

PHARMACOGNOSTICAL STUDIES ON THE LEAVES OF *COMMIPHORA MUKUL* HOOK EX STOCKS

H.K. KAKRANI*, G.A. KALYANI*, G.P. BALAIDAVAR**, D. SATYANARAYANA+
& F.V. MANVI+

College of Pharmacy, J.N. Medical College, Belgaum – 590 010, India.

*** Department of Botany, R.L. Science Institute, Belgaum, India.*

+ N.G.S.M. College of Pharmacy, Nitte (D.K), Karnataka, India.

Received: 27 May, 1988

Accepted: 20 May, 1989

ABSTRACT: *This communication deals with the detailed pharmacognostical aspects of commiphora mukul leaves which include morphological and anatomical characters and preliminary phytochemical analysis of the leaves. The microscopical characters of leaf powder are also reported with its salient features. The fluorescent behaviour of powdered drugs with some chemical reagents is also examined.*

INTRODUCTION

Commiphora mukul (Syn.: *Balsamodendron mukul*) belonging to family – Burseraceae is the source of GUGGUL reputed for various medicinal properties^{15,17,2}. The survey of literature revealed that considerable work had done on various aspects of its volatile oil^{5,9,10a,14,4}, gum^{6,7,11,12} and resin¹⁸. The flowers^{10b} seed oil¹³ and leaves¹ are also investigated earlier. The work by Amjad Ali and Mashooda¹ was to find the amino acid composition only. So, it was thought worthwhile to carry out the detailed pharmacological investigations on the leaves of *C. mukul*.

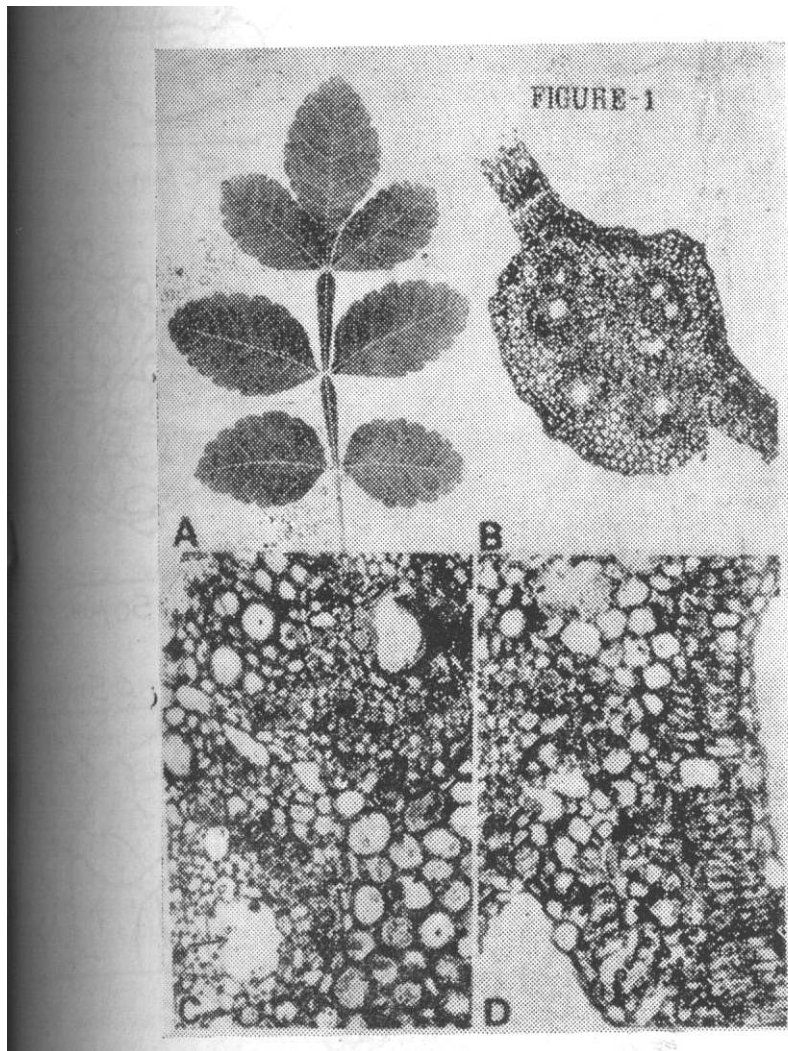
MATERIALS AND METHODS

The fresh material was collected from Bellary (Karnataka). The microtome sections (8-10 μ m) were stained with safranin and used for present studies. Phloroglucinol, iodine and ferric chloride

