

Pharmacognostic studies on *Premna corymbosa* (Burm. f.) Rottl. & Willd.**M. Chitra^{*1} and S. Prema²**¹Dept. of Biochemistry, Sengamala Thayaar Educational Trust Women's College, Mannargudi - 614 016, Tamil Nadu, India²Dept. of Siddha Medicine, Tamil University, Thanjavur, Tamil Nadu, India**Abstract**

Premna corymbosa is used in the Indian systems of medicine. The roots are used as a laxative, stomachic, cordial tonic and in the drugs to treat rheumatism and neuralgia. This paper describes macroscopic and microscopic features of its different parts for pharmacognostic applications.

Keywords: ayurvedic medicine, pharmacognosy, *Premna corymbosa*, microscopic features, root

INTRODUCTION

Traditional medicine is largely based on herbal knowledge and their use in developing countries like India is due to their affordability, accessibility and widely embedded beliefs. In developed countries, they are considered as complementary and alternative to modern medicine (CAM). The lack of quality standards in CAM is one of the important problems associated with the use of herbals. One of the reasons for this is the lack of standards and a wide variation in the composition of actual drug. Such a poor content variability leads to suspicions on their efficacies. Another area of concern is the problems of contamination of the raw materials used in the manufacture or even willful adulteration to enhance their therapeutic usefulness. As a result it is a matter of record that large percentage of herbal drugs are not of commercial successes.

Premna corymbosa (Munnai) is distributed in Peninsular, India and Srilanka. The plant is a scandent shrub and it emits mild odour when crushed. It grows upto 6 m height. The leaves are broadly elliptic-oblong 4-9 cm long and 2-4 cm broad; the lamina has margins serrate-gestate, apex acute; petiole upto 2 cm long (Yoga Narasimhan, 2000). The flowers of the plant are zygomorphic, fivemorous, aggregated into axillary's corymb. The calyx is gamosepalous, four lobed, 2 lipped and the corolla is cream to brownish, five lobed, 2 lipped, upperlip hooded. The fruits are drupe and the seeds are oblong (Ambasta, 1986). The roots form a constituent of the well known Ayurvedic medicine, *Dasamala* (Vaidya, 1975), which is a cure for obstinate fever. It was also prescribed as a laxative, stomachic and cordiac tonic. In traditional plant medicine, this tender plant is used for rheumatism and neuralgia (Anonymous, 1969). It is also regarded to have anti-inflammatory, anti-arthritis activities,

hypoglycemic action (Rathor *et al.*, 1977) and to cure liver disorders (Dhar *et al.*, 1968).

Its root contains lots of secondary metabolites of phytochemical importance. It may be adulterated with other roots during sales in the market. To formulate authentication of the root sample of this plant a pharmacognostic study was carried out and the results are presented in this paper. In this examination of root i.e., the physical characteristics, including occurrence, size, shape, color, surface markings, fracture, internal appearance, odor and taste were first taken into consideration. Following the study of physical characteristics, an inquiry into the microscopic structure of the root has been made (Youngken, 2003).

MATERIALS AND METHODS**Collection of specimens and embedding**

The plant specimens for the proposed study were collected from the hill regions of Palayamkottai, Thirunelveli, India (Fig. 1). The required samples were cut and removed from the plant and fixed in FAA (Formalin (5 ml) + Acetic acid (5 ml) + 70% ethylalcohol (90 ml)). After 24 hrs of fixing, the specimens were dehydrated with graded series of tertiary butyl alcohol. Infiltration of the specimens was carried out by gradual addition of paraffin wax (m.p. 58-60°C) until Tertiary Butyl Alcohol (TBA) solution attained supersaturation. 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 10-12 µm. Dewaxing of the sections was done by customary procedure. The sections were stained with Toluidine blue. Since toluidine blue is a polychromatic stain, the staining results were remarkably good and some cytochemical reactions could also be obtained. The dye rendered pink colour to the cellulose walls, blue to the lignified cells, dark green to violet for the mucilage. Necessary sections were also

