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Preliminary phytochemical screening, pharmacognostic and physicochemical evalution of leaf of *Gmelina arborea*

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ABSTRACT

Objective: The aim of present study was to carry out preliminary phytochemical screening, detailed pharmacognostic profile and physicochemical evaluation of leaf of *Gmelina arborea*. Methods: Fresh leaf and dried power of the leaves were studied by morphology, microscopy, preliminary phytochemical screening, and florescence analysis of powdered drug. Other physicochemical parameters were also performed as per WHO guide lines. Result: The detailed microscopy revealed that the presence of anomocytic stomata and covering uni–multicellular trichome. Leaf constant such as stomatal number, stomata index, vein islet number, vein termination number were also determined. Physicochemical parameters and florescence analysis were also studied. The preliminary phytochemical screening showed the presence of steroid, triterpenoid, saponin, protein, phenolic compound, flavanoid and carbohydrates. Conclusions: The result of these studies could be useful for correct identification and detection of adulterants of this plant material.

1. Introduction

Gmelina arborea is an unarmed, moderately sized to large deciduous tree, about 30 m or more in height and a diameter of up to 4.5 m. Leaf is used as carminative, in headache, in anasarca, asthma, bronchitis, cholera, colic pain, dropsy, epilepsy, phthisis, rheumatism, small pox, sore, spleen complaints, syphilis, throat swelling, urticaria, as antidote to snake bite and some other poisons, cough, gonorrhea. Leafpaste is applied in on wounds. Charaka prescribed a paste of the leaves as ingredients of a medicated clarified butter for stiffness of the back, facial paralysis; prescribed the soup of fruits in diarrhoea. A paste of leaf is applied to the head for the relief of headache in fever. [1-3] The leaves are used in dyspepsia, cough, wound treatment, [4] Leaf paste in cephalegia and foul ulcer. [5] The juice of leaf is used as foetid discharge, worm from ulcers, demulcent, [6] diabetes and antidote. [7] Leaf has reported anthelmintic activity [8] and antimicrobial activity.[9] The current study was

carried out to provide requisite pharmacognostic details, phytochemical aspects and preliminary phytochemical screening of leaf of *G. arborea*.



Figure 1. Plant of Gmelina arborea

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2. Material and methods

2.1 Plant material

Fresh leaves of *G. arborea* were collected from Vadodara in the month of August–September 2011. Plant was identified at Botany Department of The M. S. University, Vadodara. Voucher specimen (DC–GM–1) was stored in herbarium of our laboratory. Leaves were separated, dried under shade and powdered.

2.2 Reagent and Chemicals

All the chemicals and reagents used were of analytical grade, purchased from Sigma chemical co. (St Louis, MQ, USA) and Merck (Darmstadt, Germany).

2.3 Pharmacognostic study [10–12]

Leaves were subjected to morphological examinations. Microscopic evaluation of leaf was carried out by taking the transverse sections using standard procedures and then subjecting them to microscopic examination. The powdered samples were also subjected to histological examinations using standard procedures and their diagnostic features were identified and recorded and observed under Zeiss microscope using Mips Olympus camera. Various leaf constant like stomatal number, stomatal index, palisade ratio, vein islet number and vein termination number were also determined. Different diagnostic features were identified and reported in the results.

2.4 Fluorescence analysis [13]

The fluorescence nature of powder drug was analyzed and the observations with different chemicals were also carried out and recorded.

2.5 Physicochemical evaluation of Leaf of Gmelina arborea [14]

The various physicochemical properties like water soluble extractive value, alcohol soluble extractive value and loss on drying) were determined as per WHO guidelines.

2.6 Preliminary Phytochemical screening [15]

Phytochemical screening was carried out by using procedure by Kokate. All the extracts were concentrated by distilling the solvent and the extracts were dried under reduced pressure. Consistency, color, appearance of the extracts and their percentage yield were noted. The extracts obtained from successive solvent extraction were then subjected to various qualitative chemical tests to determine the presence of various phytoconstituents like alkaloids,

glycosides, carbohydrates, phenolics and tannins, proteins and amino acids, saponins, and phytosterols using reported methods.

Results

3.1 Morphological study

Morphological character of leaf plant is reported in Table 1. and compared with reported character in Figure-2.

