PHARMACOGNOSTIC, PHYSICOCHEMICAL AND PHYTOCHEMICAL STUDIES OF SEBASTIANIA CHAMAEOEA (L).MUELL.ARG

Anjali Asok*, Mahesh C.D, Shiva Manjunatha M.P, Seema Pradeep

*Post Graduate Scholar, 1Professor, 3Botanist, 1Professor and HOD, Department of Dravyaguna, Sri Sri College of Ayurvedic Science and Research, Karnataka, India.

KEYWORDS: Sebastiania chamaelea (L).Muell.arg, Euphorbiaceae, Standardization.

ABSTRACT

Background: The plant Sebastiania chamaelea (L).Muell.arg belongs to Euphorbiaceae family and is a native to Africa, southern Asia and Australia. It is used as a folk medicine for the management of pain in various parts of the world. Very few studies have been conducted on the S.chamaelea. This present study was conducted to establish quality control parameters of the plant.

Methodology: The study deals with detailed macro morphology, microscopy of transverse section of root, stem, leaves and fruit. The powder microscopy of seeds was carried out as well. The other parameters like physicochemical constants and phytochemical screening were studied using the standard protocol.

Results: The study provides referential information for the correct identification and standardization of crude drug of Sebastiania chamaelea (L).Muell.arg.

INTRODUCTION

Medicinal plants are gaining more popularity in treating various ailments of the society. People are aware about the adverse effects caused by usage of mainstream medicines. The main challenge faced by Ayurvedic medicines is lack of quality control and poor evidence of documentation. Hence there is a need for standardization of plants which are used as medicines. This is achieved through prompt identification by understanding characterization, pharmacognostical studies and phytochemical studies. Quality assurance of starting material is an essential step to ensure reproducible quality of herbal medicine which will help us to justify its safety and efficacy. The main objective of this study is to supplement constructive information with regards to its identification, characterization and standardization of Sebastiania chamaelea (L). Muell. arg.

The plant Sebastiania chamaelea(L). Muell. arg belonging to Euphorbiaceae group is commonly known as 'Bhumi eranda'/ 'Kodiavanakku' among the traditional healers. The plant is a main ingredient in many preparations given for pain management. It is quoted by a well-known author D. Sriman Namboothiri in his books 'Yogamrutham' and 'Chikitsamanjari' in the name 'Kodi avankku'. The plant has no references in Ayurvedic classics. The plant is quoted in various textbooks of Indian floras. The Wealth of India, Indian Medicinal Plants, Glossary of Indian Medicinal Plants, Referenced checklist of Medicinal Plants of India and many other texts describes the plant.

MATERIALS AND METHODS

Botanical Description

Botanical name- Sebastiania chamaelea (L).Muell.arg
Synonyms- Microstachys chamaelea, Tragia chamaelea. L.
Family - Euphorbiaceae.
Collection and identification of Plant

The plant material Sebastiania chamaelea (L).Muell.arg (Euphorbiaceae) was collected from the campus of Sri Sri College of Ayurvedic Science & Research, Bangalore and preserved as per the standard method. The taxonomic identity was confirmed by DR.Shivamanjunatha M.P., Botanist, Department of Dravyaguna, Sri Sri College of Ayurvedic Science and Research, Karnataka, India.
Ayurvedic Science & Research, Bangalore. The voucher specimen DGMP5001 was preserved in the Herbarium of Department of Dravyaguna. The plant were thoroughly washed and then dried under shade for one week. The dried material were ground in a mixer grinder and sieved. The powder was stored in air sealed polythene bags at room temperature until further use.

**Macroscopic Evaluation [17]**

The morphology of the plant was studied with the help of available literatures. The macroscopic characters of *Sebastiania chamaelea* (L).Muell.arg were observed for the following features- Colour, Texture, Taste and Odour.

**Microscopic evaluation [18]**

For qualitative microscopic analysis transverse section of root, stem, leaf and fruit were made. Staining procedure was performed as per standard procedure. Various identifying characters were studied with staining and the pictures were taken in Digital camera microscope. Powder microscopy of the seeds was also conducted according to standard procedure.

**Physicochemical Evaluation [19]**

Physico-chemical parameters such as foreign organic matter, moisture content, ash value, extractive values, pH and specific gravity were determined according to standard procedures done for medicinal plants.

**Phytochemical Evaluation [20]**

The qualitative chemical tests carried out for the identification of the natural phyto-constituents present in the powdered crude drug. The tests were carried out using conventional protocols. The quantification of each phyto constituent was also carried out.

**Thin Layer Chromatography [21]**

Thin layer chromatography was carried out with a Solvent system - Ethyl acetate (9): methanol (1) were used. The procedure was carried out using standard protocol and the Rf value was calculated and recorded.