*Corresponding Author
email: mschitra21@yahoo.com

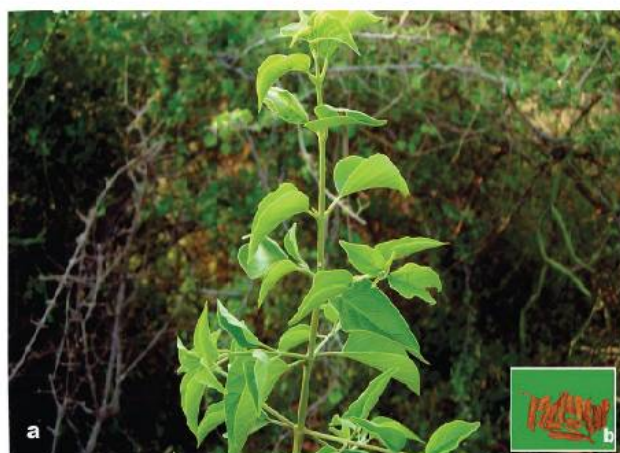


Figure 1. *Premna corymbosa*, a = whole plant, b = root

stained with safranin and fast-green and Potassium Iodide (for starch) (Johansen, 1940; Anonymous, 1985).

For the stomatal morphology, venation pattern and trichome distribution of paradermal sections was done and clearing of leaf with 5% sodium hydroxide or epidermal peeling by partial maceration by employing the Jeffrey's maceration fluid, were done. 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 by following the method of Esau (1979).

Photomicrographs

Microscopic descriptions are supplemented with photo-micrographs wherever necessary. Photographs of different magnifications were taken with Nikon Laphot and microscopic unit. For normal observations bright field microscope was used. For the study of crystals, starch grains and dignified 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 (Esau, 1965).

RESULTS AND DISCUSSION

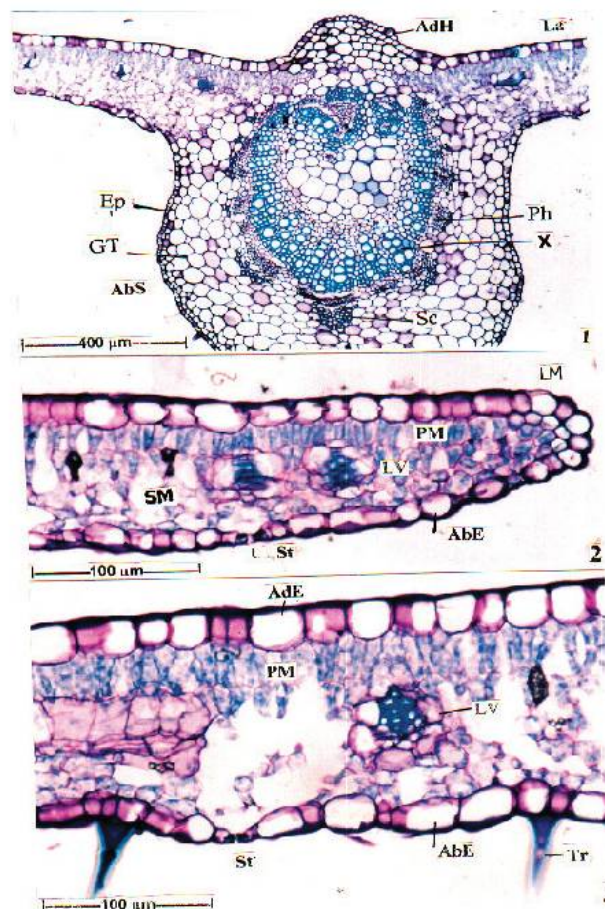
The leaf has prominent midrib, uniformly thin and even lamina. The midrib is thick and broad projecting to a hemispherical abaxial body and a short, broad adaxial hump (Figs. 2.1 to 2.3). The epidermis of the midrib is thin and distinct. The epiermdal cells are small and squarish. Remaining portion of the midrib consists of thin walled compact parenchyma.

