

## Pharmacognostic studies on Plaksa (*Ficus virens* Ait.) stem bark

Manoj Tripathi and R L S Sikarwar

Arogyadham, Deendayal Research Institute, Chitrakoot, District Satna-485 334, Madhya Pradesh, India

Received 22 April 2014; Accepted 7 November 2014

*Ficus virens* Ait. of family Moraceae, is an important plant employed in various indigenous systems of medicine against several diseases. The current communication provides a detailed account of the pharmacognostical investigation carried out on *Ficus virens* stem bark. The study includes macro and micromorphological characters including powder characteristics, physicochemical studies, HPTLC fingerprinting and preliminary phytochemical aspects. The results of the study could be useful for the identification and preparation of a monograph of the plant.

**Key words:** *Ficus virens*, Plaksa, Moraceae, Stem bark, Pharmacognosy, HPTLC fingerprinting.

**IPC code; Int. cl. (2014.01)**–A61K 36/00

### Introduction

*Ficus virens* Ait. syn. *F. lucescens* Alston, *F. infectoria* Roxb., *F. lacor* auct non Buch.-Ham., of family Moraceae, is commonly known as *Kahimal*, *Kaim*, *Keol*, *Pakar*, *Pakad* in Hindi and *Plaksa* in Sanskrit. It is a large spreading tree with occasional aerial roots spreading over large boulders, found nearly throughout the country and commonly planted as an avenue and ornamental tree<sup>1</sup>. It is a deciduous tree, 12-30 m high. Stem bark whitish grey, rough, irregularly in flakes. Leaves simple, alternate, 6-16 X 5-10 cm, acute-acuminate apex, shining surface, ovate-elliptic-oblong with entire, often undulate margins. Receptacles axillary, paired, sessile or sub sessile, globose white to cream coloured on ripening. Male flowers shortly pedicellate. Tepals 4. Stamen 1. Female flowers sessile. Gall flowers pedicellate<sup>2</sup> (Plate 1).

Traditionally, various parts of the plant are used in different ailments and diseases such as leaves are used in intrinsic haemorrhage, erysipelas and wound healing<sup>3</sup>. Stem bark is used in dysentery and menorrhagia and has antibacterial and antifungal activities<sup>4</sup>; applied in single and compound Ayurvedic formulations such as *Nyagrodhadi Kvatha Curma*, *Nalpamaradi Taila* and *Marma Gutika*. Review of literature reveals that no systematic pharmacognostical studies on the stem bark of *F. virens* Ait. has so far been carried out. Therefore,

the present work was planned to study the detailed macroscopical, microscopical, powder microscopical, physicochemical and chromatographic characteristics of the bark of this plant, which would serve as a standard reference for identification, authentication and for distinguishing the plant from its adulterants.

### Materials and Methods

#### Plant material

The fresh stem bark was collected from the Bagdara Ghati, Chitrakoot forest of Satna district in the month of October. Voucher specimens were collected and placed in the herbarium of Department of Pharmacognosy, Ayurveda Sadan Research Laboratory, Deendayal Research Institute Chitrakoot.



Plate 1–*Ficus virens* tree

\*Correspondent author:  
E-mail: trimanoj391@gmail.com  
Mob. +91- 09179147552

Fresh material was used for anatomical studies whereas shade dried material was powdered in electric grinder for physico-chemical, phytochemical and HPTLC studies.

#### Macroscopy and microscopy

Macroscopic or organoleptic characters like appearance, colour, odour and taste were evaluated. Bark section were cut by free hand sectioning and numerous sections were examined microscopically. Histochemical tests were carried out using hydrochloric acid-phloroglucinol to reveal lignified elements, Dragendroff's reagent for alkaloidal substance, ruthenium red for mucilage, iodine-iodide for starch<sup>5,6</sup>. Photomicrographs of the microscopical sections were captured with the help of Olympus trinocular research microscope CX- 21I with Digieye camera using Caliper plus version 4.2 software.

#### Powder characteristics

The dried bark was powdered till it completely passes through 355  $\mu\text{m}$  IS Sieve (old sieve number 44) and not less than 50% pass on through 180  $\mu\text{m}$  IS Sieve (old sieve number 85). About 2 g of powder was washed thoroughly with potable water, pour out the water without loss of material. Mounted a small portion in glycerine, warmed a few mg with chloral hydrate solution, washed and mounted in glycerine, treated a few mg with iodine solution and mounted in glycerine, about 1 g of powder, warmed over water bath with 50% conc. nitric acid till brown fumes appear, cooled and washed with water thoroughly and mounted a small portion in glycerin and seen under microscope at 40  $\times$ 10 $\times$  magnification of the trinocular research microscope<sup>7,8</sup>.

