Research Article

Preliminary Phytochemical Screening and Thin Layer Chromatography of Plant *Mimusops elengi* L Leaf Extracts

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ABSTRACT

To study the Physico and phyto chemical characters of the plant *Mimusops elengi* L (family *Sapotaceae*) and also to carry out the thin layer chromatographic fingerprint. Phytochemical investigations and fluorescence analysis were carried out as per the standard techniques. Various quantitative parameters like ash values, extractive values and flavonoid content can be used as quality control parameters for the plant *Mimusops elengi* L were determined. The ethyl acetate crude extract fraction with methanol and fingerprinting pattern was developed by using Thin Layer Chromatography (TLC) technique equipped with pre coated Silica plate of 12cm height using automatic TLC scanner and detected in Visual, UV light and fluorescent light. Preliminary phytochemical studies confirmed the presence of alkaloid, carbohydrate, glycoside, protein, tannin, flavonoid, triterpenoid and phenol. TLC analysis of ethyl acetate crude extract fraction methanolic extract of *Mimusops elengi* L shown that there was many phytoconstituents with different composition of mobile phase. TLC fingerprint analysis of the plant *Mimusops elengi* L can be used as a diagnostic tool for the correct identification of the plant and it is useful as a phytochemical marker and also a good estimator of genetic variability in plant populations.

Keywords: Plant extract of *Mimusops elengi L*, Phytochemical screening, Physico chemical character, TLC fingerprinting.

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INTRODUCTION

Since ancient times, plants and herbal preparations have been used as medicine. During the past decade, traditional systems of medicine have become a topic of global importance. Current estimates suggest that, in many developing countries, a large proportion of the population relies heavily on traditional practitioners and medicinal plants to meet primary health care needs. Concurrently, many people in developed countries have begun to turn to alternative or complementary therapies, including medicinal herbs.[1] Phytochemicals are in the strictest sense of the word, chemicals produced by plants. The plants generally contain 10 phytoconstituents namely anthraglycosides, arbutin, bitter drugs, flavonoids, alkaloids, saponins, coumarins, phenol carboxylic acids, terpenes and valepotriates. These phytoconstituents confer specific characteristics and properties to plants. Therefore, the analysis of these constituents in plants would help in determining various biological activities of plants.[2,3] Thus, the aims of present study are to investigate the phytochemical profile and thin layer chromatography (TLC) studies of the plant of *Mimusops elengi* L so as to know various components present in it and hence to assess the medicinal potential of the plant and justify its folklore use.

Mimusops elengi L[4, 5] is an evergreen ornamental tree of the family *Sapotaceae* with pleasant fragrant flowers. It carries a variety of names such as Bakul (Hindi and Bengali), Spanish cherry, West Indian Medlar or Bullet wood tree (English), Bakula (Sanskrit) etc. in different languages. *M. elengi* is regarded as one of the best medicinal plants since each and every part of it is used in various ways to cure a variety of human diseases. The bark is used as a tonic, and in gargles to cure odontopathy, inflammation and bleeding of gums. It is also useful in urethrorrhoea, cystorrhoea, diarrhoea and dysentery. The bark and seed coat are used for strengthening the gum and are utilized along with tannin rich substances like catechu (Acacia catechu), pomegranate (Punica granatum) bark etc. in various herbal tooth powders, such as "Vajradanti". Further, it is one of the constituents in the preparation of "Mahakhadiradivati" prescribed for stomatitis, halitosis, appetizer, anorexia, spongy gums and pharyngeal problems. The bark is also useful in painful high fever. The bark of *M. elengi* produces a commercial dye. The chemical constituents of the color components responsible for dyeing have been identified. The dyeing behavior of these color components on wool has also been evaluated. The

color components isolated from the bark mainly contain flavonoid moiety. The leaves have been considered as an antidote for snakebite. The flowers and unripe fruits are used as an ointment for treating wounds and ulcers. The powder from dried flowers is a brain tonic and relieves from cephalalgia. The flowers are used as expectorant, to cure problems of liver, nose, and are smoked in asthma. Further, the flowers are used to make garlands and for stuffing pillows. The fruits are aphrodisiac, diuretic, astringent to the bowels and good in gonorrhoea. The pulp of the ripe fruits has been successfully used to cure chronic dysentery. The immature fruits are chewed to protect loose teeth. The ripe fruits are given orally to pregnant women to facilitate delivery. The hot aqueous extract of fruits is given orally to human as diuretic which also acts as antipyretic. The ripe fruits rich in carbohydrates are good source of food.

