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# Physico-chemical and HPTLC studies on leaf and root of *Mukia maderaspatana* (L.)M. Roemer.

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**ABSTRACT**

*Mukia maderaspatana (L.) M. Romer., a plant drug of Siddha medicine, is an annual monoecious tendril climber, belonging to the family Cucurbitaceae, mostly prevalent in South India. It is commonly called Musumusukkai in Tamil. The leaf and root part of the plant were collected and investigated for their physico-chemical properties. The leaf and root showed distinct variations. High performance thin layer chromatography analysis of ethyl alcohol extract of leaf and root samples gave three and five bands respectively. Both the leaf and root samples shows the $R_f$ value 0.20 (β-sitosterol), the root sample shows the $R_f$ value 0.56 (columbin).*

**Key words:** *Mukia maderaspatana*, Cucurbitaceae, HPTLC.

## INTRODUCTION

Plants are the traditional source for many of the chemicals used as pharmaceuticals, biochemicals, fragrance, food coloures and flavors. Most valuable phytochemicals are the products of secondary metabolism and possess sufficient chemical or structural complexity [14]. Many plants synthesize substances that are useful to the maintenance of health in humans and other animals. These include aromatic substances, most of which are phenols or their oxygen-substituted derivatives such as tannins. Many are secondary metabolites, of which at least 12,000 have been isolated — a number estimated to be less than 10% of the total. In many cases, these substances (particularly the alkaloids) serve as plant defense mechanisms against predation by microorganisms, insects, and herbivores. Many of the herbs and spices used by humans to season food yield useful medicinal compounds [8].

*Melothria madraspatana* (Syn. *Mukia maderaspatana* L.*)* belongs to the family Cucurbitaceae. The plant is a tendril climber/prostate herb. The plant was reported to have activities such as

hepatoprotective [23], antirheumatic [19], diuretic, stomachic (a digestive tonic), gentle aperient, antipyretic and antiflatulent, antiasthmatic, anti-inflammatory, antidiabetic and antibronchitis and is used for tooth-ache besides its use in vertigo and biliousness [3; 10].

Identification of plants with botanical verifications is essential as contamination due to misidentification of plant species or parts in common. Characterizing compound or biomarker is identified from the plant part to assure the identity and quality of the preparation, this need not be responsible for the therapeutic activity. Thus, the present investigation was, therefore, undertaken to study the physicochemical bioactive properties of leaf and root of *Mukia maderaspatana.*

## EXPERIMENTAL SECTION

The leaf and root part of *Mukia maderaspatana(L.)* was collected from different places of Thanjavur district (Enathi village, Town Karambai, Keelamangalam). A voucher specimen has been deposited in the Tamil University Herbarium for future reference. They were identified with the help of regional Floras [5; 6; 15 & 18]. They were cleaned, air dried and powdered. The powdered leaf and root parts were subjected to physicochemical analysis by the methods described by Indian Pharmacopoeia [2]. The various fractions of successive extracts were weighed and preliminary phytochemical screening [4; 11& 12].

**High Performance Thin Layer Chromatography** [21]
HPTLC was performed on aluminum packed silica gel $60F_{254}$ HPTLC plates (Merck). The mobile phase was acetone - alcohol (1:1) for Ethyl alcohol extracts of both the samples. Samples were applied to the plates as sharp bands by means of Camag Linomat IV samples applicator. After drying the spots in a current of air the plats were placed in one trough of Camag twin trough glass chamber. The mobile phase was poured into the chamber left to equilibrate for 30 min. the plate was then developed until the solvent front had traveled a distance of 7 cm above the position of sample application. The plate was removed from the chamber and dried in a current of air. Detection was performed with a Camag TLC Scanner.

Chromatographic Condition

| | | |
|---|---|---|
| Stationary Phase | **:** | HPTLC Aluminum plate percolated with silica gel $60F_{254}$. |
| Solvent system | **:** | Acetone - Alcohol (1:1) |
| Separation Technique | **:** | Ascending. |
| Migration distance | **:** | 70mm. |
| Detection | **:** | UV. |
| Wave length | **:** | 270nm. |

## RESULTS AND DISCUSSION

Physicochemical values like total ash, water-soluble ash, acid-insoluble ash, sulphated ash, loss on drying, solubility in alcohol, water and extractive values of leaf and root of *Mukia maderaspatana* are given in Table-1.

Successive solvent extracts of leaf and root of *Mukia maderaspatana* was subjected to qualitative phytochemical screening (Table -2).

Ethyl alcohol extracts of leaf and root of *Mukia maderaspatana* were characterised by HPTLC. Numbers of major compounds were identified from its chromatogram. The percentage yields were calculated and the results were given in the Table -3.

In high performance thin layer chromatography analysis, ethyl alcohol extract of leaf sample gave three bands with $R_f$ value 0.02 0.20 and 0.87 (Fig. 1). Root sample gave five bands of $R_f$ value 0.02, 0.20, 0.56, 0.75 and 0.87 (Fig. 2). It is to be noted that 3 bands with $R_f$ value 0.02, 0.20 and 0.89 were similar in the leaf and root extracts.