were used to test lignin, starch and tannin respectively. Physico-chemical studies were performed with the shade dried powdered material. The microscopic features of powder were observed after cleansing the same with chloral hydrate and staining with a mixture of phloroglucinol ; HCl 1:1. The extractive and ash values were determined as per I.P. 1966³.

Morphological Characters

The leaves (Fig. 1A) are pinnately compound (imparipinnate) ovate-rhomboid in shape with winged rachis. Terminal leaflets are 6.5 to 6.9 cm long and 3.0 to 3.5 cm broad. The lateral leaflets pass short stalk, they are 3.5 to 4.5 cm. in length and 2.5 to 3.0 cm in breadth. Their margin is serrate, apex acute; base symmetrical and lower surface dull green, almost glabrous. They possess characteristic odour and taste.



Microscopical Characters

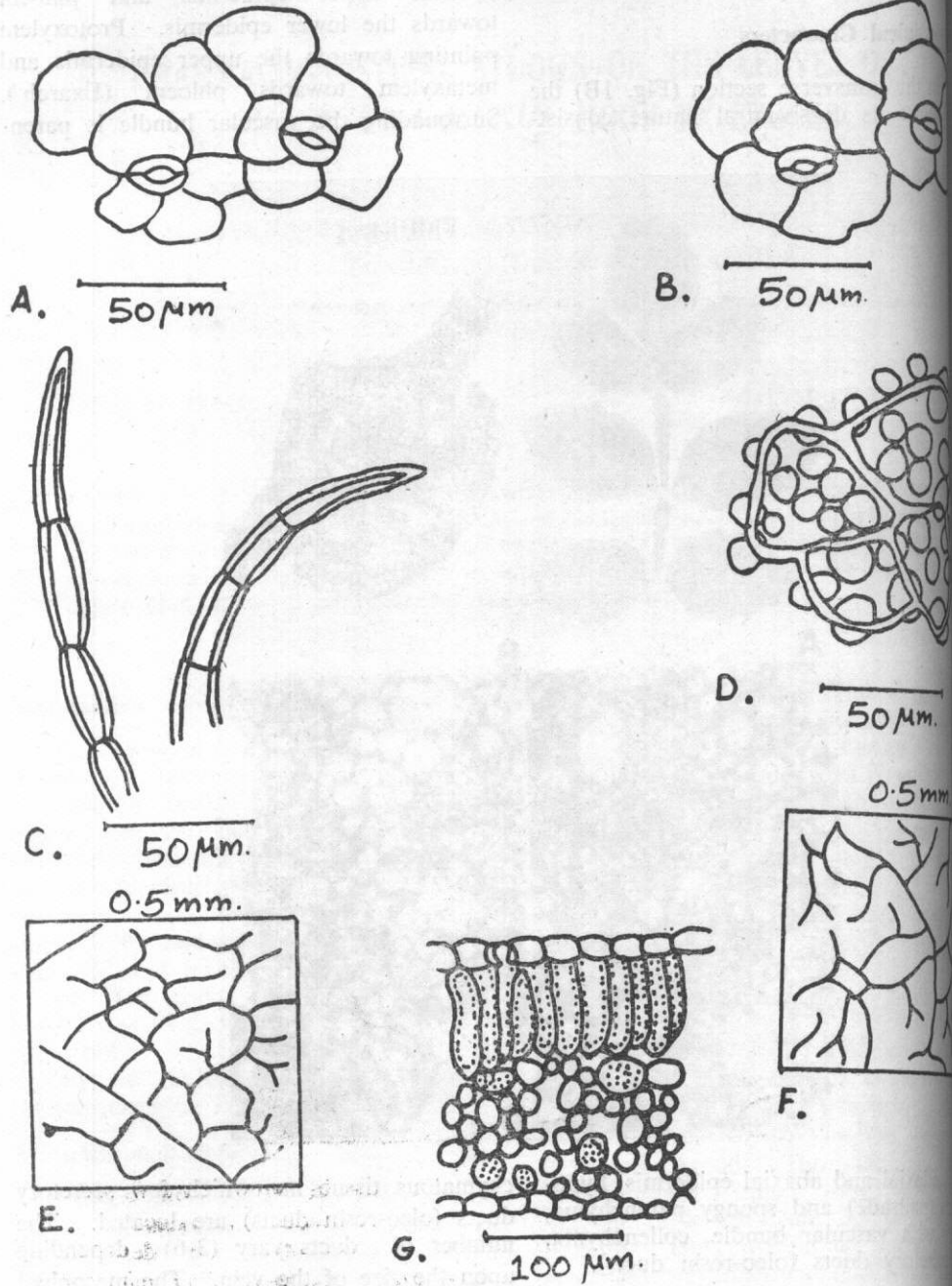
In the transverse section (Fig. 1B) the leaf shows its dorsiventral nature consisting of adaxial and abaxial epidermis, hypodermis (palisade) and spongy parenchyma, midrib with vascular bundle, collenchyma, and secretory ducts (oleo-resin ducts).