Collection and authentication of the plant specimen

The leaf of *Mimusops elengi* L. were collected from the local areas of Sivakasi, Tamilnadu and were authenticated by DR.N.SENTHIL KUMAR Department of Botany, Ayya nadar janakiammal college, Sivakasi, Tamilnadu. A voucher specimen was deposited to authentication office for future reference.

Preparation of plant extract and evaluation of percentage yield

The dried leaves were subjected to size reduction to a coarse powder with the help of grinder and powdered material was then subjected to Soxhlet extraction employing ethanol as solvent. The extract was concentrated to dryness with the help of rotavapor and finally air dried thoroughly to remove all traces of the solvent. The obtained dried extracts were weighed and extractive value was calculated[6].

Physicochemical Parameters

Physico chemical constants such as percentage of total ash, acid insoluble ash, water soluble ash, water soluble and alcohol soluble extractive values were calculated according to the described methods.[7]Fluorescence analysis was conducted by using methods.[8,9]

Determination of Total Flavonoid Content[11]

Total flavonoids were estimated using the prescribed method. To 0.5ml of sample, 0.5ml of 2% AlCl3 ethanol solution was added. After one hour at room temperature, the absorbance was measured at 420 nm. A yellow color indicated the presences of flavonoid. Total flavonoid content was calculated as quercetin equivalent (mg/g) using the regression equation of the calibration curve: y = 0.41x + 0.087, R2 = 0.425

Phytochemical Screening

The whole plant extracts like petroleum ether, chloroform, ethyl acetate, Methanol extracts and the powder were subjected to qualitative chemical analysis as per the following procedure. The results were presented in Table 1. The extract was subjected to phytochemical analysis to test the presence of carbohydrates, phenolic compounds, flavonoids, tannins, sterols, and triterpenoid in plant extracts.[10,11]

Thin Layer Chromatography[12, 13]

To identify the major components, present in the Extract a different number of solvent systems were tried and the solvents Toluene: Acetone: Formic acid was identified. Thin layer chromatographic studies of the hexane extract of *Mimusops elengi* L.

Evaluation of Ethyl acetate fraction by TLC

Stationary Phase	: Silica gel G
Mobile Phase	: Toluene: Acetone: Formic acid
Proportion	: 60:60:10
Detection	: Ferric chloride reagent
Solvent front	*: 10cm*
No of spots	: Three (8.8cm, 6.3cm,5.6cm)
R_f Values	**:** 0.88, 0.63,0.56

RESULTS AND DISCUSSION

The Phytochemical test on petroleum ether, chloroform, ethyl acetate and methanolic extracts of *Mimusops elengi* L showed the presence of various Phytoconstituents like alkaloid, carbohydrate, tannin, phenolic compounds, Saponin, terpenoid, flavonoid, Coumarin and phenol are present in Table 1.

Table 1: Preliminary phytochemical tests of various extracts of *Mimusopselen* L leaf.

Nature	Successive Extraction.				
	PET.ETHER	CHCl3	MeOH	AQ	EA
Alkaloids	-ve	--ve	--ve	--ve	--ve
Steroids	-ve	--ve	--ve	--ve	--ve
Carbohydrates	-ve	--ve	--ve	--ve	--ve
Phenolic compounds	-ve	--ve	+ve	+ve	--ve
Flavonoids	-ve	--ve	+ve	--ve	+ve
Glycoside	-ve	--ve	--ve	--ve	--ve
Triterpenoid	-ve	+ve	--ve	--ve	--ve
Tannins	-ve	--ve	+ve	+ve	+ve

