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GC-MS ANALYSIS OF ETHANOLIC AERIAL PART EXTRACT OF IMPORTANT MEDICINAL PLANT CYCLEA PELTATA (LAM.) HOOK. F. THOMS. (MENISPERMACEAE)

Biological Science

R. Shyam Praveen Department of Botany, Kongunadu Arts And Science College, Coimbatore-641029, Tamil Nadu, India.

Sathish Kumar Department of Botany, Kongunadu Arts And Science College, Coimbatore-641029, Tamil Nadu, India.

S. Jamuna Department of Botany, Kongunadu Arts And Science College, Coimbatore-641029, Tamil Nadu, India.

A. Venkatachalapathi* Department of Botany, Kongunadu Arts And Science College, Coimbatore-641029, Tamil Nadu, India. *Corresponding Author

ABSTRACT

The present study was aimed at to identify the GC-MS analysis of ethanolic aerial part extract of *Cyclea pelata*, revealed the presence of 24 compounds. The major chemical constitutes are Ethyl 9,12,15 - Octadecatrienoate (18.6%), 13-Docosenamide, (z)-(11.86%), Diethyl 3,4-bis(methylene)cyclopentane-1,1-dicarboxalate (7.88%), Hexadeccanoic acid, ethyl ester (CAS) (6.98%), 1-Heptatriacotanol (6.52%), 5-(hydroxymethyl)-2-(1-methy1-2- imidazoly1)-1H-benzimidizole (5.90%), 3,3,4,4-Tetracyano-6-pheny1-5-(p-toly1)-2-(p-methoxyphenyl imino)-2,3,4,5-tetrahydropyridine (4.15%). However, these findings, still studies are lacuna in the fields of phytochemisty and pharmacology which provides a better pharmacopoeial standard of a novel drug leads.

KEYWORDS

Cyclea pelata, GC-MS analysis, ethanolic extract, 24 compounds.

INTRODUCTION

India is one of the richest heritage centers of the world due to a wide range of climate in terms of latitude, longitude, altitude and geology (Rojers, 1991). There are over 17,500 species of higher plants, 64 gymnosperms, 1,200 pteridophytes, 2,850 bryophytes, 2,021 lichens, 15,500 fungi and 6,500 algae were reported. India has rich in its own flora that is, endemic plant species (5,725 angiosperms, 10 gymnosperms, 193 pteridophytes, 678 bryophytes, 260 liverworts, 466 lichens, 3,500 fungi and 1,924 algae) (Sanjappa, 2005). The Nilgiris is the hotspot of Western Ghats and have numerous endemic plants and generally accepted that nearby 2000 species of higher plants. 84 of fishes. 92 species of amphibians, 89 species of reptiles, 15 species of birds and 12 species of animals (Ahemedullah and Nayer, 1987). 75 percent of endemics of peninsular India are confined to the Western Ghats. It has been surveyed that the entire Western Ghats contained 44 rare and endangered spices which were published in British fern gazette in 1995 (Manickam, 2004). According to the World Health Organization (WHO) more than 80% of the world population roles on additional medicine for their primary health care and needs (Vijayan et al., 2007). The disease preventive properties and medical plants are by holding different phytochemicals, which are non-nutritive chemicals. Due to this huge medicinal values and rapid exploitation of their community are attached bio diversity loss, pollution, environmental degradation etc. Conversation of such important plants is necessary to meet the demand and protect the wild by adopting in vitro techniques. Mass propagation of plant species through in vitro culture is one of the best and most successful examples of commercial application plant tissue culture technology in tissue culture methods was provides new means of conserving and rapidly propagating the valuable rare and endangered medicinal plants. Plants are the resource of primary and secondary metabolites namely alkaloids, terpinoids, flavanoids, saponins, coumarins, glycosides, phenolics, carboxylic acids, amino acids, sugars, proteins etc. these phytochemicals have significant biological functions and also which contribute specific characteristic and property of the plant. The therapeutic use of the plant is due to presence of these compounds, Hence the study and characterization of such phytocompounds have great significant in pharmacological, antimicrobial and clinical research.

