



Research Article

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Pharmacognostic and phytochemical analysis of *Agnimantha* (*Premna corymbosa* Rottl.) root

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ABSTRACT

Agnimantha (*Premna corymbosa* Rottl. Verbinaceae), also known as Arani and Nadeyi in Sanskrit is very important plant in use since Vedic period. It is known as Wind killer/ Indian headache tree in English, Munja in Malayalam and Taggiberu in Kannada. It is a large perennial shrub which grows up to 9 to 10 m in height and is found occurring throughout India in the plains. It is one among the combination drug *Bhratpanchamoola* (5 root drugs) of *Dasamoola* (10 root drugs). Its roots are used against asthma, and bronchitis; as an expectorant, in cold, in catarrh and fever. On these grounds, this work was an attempt to establish pharmacognostic and preliminary phytochemical standards of roots including HPTLC. Pharmacognostical parameters for the root of *P. corymbosa* using parameters like macromorphology, microscopy, physio-chemical constants and phytochemical screening were done using standard methodology. Except loss on drying, other physico-chemical results were under the limits of standards given in API. It showed presence of alkaloids, steroid, tannins, phenols and flavanoids. HPTLC densitometric scan showed six chemical components at 254 nm, five at 366 nm and fourteen at 620 nm. In the present pharmacognostic analysis, chemical constituents, TLC and HPTLC of the roots of *Agnimantha* co-relates with API standards and justifying its identity and authenticity.

Keywords: *Agnimantha*, *Bhratpanchamoola*, *Dasamool*, *Premna corymbosa* Rottl., Standardisation.

INTRODUCTION

Agnimantha, *Premna corymbosa* Rottl. (Verbinaceae) is very important since Vedic period.^[1] *Agnimantha*, is called as *Agnimathanaha*,^[2-4] *Arani*,^[5-7] and *Nadeyi*^[4-9] in Sanskrit. It is called as Munja in Malayalam and Taggiberu in Kannada.^[7,10] Its usefulness or its importance can be seen from the name itself i.e., it is a tree, the twigs of which were used to light fire during the sacrificial ceremonies and other rituals by rubbing the sticks together.^[1] It is a drug with better *Sothahara* (anti-inflammatory) property and also *Kapha Vatahara*.^[11] The importance of *Agnimantha* not only lies in these features but also that, it was such a drug which is included in the *Dasamoola* and also in *Bhratpanchamoola*,^[12] and is a best Tridosahara^[11, 12-14] drug. Literary review reveals that it is an effective medicine for the diseases of digestive system like *Aruchi* (tastelessness)^[15] *Agnimandya* (loss of appetite)^[11,13,15-17]. It is a good anti-inflammatory^[11,15,18-20] drug and it is used in swellings,^[18] and scrotal enlargement^[19] and cough^[12-14,21,22]. In Ayurveda, more than the usage of *Agnimantha* leaves,^[2,9,23] roots are mentioned in asthma^[2], bronchitis^[3,5,10] as an expectorant,^[9,2,23] in cold,^[23] catarrh and fever^[10]. Hydro-alcoholic (ethanol 50% v/v) of roots are reported to have dose dependant anti-hyperglycaemic.^[24] *In-vivo* antioxidant activity of its roots ethanolic extract against streptozotocin induced oxidative stress in different organs (liver, kidney, brain, heart and pancreas) showed significant decrease in lipid peroxidation, suggesting its role in protection against lipid peroxidation induced membrane damage during diabetes.^[25] Chemical constituents present are aphelandrine, premnine, botulin, ganiarine, ganikarine, caryophellen, premnenol, premnaspirodien, sterols. Ganiarine is an alkaloid which acts as an antibiotic which is reported to be active against gram positive organisms^[23], the other chief alkaloids among it are premine, and ganikarine. The object of present study is to evaluate various pharmacognostical parameters such as macroscopic, microscopy, physicochemical, phytochemical and chromatographic studies of the roots.