The vascular system of the midrib comprises of a circular, closed, hollow cylinder. The cylinder encloses

a central, fairly wide parenchymatous ground tissue. Xylem elements include vessels and fibres which are arranged in compact radical parallel rows.

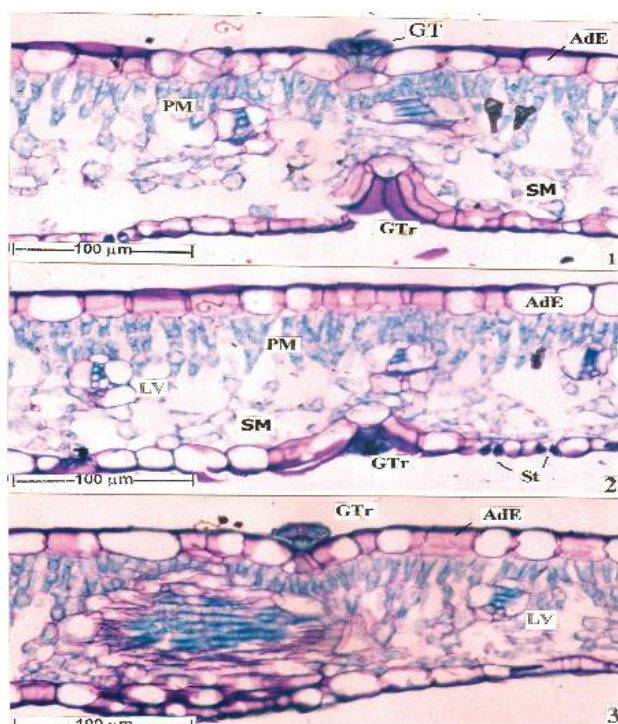
Phloem occurs as small discrete patches all around the cylinder. Small nests of fibres are seen associated with each phloem strands. The midrib is 800 μm in vertical plane and 700 μm in traverse plan.

The lamina is bilateral, mesomorphic and hypostomatic (Figs 3.1 to 3.3). The adaxial epidermis is 20 μm and apostomatic (Metcalf, and Chalk, 1950). The abaxial epidermis is stomatiferous (Figs 4.1 to 4.2).



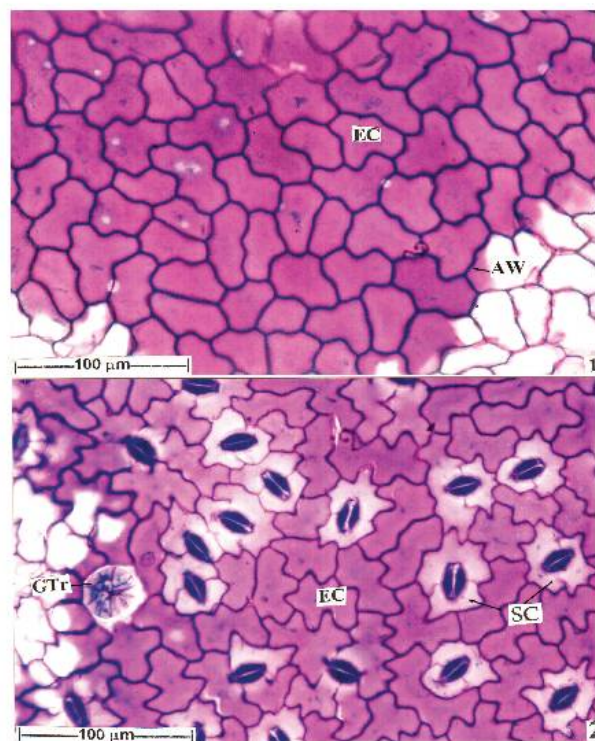
Figures 2.1 to 2.3. Cross Section of Petiole of *P. corymbosa*

