

# PHARMACOGNOSTICAL AND PRELIMINARY PHYTOCHEMICAL SCREENING OF THE LEAF *SWIETENIA MACROPHYLLA* KING (MELIACEAE) COLLECTED FROM TERAI AND DUARS REGION OF WEST BENGAL, INDIA

U. Sharma<sup>1</sup>\*, S. Deb<sup>1</sup>, S. Das<sup>2</sup>, A. Choudhury<sup>3</sup>.

<sup>1</sup>Institute of Pharmacy, Jalpaiguri, W.B., India <sup>2</sup>Calcutta Medical College and Hospital, Kolkata, W.B., India <sup>3</sup>North Bengal University, Darjeeling, W.B., India.

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## **ABSTRACT:**

*Swietenia macrophylla* King belonging to the family Meliaceae is a common plant in Terai and Duars. Traditionally the plant is used to cure malaria, anaemia, diarrhoea, fever, dysentery, hypertension, cancer, coughs, chest pains, intestinal parasitism, and anti-ulcer activity. The current work is oriented towards exploring the microanatomical, physicochemical and phytochemical aspects of the plant collected from Terai and Duars areas of West Bengal, India. Macroscopical and microscopical characters were studied. Different Physico-chemical parameters were observed. Preliminary phytochemical studies show the presence of important phytoconstituents. The presence of phytoconstituents explains that the plant must have valuable medicinal properties which must be explored.

**KEYWORDS:** *Swietenia macrophylla* King, Macroscopical and microscopical studies, Phytochemical screening, Flavonoid.

Corresponding Author: Uttam Sharma, Email: <u>uttam\_20022k@yahoo.co.in</u>

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## INTRODUCTION

*Swietenia macrophylla* King, commonly known as Mahogany, Honduran mahogany, or Bigleaf Mahogany. The Asian countries which grow the majority of *Swietenia macrophylla* King are India, Indonesia, Malaysia, Bangladesh, Fiji, Phil ippines, Singapore. *S. macrophylla* is located in more than 40 countries including in Brazil, Bolivia, Mexico, Guatemala, Peru and other central American countries, South Africa <sup>[1,2,3,4,5,6]</sup>. In India it is found in almost all part of India.

The tree of S. *macrophylla* King is usually taller than 30 m, with straight trunk and cylindrical with 100 to 200 cm at breast height. The bark is dark reddish brown, entirely rosy, thick and deeply furrowed. The leaves are alternate with leaflets opposite or occasionally changed. The small flowers have yellow-cream colored panicles. The fruit is woody, consisting of capsule, ovoid, color light brown, which opens on 5 shares, with 10 to 14 winged seeds <sup>[2,5,7]</sup>.

*Swietenia macrophylla* King contained various chemical compounds. A number of limonoids have been reported from the genus *Swietenia* with structures assigned on the basis of spectral data <sup>[5,8]</sup>.

Some chemical constituents were found from the terminal shoots, senescent and mature leaves as the essential oil components in form of fatty acids and terpenoids such as  $\gamma$ -himachalene, germacrene D, germacrene A, cadina-1,4-diene, hexadecanoic acid and ethyl hexadecanoate <sup>[5,9,10]</sup>.

Leaves and Seed also have medicinal value. In general, *S. macrophylla* King extracts especially the plant seed have many medical efficacy. Which is proven traditionally and scientific as used to cure malaria, anaemia, diarrhoea, fever, dysentery, hypertension, cancer, coughs, chest pains, intestinal parasitism, and anti-ulcer activity <sup>[4,11,12]</sup>. On the other hand, *S. macrophylla* King leaves can be used for dyeing agent while the bark extract has been used as an astringent for wounds and used occasionally for tanning because of the rich red colour <sup>[5,9]</sup>.

Few of the recent scientific study on *S. macrophylla* King collected from North East India proved that seed have potent medicinal value but in case of leaves through study has not done. So that in present study young leaves are taken from Terai and Duars areas of West Bengal, India for systematic evaluation to discover some pharmacognostical and medicinal value.

## MATERIAL AND METHODS Collection of the specimen:

The plant species for the proposed study has been collected from Terai and Duars areas of West Bengal, India. Proper care was taken to select healthy plants with normal organs. The species for the proposed study was identified as Swietenia macrophylla King by Professor, Dr. A.P. Das, MSc, DIIT, PhD, FIAT, FNScT, Ex-Head. Department of Botany, Ex-Director. Centre for Life Sciences, Ex-Coordinator: Department of Biotechnology, Taxonomy & Environmental Biology Laboratory, Department of Botany, North Bengal University, Darjeeling 734 013, W.B., India and voucher specimen (Accession No. 09691) was deposited in the Herbarium of the same department.