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MATERIALS AND METHODS

The root of *P. corymbosa* was collected from the Garden of *Shri Dharmasthala Manjunatheshwara College of Ayurveda and Hospital, Hassan*. Authentication was done at the Department of Dravyaguna, *Shri Dharmasthala Manjunatheshwara College of Ayurveda and Hospital, Hassan*. Authenticated samples were subjected for pharmacognostic, physico-chemical, phytochemical analysis and HPTLC profiling at Department of pharmacognosy, SDM Centre for Research in Ayurveda and Allied Sciences, Udupi 574118.

Pharmacognostic Study: Macroscopic / organoleptic evaluation of root was done for evaluation of external morphology, its shape and size, colour and external markings, fracture, internal colour, odour and taste of the drug.^[10] A pinch of powder was warmed with drops of chloral hydrate on a microscopic slide and mounted in glycerine. Slides observed under microscope and diagnostic characters were observed and photographed using Zeiss AXIO trinocular microscope attached with Zeiss Axiocamera under bright field light. Magnifications of the figures are indicated by the scale-bars.^[27-30]

Physico-Chemical Constants: Physico-chemical constants of roots were determined to elicit loss on drying, total ash, acid Insoluble ash, water soluble ash, alcohol soluble extractive value and water soluble extractive value.^[10]

Phytochemical Screening : Phytochemical analysis of roots were determined to see the presence of alkaloids, steroids, carbohydrate, tannin, flavanoids, saponins, triterpenoids, coumarins, phenols, carboxylic acid, amino acid, resins and quinone.^[26]

HPTLC Profile: One gram of powdered samples were dissolved in 10 ml of ethanol and kept for cold percolation for 24h and filtered. 3, 6 and 9 µl of the root samples of the drug *P. corymbosa* Rottl. were applied on a pre-coated silica gel F254 on aluminium plates to a band width of 7 mm using CAMAG (Muttenez, Switzerland) Linomat 5 TLC applicator. The plate was developed in the solvent system of toluene ethylacetate and formic acid (7.0: 3.0: 0.3) in CAMAG twin trough chamber. The developed plates were visualized in under short and long UV and then derivatised with vanillin sulphuric acid reagent, prior to derivatisation in CAMAG Photodocumentation unit. The plate was scanned under UV 254 and 366 nm using CAMAG Scanner 4. R_f, colour of the spots and densitometric scan were recorded.^[27]

RESULT AND DISCUSSION

The study on the pharmacognostic features of medicinal plants is a process to know its quality, purity and also to check the presence of adulterants and substitutes.

Macroscopical/ Organoleptic: Macroscopically the roots were cylindrical and occasionally branched; its surface was easily peeling off, yellowish brown in colour, with aromatic odour and slightly astringent taste, fracture is hard fractured surface rough.

Powder microscopy: Root powder was rough, whitish yellow, slightly aromatic and astringent in taste. In microscopic view, the powder showed the presence of cork and pitted vessel fragment, cortical parenchyma with starch, fragment of pitted vessels, transversely cut cork, bundle of fibres and parenchyma, thick walled parenchyma with content, radially cut medullary rays, vessel fragment, pitted fibres, pitted sclereids, and cell with contents (Figure 1).

Physico- chemical evaluation: Determination of loss on drying is the method to find the moisture content of a drug. It aids to prevent the decomposition of the drugs either due to chemical change or microbial contamination. From the results obtained for loss on drying, *Agnimantha* root is having 16.58% of moisture content.