Ep - Epidermis, AdH - Adaxial hypodermis, La - Lamina, Ph - Phloem, X - Xylem, GT - Ground Tissue, AbS - Abaxial Strand, Sc - Sclerenchyma, LM - Leaf Margin, PM - Parenchyma, SM - Spongy Mesophyll, St - Stomata, AbE - Abaxial Epidermis, AdE - Adaxial Epidermis, LV - Lateral Vein, Tr - Trichome



Figures 3.1 to 3.3. Microscopic features the leaf lamina of *P. corymbosa*

AdE - Adaxial Epidermis, GT - Ground Tissue, GTr - Glandular Trichome, PM - Parenchyma, LV - Lateral veins, SM - Spongy Mesophyll.



Figures 4.1 to 4.2. Views of the adaxial and abaxial epidermis of the leaves of *P. corymbosa*

AW - Anticlinal Walls, EC - Epidermal Cells, GTr - Glandular Trichome, SC - Subsidiary Cells.

Petiole

The distal part of the petiole has wide, shallow adaxial concavity and widely triangular abaxial part. The epidermal layer is thin and consists of thick walled small rectangular prominent cells (Fig 5).

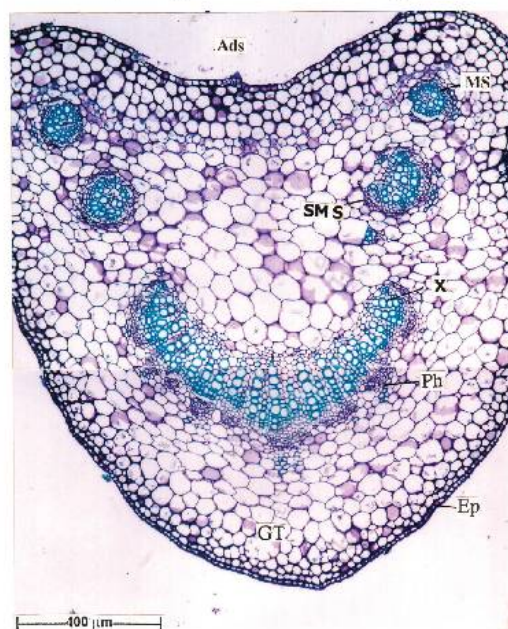


Figure 5. Cross Section of distal part of the petiole of *P. corymbosa*

AdS - Adaxial Strand, Ep - Epidemmis, GT - Ground Tissue, X - Xylem, Ph - Phloem, SMS - Submarginal Strands.

