PHARMACOGNOSTICAL STUDIES ON TRAGIA PLUKENETII R. SMITH

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ABSTRACT: The global market for herbal products is exploding. The major hindrance in the amalgamation of herbal medicines into modern medical practices is the lack of scientific and clinical data and a better understanding of efficacy and safety of the herbal products. The evaluation or standardization of a crude drug involves Pharmacognostic methods such as organological, physicochemical, phytochemical and pharmacological analysis. The experimental material selected for the present study is Tragia plukenetii R. Smith belongs to the family Euphorbiaceae. This herb is distributed in hill slopes of southern peninsular India. The root is diaphoretic and alternative and is given for fevers to cause perspiration. The present paper deals with anatomical characters of leaf, stem, and roots. The dried powders of stem, leaves and roots and their extracts in various solvents were examined with ordinary light and U.V. light (365nm). Quantitative determination such as moisture content, total ash, water-soluble ash, acid insoluble ash, water extractive values, and sulfated ash have also been made. Preliminary phytochemical analysis of the extracts was done and the results showed that tannins, steroids, triterpenoids and phenols were predominantly present in all the extracts.

INTRODUCTION: The use of medicinal plants is as old as human civilization. India has a glorious tradition of health care systems based on plants. The role of medicinal plants as the primary tool in the preservation of health as well as prevention and management of the disease is realized with alarming concern in recent days. This is mainly because of our hasty departure from nature and dependence on synthetic drugs which often produce harmful effects.

The experimental material selected for the present study is Tragia plukenetii R. Smith belongs to the family Euphorbiaceae. Tragia plukenetii R. Smith (Tamil name: Karunkanchori) the root is diaphoretic and alternative and is given for fevers to cause perspiration.

MATERIAL AND METHODS: Mature and healthy plants of Tragia plukenetii were collected from Southern Western Ghats in the district of Tirunelveli, South India. The specimens were identified, comparing the characteristics of floral and vegetative characters in the ‘Flora of the Presidency of Madras’ 2 and ‘Flora of Tamilnadu Carnatic’ 3. Voucher specimens are documented in the herbarium of St. Xavier’s College (XCH), Palayamkottai, Tamil Nadu, India. The plant parts...
were soaked in 70% alcohol, free hand sections of the leaf, stem, and root were taken for detailed microscopic observations and figures were drawn. Fixing the specimens was done as per the schedule is given by Sass. The specimens were cast into paraffin blocks by usual method. The sections were stained with toluidine blue as per the method published. Microphotographs of different magnifications were taken with Nikon Labphot 2 Microscopic Unit. For the study of crystals, starch grains, and lignified cells, polarized light was employed. A dry powder of the aerial part was used for chemical analysis. The physicochemical analysis was carried out by standard procedure. The fluorescence analysis of the powder drug under Ultra Violet was done according to the methods described. The preliminary phytochemical analysis was done by the methods described.

RESULTS AND DISCUSSION:
Macroscopic Studies: The plant is sparsely pubescent climbing herbs. Leaves are palmatifid. Male flowers have 3 perianth lobes and female flowers have six perianth lobes. Fruit are three-lobed with globose seeds.


Microscopic Studies:
Epidermal Cells and Stomata: The epidermal cells are polyhedral, thin-walled and the anticlinal walls are straight and smooth. Some of the cells have dense tannin content. Stomata occur only on the abaxial side. They are the paracytic type. The guard cells have two lateral subsidiary cells of unequal size; they are wing-like, expanded on either side of the guard cells. The guard cells are narrowly elliptical, measuring 15 x 30 µm in size.

Leaf: The leaf has bilateral symmetry with the fairly prominent midrib on the abaxial side. The midrib is planoconvex with flat adaxial side and small semicircular abaxial side. It is 300 µm in the vertical plane and 350 µm in the horizontal plane. The adaxial epidermis is fairly wide and rectangular; the abaxial epidermis is narrow with small circular cells. The vascular strand is single, triangular in outline and the xylem and phloem are collateral. The xylem elements are quite thick walled and occur in compact parallel rows without intervening parenchyma or xylem fibers. Phloem occurs in small discrete radial files; phloem elements are narrow and less prominent. Beneath the vascular strand, the abaxial part of the midrib has circular compact parenchyma cells which extend in the adaxial part in their vertical rows of cells. The palisade cells are seen up to the lateral sides of the parenchyma extensions.