#### Physico-chemical parameters

Physico-chemical parameters such as moisture content (loss on drying at 105°C ), water soluble extractive value, alcohol soluble extractive value, total ash value, acid insoluble ash value and water soluble ash were calculated<sup>9</sup>.

#### Preliminary phytochemical studies

Preliminary tests were carried out on ethanolic and water extract for the presence/absence of phyto-constituents like alkaloids, flavonoids, tannins, resins, carbohydrates, proteins and saponins<sup>10, 11</sup>.

#### High Performance Thin Layer Chromatography (HPTLC)

For HPTLC, the powdered bark 5 g of sample was extracted with 100 mL of ethanol overnight, filtered and concentrated. It was applied by spotting extracted

sample on pre-coated silica-gel aluminium plate 60 F<sub>254</sub> (5  $\times$  10 cm with 0.2 mm layer thickness Merk Germany) using Camag Linomat -5 sample applicator and a 100  $\mu\text{L}$  Hamilton syringe. The samples, in the form of bands of length 6 mm, were spotted 15 mm from the bottom, 15 mm from left margin the plate and 10 mm part. Plates were developed using mobile phase consisting of *Toluene: Ethyl acetate* (7:3 v/v). Linear ascending development was carried out in 10  $\times$  10 cm twin through glass chamber equilibrated with mobile phase. The optimized chamber saturation time for mobile phase was 30 min. at room temperature. The length of chromatogram run was 8 cm. 20 mL of the mobile phase. Subsequent to the development, TLC plates were dried with the help of Hot Air Oven. The peak area for samples and standard were recorded with Camera photo documentation system Camag Reprostar 3. Visualization of spot was made before and after derivatization (with 5% methanolic-sulphuric acid reagent) at 254 nm, 366 nm and day light with Win cat software and R<sub>f</sub> values noted<sup>12</sup>.

## Results and Discussion

#### Macroscopy and Microscopy

The bark is creamish brown to buff in colour and rough externally, internally reddish brown or very dark brown coloured and smooth, fibrous, longitudinally striated. Occurring in flat to curved, quilled pieces, measuring 1 to 2 cm thick pieces and 4 to 8 cm long, fracture, fibrous, odour faintly aromatic, taste not characteristics (Plate 1 & 2).

The transverse section of the stem bark showed cells of the dark coloured rhytidoma, 6-10 rows of rectangular radially arranged suberized cork cells, a continuous band of 3 to 5 layered, stone cells



Plate 2—*Ficus virens* dry stem bark

embedded with few fibrous sclereids, the remaining cortical cells being parenchymatous, filled with tannins and prismatic crystals of calcium oxalate, outer cortical region cells get obliterated and tangentially elongated bands. Band of ceratenchyma running throughout the phloem tissue divides it into 2 distinct zones. Outer zone

is devoid of resin canals and inner zone with resin canals. Medullary rays are multiseriate, the outer phloem runs straight and almost parallel in the inner zone. Lignified phloem fibres, thin walled, rectangular, a few phloem parenchyma containing prismatic crystals of calcium oxalate (Plate 3).