Ash of any organic material is composed of their non-volatile inorganic components. Controlled incineration of crude drugs results in an ash residue consisting of an inorganic material (metallic salts and silica). This value varies within fairly wide limits and is therefore an important parameter for the purpose of evaluation of crude drugs. Therefore, percentage of the total ash, acid insoluble ash and water-soluble ash were carried out. The determinations such as loss on drying and ash values indicate the status of air-dried drugs used for studies. The total ash values when comes in acceptable range it simply shows that no inorganic adulteration is present. The extraction of any crude drug with a particular solvent yields a solution containing different phyto-constituents. Extractive value is also useful for evaluation of crude drug, which gives an idea about the nature of the chemical constituents present in a crude drug and is useful for the estimation of specific constituents, soluble in that particular solvent used for extraction. Total ash value, acid insoluble ash and water-soluble ash were determined and results were in acceptable limits. Ash value, Extractive value and Loss on drying is the loss of mass expressed as percent w/w results are tabulated in Table 2. The Percentage yield and physical characteristics of various extracts of *Mimusops elengi* L leaf. Successive Extraction is tabulated in Table 3.The ethyl acetate fraction was found to contain three spots in TLC. The physical parameters, chemical test and evaluation of chromatogram are shown in Table 4. The total flavanoid content in ethylacetate extract is 6.2352 mg/g Table 5. The estimation of Cyanidin in *Mimusops elengi* L. leaf absorbance at 530 nm is 0.635. Table 6. The isolated compound was further characterized by physicochemical tests, chromatography and spectral analysis such as, UV, FT-IR, ^{1}H-NMR, ^{13}C-NMR and Mass spectroscopy. The isolated COMP-A: revealed following analytical data. Table 7.

Table 2: Proximate values of *Mimusops elengi* L leaf.

Sl. No.	Parameter	Determined Value % w/w
A	Extractive values	
1	Alcohol soluble extractive value	26.00
2	Water soluble extractive value	30.00
3	Pet ether soluble extractive value	1.25
B	Moisture content	12.85
C	Ash Values	
1	Total ash	6.9
2	Water soluble ash	1.09
3	Acid insoluble ash	1.6
4	Sulphated ash	5.2

Table 3: *Percentage yield and physical characteristics of various extracts of Mimusops elengi L leaf. Successive Extraction*

Extract	% Dry wt in gms.	Colour	Odour	Consistency
Petroleum ether	3.87	Reddish brown	Characteristic	Sticky mass
Chloroform	3.85	Brownish black	Characteristic	Sticky mass
Methanol	26.9	Brown	Characteristic	Powder
Aqueous	4.35	Reddish brown	Characteristic	Powder

Table 4: TLC evaluation of isolated COMP-A

Isolated Compound	Evaluation of the Chromatogram	
	Under UV range	R_f value
COMP-A	Whitish spot	0.66

TLC of ethyl acetate soluble fraction of methanolic extract. *EA Fraction*

Table 4: Yield of isolated COMP-A

Isolated Compounds	Yield from column
COMP-A	110 mg

Table 4: Physical parameters of isolated COMP-A

Parameters	COMP-A
Physical State	Solid
Colour	Reddish Brown
Odour	Characteristic
Solubility	Methanol, DMSO

Table 4: Chemical examination of isolated COMP-A

Chemical Tests	Observation	Inference
FeCl3	Greenish ppt	Flavonoids may be present
Shinoda Test	Pink Color	Flavonoids may be present
Zinc-HCl test	Red Color	Flavonoids may be presents

Mobile phase : Toluene: Acetone : Formic acid

Proportion : 60:60:10

Detection : UV-254

TLC Photograph of Ethyl acetate extract and Isolated COMP-A.

Table 5: Total flavonoid content extract of the plant *Halodule uninervis* (Forsk.) Asch.

Extract	Total Flavonoid content(mg of quercetin/g)
Ethylacetate fraction	6.2352 mg/g
Petroleum ether extract	9.251 mg/g

Table 6: Estimation of Cyanidin in *Mimusops elengi* L. leaf.

