to the mucilage, blue to the protein bodies etc. Wherever

necessary sections were also stained with safranin and Fast-green and IKI (for starch)

For studying the stomatal morphology, venation pattern and trichome distribution, paradermal sections (sections taken parallel to the surface of leaf) as well as clearing of leaf with 5% sodium hydroxide or epidermal peeling by partial maceration employing Jeffrey's maceration fluid¹⁴ were prepared. Glycerine mounted temporary preparations were made for macerated/cleared materials. Powdered materials of different parts were cleared with NaOH and mounted in glycerine medium after staining. Different cell components were studied and measured.

Photomicrographs

Microscopic descriptions of tissues are supplemented with micrographs wherever necessary. Photographs of different magnifications were taken with Nikon labphoto 2 microscopic units. For normal observations bright field was used. For the study of crystals, starch grains and lignified cells, polarized light was employed. Since these structures have birefringent property, under polarized light they appear bright against dark background. Magnifications of the figures are indicated by the scale-bars. Descriptive terms of the anatomical features are as given in the standard anatomy books¹⁵.

Extraction procedure

The authenticated fresh leaves were dried under shade and used for the preparation of extract. These leaves were coarsely powdered with the help of mechanical grinder and passed through sieve no.40. The powder was stored in an airtight container for further use.

Petroleum ether extract

The shade dried coarsely powdered leaves of *Pedallium Murex linn* (1 kg) were extracted with petroleum ether (60-80°C) until the extraction was completed. After completion of extraction, the solvent was removed by distillation. Dark green colour residue was obtained. The residue was then stored in dessicator.

Chloroform extract

The marc left after petroleum ether extraction was dried and then extracted with chloroform (55-56°C), until the extraction was completed. After completion of extraction, the solvent was removed by distillation. Dark greenish yellow colour residue was obtained. The residue was then stored in dessicator.

Ethanol extract

The marc left after chloroform extraction was dried and then extracted with ethanol 95% v/v (75-78°C), until the extraction was completed. After completion of extraction, the solvent was removed by distillation. Dark brown colour residue was obtained. The residue was then stored in dessicator.

Aqueous extract

The marc left after ethanol extraction was dried and then extracted with chloroform water by cold maceration process for 7 days. At the end of 7th days, it was filtered through muslin cloth and the filtrate was concentrated. The remaining solution was evaporated by heating on a water bath. The brown colour residue was obtained. The residue was then stored in dessicator.

All the extracts of *Pedallium murex linn* were subjected to qualitative tests for the identification of various active constituents. The phytochemical^{16, 17} constituents present in various extracts were presented in **Table 1**.

RESULTS AND DISCUSSION**Microscopic studies of the leaf**

The leaf thick and prominent midrib (**Fig 1.1, 2**) and slightly thicker lateral veins (**Fig 1.1, 2**). The lamina is thin and dorsiventral. Midrib is Plano convex is sectional view with flat adaxial side and semicircular abaxial part (**Fig 1.2**). The epidermal layer is thin, comprising thick walled circular cells which are 12 μ m in thickness. The ground tissue of the midrib is homogeneous, Parenchymatous, thin walled, angular and compact. There is a thick mass of collenchymas beneath adaxial part. The vascular tissues occur in wide, bowl shaped outline and are divided into three wedged shaped segments. The vascular strands possess short and narrow parallel rows of circular thick walled xylem segments and thin layer of phloem elements. The midrib is 600 μ m thick and 800 μ m wide.

Lamina and lateral vein (Fig 2.1, 2)

The lateral vein is slightly thick on the abaxial side with shallow depression on the adaxial side. The vascular strand of the midrib is thin, conical, and collateral and occurs towards the adaxial part (**Fig 2.2**). It has small group of adaxial xylem and then are of abaxial phloem.

The lamina is uniform in thickness with smooth and even surfaces. It is 170 μ m thick, it exhibits bilateral symmetry. The adaxial and abaxial epidermal layers are thin and less prominent, the epidermal cells are spindle shaped and thin walled. The mesophyll is differentiated into adaxial zone of palisade tissue and abaxial spongy parenchyma. The palisade cells are in two layers, the upper layer having longer cells than lower layering cells. The palisade zone is 100 μ m thick. The spongy mesophyll consists of four or five layers of compact parenchyma cells (**Fig 2.1, 2**).

