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PHARMACOGNOSTICAL AND PRELIMINARY PHYTOCHEMICAL EVALUATION OF *TOXOCARPUS BEDDOMEI* GAMBLE (L)

M.Purushothaman*[1], J.Karthi [2]

[1] Scient Institute of Pharmacy, Ibrahinpatnam, Hyderabad-501506, India.
[2] Sun Rise University, Alwar, Rajasthan – 301030, India

***Corresponding author:** vedpurushoth@gmail.com

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Key Words

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ABSTRACT

The whole plant material of Toxocarpus beddomei Gamble (L) was collected and powedered. The powdered material was subjected to successive soxhlet extraction with petroleum ether, chloroform, ethanol and finally macerated with water so as to get respective extracts. Physicochemical parameters such as total ash value, acid insoluble ash value, water soluble ash value and sulphated ash value were determined which were 12.31, 11.28, 7.54, and 8.57% respectively. Moisture content, foreign organic matter, crude fibre content, alcohol soluble extractive and water soluble extractive were also determined. The percentage yield of petroleum ether, chloroform, ethyl acetate, ethanol and water were 4.5, 8.2, 4.6, 9.2 and 4.6% respectively. Preliminary phytochemical analysis of different extracts was carried out. The results were positive for alkaloid, glycoside, sterols, flavonoids and phenolic compounds in petroleum ether extract. Chloroform extract showed positive test for alkaloid, sterols and saponins only, ethyl acetate extract showed positive test for alkaloids, tannins and flavonoids, ethanolic extract exhibited positive test for alkaloids, flavonoids, glycosides, tannins, amino acids and saponins whereas aqueous extract was found to be positive for flavonoids, carbohydrates, glycosides and amino acids. These secondary metabolites are the active constituents of Toxocarpus beddomei Gamble (L). and may be responsible for its pharmacological activities.

INTRODUCTION:

Herbal drugs play an important role in health care programs especially in developing countries. Ancient Indian literature incorporates a remarkably broad definition of medicinal plants to be potential sources of medicinal substances [1]. For centuries, plant and plant products have been used for treating various illnesses. Today, several medicinal plants and their products are still in use, being employed as home remedies, over the counter drugs as well as raw materials for the pharmaceutical industry and they represent a substantial proportion of the global drug market [2]. However a key obstacle, which has hindered the acceptance of the alternative medicines in the developed countries, is the lack of documentation and stringent quality control. There is a need for documentation of research work carried out on traditional medicines [3]. Therefore it has become extremely important to make an effort towards standardization of the plant material to be used

as medicine. The process of standardization can be achieved by stepwise pharmacognostic studies [4]. These studies help in identification and authentication of the plant material. Correct identification and quality assurance of the starting materials is an essential prerequisite to ensure reproducible quality of herbal medicine which will contribute to its safety and efficacy [5].

Toxocarpus beddomei Gamble (L) under the family Apocynaceae, which has slender climbers. Leaves opposite, up to 10 x 4cm, ovate, lanceolate or elliptic-lanceolate, acuminate, cuneate at base, glabrous, smooth and pale when dry; main nerves 7 pairs. Calyx very small, 5-lobed, with or without minute scales at the base within; corolla-tube short, .8 cm long, villous within; corona scales longer than the stamens, obtuse. Stamens inserted at the base of the corolla; filaments connate; anthers small, minutely fimbriate at tip. Fruit of 2 divaricate follicular mericarps; seeds oblong, flattened, tipped with a silky coma [6-9]. The present study was designed to investigate the pharmacognostic and phytochemical properties of *Toxocarpus beddomei Gamble (L)*

MATERIALS AND METHODS

Collection of plant material and authentication:

Toxocarpus beddomei Gamble (L) was collected from the tribal belts of the local area of Kanniyakumari district, Tamilnadu, India. The plant was identified, confirmed and authenticated by Dr. Madhava Chetty, Assistant Professor, Department of Botany, Sri Venkateswara University, Andhrapradhesh. After authentification the whole plant of *Toxocarpus beddomei Gamble* were collected in bulk and washed under running tap water to remove adhering dirt. Then leaves were shade dried. The dried materials were made into coarse powder by grinding in mechanical grinder and stored in a closed air tight container for further use.