The lamina has wide, mucilaginous adaxial epidermis cells which are rectangular and are 40 µm thick. The abaxial epidermis is thin comprising of narrowly cylindrical cells, some them possessing mucilage masses. The mesophyll tissue is differentiated into the adaxial zone of the single row of cylindrical compact palisade cells and abaxial zone of wide spherical and lobed spongy parenchyma cells which form small air-chambers. Plate 2. The leaf-margin is bluntly conical thick and slightly bent upward. It is 100 µm thick. The structure of the leaf-margin is similar to that of the mid-part of the lamina. The marginal portion has wide adaxial epidermis with dilated barrel-shaped cells, thin abaxial epidermis, and palisade-spongy tissues.

Venation Pattern: The primary, secondary and tertiary veins are thick and prominent. The leaf-margin has intramarginal venation which gives rise shortly, branched vein-terminations which spread in the intramarginal space. Vein-islets are distinct from the small area occupied by thick vein-terminations. The terminations are mostly branched once or more forming dendroid outlines.

Petiole: Structure of the petiole at proximal as well as distal regions was studied. The proximal part of the petiole is dorsiventral. The abaxial side is semicircular, and the adaxial side is flat with an uneven surface. Plate 3. It is 1.2 mm horizontally and 1mm vertically. The epidermal layer is with small squarish cells. The ground tissue is parenchymatous thin-walled, circular and less compact along the periphery and larger, angular and compact in the center. The vascular system is multistranded and consists of a circle of discrete, five vascular strands; of the five strands, two are abaxial, two lateral and the fifth one is adaxial in position. The vascular bundles are collateral,
wedge-shaped and have a dense mat of xylem elements and a thin layer of phloem elements. Glandular capitate trichomes are occasionally seen on the petiole. The trichome has long biseriate stalk and club shaped secretory body. It is 80 µm long; the body of the trichome is 30 µm thick. The distal part (the part near the leaf) of the petiole has wide, shallow adaxial concavity and semicircular abaxial part with an undulate outline.

It is 750 µm in vertical plane 1.3 mm in the horizontal plane. The petiole has a thin epidermal layer, outer zone of collenchyma and central zone of wide, angular, compact parenchyma cells. The vascular system has a circle of seven discrete strands; of the seven bundles, one is median abaxial, two are lateral, two are adaxial lateral, and one is the adaxial median. The vascular strand has wide band of xylem elements, and thin are of phloem.

**Stem Plate 4:** The stem measuring nearly 1mm was studied. It has an undulate outline with shallow ridges and furrows. The stem has a thin epidermal layer of small circular cells with the papillate outer cuticular layer. Cortex is fairly wide and parenchymatous and measures 100 µm wide. Inner to the parenchymatous cortex, there is a discontinuous cylinder of thick, are-shaped gelatinous fibers. Inner to the segment of g-fibers is a narrow zone of outer phloem-parenchyma and inner phloem element. The elements of the phloem are arranged in parallel series; the cells are small, and the size of the cells increases towards the periphery. Secondary xylem is a hollow cylinder with a wavy outline. The inner part of the xylem cylinder is the primary xylem, and the outer part is the secondary xylem. Secondary xylem consists of regular radial parallel lines of xylem fibres, diffusely distributed solitary and clusters of vessels and one or two cells wide, straight rays. The inner primary xylem consists of two or five parallel chains of xylem elements, surrounded by a wide zone of thick-walled angular parenchyma cells. The pith is wide and comprising of central thin walled, circular empty cells and an outer zone of circular, thick-walled cells with dense accumulation of starch grains.

**Root:** The root measuring 1.25 mm thick was studied. It consists of well-developed periderm; wide cortex, secondary phloem, and circular central core of secondary xylem. Periderm **Plate 5** is wide and uniformly thick all around the root. These are shallow, wide irregular fissures at certain places. The periderm cells are thin-walled, subsided and tabular in shape; they are arranged in regular radial and circular rows. The periderm zone is 100 µm thick. Cortex is inner to the periderm. The cortical cells have a dense accumulation of calcium oxalate druses. The inner boundary of the cortex consists of small masses of gelatinous fibers. Secondary phloem is narrow, and it is not quite distinct from the cortex. Crystals are abundant in the phloem tissue also. Secondary xylem is roughly circular in sectional view. It consists of wide and narrow vessels scattered in the xylem. Most of the outer vessels are wider. They are circular and thick walled. The narrow vessels are 20 µm wide; the wider ones are 70 µm in diameter. The xylem fibres are thick walled and lignified with the narrow lumen. The fibres are random in alignment.