**TABLE 1 Physicochemical analysis and extractive values (Percentage) of leaf and root powders of *Mukia maderaspatana***

| Sl. No | Parameters | Result/ Percentage of leaf | Result/ Percentage of root |
|---|---|---|---|
| 1 | **Organoleptic characteristics**<br>a. Appearance<br>b. Colour<br>c. Smell<br>d. Taste | <br>Powder<br>Greenish<br>No smell<br>Slightly Bitter | <br>Powder<br>Yellow<br>Aromatic<br>Bitter |
| 2. | Loss on drying 105 $^oC$ | 8.10 | 9.23 |
| 3. | Total Ash value | 10.72 | 12.52 |
| | Acid insoluble ash | 9.21 | 8.10 |
| | Sulphated ash | 20.66 | 24.64 |
| 4 | **Solubility values**<br>Alcohol | <br>8.64 | <br>8.90 |
| | Water | 8.65 | 7.98 |
| 5 | **Extractive values in**<br>Pet.ether | <br>2.016 | <br>3.082 |
| | Benzene | 2.020 | 2.130 |
| | Chloroform | 3.231 | 2.110 |
| | Ethanol | 7.217 | 6.731 |
| | Water | 8.282 | 7.391 |

**TABLE 2: Qualitative phytochemical screening of *Mukia maderaspatana* leaf and root**

| S. No | Compound Tested | Test applied/ Reagent used | LEAF | | | | | ROOT | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Pet.ether | Benzene | Chloroform | Alcohol | Water | Pet.ether | Benzene | Chloroform | Alcohol | Water |
| 1. | Carbohydrate | Fehilings | - | - | - | + | + | - | - | - | + | + |
| | | Benedicts | - | - | - | + | + | - | - | - | + | + |
| 2. | Alkaloids | Mayer's | - | - | - | - | + | - | - | - | - | + |
| | | Wagner's | - | - | + | + | + | - | - | + | + | + |
| | | Hager's | - | - | + | + | + | - | - | + | + | + |
| | | Dragendroff's | - | - | + | + | + | - | - | - | - | - |
| 3. | Phytosterols | L.B. Test | - | + | + | + | - | - | - | - | + | - |
| 4. | Tannins & phenols | 10% lead Acetate | - | - | - | + | + | - | - | - | + | + |
| 5. | Fixed oil & fats | Spot test | + | + | - | - | - | + | + | - | - | - |
| 6. | Saponins | Foam test | - | - | - | + | + | - | - | - | + | + |
| 7. | Gum & mucilage | Alcoholic Precipitation | - | - | - | - | + | - | - | - | - | + |

***Constituents:*** *+ = Present; – = Absent*

**TABLE 3 HPTLC profile of extracts of leaf and root of *Mukia maderaspatana***

| S.No | Sample | Total number of spots | Number of major spots | Retention factor ($R_f$) | % of Area | % Yield (100gm) |
|---|---|---|---|---|---|---|
| 1 | Leaf | 3 | 3 | 0.02 | 18.04 | 0.76 |
| | | | | 0.20 | 51.61 | 2.17 |
| | | | | 0.87 | 30.35 | 1.27 |
| 2 | Root | 5 | 5 | 0.02 | 11.24 | 2.10 |
| | | | | 0.20 | 11.14 | 1.92 |
| | | | | 0.56 | 51.61 | 2.17 |
| | | | | 0.75 | 12.20 | 2.10 |
| | | | | 0.87 | 30.40 | 5.23 |

*Solvent system used : Acetone: Alcohol (1:1)*
*Amount applied : 10µl of 20 mg/ml solution.*
*Spray reagent : Pankal –D*

Physiochemical values of leaf and root samples show distinct values. All the values can be used as diagnostic characters of the both the samples.

Qualitative phytochemical analysis of various extracts of leaf and root of *Mukia maderaspatana* showed distinguished values and can be used as diagnostic value characters. In quantitative estimation, total alkaloids, total glycoside and ascorbic acid had shown more or less similar concentration. However, total terpenoid concentration is higher in root and β-sitosterol concentration is higher in leaf. Hence, concentration of β-sitosterol and total terpenoids are used as diagnostic feature for leaf and root respectively. Phytochemicals such as alkaloids, total glycoside, ascorbic acid, β-sitosterol and total terpenoids are known to have therapeutic value [13; 20].

HPTLC finger print profile helps for the identification of active biological compounds and further isolation and characterization of various bands are to be carried out.

Both the leaf and root samples shows the $R_f$ value 0.20, which is similar to that of β-sitosterol standard [22], the root sample shows the $R_f$ value 0.56 which is similar to that of columbin standard [17]. HPTLC finger print profiles of leaf and root of *M. maderaspatana* with similar $R_f$ values to that of previous works reveal the possibility of presence of β-sitosterol and columbin in *M. maderaspatana.* However it remains to be confirmed.