Sl. No.	Sample	Absorbance at 530nm
1	Ethyl acetate fraction	0.635
2	Methanol extract	0.404

Table 7: Characterization of Isolated COMP-A

Spectra	Characters
UV	**One peak with λ_{max} at 282 nm**
FT-IR	Peaks at following wave number are observed Wave number 3369.77 – OH Stretching 2924.07 – C-H Stretching (Aromaticity) 2855.13 – C-H Stretching 2363.93 – OH Stretching
^{1}H-NMR	Peaks at following values are observed Value 6.7-7.2 – Ar-H(5H) 4.5-4.62 – OH (3H),Phenolic 3.75 – OCH_3 (6H)
^{13}C-NMR	Peaks at following values are observed Value 175.55 - C=O, Carbon 111.09 – 162.88 - C=C, Aromatic 47.40-79.10 - C-O, Carbon 103.81 - C=C, Un saturated carbon
MS	Base peak at 302 Molecular ion peak 381

UV spectra: one peak with λ_{max} at 282 nm.

IR spectra: wave numbers at, 3369.77 for OH Stretching, 2924.07 for C-H Stretching (Aromaticity), 2855.13 for C-H Stretching, 2363.93 for OH Stretching.

^{1}H-NMR spectra: Values at 6.7-7.2 for Ar-H (5H), 4.5-4.62 for OH (3H) of phenolic, 3.75 for OCH_3 (6H).

^{13}C-NMR: Values at 175.55 (C=O, carbon), 111.09 – 162.88 (C=C, Aromatic), 47.40 – 79.10 (C–O, Carbon), 103.81 (C=C, Unsaturated carbon).

MS spectra: Base peak at 302 and molecular ion peak at 381.

CONCLUSION

Thus, the physicochemical, fluorescence study, preliminary phytochemical screening, TLC analysis, flavonoid content, cyaniding content and chromatographic details can be used as a diagnostic tool for the correct identification of the plant. The ethyl acetae extract fraction of the plant of *Mimusops elengi* L. leaf has a significant flavonoid content it is useful for further antioxidant and anticancer studies. The adulterants if any in this plant material can be easily identified by using these results.

REFERENCES

1. J Bruneton, Pharmacognosy, Phytochemistry, Medicinal Plants, 2nd edition, Lavoisier, Publication, 1999, France.
2. V.Sharma, R Paliwal, Preliminary phytochemical investigation and thin layer chromatography profiling of sequential extracts of *Moringa oleifera* pods; Int J Green Pharm, 2013; 7: 41-5.
3. R Elango, U Jadhav. Phytochemical screening of *Moringa oleifera* using high performance thin layer chromatography; Plant Arch, 2010; 749-51.
4. Kirtikar KR, Basu BD. Indian Medicinal Plants. 2nd ed Vol- II, Popular Publications Dehradun, India. 1999: 1224-1227.
5. Chopra RN, Nayar SL, Chopra IC. Glossary of Indian Medicinal Plants. National Institute of Science Communication and Information Resources (CSIR), New Delhi, 2000: 167.
6. Rahman MA, Hussain A.Anticancer activity and apoptosis inducing effect of methanolic extract of *Cordia dichotoma* against human cancer cell line. Bangladesh J Pharmacol.2015; 10: 27-34.
7. PK Mukharjee, Quality Control of Herbal Drugs: An Approach to Evaluation of Botanicals.2005, India: (Business Horizones).
8. Kokoski J, Kokoski R, and Salma FJ, Fluorescence of powdered vegetable drugs under ultraviolet radiation. J. Am. Pharm. Ass, 1958; 47 (10): 715-717.
9. CR Chase, RJ Pratt, Fluorescence of powdered vegetable drugs with particular reference to development of a system of identification. J. Am. Pharm. Ass, 1949; 38: 324-333.
10. AAL Ordonez, MA Vattuone, MI, Isla, Antioxidant activities of *Sechium edule* (Jacq.) Swart extracts. Food Chem.2006; 97: 452-458.
11. KR Khandelwal, Techniques and Experiments, Practical Pharmacognosy; 17th edition, Nirali Prakashan, Pune, 2007; 149- 156.
12. Stahl Ergon. Thin Layer Chromatography. A laboratory Handbook, 2nd ed. New York: Springer-Verlag Berlin Heidelberg; 1990.
13. Wagner H. Bladet S, Zgainski EM. Plant Drug Analysis**,** A TLC Atlas;1st ed. New York: Springer Verlag Berlin Heidelberg; 1994.

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