Table 1Morphological characters of leaf of *G. arborea*

| Parameters | Characters |
|------------|---|
| Color | Dark green |
| Size | 7 21cm length and 7-13 cm width |
| Shape | Broadly Ovate or cordate |
| Apex | acuminate or caudate |
| Margin | entire on mature plants but strongly toothed or lobed |
| | on young leaf |
| Venation | reticulate |
| Surface | Smooth |
| Base | Cordate |
| Petiole | 5–15 cm |
| Midrib | Prominent on lower surface |
| Odour | Characteristic and slightly disagreeable |
| Phyllotaxy | Opposite |
| Taste | Slight |



Figure 2. Leaf of G. arborea

3.2. Microscopical Characters:

3.2.1 Microscopy of leaf

Transverse section of leaf shows following characters (Figure 3, 4)

Lamina: It is a dorsiventral. Upper epidermis is Single layered with polygonal cells covered outside with a thick walled cuticle, covering trichome and anomocytic stomata are present. Mesophyll is a differentiated into palisade and spongy parenchyma. Palisade single layered present below the upper epidermis. Vascular strand are in mesophyll. Spongy parenchyma is a thin, 3 to 6 layers loosely arranged with intercellular space. Lower epidermis is very similar to upper epidermis but more number of anomocytic stomata

and uni-multicellular (2-3 celled) trichomes are present on the lower epidermis.

Midrib

Epidermal layers of lamina are in continuity with that of midrib. The dorsal surface and ventral surface are bulged. A 2 to 4 layered collenchyma can be seen below the upper epidermis and above the lower epidermis. Two small vascular bundles are present below the upper collenchymatous layer of midrib. The rest of midrib is occupied by the cortical parenchyma with the collateral vascular bundle embedded in the middle. Xylem is towards the centre and phloem towards the periphery. Parenchymatous tissue is thin walled with prominent intercellular spaces. The vascular bundles are surrounded by incomplete sheath of pericycle. Ground tissue is present in the centre of vascular bundle.

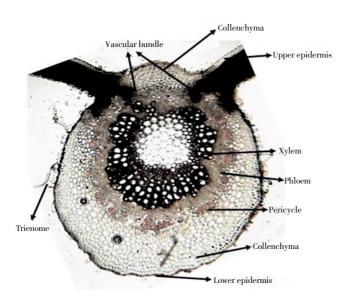


Figure 3. T.S of G. arborea leaf

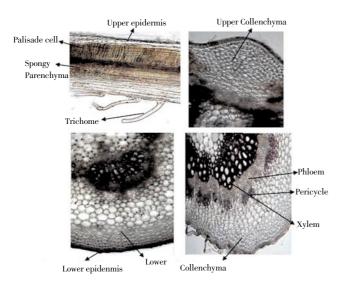


Figure 4. Microscopical character of leaf of G. arborea

3.2.2 Microscopical character of Petiole

It is more or less concave —convex, having single layered epidermis with cuticle. Trichomes are present on epidermis. Dorsal surface is convex and grooved. Below the each groove vascular bundle is present. Outer 3—5 layer of cortex is collenchymatous while inner 3—4 layers are parenchymatous cells contain chlorophyll. Endodermis is indistinct. Vascular bundle are collateral arranged in ring, groped of lignified pericycle fibre crown the phloem. Ground tissue is made up of parenchymatous cells with intercellular space.(Figure 5.)

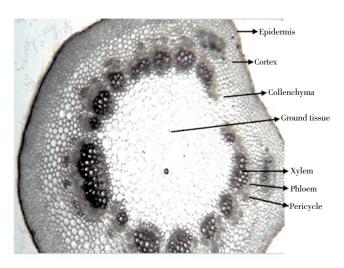


Figure 5 T.S. of petiole of leaf of G. arborea

3.3 Leaf powder characteristic

It is dark greenish color, bitter taste and characteristic odour. Leaf powder microscopy shows presence of anomocytic stomata, covering trichome, spiral xylem vessels, lamina fragments and mesophyll are reported in Figure 6.

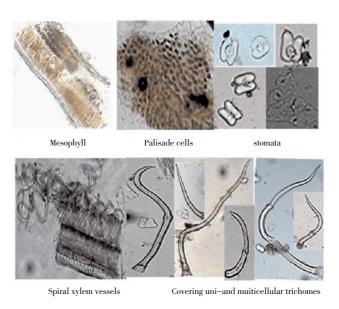


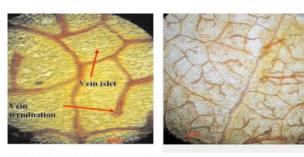
Figure 6. Powder characteristic of leaf of G. arborea

3.3 Quantitative Microscopy of leaf

Various leaves constant like stomatal number, stomatal index etc. are reported in table 2 and Figure 7.