In the midrib between the adaxial and abaxial epidermis is a large vascular bundle.

It is conjoint and collateral with xylem towards upper epidermis and phloem

towards the lower epidermis. Protoxylem pointing towards the upper epidermis and metaxylem towards phloem (Exarch.). Surrounding the vascular bundle is parenchymatous tissue in which few secretory ducts (oleo-resin ducts) are located. The number of ducts vary (3-6) depending upon the size of the vein. The mesophyll of the midrib is composed of two thin zones of collenchyma immediately within the epidermis and ground mass of colourless parenchyma.

FIGURE-II



A) Anisocytic Stomata, B) Anomocytic Stomata, C) Covering Trichomes, D) Palisade Cells, E) Vein Islets, F) Vein Terminations & G) T. S. of Lamina (Diagrammatic).

The oleo-resin ducts are lined by small rectangular parenchymatous epithelial cells with dense protoplast (Fig. 1C).

On the either side of the midrib is the typical lamina (210 – 230 μm thick) (Fig 1D) covered by adaxial and abaxial epidermal cells. The both epidermal layers consist of barrel-shaped, elongated cells (12.5 – 15.4 – 18.4 μm). Adaxial stomata are anisocytic (Fig. 2A) and abaxial ones, mixture of anisocytic and anomocytic types (Fig. 2B). Stomata are almost oval on both the surfaces, guard cells are kidney shaped (length 25 – 26.2 – 28.2 μm , breadth 6.6 – 7.6 – 8.1 μm), water pore is present. Stomatal index is 8.9 (Upper surface) and 11.1 (Lower surface). Epidermis bears typical covering trichomes, which are 3-5 cells long, curved, 267.5 – 625.0 μm in length (Fig. 2C). They are uniseriate, thick-walled and some of them contain yellowish brown matter.

Between the two epidermal layers, in lamina region, the mesophyll tissue is differentiated into:

- i) Upper palisade tissue (Hypodermis) and ii) Lower spongy tissue.
- ii) The hypodermis consists of single layer of columnar palisade, compactly arranged with elongated cells filled with chloroplasts, 65 – 74.7 – 84.5 μm long and 12.5 – 18.3 – 24.2 μm broad. The palisade ratio was found to be 2.75 to 3.35 (Fig. 2D).
- iii) The lower portion of spongy parenchyma (3 – 4 layers) is composed of loosely arranged cells (15 – 30 μm in anticlinal direction and 120.0 – 144.0 μm in periclinal

direction) with large or small intercellular spaces. These cells contain few chloroplast.

The central parts of the periole in T.S. reveals almost the same structures as the midrib.

The vein-islet number is 8.5 – 11.5 (Fig. 2E), veins branched with spirally thickened trachoids. Veins and vein termination are sheathed by a layer of long parenchymatous cells. Vein termination number was 16.8 to 19.9 (Fig. 2F).