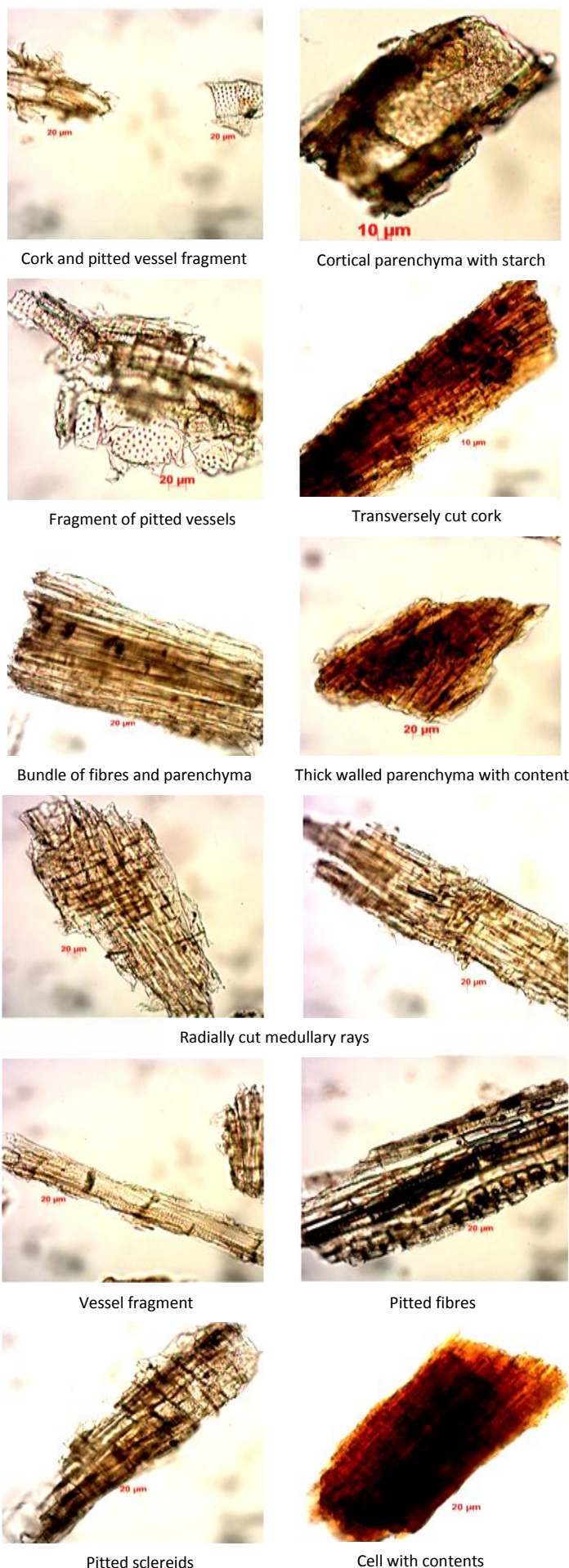


Figure 1: Powder microscopy of *Agnimantha* root

Determination of ash values is the criterion to judge authenticity and purity of crude drugs. The residue remaining after incineration is the ash content of the drug. These could be inorganic salts such as carbonates, sulphates, phosphates, silicates etc. naturally occurring in the drug or adhered to it or deliberately added to it in order to adulterate the drug. Since the drugs were collected personally from their natural habitat there was no scope of any adulteration. Total ash is to measure the total amount of plant material remaining after ignition of the drug. Acid insoluble ash or water soluble ash content is the residue obtained after boiling the total ash either with dilute hydrochloric acid or water which measures the amount of sand and silica matter present in the drug. From the above results the ash value for roots of *Agnimantha* showed total ash content of 1.79% which is within the standard limits, acid insoluble ash was 0.0%.while water soluble ash content was found to be 0.89 %.

Determination of extractive value measures the nature of the chemical constituents present in a crude drug. It is essential for the estimation of specific chemical constituents soluble in that particular solvent used for extraction. The results of ethanol soluble and water soluble extractive values of *P. corymbosa* Rottl. roots showed 12.23 % and 5.44 % respectively (Table 1).