Therefore, in recent years the demand for herbal medicines and several natural products from a variety of plant species is consistently increasing. One of the most medicinal plants is *Cyclea peltata* (Lam.) Hook and Thomas, is a slender twining shrub belonging to the family, Menespermaceae and commonly called as Patakizhangu (or) Malathangi, by tribal Toda's. The ethnic people of Toda's using this plant to treat the disease like ulcer, stomach ache, jaundice, antiprotozoal and antimalarial. The National Medicinal Plants Board of India identified this plant as medicinal plant in high trade sourced from the tropical forest in spite of common claims their remain a poorly investigated plant. Therefore studies were undertaken to identify the medicinally important active principles through GC-MS analysis and as well as the conservations of strategies are need up to day. The major objective of present study is profiling of medicinally active principles via GC-MS analysis, in ethanolic leaf extract of *Cyclea peltata*. Conservation of medicinal plant species, *Cyclea peltata* by in vitro propagation techniques of three explants such as leaf, node and intermodal explants.

MATERIALS AND METHODS

Species Description

The plant *Cyclea peltata* is climbing shrub to twine and distributed in various parts of India including Sri Lanka and Andaman Nicobar islands. Within India, It is mainly recorded in Western Ghats of Maharashtra, Karnataka, Kerala, and Tamil Nadu and also in other moist deciduous forests of peninsular India.

Extract preparation

Fresh and healthy aerial parts of the study species, *C. pellata* were washed with running tap water to remove the dust particles. Then, they were shade dried, coarsely powdered and extracted with ethanol (50g/250mL) by cold extraction. Further, the extract was filtered and concentrated to dryness and stored at 4°C for further use.

GC-MS analysis

GC-MS analysis were carried out on a GC-MS 5975C (AG1LENT) instrument employing the following conditions, column DB-5ms Agilent (30mX 0.25mm, 0.25gm film thickness), operating in a electron impact mode at 70eV. Helium (99.9995%) was used as a carrier gas at a constant flow of 1.51 mL/min in the split mode (split ratio-10:1), injector temperature 240°C. the oven temperature was programmed from 70°C (hold time for 2 min) with an increase of 10°C/min to 300°C/ impending with a 9 min isothermal. Mass spectrum was taken at 70eV, scan range of 40-1000En/z (mass/charge) and a scan interval of 5nnin.

GC analysis separates all of the compounds in a sample and provides a representative spectral output the sample was injected into the injection ort of the GC device. The GC instrument vaporizes the sample and then separates the compounds the separated compound. Were eluded from the column and enter a detector which was capable of creating an electronic signal. Each components ideally produces a specific spectral peak that may be recorded on a paper chart are electronically. The time collapsed between injection and elution is called the 'retention time'. The size of the peaks is proportional to the quantity of the corresponding substances in the specimen analyzed. The peak is measured from the base line to the tip of the peak. The X

axis showed the RT (retention time) and the Y axis measured the intensity of the signal to quantify the components in the sample injected. As individual compounds eluted from the Gas chromatography column, the enter the electro ionization (Mass spectroscopy) detector, where they were bombarded with a stream of electrons causing them to break apart fragments the fragments obtain were actually charged ions with certain mass. The m/z (mass/charge) ratio obtained was calibrated from the graph obtained, which was called as the mass spectrum graph which is the fingerprint of molecule.

DATA ANALYSIS

Identification of bio active compounds from ethanolic extract of aerial parts of *C. peltata* was based on the molecular structure, molecular mass and calculator fragmentations. The mass spectrum of the unknown compound was compared with the spectrum of the known components stored in the national institute standard and technology (NIST-11.LIB (Stein, 1990)] library, having more than 62000 patterns. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. The name, retention time (min), molecular formulae, molecular weight (data), peak area (%), nature of the compound, structure and activity were as certain. The biological activities have constituents are based on the data obtained from Pubchem an online database and published literature.

STATISTICAL ANALYSIS

The results are presented as mean ± SD of triplicates. The various parameters analyzed were subjected to statistical analysis using SigmaStat (Version 3.1). Statistical significance was determined by one-way ANOVA, with $p<0.05$ being considered significant.

RESULTS

GC-MS chromatogram of the ethanolic aerial part extract of the *Cyclea pellata* was given in Table 1. The result showed 24 peaks, indicating the presence of phytochemical compounds, these compounds were identified through Mass Spectrometry. The identification phytochemical compounds is based on the active principles with their Retention Time (RT), molecular formulae, molecular weight, peak area (%), nature of the compound, and activities.