**Cell Inclusions:** Calcium oxalate crystals and starch grains are major inclusions. Crystals are either druses or prismatic type. Druses are seen in the phloem region of the midrib, inner cortical cells of the stem, secondary phloem and pith cells of the stem. Prismatic type crystals are seen in the xylem rays of the stem. Starch grains are abundant in the pith parenchyma. The grains are spherical with central hilum. They are a simple type.

**Powder Microscopy:** In the powdered preparations of the stem are seen epidermal peelings of the stem, vessel elements, and fibers. Epidermal peelings of the stem are seen in surface view. The epidermal cells are narrow, rectangular, thick walled and are arranged in parallel vertical files. Stomata are seen in wide vertical band in between the nonstomatiferous regions. The epidermal cells in the stomatiferous region are polygonal in outline. The cells walls are thick and possess dense simple pits. The stomata are paracytic type as in the leaf. Long, cylindrical vessel elements are frequently seen in the powder **Plate 6.** The vessel elements have wide, circular, horizontal perforations at the end walls. Their lateral walls have dense elliptical scalariform or reticulate pits. The vessel elements are 200 µm long and 70 µm wide. Broken and fragmentary fibres are often seen in the powder. Some of the
fibres narrow and thick-walled; others are wide and thin walled. Parenchyma cells are also frequently seen in the powder.

**Fluorescence Analysis:** The behavior of the powdered drug in different solution and their extracts towards ordinary light and U.V light were observed, and the results were recorded in Table 1. It can be used as a diagnostic tool for testing adulteration if any. Under fluorescent light leaf powder showed different colors in various extracts.

<table>
<thead>
<tr>
<th>Light source</th>
<th>Powder + aqueous NaOH</th>
<th>Powder + HCl</th>
<th>Powder + 50% HNO₃ distilled water</th>
<th>Chloroform Pet. ether Benzene Ethanol Acetone</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visible light</td>
<td>Dark brown</td>
<td>Light yellow</td>
<td>Yellow</td>
<td>Yellow</td>
</tr>
<tr>
<td>U.V.</td>
<td>Green</td>
<td>Green</td>
<td>Green</td>
<td>Green</td>
</tr>
</tbody>
</table>

**Quantitative Determination:** The determination of ash is useful for detecting low-grade products, exhausted drugs, and excess of sandy or earthy matter. The total ash value is useful to exclude drugs, which have been coated with chalk, lime or calcium sulphate. The physicochemical parameters like total ash, acid soluble ash, water soluble ash, moisture content, extractive values were determined by standard methods and presented in Table 2.

<table>
<thead>
<tr>
<th>Extracts</th>
<th>Steroids</th>
<th>Triterpenoids</th>
<th>Reducing Sugars</th>
<th>Alkaloids</th>
<th>Phenols</th>
<th>Flavonoids</th>
<th>Catechins</th>
<th>Saponins</th>
<th>Tannins</th>
<th>Anthroquinones</th>
<th>Amino acids</th>
</tr>
</thead>
<tbody>
<tr>
<td>Petroleum ether</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
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<td>-</td>
</tr>
<tr>
<td>Chloroform</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
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<td>-</td>
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<tr>
<td>Benzene</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
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<td>+</td>
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<tr>
<td>Ethanol</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
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<td>+</td>
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</table>

Tannin is medicinally used as an astringent, hemostatic to treat burns and as a heavy metal antidote\(^{11}\). Similarly, this species also contains tannin, and this may be the reason for the wound healing property of the plant. Many studies reported that phenolic compounds in herbs significantly contributed to their antioxidant and pharmaceutical properties \(^{12, 13, 14}\). Some studies claim that the antioxidant phenolic compounds present in herbs might also play a major role in their antimicrobial effect \(^{15}\). The presence of the secondary metabolites in this selected species may involve in treating a variety of human ailments.

The plant studied here can be seen as a potential source of useful drugs. Further studies are going on this plant to isolate, identify, characterize and elucidate the structure of the bioactive compounds. Bioactivity guided chemical studies may yield the relation between the pure compounds and defined...
biological activities. This will lead to the utilization of this plant, which can be cultivated in most parts of our country, for different commercial pharmaceutical products.

CONCLUSION: The macroscopic and microscopic characters, fluorescence analysis, physicochemical determination, and preliminary phytochemical screening can be used as a diagnostic tool in the correct identification of plants. The adulterants if any in the plant material can also easily identified by these studies. The result of the present phytochemical study may serve as a guide in the selection of plant for further work on the isolation and elucidation of the active compounds

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