Leaf Powder Analysis

The leaf powder (40 mesh) cleared by boiling with chloral hydrate and treating with mixture of phloroglucinol: HCl (1:1) revealed following structures on microscopic examination.

Fibres spares, xylem with spiral thickening, members of vessels, xylem parenchyma cells. Starch grain (simple & compound) were detected in mesophyll. Lamina fragments, stomata, covering trichomes, epidermal cells, few oleo-resin ducts (entire as well as fragments). Vascular region was stained pink.

The behaviour of powdered leaves with different reagents was also studied and the same is depicted in Table 1.

The fluorescence analysis of powder was also carried out following the method of Chase and Pratt (1949)⁸ and Kokosi et. al. (1958)¹⁶. The observations are recorded in Table.2.

Phytochemical Studies

The ash and extractive values² were determined using air dried materials. The results are depicted in Table 3.

About 50 g of air dried powdered leaves were extracted separately in soxhlet apparatus with hexane, chloroform, benzene, ethyl acetate, alcohol and water successively. These extracts were screened

for presence or absence of steroids and triterpenoids (L.B. Test; Peach & Tracy, 1955)¹⁹, flavonoids (Shinoda's Test, Loc. Cit), alkaloids (Mayer' reagent Loc. Cit), tannins (Ferric chloride test Loc. Cit) and proteins (Million's reagent, Youngken 1951)²⁰. The results are depicted in Table 4.

The detailed chemical composition will constitute separate communication, so not reported here.

TABLE 1

Behaviour of powder with different Chemical reagents

S. No.	Treatment	Observation
1	Powder + 1 N NaOH	Yellowish brown
2	Powder + Saturated picric acid	Yellowish green
3	Powder + Acetic acid	Orange yellow
4	Powder + Conc. Hcl.	Reddish brown
5	Powder + Conc. HNO ₃	Chocolate brown
6	Powder + Iodine (5%)	Blackish brown
7	Powder + Sakuwabiff's Reagent	Yellowish brown
8	Powder + Ferric chloride (5%)	Dark Blackish brown
9	Powder + 40% NaOH + Few drops of 10% Lead acetate	Blackish brown
10	Powder + Sudan III (ALCOHOLIC)	Dark reddish orange
11	Powder + Conc. HNO ₃ + Ammonia	Yellowish orange
12	Powder + 35% HCl	Brownish
13	Powder + 5% KOH	Yellowish green
14	Powder + Phloroglucinol : HCl (1:1)	Yellowish brown with purple spots.

TABLE 2**Flourescence under U.V. Light***

S. No.	Treatment	Observation
1	Powder as such	Purplish brown.
2	Powder + Nitrocellulose in Amyl acetate	Olive green
3	Powder + 1N Hcl	Blackish brown
4	Powder + 1N Hcl + Nitrocellulose in Amyl acetate	Dark brown
5	Powder + 1N Aq. NaOH	Dark brown
6	Powder + 1N Aq. NaOH + Nitrocellulose in Amyl acetate	Blackish brown
7	Powder + Methanolic 1N NaOH	Brown
8	Powder + Methanolic 1N NaOH + Nitrocellulose in Amyl acetate	Greenish brown.
9	Powder + 50% HNO ₃	Reddish brown
10	Powder + 50% H ₂ SO ₄	Greenish brown with purple tinge

* The powder itself possessed yellowish – green colour.

TABLE 3**Ash & Extractive values of C. Mukul Leaves**

Type	Value
Total Ash	.. 14.372
Acid insoluble ash	.. 04.328
Sulphated ash	.. 03.212
Water soluble extractive	.. 20.419
Alcohol soluble extractive	.. 26.118

Results and Discussion

The various standards for the identification of the *Commiphora mukul* (*Balsamodendrom mukul*) family :

Bursearceae leaves are established by morphological, microscopical and

phytochemical evaluation techniques. The leaf powder was also characterized.

The microscopical examination of the leaf T.S. showed characteristic vascular bundle, oleo-resin ducts, uniseriate and multicellular covering trichomes. The mesophyll is devoid of any crystals. The mesophyll is devoid of any crystals. The anisocytic and anomocytic stomata were detected with typical kidney – shaped guard cells.