Petiole (Fig 3.1, 2, 3)

The petiole is flat with slight concavity on the adaxial. The cross sectional outline is rectangular (**Fig 3.1**). It is 850 μ m thick and 2.65mm wide. It has thin and distinct layer of epidermis with thin cuticle. Inner to the epidermis is a thin zone of two or three layered

collenchyma cells. Remaining ground tissue is parenchymatous with circular, thin walled compact cells. The petiole has multistranded vascular system. There is an arc of fine vascular strands of which the median strand in the largest, the two lateral strands are smaller and another pair of strands located within adaxial lateral part in the smallest (**Fig 3.1**). The vascular strands are collateral. They have wide, circular thick walled xylem elements which is random elusters (**Fig 3.2, 3**). The Meta xylem elements are 30 μ m wide. Phloem elements are in small discrete groups located at the outer part of the xylem strands.

Lamina (Fig 4.1)

The leaf blade bears glandular type of trichomes both on the abaxial and adaxial sides. The trichomes have short, thin walled, unicellular stalk and spherical multicellular stalk (**Fig 4.1**). The glandular head is circular in surface view with four triangular, thick or thin walled cells.

Leaf margin (Fig 4.2)

The leaf margin is thick and blunt. It is similar in structure as the middle part of the lamina. It consists of an adaxial zone of palisade and abaxial compact spongy parenchyma cells.

Venation system of the leaf (Fig 5.1, 2)

The venation of the lamina is densely reticulate with thick and straight veins. The vein islets are wide, elongated or polygonal in outline (**Fig 5.1**). Vein terminations are present invariably in all islets. The terminations are well developed. The terminations branched repeatedly forming blendroid outline; some of the terminations forked only once or not forked at all (**Fig 5.2**)

Stomata (6.1, 2)

Stomata occur only on the lower surface of the lamina. The lower epidermal cells are reduced in area; they have thick wavy anticlinal walls so that the epidermal cells are amoeboid in outline (**Fig 6.1**). The stomata are anisocytic type (**Fig 6.2**). Each stoma is surrounded by three unequal subsidiary cells.

The guard cells are circular or elliptical in outline. The circular guard cells are 15 X 15 μ m in size; the elliptical cells are 15 X 20 μ m.

Powder microscopic results

The leaf powder shows some specific inclusions when viewed under the microscope.

Epidermal peelings bearing the glandular trichomes are seen in surface view (**Fig 7.1**). The glands are perfectly circular; invariably 4 celled and are randomly distributed. Some of the glands are thin walled. Both types are 20-25 μ m in diameter. The thick walled trichomes have dense, darkly staining cells (**Fig 7.2**).

Broken triangular fragments of the thick walled trichomes are seen scattered in the powder (Fig 7.3). The thick glands easily break into four triangular cells, where as the thin walled glands do not break.

Adaxial epidermis (Fig 8.1): the powder consists of small fragments of adaxial and abaxial epidermis. The adaxial epidermis consists of narrow polygonal cells with thin straight anticlinal walls. Proto plasmic contents are seen in some of the cells.

Abaxial epidermis: Abaxial epidermal peelings are stomaliferous and the cells are amoeboid in outline. The stomata are anisocytic type; some of the stomata are anomocytic with disture subsidiary cells. The epidermal cells have thin undulate anticlinal walls (Fig 8.1,2)

CONCLUSION

The present paper reports a group of ethnobotanical, morphological, chemical, and molecular studies performed with *Pedallium murex linn leaves* obtained by collection in the pudukottai district in India. Macro- and micro-morphological parameters were established to authenticate the genuine drug that allowed detection of adulterants usually found in commercial samples of this plant material. These morphological characteristics can be used for rapid identification of the drug and are particularly useful in the case of powdered materials. The chemical studies performed demonstrated that the presence of Carbohydrates, Glycosides, Alkaloids, Steroids and Flavonoids represent the major component group in the bark.

Pedallium murex is a valuable source of medicinally useful compounds that have been used traditionally for various ailments. Leaf extracts of this plant showed good source for the bioactive compounds. Thus plant studied here can be a potential source for useful drugs, if it is involved in further research.