**RESULTS & DISCUSSION**

**Morphological features of Sebastiania chamaelea (L).Muell.arg [4-15]**

**Habit:** *Sebastiania chamaelea* (L).Muell.arg is an erect to sprawling annual to perennial glabrous herb or many stemmed shrub with slender stem, growing from a yellowish-brown taproot around 15cm long. It usually grows up to 50cm tall, occasionally to 100cm.

**Roots:** Strong, long thin tap root, yellowish brown, with a smooth surface, about 15cm in length without odour and taste.

**Stem:** Stem green to brown, at maturity it appears brown, smooth surface with longitudinal rims and no odour and bitter taste. Usually many from the root, ascending 1-2ft, slender, grooved, ribbed, or terete, dichotomously branched.

**Leaves:** Small, regularly alternate; symmetric. The ventral side is dark green and dorsal light green. The margin of leaves is very dense with glandular brown coloured teeth, almost touching apex. Distant ½-3in, by ¼-½in, sessile, petiole short (less than 1 cm long; absent to long in some Neotropical species), glandless; blade elliptic, ovate or linear, finely serrulate, base tapering, mid-nerve strong, 0.3–2 cm wide, base acute to sub-cordate, margin with very dense, minute and persistent glandular teeth, 0.3–0.6 mm apart and often nearly touching each other (rarely fused into an entire, glandulous margin), apex rounded, acute or mucronate, above glandless and glabrous, lower surface paler and smooth to papillate but never white, glabrous to pilose, at base often with few marginal to sub-marginal glands, basal ones slightly larger, secondary veins arching and usually looped but often hardly visible, tertiary veins not visible but presumably reticulate.

**Inflorescence:** Flowers are yellowish, simple, without sterile basal region, monoecious terminal and axillary, often opposite to leaves. Appears in 5-12 mm long spikes, laterally or at the end of branches. Male flowers are numerous, spirally arranged at upper part of inflorescence axis, female ones 1 or several inserted at lower part of inflorescence axis. Disk 0. Male flower calyx minute, membranous, unequally 5-lobed or partite. Stamens 2-4; filaments short free or nearly so; anther cell distinct, contiguous, distinct, parallel. Pistilloid 0. Female flower calyx 3 lobed or partite, longer than male, obovate, acute, lacerate anddidiate. Ovary exerted 3-celled; styles free or connate at the base, entire revolute or spreading; cells 1-ovuled, capsule 6-8 mm, long, globose of 3 cocci separating from a columella, endocarp crustaceous.

**Fruits:** Fruits glabrous, smooth except for the two dorsal rows of spinesules, thinly crustaceous, with short (0–2 mm long) pedicel; 3-seeded, sub-globosely oblong with very regular shape, usually with 6 rows of spine-like excrescences, dry, dehiscing regularly along the septa, glabrous to hirsute; mericarps with a thin exocarp (fruit length/pericarp thickness > 10/1), septa very regular, with a small separate basal triangle and 1 furcate
vascular strand; remaining central columella very regular, slightly alate with parallel margins over its whole length.

**Seeds:** Carunculate, oblong or subglobose, rounded at both ends, strophiolate, mottled. Endosperm fleshy; testa smooth, cotyledons broad, flat.

**Table 1: Organoleptic Features**

<table>
<thead>
<tr>
<th>Sl. No</th>
<th>Organoleptic Characters</th>
<th>Powder</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Colour</td>
<td>Light to dark green</td>
</tr>
<tr>
<td>2</td>
<td>Texture</td>
<td>Smooth</td>
</tr>
<tr>
<td>3</td>
<td>Taste</td>
<td>Bitter</td>
</tr>
<tr>
<td>4</td>
<td>Odour</td>
<td>Characteristic odour</td>
</tr>
</tbody>
</table>

**Microscopic evaluation**

**Root:** Transverse and vertical section of root having uniseriate epidermis, absence of root hairs. Parenchymatous cortex with intercellular spaces. The secondary vascular tissues forms continuous cylinder and the primary xylem gets embedded in it. Primary phloem is in crushed condition. Medullary rays are transversed in the xylem and phloem through cambium, which is characteristic feature of the root. Pith is completely absent.

**Stem:** Transverse and vertical section of stem shows epidermis with single layer and no trichomes. Cells appears almost rectangular with compact arrangement. Cortex is made up of collenchyma. Stele contains sclerenchymatous cells in the patches forming pericycle in outer part. Pith rays can be seen, separating the vascular bundles. Presence of sclerenchymatous patches in pericycle and with a large portion of pith.