Table 1: Showing results of physical analysis of roots

Parameter	<i>Agnimantha</i> root	API standards
Loss on drying	16.58	not more than 2%
Total Ash	1.79	not more than 6%
Acid Insoluble Ash	0.0	not more than 1%
Water soluble Ash	0.89	-
Alcohol soluble extractive value	12.23	not less than 2%
Water soluble extractive value	5.44	not less than 5%

Phyto-chemical evaluation:

The aqueous extract of *P. corymbosa* root had alkaloids, steroids, carbohydrates and quinone, tannins, flavanoids, coumarins (Table. 2).

Table 2: Showing chemical constituents reported during preliminary phytochemical screening

TEST	<i>Agnimantha</i> root
Alkaloid	+
Steroid	+
Carbohydrate	+
Tannin	+
Flavanoids	+
Saponins	-
Terpenoid	-
Coumarins	+
Phenol	+
Carboxylic acid	-
Amino acids	-
Resins	-
Quinone	+

HPTLC: Retention factors (R_f) values of bands obtained were calculated by exposing the plates to different wavelengths of light (Figure 2). Under short UV, root of *P. corymbosa* showed green coloured spots at 0.50, and 0.89. The R_f values in long UV showed blue coloured spots at 0.68, 0.75, 0.85, 0.93. After derivatisation, when the plate is observed under white light, it showed violet and blue coloured spots for roots. R_f values in derivatised plate were 0.04, 0.16, 0.20, 0.53, 0.66, 0.77 in that the R_f value 0.53 showed blue coloured spot and the rest was violet coloured (Table 3).

Table 3: R_f values of *Agnimantha* root

Short UV	Long UV	Post derivatisation
-	-	0.04 (L. violet)
-	-	0.16 (L. violet)
-	-	0.20 (L. violet)
0.50 (L. green)	-	-
-	-	0.53 (Blue)
-	-	0.66 (D. violet)
-	0.68 (FL. blue)	-
-	0.75 (FL. blue)	-
-	-	0.77 (L. violet)
-	0.85 (FL. blue)	-
0.89 (L. green)	-	-
-	0.93 (FL. blue)	-

From the results of HPTLC densitometric scan, six chemical components at 254 nm, five chemical components at 366 nm and fourteen chemical compounds at 620 nm were found in roots of *P. corymbosa* (Figure 3).