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Table 1: Phytochemical studies on *Pedallium murex linn*

Extract	Carbohytrates	Glycosides	Fixed oils & fats	Proteins	Saponins	Tannins	P.sterols &steroids	Alkaloids	Flavanoids
Pet.ether	-	+	+	+	-	-	+	+	+
Ethanol	-	+	-	+	-	-	+	-	+
Chloroform	-	+	-	-	-	-	+	-	+
Aqueous	+	+	-	-	-	-	+	-	-

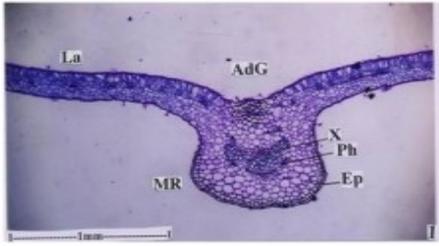


Fig 1.1: TS of leaf through Midrib and lamina

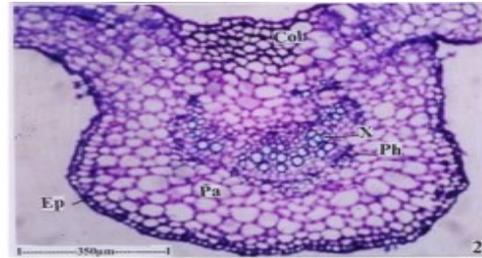


Fig 1.2: TS of Midrib-enlarged.

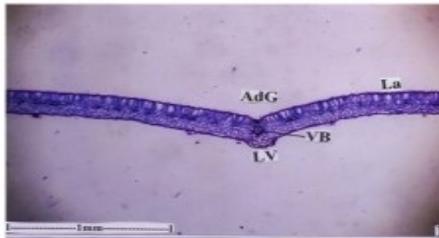


Fig 2.1: TS of leaf through lateral vein

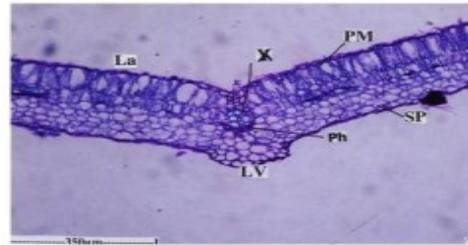


Fig 2.2: Lateral vein and lamina-enlarged

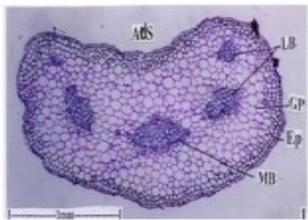


Fig 3.1: TS of Petiole-entire view

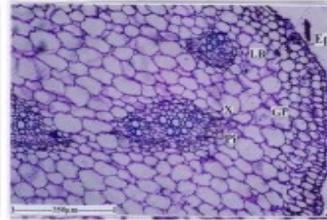


Fig 3.2: One sector enlarged

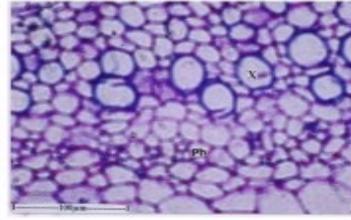


Fig 3.3: Vascular elements-enlarged

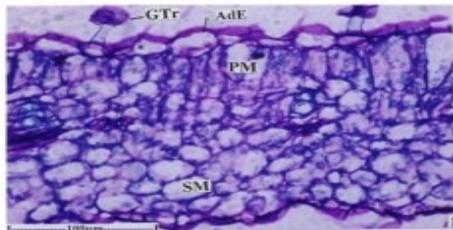


Fig 4.1: TS of lamina showing mesophyll tissues and glandular

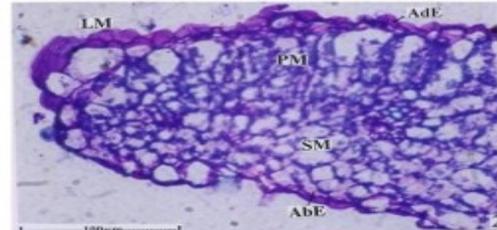


Fig 4.2: TS of leaf through marginal part

(AdG: Adaxial Groove; col: collenchymas; Ep: Epidermis; La: Lamina; MR: Midrib; Pa: Parenchymatous ground tissue; Ph: Phloem; X: Xylem; LV: Lateral Vein; PM: Palisade mesophyll; SM: Spongy mesophyll; VB: Vascular Bundle; AdS: Adaxial Side; GP: Ground Parenchyma; LB: Lateral Bundles; MB: Median Bundle; Pi: Pith; AbE: Abaxial Epidermis; AdE: Adaxial Epidermis; GTr: Glandular Trichome)