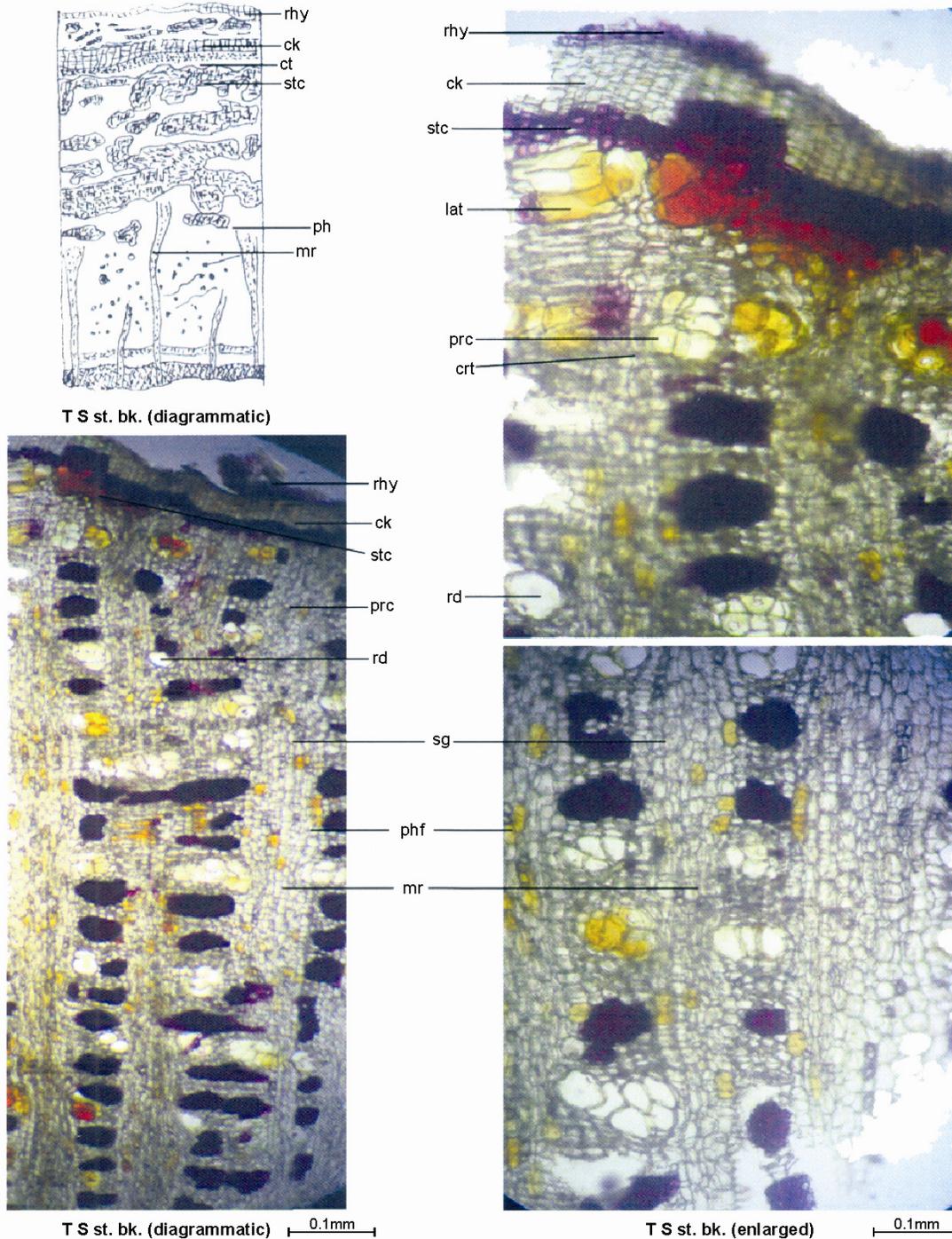


Plate 3—Microscopic characters of bark (Abbreviations: rhy, rhytidoma; ck, cork; ct, cortex; stc, stonecells; ph, phloem; mr, medullary rays; lat, laticiferous rubes; rd, resin duct; sg, starch grains; prc, prismatic crystals of calcium oxalate; phf, phloem fibres; crt, ceratenchyma)

**Diagnostic characters of powder**

The powder colour is pale pinkish brown, not characteristic odour and astringent taste. Under microscope the powder showed abundant prismatic crystals of calcium oxalate scattered as such and embedded in medullary rays, parenchyma and stone cells, starch grains simple, oval to circular in outline, scattered as such or filled in the parenchymatous cells, the fragments of laticiferous canal filled with light

brown granular contents often associated with stone cells, fragments of lignified cork cells in surface view, stone cells various shape and sizes, few containing prisms, elongated phloem fibres with wide lumen and pointed ends, tangential-longitudinally cut medullary rays embedded with prisms, associated with phloem parenchyma and sieve tissue, radially-longitudinally cut sclerosed medullary rays crossing the fibres (Plate 4).