Microscopic study

Microscopy of plant material is performed to distinguish it from the allied drugs and adulterant. The dried leaf was soaked overnight in water to make it smooth enough for transverse section. Paraffin wax embedded specimens were sectioned using the rotatory microtome. The thickness of section was 10-12 µ m. Very fine section was selectively subjected to staining reaction with staining reagent safranin one precent solution and light green 0.2% solution. Slides were cleaned in xylol and mounted in mountant (DPX). Photomicrographs were taken using trinocular microscope [10-14].

Powder studies

Microscopic study

The shade dried plant material were mechanically pulverized to coarse powder and sifted through 40 mesh sieve. Take a pinch of powder was taken on slide and mounted with phloroglucinol, hydrochloric acid and glycerine. Slide was seen under microscope.

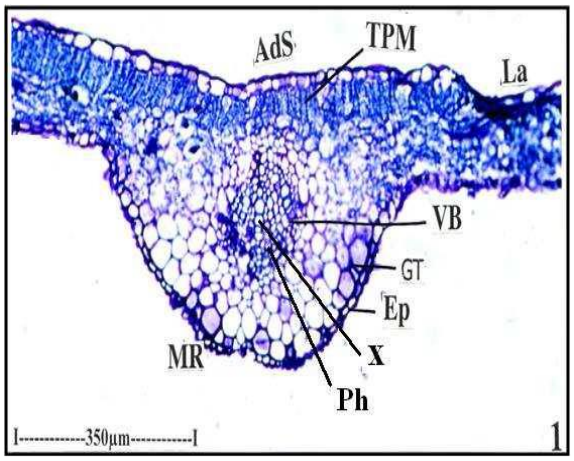

Figure 1. T.S of Leaf through midrib with lamina

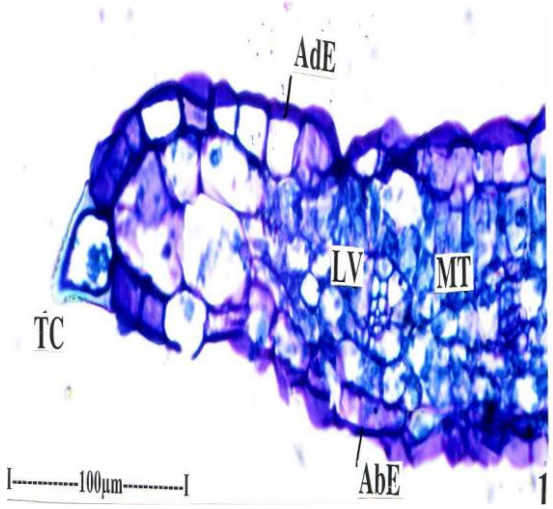

Figure 2. T.S. of the leaf margin

Determination of physicochemical parameters [15-18]:

The dried plant material was subjected for determination of physicochemical parameters such as total ash value, acid insoluble ash value, water soluble ash value, moisture content, pet ether soluble, chloroform soluble, ethyl acetate soluble, alcohol soluble extractive and water soluble extractive, Crude fiber content, Loss on drying, Swelling index, Foaming index, Tanins contents, Bitterness value, and Haemolytic value.