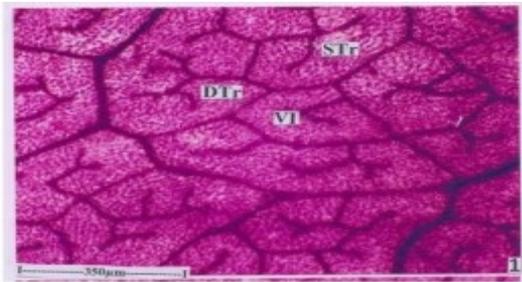


Fig 5.1: Venation pattern of the leaf

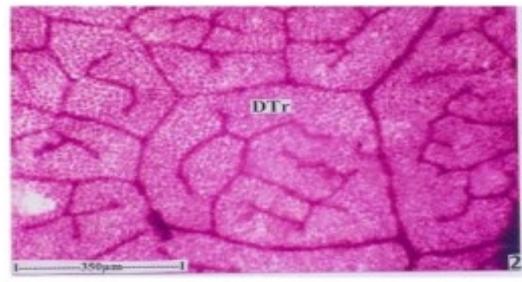


Fig 5.2: Veins forming vein islets and vein termination

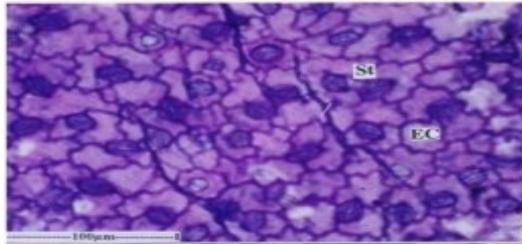


Fig 6.1: Paradermal sections, Abaxial stomatiferous epidermis

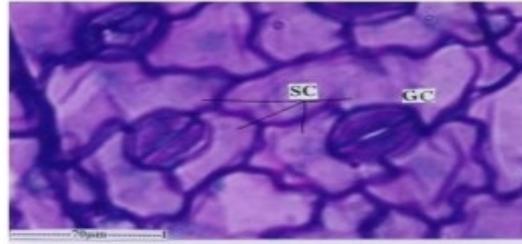


Fig 6.2: Stomata and epidermal cells-enlarged

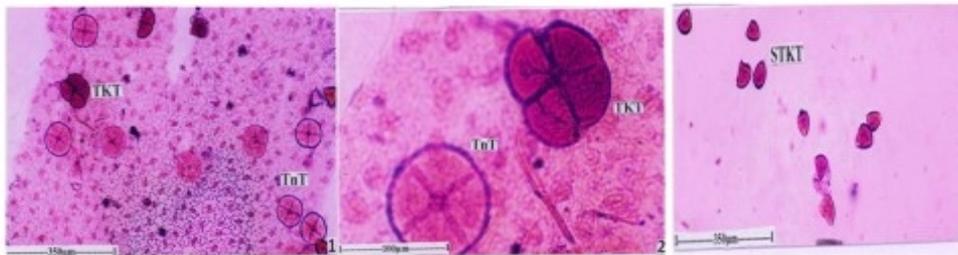


Fig 7.1: Abaxial epidermis with stomata and glandular trichomes – in surface view

Fig 7.2: Thick walled and thin walled glandular trichomes-enlarged

Fig 7.3: Broken cells of the thick walled glandular cells.

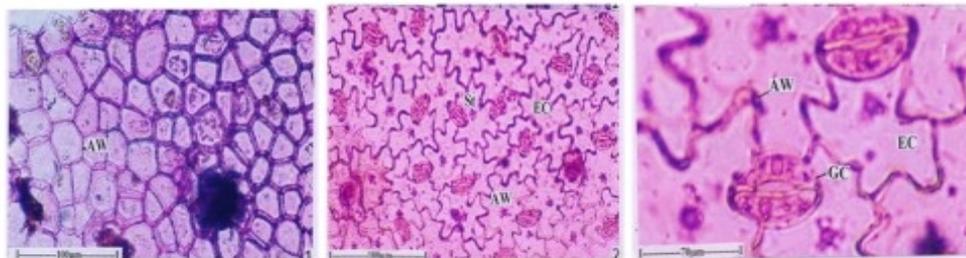


Fig 8.1: Adaxial epidermal peeling in surface view

Fig 8.2: Abaxial epidermal peeling with stomata

Fig 8.3: Stomata and amoeboid epidermal cells

(DTr: Dendroid Trichome; STR: Simple Trichome; VI: Vein islet; EC: Epidermal Cells; GC: Guard Cells; SC: Subsidiary Cells; St: Stomata; TKT: Thick walled Trichome; TnT: Thin walled Trichome; STKT: Segments of the Thick walled Trichome; AW: Anticlinal Walls)

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