**Leaf:** Transverse and vertical section of leaf shows single layered epidermis on both surfaces, upper having cuticle. Mesophyll with palisade parenchyma as the major portion, than the spongy parenchyma. Vascular bundle in crescent shape at the mid rib and possess xylem towards upper surface and phloem towards lower surface. Parenchyma is found in the middle.

**Fruit:** The pericarp consists of uniseriate exocarp and multicellular appendages. The mesocarp consists of 3 tissues regions: the outer Parenchymatous, the middle elongated and thin walled cells and inner with palisade like cells. The endocarp is similar to middle mesocarp with sclerenchymatous cells. In the central region of the septum, ventral vascular bundles, parenchyma and sclerenchyma.

**Powder microscopic study of seeds:** Powder microscopy of the seeds of *Sebastiania chamaelea (L).Muell.arg* showed presence of Scleroid cells, Mucilaginous cells, Epithelial cells, Calcium oxalate crystals, Tannins, Resinous cells, Oil globules, Embryo and Starch grains.

**Table 2: Physicochemical evaluation**

<table>
<thead>
<tr>
<th>S. No</th>
<th>Parameters</th>
<th>Sebastiania chamaelea (L).Muell.arg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Foreign matter</td>
<td>Nil</td>
</tr>
<tr>
<td>2</td>
<td>Loss on drying</td>
<td>11.49%</td>
</tr>
<tr>
<td>3</td>
<td>Total ash</td>
<td>8.18%</td>
</tr>
<tr>
<td>4</td>
<td>Acid insoluble ash</td>
<td>3.31%</td>
</tr>
<tr>
<td>5</td>
<td>Water soluble extractive value</td>
<td>25.5%</td>
</tr>
<tr>
<td>6</td>
<td>Alcohol soluble extractive value (25% alcohol)</td>
<td>18.8%</td>
</tr>
<tr>
<td>7</td>
<td>pH value</td>
<td>3.05</td>
</tr>
<tr>
<td>8</td>
<td>Specific gravity</td>
<td>1.0005g/ml</td>
</tr>
</tbody>
</table>

**Table 3: Phytochemical evaluation**

<table>
<thead>
<tr>
<th>Constituents</th>
<th>Tests</th>
<th>Sebastiania chamaelea (L).Muell.arg</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Aqueous</td>
<td>Alcoholic</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>Wagner's test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Dragendroff's test</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Ferric chloride test</td>
<td>+</td>
</tr>
<tr>
<td>Saponins</td>
<td>Foam test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Froth test</td>
<td>+</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>Molisch's test</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Benedict's test</td>
<td>+</td>
</tr>
<tr>
<td>Sl no.</td>
<td>Constituents</td>
<td>Percentage</td>
</tr>
<tr>
<td>-------</td>
<td>------------------------------</td>
<td>------------</td>
</tr>
<tr>
<td>1</td>
<td>Total Alkaloids</td>
<td>1.61%</td>
</tr>
<tr>
<td>2</td>
<td>Total Flavonoids</td>
<td>2.33%</td>
</tr>
<tr>
<td>3</td>
<td>Total Poly phenols</td>
<td>15.84%</td>
</tr>
<tr>
<td>4</td>
<td>Total Carbohydrates</td>
<td>72.83%</td>
</tr>
<tr>
<td>5</td>
<td>Total Proteins</td>
<td>9.77%</td>
</tr>
<tr>
<td>6</td>
<td>Total fats</td>
<td>2.25%</td>
</tr>
<tr>
<td>7</td>
<td>Total Tannins</td>
<td>5.23%</td>
</tr>
<tr>
<td>8</td>
<td>Total sugar</td>
<td>7.9%</td>
</tr>
<tr>
<td>9</td>
<td>Total starch</td>
<td>10.54%</td>
</tr>
</tbody>
</table>

**Table 4: Quantification of the Phyto-Constituents**

**Thin Layer Chromatography**

Under UV 366nm at the Rf values obtained are 0.45 (Pink), 0.51 (Light pink), 0.63 (Blue), 0.87 (Light blue), 0.93 (Light pink), 0.97 (Dark pink). On exposure to Sodium vapour, the Rf values obtained are 0.45(Brown), 0.76&0.87 (Pale yellow), 0.97 (Green). After spraying with 8% methanolic Sulphuric acid reagent, 0.47(Yellow), 0.55(Green), 0.59(Yellow) 0.66 (Light violet), 0.76 (Light green) and 0.86 (Dark green).