# Sectioning:

The macroscopic characters of the leaf such as colour, odour, taste, nature, texture were studied for morphological investigation. For anatomical studies free hand sectioning was performed to obtain a thin transverse section of leaf with the help of 7 o'clock blade and central portion of mature leaflets were taken. The thick sections were stained with safranin.

Photomicrographies were taken with Nikon lab photo – microscopic unit. The quantitative microscopy was studies as per the procedure given by Wallis <sup>[13,14]</sup>. The powder analysis has been carried out according to the method of Brain and Turner <sup>[14]</sup>.

Descriptive terms of the anatomical features are taken from the standard Anatomy books <sup>[16-20]</sup>.

# Physicochemical parameters:

The residue left after incineration of a drug is designated as ash. The residue originate from inorganic elements present in the plant is called as physiological ash. It varies with in definite limits according to types of soil, dust, sand and mineral impurities and admixture of other drugs may alter the ratio. Ash value represents the inorganic salts naturally occurring in the drug and adhering to it. Total ash is the residue remaining after incineration. The acid insoluble ash is the part of total ash which is insoluble in dilute hydrochloric acid. Mixing of sulphuric acid with powdered crude drug before ashing and this sulphated ash is normally less fusible than ordinary ash.

Extractive value which is an indicative of approximate measures of chemical constituents and nature of the constituents was performed using ethanol and distilled water as solvents.

The moisture content was determined in reference to air-dried sample by loss on drying method.

Many plant (medicinal) materials contain saponins that can cause persistent foam when an aqueous decoction is shaken. The foaming ability of an aqueous decoction of plant materials and their extracts are measured in terms of 'foaming index'.

All the procedure taken from the standard books <sup>[20-22]</sup>.

#### Fluorescence analysis and behavioral change:

A very small quantity of powdered drug was kept in a watch glass in an accumulated form. Then 2-3 drop of respective reagent was added and the fluorescence character of the plant powders was studied both in daylight and UV light as such and after treatment with the reagent like sodium hydroxide, picric acid, acetic acid, Hydrochloric acid, nitric acid, iodine, ferric chloride etc. similarly the fluorescence analysis of the plant extract was observed under visible and UV light <sup>[23]</sup>.

#### **Extraction:**

Successive extraction was done in soxhlet

extractor using the following solvents: Petroleum Ether, Ethyl Acetate, Methanol, and Water. All the extracts obtained by extraction were subjected to various qualitative tests for the identification of various plant constituents present in the species <sup>[20]</sup>.

## Thin layer chromatography:

Ascending Thin layer chromatography was performed for the separation of phytocontituents. As Ethyl acetated extract gives flavonoids fractions and now a day flavonoids is an important potent phytoconstituents so it was subjected for this study with different ratios of various solvents, and it's showed clear isolation and resolution of spots in different solvent ratio on the TLC plates. Authentic flavonoid samples of also run in the TLC plates. Different spots developed in each solvent system were identified through UV light (510 nm) and the Rf values were accordingly calculated <sup>[24-26]</sup>.

## RESULTS

In macroscopical studies of the leaf, it were found that it's shape was usually oblong to oblonglanceolate or ovate-lanceolate with acute apex and entire margins, varying in size from 5-7cm wide and 16-30 cm in long. The leaves are alternate with leaflets opposite or occasionally changed. Colour was dark glossy green (upper surface), light green (lower surface), upper surface was smooth and lower surface was slightly rough, odour was characteristic, taste was pungent bitter (Fig. 1).



Figure 1: Leaf of Swietenia macrophylla King

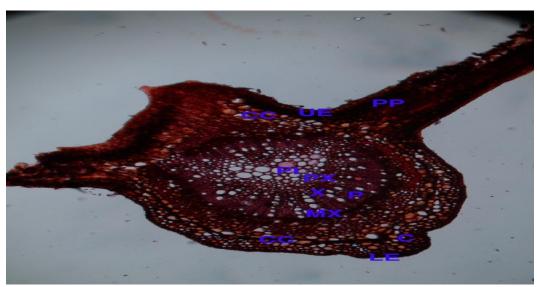


Figure 2: Microscopy of the leaf of *Swietenia macrophylla* King UE: Upper epidermis, LE: Lower epidermis, CC: Collenchyma, C: Cortex, X: Xylem, PX: Proto xylem, MX: Meta xylem, P: Phloem, PI: Pith, PP: Palisade parenchyma.

