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Phytochemical screening and GC-MS analysis of *Mukia maderaspatana (L.)* leaves.

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ABSTRACT

The investigation was carried out to determine the qualitative analysis of phytochemical screening and possible chemical components of *Mukia maderaspatana* (L.) (family: Cucurbitaceae), leaves GC-MS. The plant is an indigenous plant; traditionally it is used as an ingredient of various cocktail preparations and for the management of severe inflammatory disorders in Indian system of medicine. GC-MS analysis of hydroalcoholic extract lead to identification of 7 compounds. This analysis revealed that contains *Mukia maderaspatana* (L.) leaves mainly Dichloroacetic acid, 4-methylpentyl ester, 2-Butyn-1-ol, 4-methoxy and also showed the presence of other constituents like flavonoids, saponins, carbohydrates, steroids, tannins and phenolic compounds.

INTRODUCTION

Plant is man's friend in survival, giving him food and fuel and medicine from the days beyond drawn of civilization (Bose, T.K and Choudhary, K., 1991). Plant continues to be a major source of medicine, as they have throughout human history (Prince, L and Prabakaran, P., 2011).*Mukia maderaspatana* (family: Cucurbitaceae), is a medicinal plant. The plant was reported to have activities such as hepatoprotective (Thabrew, et al., 1995) antirheumatic (Ramakrishnamacharya, C.H et al., 1996), diuretic, stomachic (a digestive tonic), gentle aperients, antipyretic and antiflatulent, antiasthmatic, anti-inflammatory, antidiabetic and antibronchitis and is used for tooth-ache besides its use in vertigo and biliousness (Chopra, R.N., 2002; Kirthikar, K.R and Basu, B.D., 1980).

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Most valuable Phytochemicals are the product secondary metabolism and possess chemical or structural complexity (Kunkel, G., 1984).The use of this herb has been reported in Indian Traditional Systems of Medicine and its modern applications are receiving wide spread attention day by day. Hence, the present investigation was carried out to determine the Possible chemical components from *Mukia maderaspatana (L.)* leave by GC-MS.

MATERIALS AND METHODS

Collection and identification of plant material

Leaves of the plant were collected from local region and District of Tiruvannamalai, TamilNadu, India in the month of March 2011. The botanical identity was confirmed by a taxonomist; the plant material was taxonomically identified and authenticated by V.Chelladurai (Research Officer) Botany (C.C.R.A.S) Government of India. Voucher specimen (AECBT-09/2011) has been retained in the Dept of Biotechnology, AEC, Tiruvannamalai.

Plant sample extraction

Leaves were cleaned, shade dried and pulverized to powder in mechanical grinder. Required Quantity of powder was weighed and transferred to Stoppard flask, and treated with hydroalcohol (70% v/v) until the powder is fully immersed. The flask was shaken every hour for the first 6 hrs and then it was kept aside and again shaken after 24 hrs. This process was repeated for 3 days and then the extract was filtered. The extract was collected and evaporated to dryness by using vacuum distillation unit. The final residue thus obtained was then subjected to GC-MS analysis.

Phytochemical screening

Table. 1: Chemical constituents of ethanol extract from *Mukia maderaspatana*

S.No	Tests	Result
1	Alkaloids	
	(i) Draggendoff's test	+
	(ii) Mayer's test	+
	(iii) Wagner's test	+
2	Flavonoids	
	(i) Lead acetate test	+
	(ii) Ferric chloride test	+
	(iii) Sodium chloride test	+
3	Carbohydrates	
	(i)Fehilings test	+
	(ii)Benedicts test	+
4	Saponins	
	(i) Emulsion test	+
	(ii) Frothing test	-
5	Tannins	
	(i) Bromine water test	-
	(ii) Ferric chloride test	+

Chemical test were carried out on the ethanolic extract and on the powdered specimens using Standard procedures to identify the constituents as described by Sofowara (Sofowara, A., 1993), Trease and Evans (Trease, G.E and Evans, W.C., 1989) and Harbone (Harborne, J.B., 1973).