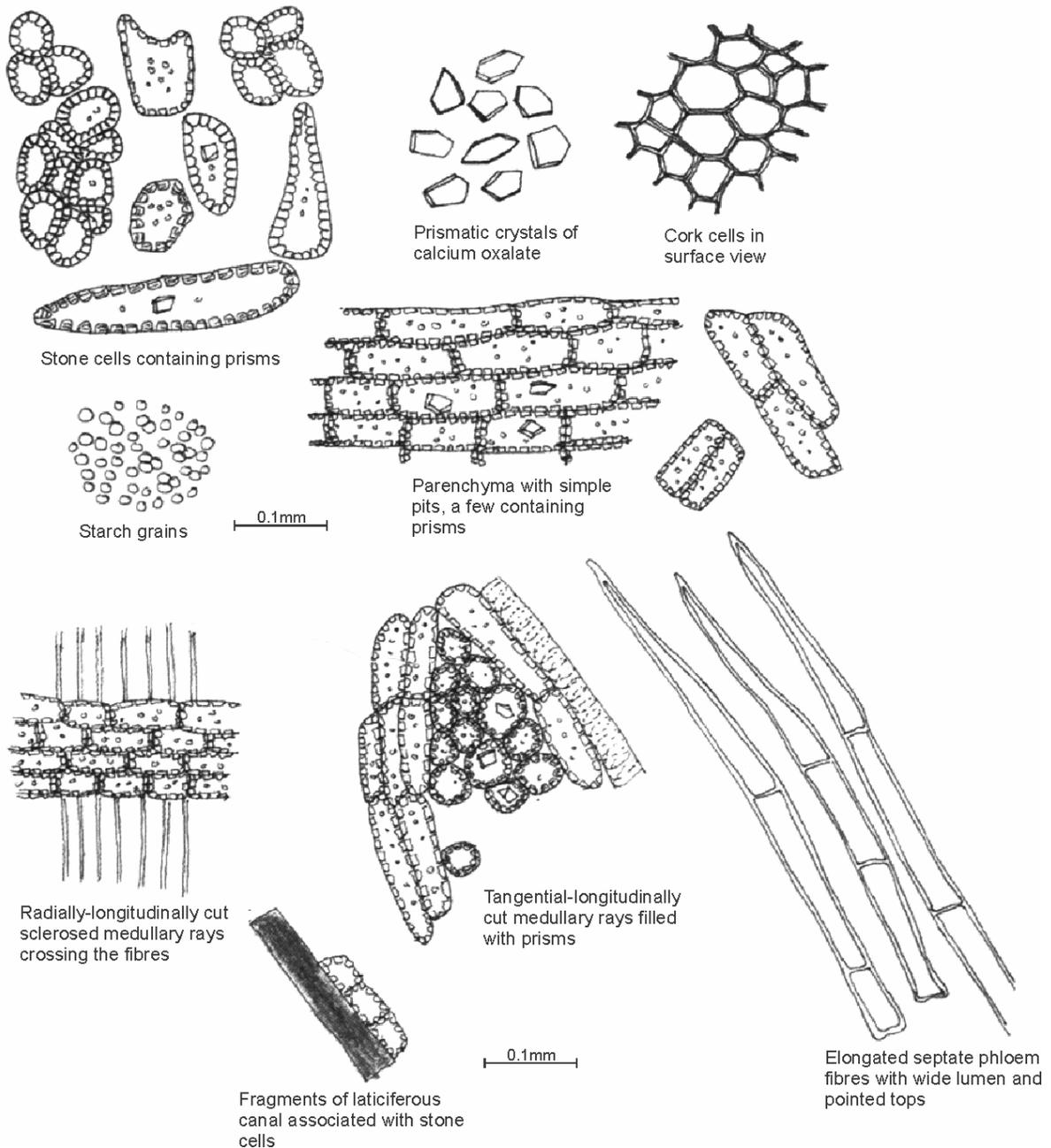


Plate 4—Powder microscopic characters of bark

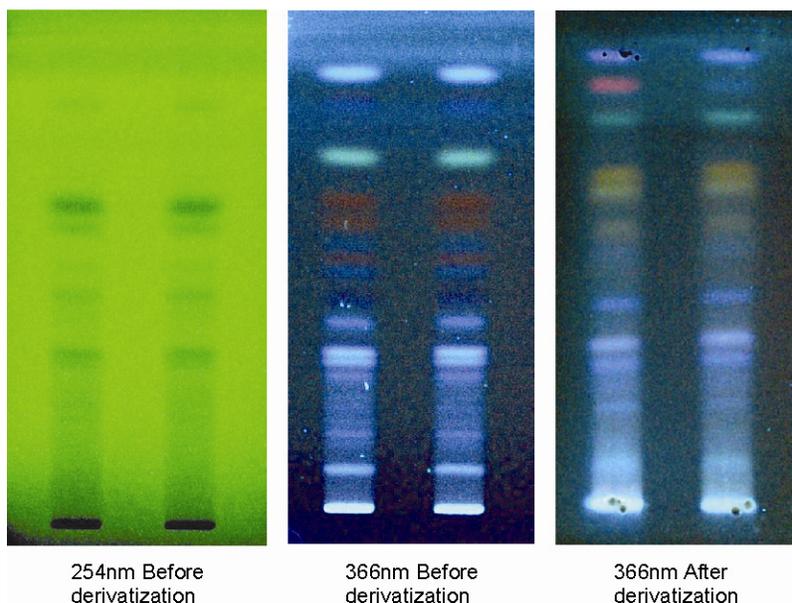


Plate 5-HPTLC Finger Print Profile

#### Physico-chemical analysis

The physico-chemical parameters such as extractive values are useful for the determination of exhausted or adulterated drug; ash values of the drug gave an idea of the earthy matter or the inorganic composition and other impurities present along with the drug. Physico-chemical results of the drug are loss on drying at 105°C is 6.8%, ethanol soluble extractive value 9.5 %, water soluble extractive value 12.6 %, total ash value 9.8 % and acid-insoluble ash value 1.4 %.