In microscopy T.S of leaf shows Upper and lower epidermis with anticlinal walls. Xylem and Phloem are found. Xylems are surrounded by Proto and Meta Xylem. Few collenchymas cell are present below upper and above lower epidermis. Layer of Palisade Parenchyma cell, Cortex and Pith was found (Fig. 2).

Physico-chemical parameters like Total ash value (7 % w/w), acid insoluble ash value (0.67 %w/w), water-soluble ash value (10.33 %w/w) and sulphated ash value (8 %w/w) were observed. Alcohol soluble extractive value (1.8 %w/w), water-

soluble extractive value (2.2 %w/w) were also observed and loss on drying was observed as 3.4 %w/w. The foaming index was found to be 111.11. The result of different physicochemical parameters are given in Table No. 1

The dried leaf of *Swietenia macrophylla* King were extracted with petroleum ether, ethyl acetate, methanol and distilled water by continuous hot soxhlet extraction and the percentage yield were found to be 0.76 % w/w, 2.83 %w/w, 1.9% w/w and 2.20 % w/w respectively.

| SL  | PARAMETERS                 | % w/w |
|-----|----------------------------|-------|
| No. |                            |       |
| 1   | Alcohol Soluble Extractive | 1.8   |
| 2   | Water soluble Extractive   | 2.2   |
| 3   | Ash values                 |       |
|     | (i) Total Ash              | 7     |
|     | (ii) Acid Insoluble Ash    | 0.67  |
|     | (iii) Water Soluble Ash    | 10.33 |
|     | (iv) Sulphated Ash         | 8     |
| 4   | L D .                      | 3.4   |
|     | Loss on Drying             |       |

| Table 1: | Physico-chemical | parameters of | powdered of | different plant materials |
|----------|------------------|---------------|-------------|---------------------------|
|----------|------------------|---------------|-------------|---------------------------|

Fluorescence analysis and behavior of the leaf powder revealed wide range of fluorescence colours. In comparison, it was observed that all the parts showed similar colour ranges with mild differences. This may be due to the presence of similar phytoconstituents. The result of fluorescence analysis and behavior of the leaf powder are given in the Table No 2,3.

| SL No. | Treatment with chemical reagents         | Observation            |  |
|--------|--|------------------------|--|
| 1.     | Powder as such                           | Brown                  |  |
| 2.     | Powder + 1N Sodium hydroxide in methanol | Yellowish Green        |  |
| 3.     | Powder + 1N Sodium hydroxide in water    | Pale Greenish<br>Black |  |
| 4.     | Powder + 50% Hydrochloric acid           | Greenish Black         |  |
| 5.     | Powder + 50% Sulphuric acid              | Black                  |  |
| 6      | Powder + 50% Nitric acid                 | Greenish Black         |  |
| 7      | Powder + Petroleum ether                 | Pale Green             |  |
| 8      | Powder + Chloroform                      | Black                  |  |
| 9      | Powder + Picric acid                     | Black                  |  |
| 10     | Powder + 5% Ferric chloride solution     | Greenish Black         |  |
| 11     | Powder + 5% Iodine solution              | Greenish Black         |  |
| 12     | Powder + Methanol                        | Greenish Black         |  |
| 13     | Powder + (Nitric acid + Ammonia)         | Green                  |  |
| 14     | Powder + Glacial acetic acid             | Pale Green             |  |
| 15     | Powder + 5% Potassium Hydroxide solution | Pale Green             |  |

Table 2: The fluorescence analysis of the powder of different plant materials

| Table 3: The behavior of the leaf powder of different plant materials when treated with |  |  |  |
|---|--|--|--|
| different chemical reagents   |  |  |  |

| SL<br>No. | Treatment with chemical reagents         | Observation     |
|-----------|--|-----------------|
| 1.        | Powder as such                           | Brown           |
| 2.        | Concentrated Hydrochloric acid           | Greenish Brown  |
| 3.        | Concentrated Sulphuric acid              | Intense Brown   |
| 4.        | Concentrated Nitric acid                 | Brown           |
| 5.        | Glacial acetic acid                      | Deep Brown      |
| 6         | 5% Sodium hydroxide solution             | Brown           |
| 7         | 5% Potassium hydroxide solution          | Deep Brown      |
| 8         | 5% Ferric chloride solution              | Greenish Black  |
| 9         | Picric acid                              | Yellowish Green |
| 10        | Ammonia                                  | Deep Brown      |
| 11        | 1N Sodium hydroxide in methanol          | Deep Brown      |
| 12        | Powder + 1N Sodium hydroxide in<br>Water | Brown           |

