

Full Length Research Paper

# Pharmacognostic study of *Tylophora dalzellii* Hook.f.

Shahla Najafi<sup>1\*</sup> and S. S. Deokule<sup>2</sup>

<sup>1</sup>Department of Biology, University of Zabol, Iran.

<sup>2</sup>Department of Botany, University of Pune, India.

Accepted 2 February, 2010

Plants are a great source of medicines, especially in traditional medicine, which are useful in the treatment of various diseases. *Tylophora dalzellii* a medicinal member belongs to the tribe Marsdenieae of the family Asclepiadaceae. Traditionally, *T. dalzellii* has been used in treatment of asthma, dermatitis and rheumatism. However, it has not yet been studied pharmacognostically. The study includes macroscopy and microscopy, anatomy, phytochemistry including fluorescence and ash analysis of the leaf and stem of *T. dalzellii*. Phytochemical screening of the plant revealed the presence of tannins, saponins and alkaloids. The detailed pharmacognostic account of *T. dalzellii* which includes macroscopic and microscopic characters will be helpful for the correct botanical identification of the drug. In addition, the values of percentage extractive and ash analysis, results of fluorescence analysis and phytochemical data will be helpful for the standardization and quality control of precious indigenous drug. The study scientifically validates the use of plant in traditional medicine.

**Key Words:** *Tylophora dalzellii*, pharmacognosy, Asclepiadaceae.

## INTRODUCTION

The use of herbal medicine for the treatment of diseases and infections is a safe and traditional therapy. Hence, medicinal plants have been receiving great attention worldwide by the researchers because of their safe utility. The curative properties of medicinal plants are mainly due to the presence of various complex chemical substances of different composition which occur as secondary metabolites (Karthikeyan et al., 2009, Lozoya et al., 1989). Medicinal plants form a large group of economically important plants that provide the basic raw materials for indigenous pharmaceuticals, (Aiyelaagbe et al., 2001; Augusti et al., 1996). *Tylophora dalzellii* Hook.f. is a twining perennial plant in India, apparently endemic in the western parts of the Indian Peninsula. It is found to be growing in scrub forest, at the edges of the forest, or on hedges and along the roadsides (Santapau and Irani, 1960). Traditionally, *T. dalzellii* has been used in treatment of asthma, dermatitis and rheumatism. It is reported in the literature that *T. dalzellii* has the same medicinal properties like that of *T. indica* in asthma. However, its chemical constituents are not known (Chopra et al., 1956).

## MATERIALS AND METHODS

Fresh plant material was collected from Western Ghat region of Maharashtra (India) in large quantities. Efforts were made to collect the plants when they started flowering and fruiting for the correct botanical identification. The plant material was brought to the laboratory and identified with the help of flora of Maharashtra State (Singh et al., 2001), Fascicals of flora of India (Jagtap and Singh, 1999) and Flora of British India (Hooker, 1872).

For microscopic studies, uniform and thin free hand section were taken from the fresh leaves and stems, dehydrated, double stained and finally mounted in Canada balsam by following the micro techniques method of Johansen (1940). Macro and microscopic characters were studied as per Wallis (1976) and Trease and Evanse (1982). For Phytochemical investigation, mature and healthy leaves were separated and dried in shade so as to prevent the decomposition of chemical constituents, powdered in blender and analysed qualitatively and quantitatively for different chemical parameters as per Trease and Evanse (1982). Detailed phytochemical studies were carried out by following Stahl (1969), Harborne (1973) and Manikam and Sadshivam (1991). Fluorescence analysis on powdered drug was carried out as per Chase and Pratt (1949) and ash analysis constants were studied as per Anonymous (1955).

## OBSERVATIONS

### Macroscopic characters

*T. dalzellii* is a frequent climber of scrub forest. Stems are much branched and pubescent. Latex is milky-white. The leaves are

\*Corresponding author. E-mail: najafi\_sh2003@yahoo.com.

simple, opposite, ovate or oblong. The young leaves are shortly acuminate at the apex and usually cordate at the base. The venation patterns of leaves are observed as camptodromous – brochidodromus.

#### Microscopic characters

Transverse section of leaf shows typical dorsiventral structure. Epiderm is a single layered with thick cuticle. Mesophyll is differentiated in two layers, armpalisade and palisade tissue. Midrib portion is bulged towards adaxial side of the leaf and epidermis is as in lamina, collenchyma cells on upper epidermis (2 - 3 rows) is less than lower epidermis (5 - 6 rows) and cortex is 6 - 7 rows of parenchyma cells. Central vascular strand is surrounded by paranchymatous cells. Xylem lays adaxially, phloem below abaxially. Midrib region consists of schizogenous cavities and few tannin cells. Stomata are rubiaceous type and present only on lower epidermis.