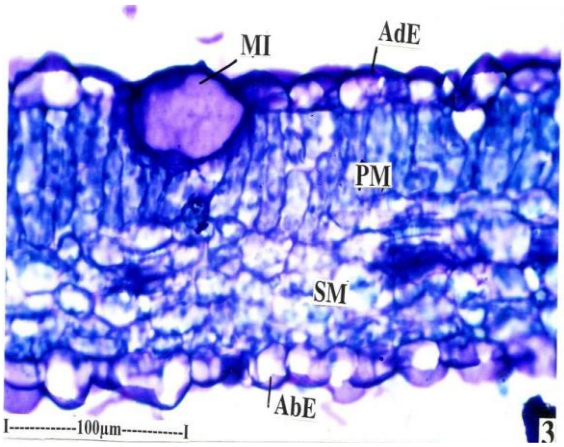

Figure 3. T.S. of the lamina showing mucilage

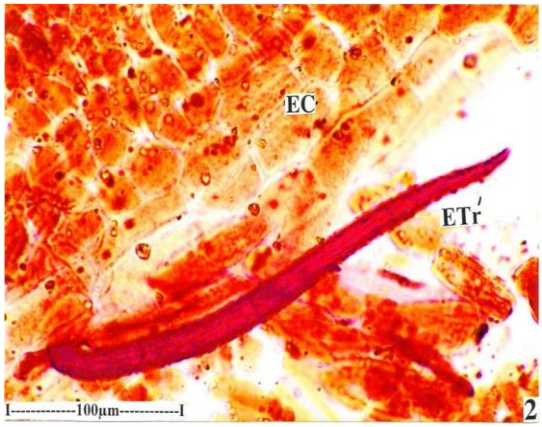

Figure 4. Epidermal trichomes

Extraction of powdered plant material:

The shade dried powdered plant material was subjected to sequential soxhlet extraction using the solvents of different polarity such as petroleum ether (40-600), chloroform, ethyl acetate, ethanol and finally macerated with water so as to get respective extracts. Cold maceration was also done using ethanol and water. The extracts were filtered individually, evaporated to dryness and the percent yields of all the extracts were determined. All the extracts were then stored in a refrigerator till further analysis.

Preliminary phytochemical analysis:

Preliminary qualitative phytochemical analysis of all the extracts was carried out by employing standard conventional protocols.

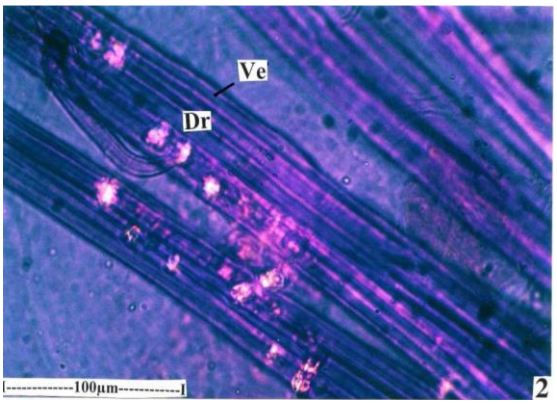

Figure 5. Druses in the leaf powder

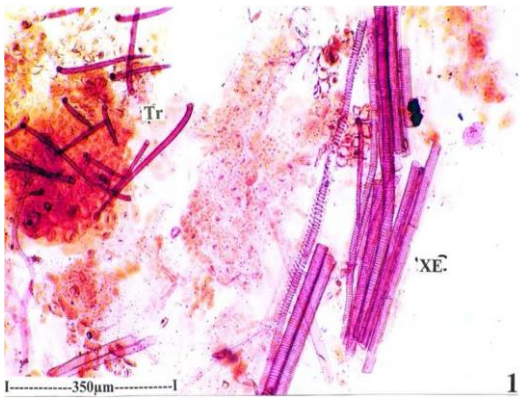

Figure 6. Trichomes and Xylem elements

Table 1. Total ash, water soluble ash, acid insoluble ash and sulphated ash of aerial plant parts of the powder

S. No.	Ash	Percentage in TB
1	Total ash	12.31
2	Water soluble ash	11.28
3	Acid insoluble ash	7.54
4	Sulphated ash	8.57

RESULTS AND DISCUSSION:

Epidermis (Upper and lower)

Polygonal tubular cells having straight anticlinical walls. The inner preclinical wall contain mucilage and can be stained with ruthenium red. The Epidermal trichomes are

simple, covering conical, thin walled with warty cuticle and are curved at the base. The stomata present are of paracytic type. (Paracytic type of stomata can only be seen in the surface view of epidermis)

Table 2. Extractive values of roots powder with various solvents

S. No.	Solvent	Extraction period (h)	TB Extractive values (%) w/w
1.	Petroleum ether (60	24	4.5
2.	Chloroform	24	8.2
3.	Ethyl acetate	24	4.6
4.	Ethanol	24	9.2
5.	Aqueous	24	4.6