GC-MS analysis

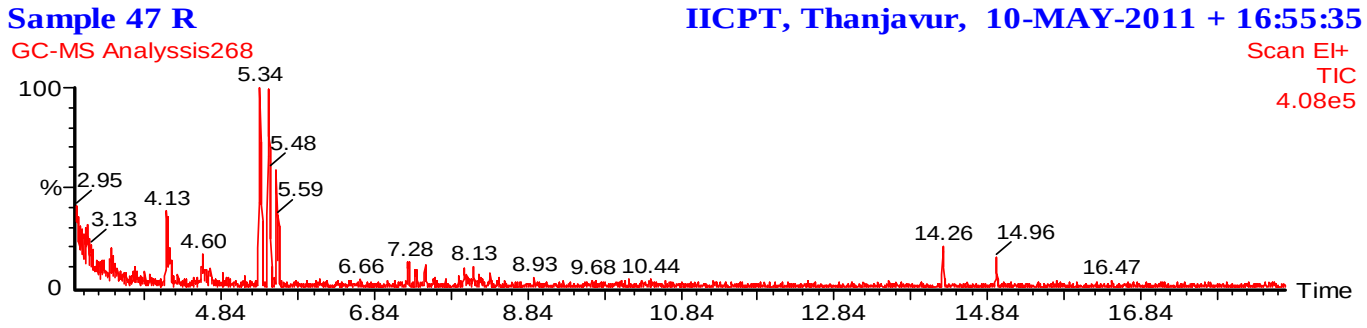
GC-MS analysis of these extracts was carried out by following the method of Hema *et al*. (Hema et al., 2010). GC-MS analysis were performed using a Perkin-Elmer GC clauses 500 system and Gas Chromatograph interfaced to a mass spectrometer (GC-MS) equipped with a Elite-1, fused silica capillary column (30 m × 0.25 mm ID × 1× df, composed of 100% Dimethyl poly siloxane). For GC/MS detection, an electron ionization system with ionizing energy of 70 eV was used. Helium gas (99.999%) was used as the carrier gas at constant flow rate 1 ml/min and an injection volume of 2 µl was employed (Split ratio of 10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min) with an increase of 10°C/min to 2000°C, then 5°C/min to 280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70eV; a scan interval of 0.5 seconds and fragments from 45 to 450 Da. Total GC running time was 36 minutes. The relative % amount of each component was calculated by comparing its average peak area to the total areas, software adopted to handle mass spectra and chromatograms was a Turbo mass.

RESULTS AND DISCUSSION

The qualitative analysis of the ethanolic extracts from the leaf sample of *Mukia maderaspatana showed* the presence of phytochemical constituents such as tannins, saponin, flavonoids, steroid, terpenoids and cardiac glycerides. At the same time, the phytochemical constituents like phlobatannin were absent (Table 1). The compounds present in the hydroalcoholic extracts of *Mukia maderaspatana* were identified by GC-MS analysis (Figure 1). The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are presented in Table 2. Seven compounds were identified in ethanolic extract by GC-MS. The major components present in leaves of *Mukia maderaspatana* were 2-Methylthiolane,S,S-dioxide, Diazeene, bis(1,1-dimethylethyl),3-Buten-2-ol,2-Butyn-1-ol, 4-methoxy, Dichloroacetic acid, 4-methylpentyl ester,2-(Chloromethyl)-2-3-dihydro-4(1H)- quinolinone, Pantolactone compounds. Phytochemical constituents which contribute to the medicinal activity of the ethanolic extract of *Mukia maderaspatana*. The leaves contains many constituents are considered mainly to be responsible for various antimicrobial properties. Eugenol is the main constituent and it is responsible for its repellent property. The presence of eugenol attributes to its antioxidative property and is also thought to be responsible for inhibition of lipid peroxidation (Gupta et al., 2002). This property helps in maintaining good health and in preventing the changes occurrence of heart diseases as well as most of the other biochemical diseases because oxidative stress is the hallmark of such diseases (Hannan, J.M et al., 2006). Eugenol is the major components found in the whole plant of *Mukia maderaspatana* which is being used for the pharmacological work. Saponins are generally regarded a antinutrients but are also believed to be useful in human diet for controlling cholesterols. It presence in this plant therefore could suggests that the plant is of medicinal value. There is evidence of the presence of saponins in traditional medicine preparations (Asl, M.N and Hosseinzadeh, H., 2008; Xu, R et al., 1996).Where oral administrations might be expected to lead to hydrolysis of glycoside from terpenoid and obviation of any toxicity associated with the intact molecules. Tannins are astringent, bitter plant polyphenols that either bind and precipitate or shrink proteins, tannins are distributed all over the plant kingdom. Tannins have traditionally been considered antinutritional but it may be employed medicinally in antidiarrheal, hemostatic and antihemorrhoidal compounds. It presence in the plant suggest it to be of medicinal value because tannins have shown potential antiviral (Lin, LU et al., 2004), antibacterial (Akiyarma, H et al., 2001; Funatogawa, K et al., 2004) and antiparasitic effects (Kolodziej, H and Kiderlen, A.F., 2005).

Table. 2: Components identified in *the Mukia maderaspatana* plant powder sample.

No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %
1.	2.91	2-Methylthiolane, S,S-dioxide	$C_5H_{10}O_2S$	134	21.28
2.	4.13	Diazene, bis(1,1-dimethylethyl)-	$C_8H_{18}N_2$	142	10.64
3	4.60	3-Buten-2-ol	C_4H_8O	72	2.13
4.	5.34	2-Butyn-1-ol, 4-methoxy-	$C_5H_8O_2$	100	29.79
5.	5.45	Dichloroacetic acid, 4-methylpentyl ester	$C_8H_{14}Cl_2O_2$	212	29.79
6.	14.26	2-(Chloromethyl)-2,3-dihydro-4(1H)-quinolinone	$C_{10}H_{10}ClNO$	195	4.26
7.	14.96	Pantolactone	$C_6H_{10}O_3$	130	2.13



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