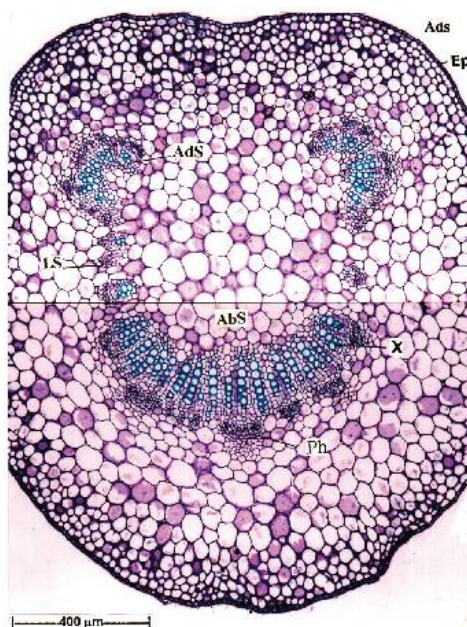
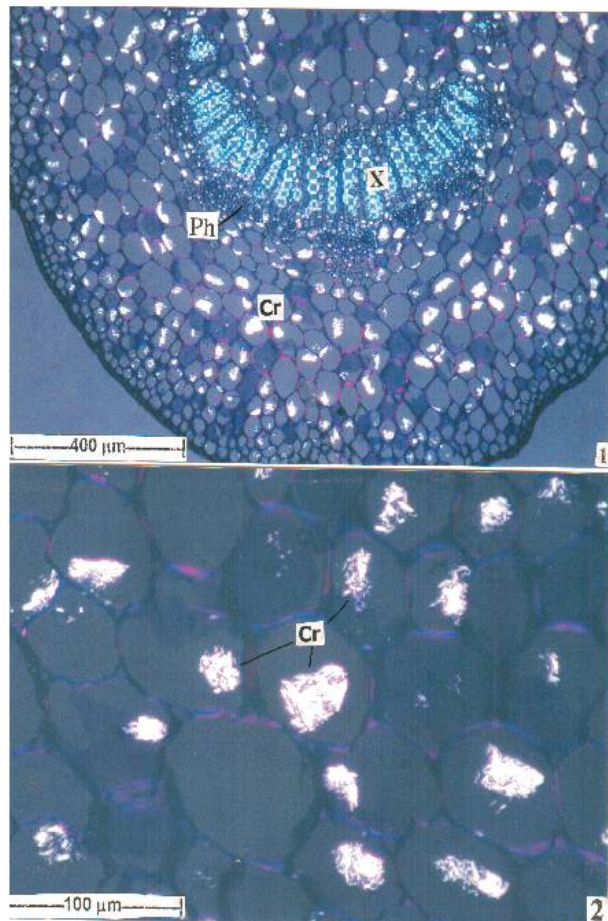


Figure 6. Cross Section of proximal part of the petiole of *P. corymbosa*

AdS - Adaxial Strand, AbS - Abaxial Strand, Ep - Epidemmis, LS - Lateral Strand, X - Xylem, Ph - Phloem.

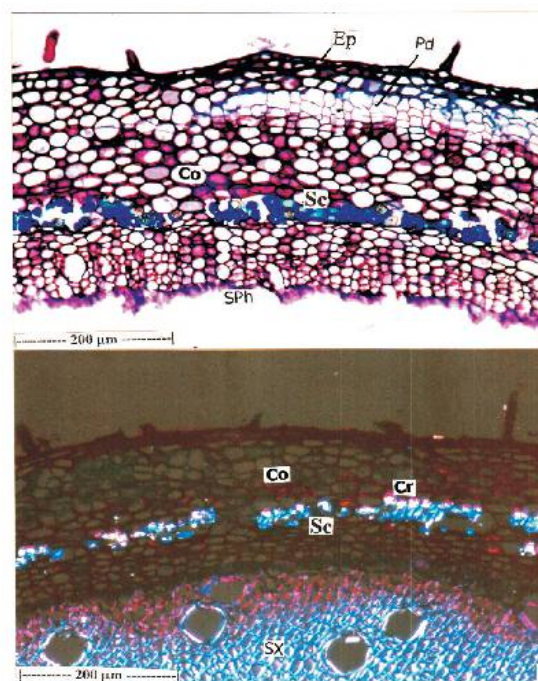
The proximal (or) basal part of the petiole is more or less circular in cross sectional outline (Fig. 6). Three or four layers of outer ground cells are collenchymatous and rest of the cells are large, circular, thin walled compact parenchyma cells (Metcalfe and Chalk, 1979). Calcium oxalate crystals were observed in the ground tissue of the petiole (Figs. 7.1 to 7.2)



Figures 7.1 to 7.2. Calcium oxalate crystals in the ground tissue of the petiole of *P. corymbosa*
Cr - Crystals, X - Xylem, Ph - Phloem.