The compound prediction is based on Dr. Duke's phytochemical and ethnobotanical databases. by Dr. Jim Duke of the agriculture research services/USDA, In the present investigation, the ethenolic extract showed the presence of 24 compounds namely, Diethyl 3,4 bis(methylene)cyclopentane-1,1-dicarboxalate (3.04%), 9-Octadecene,(E) (11.12%), 5- lsopropy1-4-(triflurомethyl)-1H-pyrimidin-2-one (13.64%), Tricholoro acetic acid, hexadecyl ester(15.29%), 2,2-Dideutero octadecanal (19.53%), 5-(hydroxymethyl)-2-(1- methyl-2-imidazoly1)-1H-benzimidizole (20.22%), 3,3,4,4-tetrocyano-6-phenyl-5-(p-to1y1)-2-(p-methoxyphenylimino)-2,3,4,5-tetrahydropyridine (23.00%), Hexadeccanoic acid, ethyl ester (CAS) (23.52%), Lucenin 2 (26.00%), Ethyl 9,12,15-Octadecatrienoate (26.87%), Octadecanoic acid, ethyl ester (CAS) (27.30%), Dotriaconate (CAS) (27.83%), hahnfett (28.68%), 17-Pentatriacontene (29.74%), 14-a-H-PREGNA (30.11%), isochiapin b (30.39%), 13-Docosenamide, (z)¬(31.35%), Tetrapentaconate, 1,54-dibromo (31.68%), 3,3,4,4-Tetracyano-6-pheny1-5-(p¬toly1)-2-(p-methoxyphenylimino)-2,3,4,5-tetrahydropyridine (32.76%), Isochiapin b %2< (34.39%), Octadecane,3-ethyl-5-(2-ethylbuty1)- (36.93%), 1-Heptatriacotanol (37.22%) and Docosanoic acid, 1,2,3-propanetriy1 ester (37.79%). The GC-MS spectrum confirmed the presence of various compounds with the different retention times, the individual fragmentation pattern of the components was illustrated in Fig. 1.

Table 1. GC-MS analysis of methanolic leaf extract of *Cyclea peltata*

S. No.	Compound Name	Retention time (Min)	Molecular Formulae	Molecular Weight	Area %	Nature of the compound	Activity
1	Diethyl 3,4 bis(methylene) cyclopentane-1 , 1-dicarboxalate	3.04	$C_{13}H_{18}O_4$	238	7.88	Carboxylic acid	Kinase Inhibitors
2	9-Octadecene, (E)	11.12	$C_{18}H_{36}$	252	1.12	Phenol	Anticancer
3	5-Isopropyl-4-(triflurомethyl) -1H-pyrimidin-2-one	13.64	$C_8H_9F_3N_{20}$	206	2.13	Phenol	Antiasthmatics
4	Tricholoro acetic acid,Hexadecyl ester	15.29	$C_{18}H_{33}C1_3O_2$	386	0.97	Carboxylic Acid	Bactericide
5	2,2-Dideutero Octadecanal	19.53	$C_{18}H_{34}D_{20}$	268	0.92	Aldehyde	Expectorant
6	5 (hydroxymethyl) 2-(1-methyl-2-imidazoly) -1H-benzimidizole	20.22	$C_{12}H_{12}N_{40}$	228	5.90	Hydroxyl	Methyl donar
7	3,3,4,4-tetrocyano-6 Phenyl-5-(p-toly) -2-(p methoxyphenylimino) -2, 3,4,5-tetrahydropyridine	23.00	$C_{29}H_{20}N_{60}$	468	0.86	Phenol	Bactericide
8	Hexadeccanoic acid, Ethyl ester (CAS)	23.52	$C_{18}H_{36}O_2$	284	6.98	Palmitic Ester	Antioxiant Hypocholesterolekic, Nematiccide pesticide anti Androenic flavor
9	2-hexadecen -1-ol, 3,7,11,15-tetramethyl, [R-[R*-€]]-CAS	25.79	$C_{20}H_{40}O$	296	2.73	Flavaniod	Antitubercular Activity against Mycobacterium Tuberclosis
10	Lucenin 2	26.00	$C_{27}H_{30}O_{16}$	610	0.92	Glycoside	Anxiolytic activities
11	Ethyl 9,12,15- Octadecatrienoate	26.87	$C_{20}H_{34}O_2$	306	18.60	Carboxylic acid	Anti-fungal activity
12	Octadecanoic acid, ethyl ester (CAS)	27.30	$C_{20}H_{40}O_2$	312	0.83	Carboxylic Acid	Flavoring agent
13	Dotriaconate (CAS)	27.83	$C_{32}H_{66}$	450	1.30	Alkane	Anti-inflammatory
14	HAHNFETT	28.68	N/A	0	1.39	-	-
15	17 pentatriacontene	29.74	$C_{35}H_{70}$	7.29	2.66	Carbonyl Group	Anti-microbial
16	14-a-H-PREGNA	30.11	$C_{21}H_{36}$	288	0.85	Steroid	Anti-HIV, Cushing's syndrome
17	ISOCHIAPIN B	30.39	$C_{19}H_{22}O_6$	346	3.01	-	Anti-cancer
18	13-Docosenamide, (Z) -	31.35	$C_{22}H_{43}NO$	337	11.86	Amide	Anti-adhesive agent
19	Tetrapentaconate, 1,54-dibromo	31.68	$C_{54}H_{108}Br_2$	914	2.27	-	-
20	3,3,4,4-Tetracyano-6-phenyl-5-(P-tolyl) -2-(p-methoxphenylimino) 2,3,4,5- tetrahydropyridine	32.76	$C_{29}H_{20}N_{60}$	468	4.15	Phenol	Anti-bacterial agent
21	Isochiapin b%2<	34.39	$C_{19}H_{26}O_6$	350	1.66	-	Anti-cancer
22	Octadecane, 3-ethyl-5-(2-ethylbutyl) -	36..93	$C_{26}H_{54}$	366	3.75	Alkane	-
23	1-Heptatriacotanol	37.22	$C_{37}H_{760}$	536	6.52	Carboxylic Acid	Dermatological Disorder
24	Docosanoic acid, 1,2,3-propanetriyl ester	37.79	$C_{69}H_{134}O_6$	1058	1.47	Carboxylic Acid	-