Palisade Layer : A single layer is present on upper and lower surface(isobilateral leaf)

Spongy Tissues : - Parenchymatous cell which are loosely arranged, clusture crystals of calcium oxalate are present in them. There are few vessels having special thickness

Midrib and Lamina : Showing collenchymas, Slerencychyma layer on both the sides of the vascular bundle consisting of xylem and phloem

Epidermal trichomes :

Unicellular, unbranched, covering – type or non – glandular type having thick lignified walls with echinate circle, 450 -600 mm long and 30mm thick

Calcium oxalate crystal:

Sphaero crystals, diffuse in distribution and occur in the mesophyll tissue and may be the associated with veins.

Broken xylem elements :

Broken xylem elements of the veins are seen scattered in the powder. The xylem elements have spiral, annual and scalariform lateral wall thickenings

Epidermal peelings :

Fragments of epidermal layer with stomata are seen into powder. The stomata are paralytic type. The epidermal cells have straight, thin walls.

Ads – Adaxial side, Ep – Epidermis, Gt – Ground tissue, La – Lamina, MR – Midrib, Pa – Parenchymatous ground tissue, Ph – Phloem, TPM – Transcurrent palisade mesophyll, VB – Vascular bundle, X – Xylem. AbE – Abaxial epidermis, AdE – Adaxial epidermis, LV – Lateral vein, MT – Mesophyll tissue, TC – Thick walled cell**,** SM – Spongy mesophyll, EC – Epidermal cells, ETr – Epidermal trichomes, Dr – Druses, Ve – Vein, Tr – Trichomes, XE – Xylem elements

Table 3. Crude fiber content, Loss on drying, Swelling index, Foaming index, Tanins contents, Bitterness value, Haemolytic value

S. No.	Parameter	Observation in TB
1.	Crude fiber content	8.25%
2.	Loss on drying	12%
3.	Swelling index	No significant result
4.	Foaming index	No significant result
5.	Tannins	24
6.	Bitterness value	2.1 unit / g
7.	Haemolytic activity	27.35%

The Total ash; water insoluble ash; sulphated ash; and acid insoluble ash (12.31%,11.28%,7.54%,8.57%). Extracting values, i.e. petroleum ether (PE); chloroform (CF); ethyl acetate (EA); ethanol (ET) and aqueous extract (4.5%,8.2%,4.6%,9.2%,4.6%). The fiber content was found to be 8.25%. Plant bitterness was found to be 2.1 unit / g. The plant also has hemolytic potential. The tannin content was found to be 24.

The preliminary phytochemical investigations of various extracts of *Toxocarpus beddomei Gamble* were studied. The results were positive for alkaloid, glycoside, sterols, flavonoids and phenolic compounds in petroleum ether extract. Chloroform extract showed positive test for alkaloid, sterols and saponins only, ethyl acetate extract showed positive test for alkaloids, tannins and flavonoids, ethanolic extract exhibited positive test for alkaloids, flavonoids, glycosides, tannins, amino acids and saponins whereas aqueous extract was found to be positive for flavonoids, carbohydrates, glycosides and

Table 4. Preliminary phytochemical screening of *Toxocarpus beddomei* Gamble

Sr. no.	Plant Constituents Test / Reagent	Pet. Ether extract	Ethyl acetate extract	Chloroform extract	Ethanol extract	Aqueous extract
1.	**ALKALOIDS** Mayer's reagent Dragendroff's reagent Wagner's reagent	+ + +	- + +	- + -	+ + +	- - -
2.	**GLYCOSIDES** Killer-Killani test Sodium nitropruside test Borntrager test	+ - +	- - -	- - -	- - -	+ + +
3.	**CARBOHYDRATES** Molisch's reagent Fehling solution	- -	- -	- -	+ +	+ +
4.	**STEROLS** Liebermann- Burchard's test Salkowski test Hesses reaction Hersch reaction	+ + + -	- - - -	+ - + +	+ - + +	- - - -
5.	**SAPONINS** Foam test Sodium bicarbonate test	- -	- -	+ +	+ +	- -
6.	**PHENOLIC COMPOUNDS & TANNINS** Ferric chloride solution Lead acetate solution	+ +	+ +	- -	+ +	- -
7.	**PROTEINS & AMINO ACIDS** Biuret test Millon's reagent Ninhydrin reagent	- - -	+ - +	- - -	+ + +	- + +
8.	**FLAVANOIDS** Shinoda/Pew test Ammonia test	+ +	+ +	- -	+ +	+ +

amino acids. These secondary metabolites are the active constituents of *Toxocarpus beddomei Gamble (L)*. and may be responsible for its pharmacological activities.