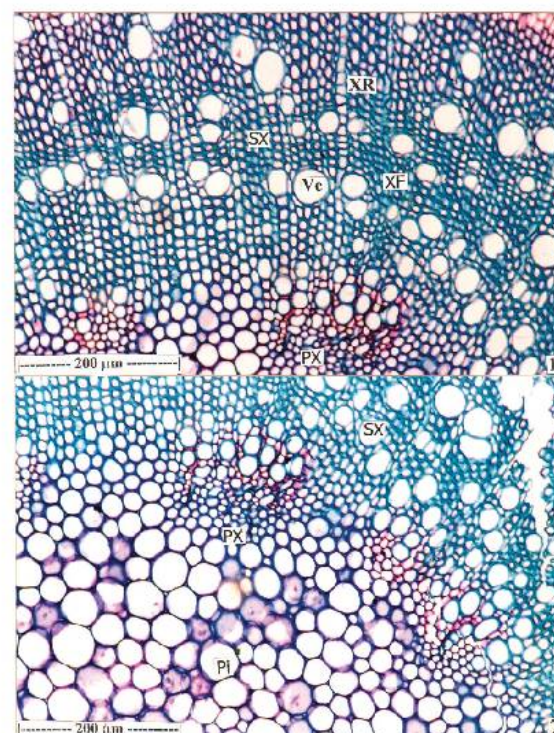
Stem

The stem in young stage has intact epidermis. The first periderm is formed in the cortex, below three or four layers and the periderm formation is not continuous (Figs. 8.1 to 8.2). The epidermal layer is thin and consisting of small tubular cells. The cortex is broad, homogenous and the secondary phloem is 100 µm wide. When the bark is viewed under polarized light microscope, small prismatic crystals are seen along the sclerenchyma (Figs. 9.1 to 9.2). Cylindrical secondary xylem is dense comprising of thick walled figures with wide circular vessels (Wallis, 1985).



Figures 8.1 to 8.2. Cross Sections of Stem of *P. corymbosa*

Ep - Epidermis, Pd - Phelloderm, Co - Collenchyma, Sc - Sclerenchyma, SPh - Secondary Phloem, Cr - Cambium ring, SX - Secondary Xylem.



Figures 9.1 to 9.2. Cross sections of the stem of *P. corymbosa*

Pi - Pith, PX - Primary Xylem, SX - Secondary Xylem, XF - Xylem Files, XR - Xylem Rays, Ve - Vessels.

Root

Thin lateral root measuring about 700 µm in diameter as well as thick root of about 4 mm in diameter were studied. The thin root has very wide, irregularly lobed, homocellular periderm, which has replaced epidermis and cortex. Secondary phloem is a thin, continuous and solid cylinder of uneven circumference with dense, thin walled fibres and has wide diffusely distributed solitary vessels.

Thick, old root has still wider, irregularly figured homocellulotic periderm which ranges from 100-350 µm wide. Secondary phloem is also wide and continuous measuring about 300 µm wide. It consists of regular radial files of rays which dilate towards periphery into wide funnel shaped structures. Phloem elements are in small radial rows around the xylem cylinder (Evans, 1996).

Secondary xylem has no growth rings. It consists of libriform fibers, vessels and fairly prominent, straight rays. The vessels are diffuse, circular and thin walled with 30-60 µm in diameter. The fibres are thick walled with wide lumen. They occur as regular radial fibres.

CONCLUSION

The above results could be used to set up commercial standards and prevent adulteration of this drug in markets.