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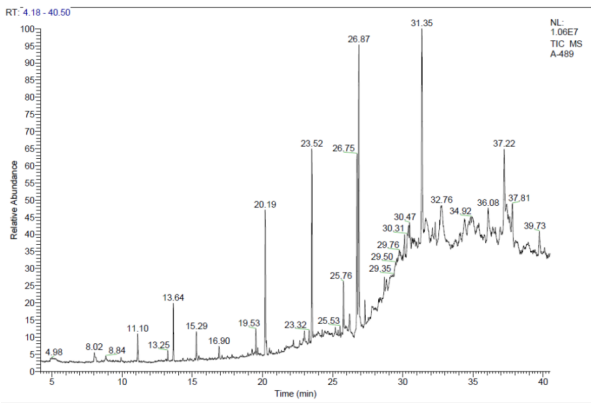

Fig. 1. GC-MS Chromatogram of methanolic leaf extract of *Cyclea peltata.*

DISCUSSION

In the present study, the GC-MS analysis of aerial part of the ethanolic extract of *C. peltata*, showed the presence of 24 compounds (Fig.1). The Mass Spectra identified for seven major compound are presented in ethanolic extract. They are Ethyl 9,12,15- Octadecatrienoate (18.6%) 13-docosenamide, (z) (11.86%), Diethyl 3,4-bis(methylene)cyclopentane-1,1-dicarboxalate (7.88%) (Fig. a), Hexadeccanoic acid, ethyl ester (CAS) (6.98%), 1-Heptatriacotanol (6.52%), 5- (hydroxymethyl)-2-(1-methy1-2-imidazoly1)-1H-benzimidizole (5.90%), 3,3,4,4- Tetracyano-6-pheny1-5-(p-tolyl)-2-(p-methoxyphenyl im ino)-2,3,4,5-tetrahydropyridine (4.15%). The compound Ethyl 9,12,15-Octadecatrienoate is reported to have Anti-fungal activity, 13-Docosenamide found to have Anti-adhesive property, Diethyl 3,4-bis(methylene)cyclopentane-1,1-dicarboxalate reported to have Kinase inhibitor, Hexadeccanoic acid, ethyl ester (CAS) have an Anti-oxidant, hypercholoestrol activity, Nematicide property (Ravi et al., 2012). 1-Heptatriacotanol have a property to treat dermatological disorder, Hexadeccanoic acid, ethyl ester (CAS) reported to regulate Methyl Donor activity, 3,3,4,4-Tetracyano-6-pheny1-5-(p-toly1)-2-(pmethoxyphenylimino)-2,3,4,5-tetrahydropyridine used as an anti-bacterial agent.

Conflict Of Interest

Conflict of interest declared none.

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