CONCLUSION:

Toxocarpus beddomei Gamble (L) powder was subjected for preliminary Pharmacognostic standardization including

phytochemical screening. The present investigation adds to the existing knowledge of *Toxocarpus beddomei Gamble (L)* and will be quite useful for development of a formulation for treating various ailments.

REFERENCES

1. Shankar D. and Ved D.K. (2003). Indian Forester. 129, 275-288.
2. World Health Organization Geneva, Quality control methods for medicinal plant materials, Type set in Hong kong, Printed in England, ISBN 92 415 45100 (NLM classification QV 766).
3. Dahanurkar S.A., Kulkarni R.A. and Rege N.N. (2000). Ind.J. Pharmacol., 32 ; 81-118.
4. Ozarkar K.R. (2005). Studies on antiinflammatory effects of two herbs Cissus quadrangularis Linn. andValeriana wallichi DC using mouse model. Ph.D. Thesis, University of Mumbai, Mumbai.
5. Thomas S., Patil D.A., Patil A.G. and Naresh Chandra, (2008). Pharmacognostic evaluation and physicochemical analysis of Averrhoa carambola L. fruit. J of Herb Med and Toxicol. 2(2), 51-54.
6. Hassler M. (2017). World Plants: Synonymic Checklists of the Vascular Plants of the World (version May 2017).
7. In: Roskov Y., Abucay L., Orrell T., Nicolson D., Bailly N., Kirk P.M., Bourgoin T., DeWalt R.E., Decock W., De Wever A., Nieukerken E. van, Zarucchi J., Penev L., eds. (2017).
8. Species 2000 & ITIS Catalogue of Life, 29th May 2017.
9. Digital resource at www.catalogueoflife.org/col. Species 2000: Naturalis, Leiden, the Netherlands. ISSN 2405-8858.
10. Gupta PC, Sharma N, Rao ChV. Pharmacognostic studies of the leaves and stem of Careya arborea Roxb. Asian Pac J Trop Biomed. 2012;2(5):404-8.
11. Azajudin, Saraf Sailendra. Evaluation of physiochemical and phytochemical properties of safoff-E-Sana, a Unani polyherbal formulation. Pharmacognosy Res. 2010; 2(5):318-322
12. Liu WR, Qiao WL, Liu ZZ, et al. (2013). Gaultheria: Phytochemical and pharmacological characteristics. Molecules 18:12071–108
13. Okhale SE, Amanabo MO, Jegede IA, et al. (2010). Phytochemical and pharmacognostic investigation of antidiabetic Scoparia dulcis Linn Scrophulariaceae whole plant grown in Nigeria. Researcher 2:7–16
14. Singh A, Saharan VA, Bhandari A. (2013). Pharmacognostic standardization with various plant parts of Desmostachya bipinnata. Pharm Biol 52:298–307
15. Prajapati DN, and Purohit SS (2003). Agro's color atlas of medicinal plants. Agrobios India: New Delhi.
16. Dobelis NI, Ferguson, James D, and Rattray D. Magic and medicine of plants. Pleasantville: NewYork, 1986
17. Ayitey SE and Addae ML. Phytochemical nutritional and medical properties of some leafy vegetables consumed by Edo people of Nigeria. J.Pharmacol.Drug Res, 1977, 4:7- 8.
18. Maobe MAG, Gatebe E, Gitu L and Rotich H. Preliminary phytochemical screening of eight selected medicinal herbs used for the treatment of diabetes, malaria and pneumonia in Kisii region, southwest Kenya, European journal of applied sciences, 2013, 5(10) : 01-06.