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REFERENCES

- Ambasta, S.P. 1986. *The Useful Plants of India*. C.S.I.R. Publication, New Delhi. P. 705.
- Anaonymous, 1985. *Pharmacopoeia of India*. Controller of publications, Delhi.
- Anonymous, 1969. *Wealth of India*. Publication and information directorate, CSIR, New Delhi, P.240.
- Caius, J.F. 1988. *The Medicinal Poisonous Plants of India*. Scientific Publishers (India) Jodhpur, India. P. 493.
- Chopra, R.N., Nayar, S.L. and Chopra, I.C. 1956. *Glossary of Indian Medicinal Plants*. National Institute of Science Communication. C.S.I.R. Publication, New Delhi, India P. 330.
- Dhar, M.L., Dhar, M.M., Dhawan, B.W., Mehrotra, B.N. and Ray, S. 1968. Screening of India plants for biological activity part I. *Indian J. Exp. Biol.* 6: P. 232.
- Esau, K. 1965. *Plant Anatomy*. John Wiley and Sons, New York, P. 767.
- Esau, K. 1979. *Anatomy of Seed plants*. John Wiley and Sons, New York, P. 550.
- Evans, W.C. 1996. *Trease and Evans pharmacognosy*. WB Saunders Company Ltd, London. Philadelphia. Toronto, Sydney, Tokyo.
- Foster, A.S. 1934. The use of tannic acid and iron chloride for staining cell walls in meristematic tissue. *Stain technol.* 9: 91-92.
- Gamble, J.S. 1935. *Flora of the Presidency of Madras*. Vol. I, II, & III. Botanical Survey of India, Calcutta, India.
- Henry, A.N. Kumari, G.R. and Chitra, V. 1987. *Flora of Tamil Nadu*. India Vol. 3. Botanical Survey of India, Southern Circle. Coimbatore, India. P.258.
- Johansen, D.A. 1940. *Plant Microtechnique*. McGraw Hill Book Co. New York. pp.523.
- Mathew, K.M. 1983. *The Flora of Tamil Nadu Karnatic Vol. I. Polypetalae. Gamopetalae and Monochlamydae*. The Rapinat Herbarium. St. Johnson's College, Tiruchirappalli, India. Vol. 3, P. 688.
- Mayuranathan, P.U. 1929. *The flowering plants of Madras City and its immediate Neighbourhood*. Bull. Madras Govt. Mus; Madras, Chennai.
- Metcalfe, C.R. and Chalk, I. 1979. *Anatomy of the Dicotyledons*. Vols. I & II. Clarendon Press, Oxford, P. 276.
- Metcalfe, C.R. and Chalk, L. 1950. *Anatomy of the Dicotyledons*. Vols. I & II. Clarendon Press, Oxford.
- Nair, N.C. and Henry, A.N. 1983. *Flora of Tamil Nadu*. India Vol. I. Bot. Sur. India. Southern Circle, Coimbatore. India. P. 184.
- O'Brien, T.P. Feder, N. and McCull, M.E. 1964. Polychromatic staining of plant cell walls by toudine blue-O. *Protoplasma*. 59: 364-373.
- Rathor, R.S., Prakash, A. and Singh, P.P. 1977. *Premna integrifolia* Linn. A preliminary study of the anti-inflammatory and anti arthritic activity. *Rheumatism* 12: 130.
- Sass, J.E. 1940. *Elements of Botanical Microtechnique*. McGraw Hill Book Co., New York. P. 222.
- Solereder, H. 1899. *Systematic Anatomy of the Dicotyledons*. Transl. L.A. Boodle and F.E. Fritich. Oxford. Clarendon Press.
- Vaidya, B.G. 1975. Some controversial drugs in the Indian medicine. Vol. VIII *J. Res. Indian. Med.* 10: 80.
- Wallis, T.E. 1985. *Text book of Pharmacognosy*. CBS publishers and distributors, shahdara. Delhi, India.
- Willis, J.C. and Airy - Shaw, H.K. 1973. *A Dictionary of the Flowering Plants and Ferns*. Cambridge University Press. London. P. 1214.
- Yoganarasimhan, S.N. 2000. *Medicinal Plants of India*. Vol. II. Tamil Nadu. Regional Research Institute (Ay.) Bangalore, India. P. 715.
- Youngken, H.W. 2003. *Natural drugs Morphologic and Taxonomic consideration*. 2nd edn. Biotech books, Delhi. P. 16-18.