Columbin is a bitter crystalline constituent of *Jateorhiza calumba* [16]. *Tinospora cordifolia* is a large climbing shrub, growing throughout tropical India; and popularly known as *Tinospora* in English. It contains diterpenoid furanolactone, tinosporidine, columbin and β-sitosterol. The bitter principle present shows adaptogenic, antispasmodic, anti-inflammatory, antipyretic, anti-neoplastic, hypolipidemic, hypoglycemic, antioxidant, immunopotentiating and hepatoprotective properties [1; 7]. *T. cordifolia* root is used in general debility, digestive disturbances, loss of appetite and fever in children. It is also an effective immunostimulant [9]. The anti-inflammatory, antipyretic and antioxidant activities of *Mukia maderaspatana* might be due to the presence of β-sitosterol and columbin.

Fig. 1: HPTLC finger print of leaf of *Mukia maderaspatana*

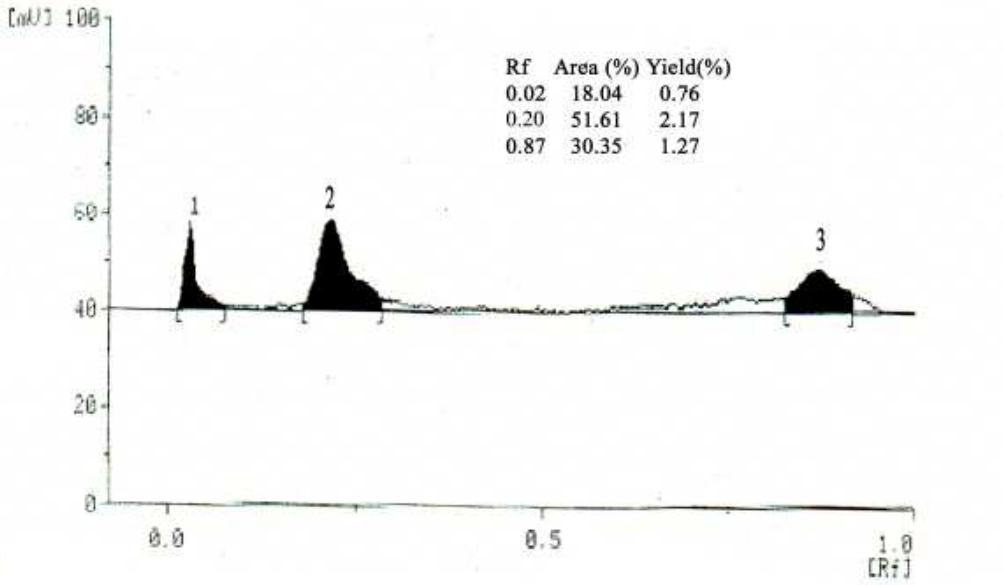
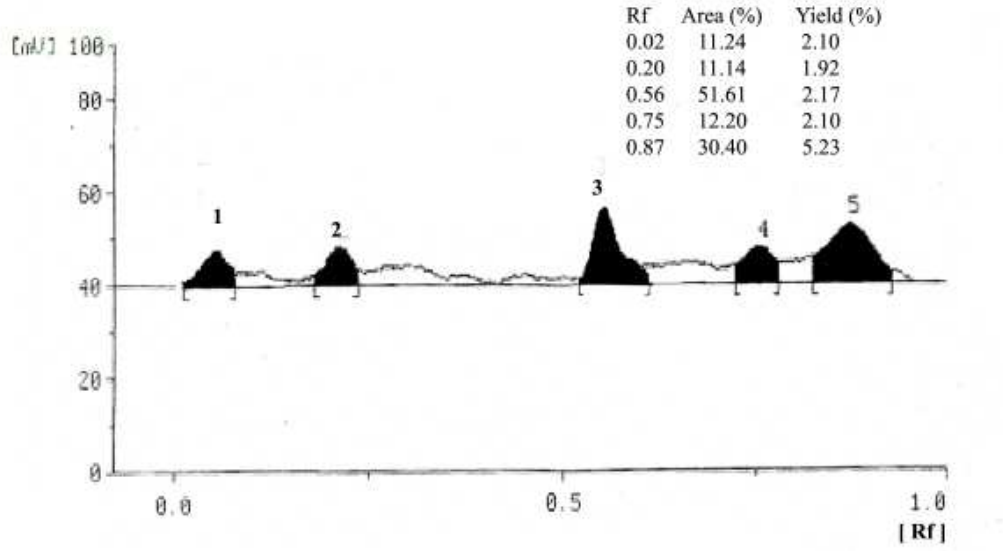



Fig. 2: HPTLC finger print of root of *Mukia maderaspatana*




## CONCLUSION

The leaf and root of *M.maderaspatana* has shows different physico-chemical characteristics and chemical composition. Thus both the plant parts are used medicinally one for the other.

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