Table 2 Leaf constant of Gmelina arborea leaf

| Loui constant of ometima around real | | | | |
|--------------------------------------|-----------------------|--|--|--|
| Parameter | Value (in I mm2 area) | | | |
| Stomatal number (lower epidermis) | 205-225 | | | |
| Stomatal index(lower epidermis) | 8-18.7-27.8 | | | |
| Palisade ratio | 4.25-5.6-6.3 | | | |
| Vein termination number | 10-20 | | | |
| Vein islet number | 30-40 | | | |







Lower Epidermis

Upper Epidermis



Figure 7. Surface preparation of *G. arborea* leaf

3.5 Fluorescence analysis

3.6 Physico chemical constant

Physico -chemical constant leaf of Gmelina arborea are given in Table 4.

Table 3 Fluorescence analysis of powder with various reagents

| Reagent | Visible light | U.V. light |
|----------------------|-----------------------|-------------------|
| Drug powder as such | Light green | NF |
| NaOH (aqueous) | Brownish yellow | Greenish yellow |
| NaOH (alcohol) | Green | Golden Yellowish |
| HCL | Light green | Light green |
| H2So4 | Light yellowish | Greenish yellow |
| powder + Nitric acid | Light yellowish-brown | Greenish yellow |
| Picric acid | Yellowish green | Yellowish green |
| Acetic acid | Light yellowish-brown | Golden yellow |
| NH3 | Yellowish green | Intense Yellowish |
| | | green |
| KOH(alcohol) | Brownish -yellow | Greenish yellow |

Table 4 Physicochemical constant of leaf of Gmelina arborea

| Parameter | *Average values %w/w | | |
|----------------------------------|----------------------|--|--|
| Water soluble extractive value | 24% | | |
| Alcohol soluble extractive value | 17.5% | | |
| Loss on drying | 2% | | |

*The values given here are expressed as percentage of air dried material. Each value is average of three determinations.

3.7 Preliminary phytochemical screening

A successive solvent extracts of leaf was studied for their phytochemical profile. Their % yield, color and consistency are recorded in Table- 5. The extracts obtained from successive solvent extraction were then subjected to various qualitative chemical tests for the identification of various plant constituents. A leaf shows the presence of carbohydrates, Saponins, steroid, flavonoid, phenolics compound. (Table 6.)

Table 5 Preliminary phytoprofile of leaf of G. arborea

| Extracts | Color and consistency | %Yield (w/w) | | |
|-----------------|-----------------------|--------------|--|--|
| petroleum ether | Dark green, Sticky | 3.76% | | |
| Toluene | Dark green, Sticky | 1.92% | | |
| Chloroform | Dark green, Sticky | 1.56% | | |
| Ethyl acetate | Dark green,Sticky | 2.26% | | |
| Methanol | Green,Sticky | 19.36% | | |
| Water | Brown.,non sticky | 26.3% | | |

4. Discussion

Pharmacognostic study is the initial step to confirm the identity and to assess the quality and purity of the crude drug. Quality control of crude drugs is very challenging task because of complex nature of chemical constituents. Microscopical evaluation is simplest and reliable tool for correct identification of herbs as well as small fragment of crude drugs or powderd drugs and

 Table 6

 Qualitative chemical test on extracts of leaves of Gmelina arborea

| Chemical constituent | P.E ext. | Tol. ext. | CHCl₃ ext | E.A.ext | Methanol ext | Water ext |
|-----------------------|----------|-----------|-----------|---------|--------------|-----------|
| Carbohydrates | - | - | - | - | + | + |
| Proteins | - | - | - | - | - | + |
| Saponins | - | - | - | _ | + | + |
| Alkaloids | _ | _ | _ | _ | _ | _ |
| flavonoids | _ | _ | _ | _ | + | _ |
| Tannin & phenolics | _ | _ | _ | + | + | + |
| Steroids & triterpens | + | + | + | + | _ | _ |

P. E. = Petroleum ether, Tol. = Toluene, E.A. = Ethyl acetate, (- = absent, + = positive)

detection of adulterants and substituents. [16-18] There is no pharmacognostic work reported on leaf of this plant. So the present work was undertaken for development of pharmacognostic standard of leaf of G. arborea. Some of the diagnostic features of the leaf drug noted from the microscopical study are anomocytic stomata, covering trichome. Physicochemical studies revealed the presence of, alcohol soluble extractive; 17.5% and water soluble extractive; 24.0%. Preliminary phytochemical study showed that the presence of steroid, saponin, carbohydrates triterpenoid and phenolic compound. The details of pharmacognostic characters, various evaluative parameters, results of preliminary and detailed phytochemical analysis established in the present study will facilitate in identifying the genuine drug and will also be useful in preparation of monographs of leaf of this plant.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgment

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