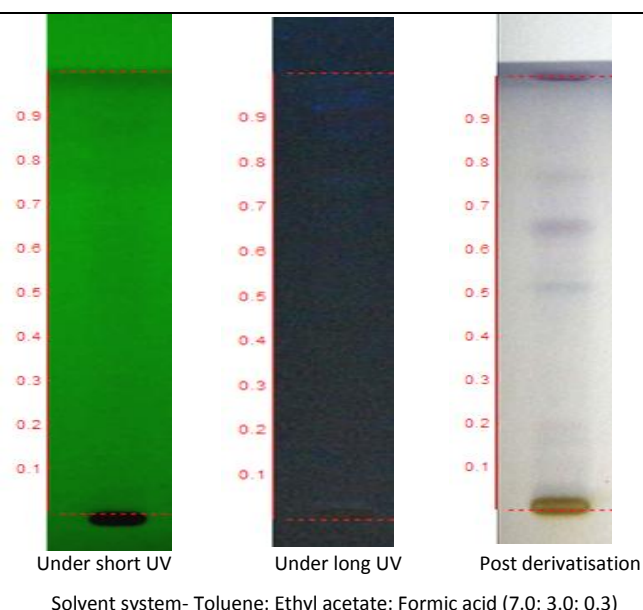


Figure 2: HPTLC of ethanoilic extract of *Agnimantha* root

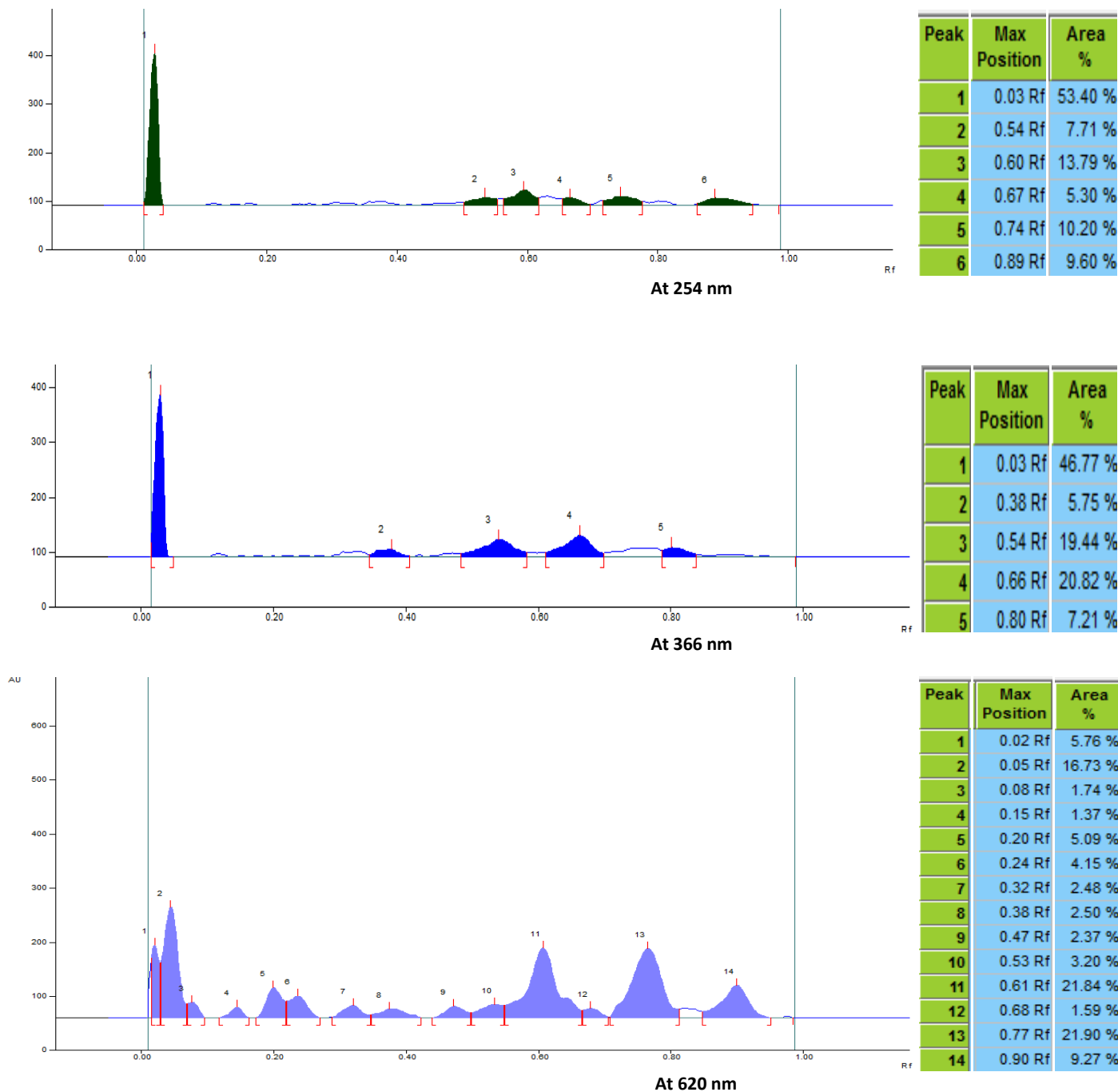


Figure 3: Densitometric scan of Agnimantha root

CONCLUSION

In the present study the pharmacognostic analysis, chemical constituents, TLC and HPTLC of the roots of *Agnimantha* co-relates with API standards and justifying its identity and authenticity.

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