**DISCUSSION**

The diagnostic feature of *Sebastiania chamaelea* (L).Muell.arg is the tri-lobed fruit with often red spikes. Flower is slender with axillary spikes. These botanical features exactly match to that of Bhoomi eranda. Hence it can be concluded both are the same. Powder microscopy of the seeds of *Sebastiania chamaelea* (L).Muell.arg shows Scleroid cells, mucilaginous cells, epithelial cells, starch grains, oil globules, calcium oxalate crystals, tannins and embryo

Ash value of the drug was found to be 8.18%, this might be due to presence of inorganic salts naturally occurring in the drug or adhering to it. This also depends on the state of collection of the plant. Loss on drying of the plant was found to be 11.49%. This may be due to probable presence of more water and volatile impurities.

It was observed that the % water-soluble extractive values were higher than alcohol-soluble extractive. Water-soluble and Alcohol soluble extractive value plays an important role in evaluation of crude drugs. Less extractive values may be due to addition of exhausted material, adulteration or incorrect processing during drying or storage or formulating. pH value is 3.05 indicative of strong acidic nature of the plant. Due to less pH most nutrients can be dissolved easily. This may be also due to poor soil. Majority of the active constituents were identified as alkaloids, flavonoids, saponins, tannins, polyphenols and carbohydrates have been reported to be promising analgesic action in animal model. Alkaloids and flavonoids are well known for their ability to inhibit pain perception. Published biological effects of polyphenols such as flavonoids and tannins appear to justify some of the traditional, folkloric uses of the plant in the control or management of painful conditions. Saponins inhibits the cyclooxygenase pathway and reduces prostaglandin E2 production.

**CONCLUSION**

The plant -*Sebastiania chamaelea* (L).Muell.arg was studied for its macro morphology and microscopy along with physico chemical and phytochemical parameters. The study has set preliminary standards which helps to establish the correct identity of the plant. Hence this study helped to conclude the authenticity of plant *Sebastiania chamaelea* (L).Muell.Larg.

**REFERENCES**


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Plate 1: Morphology of S. Chamaelea

Fig1: Root of Sebastiania chamaelea (L).Muell.arg
Fig2: Stem of Sebastiania chamaelea (L).Muell.arg
Fig3: Leaves of Sebastiania chamaelea (L).Muell.arg
Fig4: Fruits with inflorescence of Sebastiania chamaelea (L).Muell.arg

Plate 2: Morphology of Plant Parts of S. Chamaelea
Plate 3: Microscopic Structure of Stem *Sebastiania Chamaelea* (L).Muell.Arg.
A. Outline of Transverse section of root  
   a: Cork  

B. Portion Enlarged.

C. Enlarged view of patches of Vascular Bundles.

D. Vascular bundles  
   b: Medullary rays  
   c: Xylem fibres  
   d: Xylem vessels

Plate 4: Microscopic Structure of Root of *Sebastiania Chamaelea* (L). Muell. Arg.
Plate 5: Microscopic Structure of Leaf of *Sebastiania Chamaelea* (L).Muell.Arg

- **A. Outline of T.S. Leaf**
- **B. Portion Enlarged – Midrib**
  - j: lower epidermis, k: upper epidermis
- **C. Portion enlarged – Midrib**
  - l: Cuticles, m: Polysade parenchyma cells, n: Spongy parenchyma cell
- **D. Portion enlarged Midrib**
- **E. Portion enlarged of Upper epidermis**
  - o: Stomatal opening
- **F. Portion enlarged**
  - l: Calcium oxalate crystals
<table>
<thead>
<tr>
<th>A. Fruits</th>
<th>B. Fruit, Cocci</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. Rigma</td>
<td>D. Seed Coat</td>
</tr>
<tr>
<td>E. Fruit and Rigma</td>
<td>F. Caurncle</td>
</tr>
</tbody>
</table>


<table>
<thead>
<tr>
<th>A. Scleroid cells</th>
<th>B. Tannins</th>
<th>C. Mucilaginous cells</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Scleroid cells" /></td>
<td><img src="image2" alt="Tannins" /></td>
<td><img src="image3" alt="Mucilaginous cells" /></td>
</tr>
<tr>
<td>D. Oil Globules</td>
<td>E. Rigma</td>
<td>F. Caruncle</td>
</tr>
<tr>
<td><img src="image4" alt="Oil Globules" /></td>
<td><img src="image5" alt="Rigma" /></td>
<td><img src="image6" alt="Caruncle" /></td>
</tr>
<tr>
<td>G. Calcium oxalate crystals</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

![Calcium oxalate crystals](image7)
Plate 8: TLC Plates of *Sebastiania Chamaelea* (L).Muell.Arg

TLC Plate of *Sebastiania chamaelea* (L).Muell.arg

On Exposure to sodium vapour

On spraying 8% Methanolic Sulphuric acid