#### Quantitative microscopy of the leaf

Stomatal index.....	12.50 per mm <sup>2</sup> area
Stomatal number.....	9 per mm <sup>2</sup> area
Vein-islets number.....	16 per mm <sup>2</sup> area
Veinlet termination number.....	6 per mm <sup>2</sup> area
Palisade ratio .....	13.19 per cell

Transverse section of the stem is almost circular in outline. A thick cuticle covers the epidermis and uniseriate, multicellular hair is also present. Below the epidermis is the cortex, a distinct endodermis with casparian strips is absent. Pericycle is represented by scattered groups of stone cells in a complete ring. The stele is represented by closely arranged vascular bundles. Cambium is 3 to 4 layered. Medullary rays are uniseriate and narrow. Intraxylary phloem present at the periphery of the pith, in the form of separate strands. Different types of crystals of calcium oxalate and stone cells are present. Laticiferous tubes are also found in this region.

#### Vessels in stems

Vessels are broad and drum shaped. Pits are many small and arranged in definite patterns. Because of climbing habit, perforations are restricted to the end walls and the end walls are with narrow or broad pitted tails. Vessel length is increased in primary stem while it is decreased in the lateral branches. From the above observations, vessels follow general trends of specialization in dicots noted by Bailey (1944).

### RESULTS AND DISCUSSIONS

In the present investigation, the detailed pharmacognostic account of *T. dalzellii* is given which includes macroscopic and microscopic characters, which will be helpful for the correct botanical identification of the drug. Phytochemical tests were carried out of water extractives for starch, tannins, saponins, proteins, anthraquinones and reducing sugars and on alcoholic extract for alkaloids, glycosides and flavanoids. Results are tabulated in Table 1. In addition, carbohydrates, proteins and alkaloids were quantitatively estimated and their results were given in Table 2. Phytochemical screening portrays that

most of the natural products tested for were present in the plant material except glycosides, anthroquinones and flavonoides which were not detected in any of the tested fractions. Analysis of alkaloids, saponins and proteins in the leaves and stem extracts was positive and the stem extract showed positive results for tannins while the leaves extract showed negative results for tannins. Reducing sugar and starch are present only in the leaves extract and both absent in the stem extract (Table 1). Results of the quantitative estimation of proteins indicated that proteins are less in quantity in the leaves while more in the stems of studied plant. It is also observed that alkaloids are more in quantity in leaves than stem and also carbohydrates and starch are present in this plant but the percentage values are very less (Table 2). Mixture of such chemicals shows a spectrum of biological effects and pharmacological properties (Felix, 1982). Saponins are a special class of glycosides which have soapy characteristics (Fluck, 1973). It has also been shown that saponins are active antifungal agents (Sodipo et al., 1991). Tannins have been reported to prevent the development of microorganisms by precipitating microbial protein and making nutritional proteins unavailable for them (Sodipo et al., 1991). The growth of many fungi, yeasts, bacteria and viruses was inhibited by tannins (Chung et al., 1998). Presence of tannins suggests the ability of this plant to play a major role for the treatment of some disease (Asquith and Butler, 1986). Alkaloids were present in the ethanolic extracts. On this premise it will be advisable to extract the leaf and stem of *T. dalzellii* with ethanol in an attempt to exploit its detoxifying and antihypertensive properties since alkaloids is known to be effective for this purposes (Trease and Evans, 1982). Percentage extractive and ash analysis were carried out and results are tabulated in Table 3. Results of ash and acid insoluble ash of *T. dalzellii* indicated that percentage extractives are less in quantity in the leaves while more in quantity in the stem, so quality of the leaves of *T. dalzellii* is more than quality of the stem of *T. dalzellii* for their folkloric use as a medicine (Anonymous, 1955). Fluorescence analysis of powdered drug was also observed and the results are shown in Table 4. Results of fluorescence analysis of the leaves and stem of *T. dalzellii* showed Green color for leaf and yellowish green color for stem in powder as such, greenish black color for leaf and light green color for stem in powder as such in UV-light, dark green color for leaf and blackish green for stem in powder mounted in nitrocellulose, blackish green color for leaf and dark green for stem in powder mounted in 1 N NaOH in methanol, blackish green color for leaf and greenish black for stem in powder in 1 N NaOH in methanol and after drying for 30 min. mounted in nitrocellulose. This analysis suggests that, stem and leaves extract of *T. dalzellii* probably contain active agent(s) and this provides the basis for their folkloric use as a cure for some human ailments (Chase and Pratt, 1949). The values of percentage extractive and ash analysis, results

**Table 1.** Phytochemical tests of *Tylophora dalzellii*.

Sr. No.	Tests	Reagents used	Results of stem	Results of leaf
<b>Water extractives</b>				
1.	Starch	I2-KI	-ve	+ve
2.	Tannins	Acidic FeCl <sub>3</sub>	+ve	-ve
3.	Saponins	H <sub>2</sub> SO <sub>4</sub> + aceticunhydride	+ve	+ve
4.	Proteins	Million's test	+ve	+ve
5.	Anthraquinones	Benzene + 10% NH <sub>4</sub> OH	-ve	-ve
6.	Reducing sugars	Benedict's	-ve	+ve
<b>Alcoholic extractives</b>				
			Mayre's	+ve
1.	Alkaloids	Wagner's	+ve	+ve
			Dragnendorff's	+ve
2.	Flavonoides	HCl + Mg turnings	-ve	-ve
3.	Glycosides	Benzene+hot ethanol	-ve	-ve

**Table 2.** Quantitative estimation of *T. dalzellii*.

Name of the plant species	Plant part used	
	Stem% / g / dry weight	Leaf% / g / dry weight
Alkaloids	1.03	1.46
Proteins	1.95	0.98
Reducing sugar	00.00	0.036
Non-reducing sugar	0.022	00.00
Total carbohydrates	0.022	0.036
Total starch	00.00	0.076

The results are mean of three different readings.