#### Preliminary phytochemical studies

Qualitative phyto-constituents were screened in the extracts taken in water, acetone, petroleum ether and ethyl alcohol. The screening exhibited presence of saponin, alkaloids, tannin, sterols and carbohydrate.

#### HPTLC finger print profile

The High Performance Thin layer Chromatography (HPTLC) plate was examined under 254 nm, 366 nm, after derivatization 366 nm. Chromatogram profiles are given in Plate 5. The  $R_f$  values and colours of the bands obtained were recorded. It shows major spots at 254 nm  $R_f$  0.36, 0.42, 0.62, 0.64 (all black), at 366 nm  $R_f$  0.11, 0.18, 0.36 (all sky blue), 0.50 (brownish red), 0.54 (sky blue), 0.62, 0.64 (both brownish red), 0.68 (flescent blue), 0.74 (deep brown), 0.80 (white) and after derivatization at 366 nm  $R_f$  0.10, 0.16 (both sky blue), 0.36 (flescent blue), 0.50 (sky blue), 0.60, 0.66 (yellow), 0.76 (sky blue), 0.80 (sky blue).

#### Conclusion

The macroscopic, microscopic and powder microscopic diagnostic features have been established to identify *Ficus virens* stem bark. The pharmacognostic and physico-chemical parameters can be used for checking the adulteration and purity of this drug. HPTLC finger print profile helps in identification of various phytochemical constituents present in the crude drug thereby authentication of crude drug. The TLC profile also helps to identify and isolate important phyto-constituents.

#### Acknowledgements

The authors are grateful to Dr. Bharat Pathak, General Secretary, Deendayal Research Institute, Chitrakoot, Satna (M.P.) for providing necessary facilities to complete this work.

#### References

- 1 Anonymous, The Wealth of India- A Dictionary of Indian Raw Material, Vol. IV, Council of Scientific and Industrial Research, PID, New Delhi, 1956, p. 36-37.
- 2 Mudgal V, Khanna KK and Hajra PK, Flora of Madhya Pradesh, Vol.2, Botanical Survey of India, Calcutta, 1997, p.625.
- 3 Anonymous, Quality Standards of Indian Medicinal Plants, Vol.3, Indian Council of Medical Research, New Delhi, 2005, 222-228.
- 4 Swami KD, Bisht NPS, Constituents of *Ficus religiosa* and *Ficus infectoria* and their biological activity, *J Indian Chem Soc* 1996, **73**, 631.
- 5 Meena AK, Sachan A, Kaur, Pal B and Singh B, *Moringa oleifera*: A Review, *J Pharm Res*, 2010, **3**(4), 840-842.
- 6 Sholapur Hasan Pasha N and Patil Basanagouda M, Pharmacognostical and phytochemical investigations on the

- bark of *Moringa oleifera* Lam., *Indian J Nat Prod Resour*, 2013, 4(1), 96-101.
- 7 Anonymous, The Ayurvedic Pharmacopoeia of India, Part-I, Vol. I, 1<sup>st</sup> Edn, The Controller of Publications Civil Lines, Delhi, 2001.
  - 8 Anonymous, The Ayurvedic Pharmacopoeia of India, Part-I, Vol. III, The Controller of Publications Civil Lines, Delhi, 2001.
  - 9 Mukherjee PK, Quality Control of Herbal Drugs, Business Horizons Pharmaceutical publishers, New Delhi, 2002, 356-357.
  - 10 Harborne JJ, Phytochemical methods: A Guide to Modern Techniques of Plant Analysis, 2<sup>nd</sup> Edn, Chapman and Hall, New York 1984, 85.
  - 11 Ansari MY, Wadud A, Ehteshamuddin and Bano H, Pharmacognostical evaluation of root of Gumma (*Leucas cephalotes* Spreng.), *Indian J Nat Prod Resour*, 2013, 4(1), 88-95.
  - 12 Anonymous, Protocol for Testing of Ayurvedic, Siddha & Unani Medicines, Pharmacopoeial Laboratory for Indian Medicines, Ghaziabad, 2007.