The quantitative microscopy provided following data: a) Stomatal index 8.9 – 11.1, b) Palisade ratio 2.75 to 3.55, c) Vein islet number 8.5 to 11.5 and d) Vein termination number 16.8 to 19.9.

The various physico-chemical constants established for the *C.mukul* leaves are a) Total ash – 14.372, b) Acid insoluble ash – 4.328, c) Sulphated ash – 3.212, d) Water soluble extractive – 20.419 and e) Alcohol soluble extractive – 26.118.

The fluorescence analysis behaviour of leaf powder with some chemical reagents is also reported with its microscopic characters.

The various extracts of the leaves revealed the presence of alkaloids, flavonoids, resins, saponins, phytosterols, terpenoids, tannins, proteins and reducing sugars.

TABLE 4

Preliminary Phytochemical Observations of *C. Mukul* Leaves

Parameters	Hexane Extract	Benzene Extract	Chloroform Extract	Ethyl acetate Extract	Alcoholic Extract	Aqueous Extract
Alkaloids	-	-	+	-	+	-
Flavonoids	-	-	-	+	+	-
Reducing Sugars	-	-	-	-	-	+
Resins	+	+	+	-	+	-
Saponins	-	-	-	-	+	-
Steroids	-	-	-	-	-	-
Terpenoids	+	+	-	-	-	-
Tannins	-	-	-	-	+	+
Proteins	-	-	-	+	-	-

REFERENCES

1. Amjad Ali and Mashooda Hasan., *Pakistan J. Ind. Res.* 10 (1), 21 – 23 (1967).
2. Anonymous, *Wealth of India (raw materials)*, Vol. II. CSIR, New Delhi, pp 313 (1950).
3. Anonymous, *India Pharmacopoeia*, 2nd Ed., Delhi, Govt. of India Publications (1966).

4. Bagi M.K., Kakrani H.K., Kalyani G.A., Satyanarayana D. and Manvi F.V. *Fitoterapia*, Vol. LVI (4), 245 (1985).
5. Bhati A., *J. Indian Chem. Soc.*, 27 (9) 437 (1950).
6. Bose S. and Gupta K.C., *Indian J. Chem.* 2(2), 57 (1966).
7. Bose S. and Gupta K.C., *Indian J. Chem.*, 4(2), 87 – 89 (1966).
8. Chase, C.R. and Pratt R.J., *J. Am. Pharm. Assoc.*, 38, 24 – 31 (1949).
9. Dennis T.J., Yadav B.B.L., Joseph T.G. and Mishra K.P., *Bull Medico Ethno. Bot. Res.*, 1(1), 72, (1981).
10. a) Kakrani H.K., *Bull Medico Ethno Bot. Res.*, 2 (1), 100 (1981).
b) Kakrani H.K., *Fitoterapia* No.5, 221 (1981).
11. Kakrani H.K and Jain N.K., *Indian J. Hosp. Pharm.* XVIII(3) 100 (1981).
12. Kakrani H.K. and Varma K.C., *Indian J. Hosp. Pharm.* XVIII (4), 134 (1981).
13. Kakrani H.K., *Indian Drugs*, 19(9), 339 (1982).
14. Kakrani H.K. and Kalyani, G.A., *Fitoterapia*, Vol. LV, No. 4, 232 – 234 (1984).
15. Kirtikar K.R. and Basu B.D., *Indian Medicinal Plants*, L.M. Basu and Co., Allhabad, pp. 525 – 29 (1933).
16. Kokosi J., Kokosi R. and Slama F.J., *J. Am. Pharm. Assoc.* 47, 715, (1958).
17. Nadkarni, A.K., *The Indian Materia Medica*, Vol. I., Popular Book Depot, Bombay, pp. 167 – 170 (1954).
18. Patil V.D., Nayak U.R., and Sukhadev, *Tetrahedron*, 28, 2341 (1972).
19. Peach K. and Tracy, M.V., *Modern Methods of Plant Analysis* (Heidelberg-Springer Verlag) 3rd and 4th Vol, (1955).
20. Youngken H.W., *Pharmaceutical Botany*, 7th Ed., Blackish Company, Toronto, (1951).