**Table 3.** Results of ash and acid insoluble ash of *Tylophora dalzellii*.

Criteria	Leaf (%)	Stem (%)
% of ash analysis	7.64	8.37
% of acid insoluble ash	1.35	2.37

**Table 4.** Results of fluorescence analysis of *T. dalzellii*.

Treatments	Leaf	Stem
Powder as such	Green	Yellowish green
Powder as such in UV-light	Greenish black	Light green
Powder + Nitrocellulose	Dark green	Blackish green
Powder + 1 N NaOH in methanol	Blackish green	Dark green
Powder + 1 N NaOH in methanol dry for 30 min. + Nitrocellulose	Blackish green	Greenish black

of fluorescence analysis and phytochemical data will be helpful for the standardization and quality control of precious indigenous drug.

#### REFERENCES

- Aiyelaagbe O (2001). Antibacterial activity of *Jatropha multifida* roots. J. Fit. 72(5): 544-546.

- Anonymous (1955). Pharmacopoeia of India. Government of India Ministry of Health Manager Publications Delhi 1st Edn. : 370: 864.
- Asquith TN, Butler LG (1986). Interaction of condensed Tannins with selected Proteins. *Phytochemistry* 25(7): 1591-1593.
- Augusti KT (1996). Therapeutic values of onion and garlic. *Indian J. Exp. Biol.* 34(3): 634-640.
- Bailey IW (1944). The development of vessels in angiosperms and its significance in morphological research. *Am. J. Bot.* 31: 421 - 428.
- Chase CR, Pratt R (1949). Fluorescence of Powdered vegetable drugs with particular reference to development of system of identification, *Am. J. Pharm. Sci. Assoc. (Sci.ed.)* 38: 324-330.
- Chopra RN, Nayar SL, Chopra IC (1956). Glossary of Indian Medicinal Plants, CS I.R Pub. New Delhi India: 94.
- Chung KT, Wong TY, Wei CI, Huang YW, Lin Y (1998). Tannins and human health. *Crit. Rev. Food Sci. Nutr.* 38(6): 421-464.
- Felix MT (1982). *Medical Microbiology* Churchill Livingstone, London: 445-459.
- Fluck H (1973). *Medicinal plants and their uses.* W. Feulshom and comp. Ltd New York: 7-15.
- Harborne JH (1973). *Phytochemical Methods.* Chapman and Hill Tokyo Japan.
- Hooker JD (1872). *Flora of British India.* 4: 28-33.
- Jagtap AP, Singh NP (1999). Fascicles of Flora of India. The Director, Bot. India 24: 1-6, 82-91: 62-71.
- Johnsen DA (1940). *Plant Microtechnique.* Mc Graw Hill Book Co. Inc. New York. p. 154.
- Karthikeyan A, Shanthi V, Nagasathaya A (2009). Preliminary phytochemical and antibacterial screening of crude extract of the leaf of *Adhatoda vasica* L. *Int. J. Green Pharm.* 3: 78-80.
- Lozoya M, Lozoya X (1989). Pharmacological properties *in vitro* of various extracts of *Mimosa pudica* Linn. *Tepescohuite Arch Invest Mex.* pp. 87-93.
- Manikam AK, Sadashivam SM (1991). *Biochemical Methods in Agriculture Science.* Wiley Eastern Indian Ltd., India.
- Sodipo OA, Akanji MA, Kolawole FB, Odutuga AA (1991). Saponin is the active antifungal principle in *Garcinia Kola* heckles seed. *Biosci. Res. Commun.* 3: 171.
- Santapau SJ, Irani NA (1960). The Asclepiadaceae and Periplocaceae of Bombay St. Xavier College Bombay. pp. 12 -14, 43 - 50, 85 - 88.
- Singh NP, Lakshminarasimhan P, Karthikeyan S, Prasanna PV (2001). *Flora of Maharashtra State. Dicotyledones.* The Director, Botanical Survey of India. 334-342: 362-364.
- Stahl E (1969). *Thin layer Chromatography A Laboratory Handbook.* Spring Verlag Berlin, Heidenberg.
- Trease GE, Evans WC (1982). *Pharmacognosy.* Baillene Tindall, London. 735-738.
- Wallis TE (1976). *A textbook of pharmacognosy.* 3<sup>rd</sup> Edn. J and A Churchill Ltd, London.