The extracts obtained were subjected to qualitative phytochemical tests to find out the active constituents. which showed presence of carbohydrates, fixed oils and fats, alkaloids, and saponins, tannins phenolic compounds, flavonoids and coumarins in petroleum ether extract, carbohydrates, fixed oils and fats, alkaloids, saponins. tannins and phenolic compounds, flavonoids and coumarins in ethyl acetate extract,

carbohydrates, fixed oils and fats, gums and mucilage, alkaloids, glycoside, saponins, flavonoids, triterpenoides and coumarins in methanolic extract, carbohydrates, fixed oils and fats, alkaloids, saponins, tannins and phenolic compounds, flavonoids, triterpenoides and coumarins in aqueous extract. Details of phytochemical tests are given in the Table No. 4.

| PLANT CONSTITUENTS             | S.M. Extract |         |            |         |
|--------------------------------|--------------|---------|------------|---------|
|                                | Pet. Ether   | Ethyl   | Methanolic | Aqueous |
|                                |              | Acetate |            |         |
| Carbohydrates                  | +            | +       | +          | +       |
| Proteins and Amino acids       | -            | -       | -          | -       |
| Fixed Oils and Fats            | +            | +       | +          | +       |
| Gums and Mucilage              | -            | -       | +          | -       |
| Alkaloids                      | +            | +       | +-         | +       |
| Glycosides                     | -            | -       | +          | -       |
| Steroids /Phytosterol          | -            | -       | -          | -       |
| Saponins                       | +            | +       | +          | +       |
| Tannins and Phenolic compounds | +            | -       | +          | +       |
| Flavonoids                     | +            | +       | +          | +       |
| Triterpenoides                 | -            | -       | +          | +       |
| Lignin                         | -            | -       | -          | -       |
| Coumarins                      | +            | +       | +          | +       |

Table 4: Qualitative phytochemical analysis of various extracts of different plant materials

(+): Present, (-): Absent, P.E.: Pet Ether, E.A.: Ethyl Acetate, M.: Methanol, A.: Aqueous

Ethyl acetate extract in n-BuOH: HOAc:  $H_2O$  (125:72:3) solvent system showed one spot with Rf values 0.90 (Florescence yellow, orange), the spots identified authentic flavonoid samples and test sample as Quericetin.

## DISCUSSION

In macroscopical amd microscopical studies of the leaf characters collected from Terai and Duars areas of West Bengal, India are slightly different with specific geographical variation. Physicochemical parameters of the leaves of this Terai and Duars areas also show some characteristic results. The extracts obtained were subjected to qualitative phytochemical tests to find out the active constituents, which showed presence of carbohydrates, fixed oils and fats, alkaloids, saponins, tannins and phenolic compounds, flavonoids and coumarins etc in different extracts. So the leave part is very much rich in different phytoconstituents. On the basis of standard sample Rf values Quericetin have been identified. Several other solvent system such as  $CH_2Cl_2$ : HOAc:  $H_2O$  (2: 1: 1), n-BuOH: HOAc:  $H_2O$  (4:1:5), EtOAc: EtOH: HCOOH:  $H_2O$  (100:11:11:26), EtOAc: HCOOH: HOAc:  $H_2O$  (100:11:11:26),  $C_6H_6$ : HOAc:  $H_2O$  (125:72:3) were also tried, but the solvent system containing n-BuOH: HOAc (125:72:3) gave best result.

#### CONCLUSION

Anatomical and physicochemical studies of the leaf characters collected from Terai and Duars areas of West Bengal, India have some characteristic geographical variation. The current study identified the one compound Quericetin (flavonol) from the Ethyl acetate extract of *Swietenia macrophylla* King. Presence of that constituent may be one of the contributing factors responsible for the different types of therapeutic activity. Further investigations are required to study the mechanism of actions of *Swietenia macrophylla* King and its constituents by which they exert their therapeutic effects.

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